

Isolation of *Aeromonas salmonicida* from Human Blood Sample: A Case Report

RACHNA TEWARI¹, MRIDU DUDEJA², SHYAMASREE NANDY³, AYAN KUMAR DAS⁴

ABSTRACT

Aeromonas salmonicida belonging to the genus *Aeromonas*, is a common pathogen that causes furunculosis and septicaemia in variety of fishes. It infects cold blooded vertebrates living at low temperatures mainly salmonid fish hence named salmonicida. Until recently *Aeromonas salmonicida* is considered to be a fish pathogen. *A. salmonicida* is considered to be non-pathogenic for humans as it cannot grow at 37°C. "However, In our laboratory culture plates and broths were incubated twice at 37°C and each time same type of colonies were isolated which were identified as *A. salmonicida* by Vitek 2 compact system bioMerieux, Inc. (Durham, N.C.)". By far no report has been received regarding its isolation from humans biological sample. Here we present the first report of *A. salmonicida* isolated from the human blood.

CASE REPORT

A 34-year-old female visited medicine Out Patient Department (OPD), at our institute Hamdard Institute of Medical Science and Research, Delhi, India (HIMSR) in April 2013 with complaints of off and on, low grade fever, associated with weakness and malaise for last 10 days. There was no history of chills and rigor, sore throat, diarrhoea, urinary discomfort, any other localised infection, or chronic illness suggestive of immunocompromised state except history of two recurrent miscarriage within last nine months for which she could not give any medically strong reason. Being nurse by profession she would remain in contact with patients in hospital settings. She took Combiflam for mild headache and fever but recurrence of fever made her consult specialist. All the routine investigations were found to be normal except leucocytes which were slightly raised.

Blood, urine and stool were sent to microbiology lab for serology, culture and sensitivity. Serology for typhoid, malaria and dengue were reported negative. Cultures for the stool and urine showed growth of non-pathogenic organism. However, after over night incubation, turbidity and haemolysis were reported in blood culture bottle. Blood culture broth was subcultured on blood agar plate and McConkey agar plate and incubated overnight. Next day, pinpoint yellowish colonies, friable, not easily emulsifiable, non-haemolytic and having entire margin grew on blood agar plate. No growth was observed on McConkey Agar. On Grams staining, gram negative bacilli with no specific arrangement were seen. The organism was nonmotile, catalase positive and oxidase positive. Triple sugar iron (TSI) was K/K, hydrogen sulphide production negative, indole negative, urea negative and citrate utilisation negative. As identification could not be clearly made by these biochemical test blood agar plate was reincubated. Next day, colour of colonies turned golden yellow where as size of colonies became larger.

Antibiotic sensitivity was determined by Kirby Bauer's disk diffusion method as per CLSI guidelines [1]. The isolate was found to be sensitive for all the drugs (disks of Himedia) chloramphenicol (30 mcg), ampicillin (10 mcg), imipenem (10 mcg), aztreonam (30 mcg), ceftazidime (30 mcg), ceftriaxone (30 mcg), cefixime (5 mcg), tetracycline (30 mcg), ciprofloxacin (5 mcg), piperacillin (100 mcg), levofloxacin (5 mcg), netilmycin (30 mcg), and amikacin (30 mcg).

As identification of organism could not be done by conventional biochemical reactions second sample was collected on her second

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visit. Sample was processed by Bact T alert (bioMerieux, New York) and subcultured on Blood and McConkey agar and incubated overnight at 37°C. Next day colonies with same morphology (as of first sample) were isolated. For second sample identification was done and confirmed by Vitek 2 compact system. To our surprise report showed isolation of *Aeromonas salmonicida*. Our patient was treated with cefixime and she responded well. She did not visit the hospital for any complaints after that.

DISCUSSION

Member of the genus *Aeromonas* have been recognised since 1891, when Sanerelli, first reported them in frog and produced septicaemia and other diseases upon reinoculation into cold and warm blooded animals. Through the use of improved techniques for isolation and identification from biological specimens their importance in human has recently become better appreciated. Much advancement has occurred regarding their taxonomy, disease spectrum and pathogenicity over many past years [2].

Aeromonas salmonicida belonging to the genus *Aeromonas*, was first discovered in a Bavarian brown trout hatchery by Emmerich and Weibel in 1894 [3]. It is facultative anaerobe, Gram negative, nonmotile bacterium which readily ferments and oxidises glucose and give catalase and oxidase test positive [4]. It is considered as primary pathogen in variety of fishes [4,5] and not in humans as they cannot grow at 37°C. Optimal temperature required for its growth has been reported as 22-25°C [6]. In 90% of the strains, its virulence was found to be lost if cultured at 30°C and above [5].

Previously other *Aeromonas* species like *A. hydrophila*, *A. caviae*, *A. veronii* etc were also considered as pathogen in cold blooded animals only, including fish, amphibians and reptiles but gradually recognised as opportunistic pathogen for humans. However these organisms have increasingly been identified as a primary pathogen for humans in normal individual as well as in immunocompromised patient mainly in gastrointestinal infections and septicaemia [7]. Major sources described for *Aeromonas* species in gastrointestinal infection are environment-water-animal complex and ingestion of contaminated foods whereas sources reported for extraintestinal infection are either direct soil or water contact or ingestion of contaminated food followed by bacteremic dissemination from gastro intestinal tract [8]. However in some cases of *Aeromonas*

septicaemia with no history of infections the origin of the organism and portal of entry is yet unclear [8], similar to our case where source could not be traced.

Compared with other pathogenic aeromonads *A. salmonicida* can also be found in environment, diseased fish and water and may be transmitted by all these sources. Also it has pathogenic factor S-layer which mediates tissue adherence as in other *Aeromonas* like *A. hydrophila*, *A. veronii* so it may also act as pathogen in humans like other species [5,9]. Although previously documented that growth is unlikely at 37°C and so in humans [5] which is contradictory to our report where blood samples were cultured by two different methods (conventional and BacT alert) and subcultured plates were incubated at 37°C and for both samples (first and second) same colonies were isolated which suggests that either *A. salmonicida* has undergone some changes and can grow at 37°C or it has been misidentified by Vitek 2 system.

Although Vitek 2 compact system is considered to be a very reliable system but there are reports in literature where misidentification of *Aeromonas* sp. has been done for other species or rather genus. Reports of misidentification of *Aeromonas* as *vibrio*, two strains of *A. schubertii* by *A. sobria* and by *Vibrio damsella* [8] and sometimes by *Elizabethkingia meningosepticum* have been documented and this again raises a question whether this type of system should be used or not in laboratories for identification of organism. We have limited resources and facilities to work on molecular basis in our setup hence this Vitek 2 report could not be confirmed further but we can throw the light on this organism and make the scientists to rethink and work more on this organism to find any change undergone by this organism in pathogenicity, virulence, disease spectrum and antibiotic resistance.

Although our isolate was found sensitive to all drugs (by Kirby Baeurs method) and our patient responded well with cefixime, however

resistance for tetracycline and quinolone has been reported in *A. salmonicida*. Recently antibiotic resistant *A. salmonicida* strains have been recognised as a serious concern owing to their potential health risk to humans and animals [3]. Has this problem been evolved by humans only who are devising techniques and facilities to cross all the boundaries and getting interacted with all types of living and non living things? To conclude, society is already overburdened with resistant organism and if nonpathogenic organisms will change their host preference, virulence and sensitivity, it will be difficult for clinicians and scientists to tackle this problem.

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PARTICULARS OF CONTRIBUTORS:

1. Assistant Professor, Department of Microbiology, Hamdard Institute of Medical Sciences & Research (HIMSR), India.
2. Professor, Department of Microbiology, Hamdard Institute of Medical Sciences & Research (HIMSR), India.
3. Demonstrator, Department of Microbiology, Hamdard Institute of Medical Sciences & Research (HIMSR), India.
4. Demonstrator, Department of Microbiology, Hamdard Institute of Medical Sciences & Research (HIMSR), India.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:

Dr. Rachna Tewari,
Assistant Professor, Department of Microbiology,
Hamdard Institute of Medical Sciences & Research (HIMSR), New Delhi, India.
Phone: 9311065999, E-mail: dr_rachna_tewari@yahoo.com

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