

## *Salmonella choleraesuis* subsp. *indica* Serovar bornheim Causing Urinary Tract Infection

SHEILA SNEHALATHA,<sup>1</sup> ELIZABETH MATHAI,<sup>1\*</sup> M. JAYASHEELA,<sup>2</sup> MAMMEN CHANDY,<sup>3</sup>  
M. K. LALITHA,<sup>1</sup> AND T. JACOB JOHN<sup>1</sup>

Department of Microbiology,<sup>1</sup> and Department of Clinical Haematology,<sup>3</sup> Christian Medical College Hospital,  
Vellore 632 004, and Central Research Institute, Kasauli 173205,<sup>2</sup> India

Received 11 February 1992/Accepted 15 May 1992

**An unusual *Salmonella* species, *S. choleraesuis* subsp. *indica* serovar bornheim, was isolated from the urine of a patient with aplastic anemia, diabetes mellitus, and a healed urethral injury. An immune response to this isolate was demonstrated by whole-bacterial-cell agglutination.**

Although infections due to salmonellae are common worldwide, serovars other than those belonging to subspecies I are rarely pathogenic to humans (3, 5). In addition, salmonellae are rarely reported as etiological agents of urinary tract infection (UTI) (1). We report a case of UTI caused by serovar bornheim (*S. bornheim*), a member of *Salmonella choleraesuis* subsp. *indica* (VI).

A 32-year-old man with aplastic anemia with leukopenia, diabetes mellitus, and a healed urethral injury after pelvic trauma in an accident 7 years earlier was admitted to the hematology unit. He was given anti-T-cell monoclonal antibodies for 5 days from day 7 of admission, in addition to prednisolone (10 mg twice a day). Twenty-two days after admission, he developed fever and increased frequency of and a burning sensation on micturition. Upon microscopic examination of his urine, approximately 20 polymorphonuclear leukocytes and 30 erythrocytes per high-power field were seen. A midstream clean-catch specimen of urine was transported without delay to the laboratory for culture; 0.01 ml was plated on blood agar for isolation and on MacConkey agar for colony counting (8). After overnight incubation at 37°C, a pure growth of nonhemolytic and non-lactose-fermenting colonies grew to a colony count of >10<sup>5</sup>/ml. This isolate was identified as *Salmonella* species, based on biochemical reactions and agglutination with *Salmonella* polyvalent O antiserum (A-I and Vi) (Difco Laboratories, Detroit, Mich.). The screening biochemical tests used included an acid butt with gas and H<sub>2</sub>S on triple sugar iron agar medium, mannitol fermentation, citrate utilization, lysine decarboxylation, and negative tests for indole and urease (3) (Table 1). Antimicrobial susceptibility tests were done by the modified Kirby-Bauer disk diffusion method (10). *Staphylococcus aureus* and *Escherichia coli* with known zone sizes for each antibiotic were used as controls. The *Salmonella* isolate was sensitive to chloramphenicol (30 µg per disk), ampicillin (10 µg), co-trimoxazole (1.25/23.75 µg), tetracycline (30 µg), gentamicin (10 µg), cephalothin (30 µg), norfloxacin (10 µg), and nalidixic acid (30 µg). Zone size was interpreted according to the National Committee on Clinical Laboratory Standards chart (vol. 7, 1988).

A blood culture done immediately after the isolation of *Salmonella* species from the urine was sterile. The patient's serum sample was tested for the presence and titer of agglutinating antibodies against the isolate. The isolate was

grown on nutrient agar, harvested, suspended in 0.85% saline, and treated with absolute alcohol to a final concentration of 33%. The opacity was adjusted to that of Browne's tube no. 3 (2). The serum was diluted in serial twofold steps in 0.85% saline. To 0.5 ml each of the serum dilutions, an equal volume of the antigen cell suspension was added, and the tubes were incubated at 37°C for 18 h. Agglutination was seen up to a dilution of 1:640.

The patient was given 400 mg of norfloxacin twice daily for 5 days, and he became symptom-free. A second urine culture 4 days after treatment was started did not yield any growth. The *Salmonella* isolate was sent to the National Salmonella and Escherichia Centre at the Central Research Institute, Kasauli, India. It was identified as belonging to subspecies VI (*indica*) by the criteria shown in Table 1. The isolate was found to have an antigenic profile of S.VI 1,6,14,25:Z<sub>10</sub>:1(2),7. Hence it was identified as *Salmonella bornheim*.

The patient was diagnosed as having UTI because of the symptoms and pyuria. A midstream clean-catch specimen of urine yielded >10<sup>5</sup> CFU of *S. bornheim* per ml in pure culture. Hence, this organism was considered to be the causative agent and not part of the fecal flora contaminating the urine.

Salmonellae are rare causes of UTI. In males, such infections are associated with urinary tract abnormalities (11). The patient reported here had an earlier injury of the urinary tract; we believe that this was a risk factor for his unusual infection. Aplastic anemia, the associated leukopenia, and the immunosuppressive treatment also could have contributed. Finding *Salmonella* species in the urine need not necessarily mean UTI caused by *Salmonella* species, since bacteruria may follow bacteremia. One blood culture done in this patient was sterile. This culture, however, was done after the isolation of *S. bornheim* from the urine. In bacteremic *Salmonella* infections like enteric fever, the blood culture is usually positive during week 1 of illness, while the urine culture becomes positive only around week 3. This patient had a fever for 4 days, 1 week before the onset of UTI. No cultures were done at this stage, and therefore a preceding bacteremia cannot be ruled out.

According to the currently accepted taxonomic classification, there are two species of *Salmonella*, *S. choleraesuis* and *S. bongori* (9). Most salmonellae causing human infections belong to *S. choleraesuis*. Many workers prefer the term *S. enterica* for this species (3, 9). This species has six subspecies, *S. enterica* subsp. *enterica*, subsp. *salamae*,

\* Corresponding author.

TABLE 1. Results of tests done for identification of *S. choleraesuis* serovar bornheim<sup>a</sup>

Test	Result	Test	Result
Motility .....	+	Methyl red .....	+
Glucose .....	AG <sup>b</sup>	Voges-Proskauer .....	-
Lactose .....	-	Urease .....	-
Sucrose .....	-	<u>Gelatinase</u> .....	+
Maltose .....	AG	Mucate .....	+
Mannitol .....	AG	H <sub>2</sub> S .....	+
Dulcitol .....	-	Indole .....	-
<u>Salicin</u> .....	-	Phenyl alanine deaminase .....	-
Adonitol .....	-	KCN .....	-
Inositol .....	-	Lysine decarboxylase .....	+
<u>Sorbitol</u> .....	-	Arginine dihydrolase .....	+
Cellulose .....	-	Ornithine decarboxylase .....	+
Xylose .....	AG	Simmons citrate .....	+
Mannose .....	AG	O-Nitrophenyl-β-D-galactopyranoside .....	-
Arabinose .....	AG	<u>Malonate</u> .....	-
Trehalose .....	AG	D-tartrate .....	-
Rhamnose .....	AG	meso-tartrate .....	-
Raffinose .....	-	<u>L-tartrate</u> .....	-
Melibiose .....	AG	Galacturonate .....	+

<sup>a</sup> Characteristic reactions for subspecies VI are underlined.

<sup>b</sup> AG, acid and gas.

subsp. *arizonae*, subsp. *diarizonae*, subsp. *houtenae*, and subsp. *indica*. Subspecies VI (*indica*) was described by LeMinor et al. (7) in 1986, based on biochemical and genomic characteristics. Biochemically, this subspecies can be identified by five properties. They produce gelatinase, do not utilize malonate and L-tartrate, and do not ferment salicin and sorbitol (7). Ten serovars are assigned to this subspecies (9). The initial isolation of *S. bornheim* was from *Cordylus cordylus* in the Frankfurt Zoo in 1964 (4). *S. bornheim* was then placed in subgenus II of Kauffmann and shifted to subspecies VI in 1988 (6). A literature search done for the word "*bornheim*" on MEDLINE for the years 1966 to 1991 did not detect any report of human infection caused by *S. bornheim*.

#### REFERENCES

- Christie, A. B. 1987. Food poisoning. Salmonellosis, p. 43-99. In *Infectious diseases: epidemiology and clinical practice*, vol. I. Churchill Livingstone, Ltd., London.
- Cruickshank, R., J. P. Duguid, B. P. Marmion, and R. H. A. Swain. 1975. Centrifuges, calorimeters, bacterial counts, p. 301-308. In *Medical microbiology. The practice of medical microbiology*, vol. II. Churchill Livingstone, Ltd., London.
- Ewing, W. H. 1986. Edwards & Ewing's identification of enterobacteria, p. 247-318. Elsevier Science Publishing, Inc., New York.
- Kelterborn, E. 1967. *Salmonella* species, p. 84. W. Junk, The Hague, The Netherlands.
- LeMinor, L. 1988. Typing of *Salmonella* species. *Eur. J. Clin. Microbiol. Infect. Dis.* 7:214-218.
- LeMinor, L., and M. Y. Popoff. 1988. Antigenic formulas of the *Salmonella* serovars, 5th revision. WHO Collaborating Centre for Reference and Research on Salmonella. Institute Pasteur, Paris, France.
- LeMinor, L., M. Y. Popoff, B. Laurent, and D. Hermant. 1986. Individualisation d'une septieme sous-espèce de salmonella *S. choleraesuis* subsp. *indica* subsp. nov. *Ann. Inst. Pasteur Microbiol.* 137B:211-217.
- Myers, R. M., and G. Koshi. 1982. Diagnostic procedures in medical microbiology and immunology/serology, p. 60-63. Christian Medical College Hospital, Vellore, India.
- Popoff, M. Y., J. Bockemuhl, and A. McWhorter-Murlin. 1991. Supplement 1990 (no. 34) to the Kauffmann-White scheme. *Res. Microbiol.* 142:1029-1033.
- Vandepitte, J. K., K. Engboeck, P. Piot, and C. C. Heuck. 1991. Antimicrobial susceptibility testing, p. 78-93. In *Basic laboratory procedures in clinical bacteriology*. World Health Organization, Geneva.
- Wilson, R., and R. A. Feldman. 1982. *Salmonella* isolated from urine in the United States. *J. Infect. Dis.* 146:293-296.