



## Original Research Article

Topic: *Shewanella* species: Case series on a rare emerging pathogenPriyanka Prasad<sup>1</sup>, Sulmaz Reshi<sup>1</sup>, Gita Nataraj<sup>1,\*</sup><sup>1</sup>Dept. of Microbiology, Seth GS Medical College & KEM Hospital, Mumbai, Maharashtra, India

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## ABSTRACT

**Introduction:** *Shewanella* is a gram negative marine bacterium of which *S. putrefaciens* and *S. algae* are pathogenic in immunocompromised patients and patients with pre-existing chronic ulcers. They can cause a broad range of human infections.

**Aims and Objectives:** To describe four cases of *Shewanella* infection in susceptible patients.

**Materials and Methods:** *Shewanella algae* was isolated from blood (2) and pus (2) of four patients admitted to the ward.

The first case was an 18 year old female, known case of osteomyelitis following hip fracture surgery 12 years back. She presented with discharging sinus from the surgical site which grew *Shewanella algae* repeatedly.

The second case was a 38 years old male patient with history of trauma to left knee at a beach. He was operated and intra-op tissue grew *Shewanella algae* resistant to 3<sup>rd</sup> generation cephalosporins and susceptible to quinolones, aminoglycoside, carbapenems and piperacillin-tazobactam.

Both patients were successfully treated based on the AMST profile.

The other two cases were dialysis patients from whom *Shewanella putrefaciens* was isolated from blood. The isolate had similar antibiogram being resistant to quinolones, 3<sup>rd</sup> generation cephalosporins, carbapenem and aztreonam and susceptible to piperacillin-tazobactam, cefepime, aminoglycosides & polymyxin. One of the patients expired while the other patient was successfully treated based on AMST profile.

**Conclusion:** Isolation of *Shewanella* species from invasive specimen highlights importance of this organism as an emerging pathogen. Though it is known to be sensitive, in the mentioned study, *Shewanella algae* was resistant to 3<sup>rd</sup> generation cephalosporins or fluoroquinolones and *Shewanella putrefaciens* was multi-drug resistant including to carbapenems.

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## 1. Introduction

*Shewanella* is a saprophytic gram negative bacillus which is naturally associated with marine and aquatic habitat. Earlier considered as a colonizer, readily living on previously injured tissue, it has emerged as an opportunistic pathogen gaining importance as an increasing cause of human morbidity and mortality.<sup>1,2</sup> It belongs to the family Shewanellaceae, which comprises of over 62 species. Only two species i.e. *Shewanella algae* and *Shewanella putrefaciens* have been reported from human infections.

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Though it is a rare opportunistic human pathogen, it has been known to cause a wide variety of infections in humans such as skin and soft tissue infections, bacteremias, endocarditis, biliary tract infections, arthritis, peritonitis, intracranial abscesses and ventilator associated pneumonias.<sup>1,3,4</sup> Rare cases of this organism have also been reported from immunocompetent patients.<sup>5</sup> The risk factors associated with the infection include chronic ulcers, burn wounds, peripheral vascular occlusive disease, hepatobiliary disease and trauma. Other risk factors include living in warm areas, living near sea coasts, contact with sea water, marine life, swimming pools, substandard living,

poor nutrition, underlying conditions like diabetic mellitus & consumption of seafood.<sup>1–3,6,7</sup> Contact with sea water is the most common source of this organism and the most common point of entry is injured skin. Other sources from where this organism has been reported include corroded pipes and soil.<sup>2</sup> The clinical presentation in case of skin and soft tissue infections includes cellulitis, wound infection and formation of abscesses. There are very few reports describing cases of *Shewanella* infections. This study tries to highlight the presentations of a series of four cases from a tertiary care hospital in a coastal metropolitan city.

### 1.1. Microbiology of case reports

Institutional ethics committee approval was taken before the paper was initiated.

Clinical samples [two pus and two blood specimens] from four patients were received in the microbiology laboratory for culture which were processed as per routine protocol. Pus and blood culture were processed as per standard protocol for aerobic bacteria. Any growth from the specimens was identified to species level.<sup>8,9</sup>

*Shewanella* colonies are easily identified because they are gram negative, motile bacilli producing tan coloured colonies on blood agar with beta hemolysis that are catalase and oxidase positive, urease negative, citrate negative, indole negative, ornithine is decarboxylated while lysine is not decarboxylated and arginine is not hydrolysed. They are the only non-fermenters producing H<sub>2</sub>S on TSI or KIA agar.<sup>10</sup> The two species, *Salga* and *S. putrefaciens* were differentiated based on the following characteristics.<sup>2</sup> (Table 1)

Conventional AMST was carried out using Kirby Bauer disc diffusion method as per CLSI.<sup>11</sup> Since breakpoints for *Shewanella* species have not been specifically mentioned in CLSI, the table for *Pseudomonas* species was used for interpretation of results. The antibiotics tested by disc diffusion method were piperacillin (100mcg), piperacillin-tazobactam (100/10mcg), ceftazidime (30mcg), cefepime (30mcg), aztreonam (30mcg), imipenem (10mcg), meropenem (10mcg), amikacin (30mcg), gentamicin (10mcg), ciprofloxacin (5mcg) and levofloxacin (5mcg).

Identification and AMST was also performed by VITEK 2 Compact (Biomérieux India Ltd.) using the GNB ID & N281 cards.

### 1.2. Case 1

An 18 year old female presented with discharging sinus from the surgical site since 15-20 days. The patient was operated for ICN fracture femur in 2005 and a nail was inserted to hold the bones in place. After a month, she developed fever, swelling and pain with restriction of left hip joint movement. Investigations revealed a total leucocyte count of 29,000/ml. Ultrasonography of left

hip revealed a septic arthritis with osteomyelitis of left proximal femur and the neck of femur. In December 2005, debridement was done and swab sent for culture yielded growth of *P.aeruginosa* and *K. pneumoniae*. Implant was removed after a year.

Between March 2006 to May 2011, repeat infections with sinus formation occurred for which repeated debridement was done. Different bacteria were isolated each time (*P. aeruginosa*, *Streptococcus spp.*, *Staphylococcus aureus*).

In December 2017, patient developed a discharging sinus. At the time of debridement, three intra-operative pus specimens from left hip joint sent for culture yielded colonies of *Shewanella algae* on both the conventional as well as VITEK 2 Compact system. (99% probability, excellent identification). The pathogenic role of the organism in this case was not confirmed, so patient was not started on any antibiotic therapy.

Two repeat specimens after one month again grew *Shewanella algae* along with *Proteus mirabilis* and MRSA. The present isolate was susceptible to all antibiotics except fluoroquinolones (VITEK 2). Patient was started on a combination therapy comprising of amikacin and ceftazidime along with linezolid for MRSA. Patient responded to this treatment and a repeat culture after 1 month showed no growth.

### 1.3. Case 2

Case 2 was a 38 yr old male patient who presented with history of trauma to left knee at a beach. He did not give any history of head or chest injury. Next day, he showed signs and symptoms of altered sensorium with two episodes of seizures for which he was admitted. CSF routine-microscopic examination was normal and the bacteriological culture revealed no growth in blood and CSF cultures. Serum electrolytes were within normal range. Patient did not give any history of diabetes mellitus or hypertension. Before surgery, patient did not have any other episodes of seizures. One week later, he was operated for internal fixation of left proximal tibial fracture and two intra- op tissue specimens were sent for culture. The WBC count was 7000/ml on the day of surgery and he was started on cefotaxime, amikacin and metronidazole. Smear of the intra-op tissue showed occasional pus cells with gram negative bacilli. Culture grew colonies of *Shewanella algae* which was resistant to only 3rd generation cephalosporins and sensitive to imipenem, meropenem, piperacillin, ciprofloxacin, levofloxacin, amikacin, gentamicin and piperacillin-tazobactam.

The patient was successfully treated on piperacillin-tazobactam and amikacin based on the antibiotic susceptibility testing.

#### 1.4. Case 3

Case 3 was a 65 yr old male, known case of chronic kidney disease with type 2 diabetes mellitus and hypertension on hypertensive drugs since 2 years, presented with signs and symptoms of sepsis and admitted. The investigation results were as follows: Procalcitonin -7.3, D-dimer >20, Fibrinogen- 610, creatinine – 4.3mg/dl and TLC- 13500 indicating bacterial sepsis. Blood was sent for culture and the patient was empirically started on ceftazidime. *Shewanella putrefaciens* was isolated from arterial port, venous port and venous peripheral blood which was resistant to aztreonam, fluoroquinolones, 3<sup>rd</sup> generation cephalosporins and carbapenams while being sensitive to piperacillin-tazobactam & aminoglycosides. The antibiotics were changed to piperacillin-tazobactam and vancomycin; however patient expired the next day.

#### 1.5. Case 4

Case 4 was a 41 old male, case of malignant hypertension with chronic kidney disease and on a hypertensive since 5 months presented with fever. Blood for culture was sent and *Shewanella putrefaciens* was isolated from arterial port, venous port and venous peripheral blood. Patient was successfully treated based on the antibiotic susceptibility testing which was similar to case 3.

## 2. Discussion

*Shewanella* species have been reported as pathogens in skin and soft tissue infections, intra-abdominal infections, lower respiratory tract infections, otitis media extending to brain and presenting as cerebellar abscess and meningitis, necrotizing fasciitis, arthritis, spondylodiscitis and septicemia.<sup>2,12</sup> In India, case reports of *Shewanella* infection from soft tissues and wound infections are rare. This maybe because of incomplete identification or rarity of this pathogen in environmental source.

Cases of osteomyelitis due to *Shewanella* species have been reported.<sup>13</sup> In the case report by Bothelho-Nevers et al, the isolate was first identified as *S. putrefaciens* by phenotypic characterization but genotyping identified the organism as *S. algae*. Another case of *Shewanella putrefaciens* associated sacroiliac osteomyelitis with bacteremia acquired after receiving subcutaneous infusions for thalassemia major has been described in a young boy.<sup>14</sup> Bacteremia is a known complication of *Shewanella* infection in at least 28% of cases.<sup>9</sup>

Amongst the four cases, two of the cases of soft tissue and bone infection grew *S. algae* whereas the two cases of septicaemia grew *Shewanella putrefaciens*. Repeated isolation of this organism with cor-relating clinical history established the pathogenic role of this organism though it is often considered as a colonizer.<sup>9</sup> Both the cases of septicaemia grew *Shewanella putrefaciens*. Repeated

isolation of this organism with cor-relating clinical history established the pathogenic role of this organism though it is often considered as a colonizer.<sup>9</sup>

Tsai et al, in 2008, reviewed 27 cases of *Shewanella* SSTI in adults. The majorities of these were associated with infections of the extremities (82%) with over half of all cases involving chronic ulcers of the legs.<sup>3</sup> Studies have reported a male preponderance which may be possibly due to more chances of occupational exposure to seawater.<sup>2</sup> In this present study too, three of the four cases were adult males.

In all the patients, some kind of exposure to a source of infection could be traced. In the present series, case 1 reported no contact with marine life, but the sewage water running beside the patients home could have been a source of the organism. The second patient with osteomyelitis got injured at a beachside where he may have come in contact with seawater. In the 3<sup>rd</sup> case, patient gave a history of regular visit to a local barber which raised the possibility of contamination of his venous and arterial dialysis ports. The fourth patient did not give any history of contact with a known source, but he was admitted in the same ward as case 3. *Shewanella* spp. have rarely been isolated from medical devices and have been known to cause health – care-associated infections (19% cases) and outbreaks.<sup>9,15</sup>

An outbreak of *Shewanella* infection occurring in a single surgical ward of Seoul National University Hospital between June 2003 and January 2004 has been recorded. 31 cases of *Shewanella* infection or colonization were observed. Blood, bile, and ascitic fluid were the most common clinical specimens that yielded *Shewanellae*. A retrospective analysis of infection control procedures identified a measuring cup used for catheter drainage which was routinely used repeatedly for many patients.<sup>15</sup> In the present study, surveillance of environment, personnel and patients did not yield *Shewanella* species from any of the sites and therefore, contamination was ruled out.

The organism has been associated with other bacterial pathogens i.e. polymicrobial infection in 60% of cases.<sup>16</sup> Ryan et al have reported an incidence of 53% cases as polymicrobial and associated with underlying conditions.<sup>5</sup> In the present case series, only the first case showed polymicrobial infection with *Proteus mirabilis* and MRSA in the later period whereas the other cases were mono-microbial.

CLSI 2018 has not described standards for *Shewanella* species disk diffusion interpretation. So the recommendation for *Pseudomonas* was used for *Shewanella* species. The role of VITEK automated system for susceptibility testing for this organism has already been documented in earlier literature.<sup>3</sup> *Shewanella* species are susceptible to commonly used antimicrobial agents particularly 3<sup>rd</sup> gen cephalosporin (95%), piperacillin (94%), piperacillin- tazobactam (98%), ciprofloxacin (94%), gentamicin (99%), imipenem (82%),



**Fig. 1:** Osteomyelitis of left proximal femur with soft tissue involvement in an 18 year old female patient due to *Shewanella* algae

amikacin (100%). *Shewanella* species showed complete resistance to penicillin which was used commonly to treat soft tissue infections.<sup>9</sup>

Studies have reported variable susceptibility to cephalosporins.<sup>16</sup> In the present study, both the *Shewanella algae* cases were sensitive to all the drugs except resistance was seen to third generation cephalosporins in one case and to quinolones in another case. The *Shewanella putrefaciens* strains were more resistant showing resistance to third generation cephalosporins, quinolones, aztreonam and carbapenems. Studies have shown that *Shewanella* can show resistance to imipenem by secreting an oxacillinase.<sup>17</sup> This shows that this organism is developing resistance to antibiotics which had earlier been reported sensitive and therefore antimicrobial resistance testing for this organism is essential for treatment.

Out of the 4 cases, three could be successfully treated while one succumbed just one day after specific antibiotic treatment was initiated. Death has been reported in 13% of *Shewanella* cases<sup>9</sup> but all these patients already had some chronic conditions which would have contributed to the high morbidity and mortality.<sup>1</sup>

**Table 1:** Biochemical characteristics of *Shewanella* species

Trait	Characteristics	
	<i>S. putrefaciens</i>	<i>S.algae</i>
Mucoid colony	-	+
Growth at 42° C	-	+
Growth at 4° C	+	-
Growth in nutrient broth	+/-	+
Growth in presence of 6-6.5% NaCl	-	+
Hemolysis on SBA	-	+
Glucose fermentation	-	v
Arabinose fermentation	+	-
Nitrite reduction	+	-

### 3. Conclusion

The isolation of *Shewanella* species from invasive specimens highlights the importance of this organism as an emerging pathogen. In the present study, *Shewanella* species was isolated as a community acquired infection in two cases and a possible hospital acquired pathogen in the other two. Proper hospital aseptic techniques along with proper hand hygiene goes a long way in controlling the spread of this infection in hospitals.<sup>2</sup> Epidemiologic studies for *Shewanella* species have not been carried out anywhere and would be helpful in the control of this organism.

Although *Shewanella* species is known to be a sensitive organism, their varied susceptibility profile mandates antibiotic susceptibility testing for which standard criteria need to be described.

### 4. Conflicts of interest

Nil

### 5. Source of funding

None

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