## Dual Emergence in Food and Humans of a Novel Multiresistant Serotype of Salmonella in Senegal: Salmonella enterica subsp. enterica Serotype 35:c:1,2

Food-borne diseases remain one of the most widespread public health problems in the contemporary world and an important cause of reduced economic productivity despite progress in food science and technology. Throughout the world, hundred of millions of people suffer from communicable diseases caused by contaminated food and water. The problem is more acute in developing countries, where there are an increased number of vulnerable people such as immunocompromised and/or undernourished individuals. Salmonella infections in humans are the primary cause of food-borne disease, resulting in considerable morbidity and occasionally death (1). Serotyping is a useful epidemiological tool to detect outbreaks and emergence of a new serotype (3). Between 29 March and 9 May 2000, the Senegalese National Salmonella and Shigella Reference Laboratory at the Pasteur Institute in Dakar received five Salmonella strains for serotyping and determination of antimicrobial susceptibility pattern. The first strain was isolated from poultry on 29 March 2000, and the remaining four strains were isolated from human samples. The first human strain was isolated on 30 March 2000 from a blood sample, and the other strains were isolated from stool samples from three patients admitted to the hospital during April and May 2000. All patients were from the same ward.

Determination of somatic and flagellar antigens was performed by the agglutination method with commercial antiserum (Bio-Rad-Pasteur, Paris, France). The strains belonged to group C of the Kauffmann-White scheme, and the definite antigenic formula was established by the French National Salmonella and Shigella Reference Laboratory at the Pasteur Institute in Paris. The antigenic formula of all five of these strains was 35:c:1,2; this serotype has never before been described (11). Confirmation of this new antigenic formula was done later by the Hamburg National Reference Center in Germany and the National Salmonella Reference Laboratory at the Centers for Disease Control and Prevention in Atlanta, Ga. Antimicrobial susceptibility testing was done by a disk diffusion method on Mueller-Hinton agar prepared according to the National Committee for Clinical Laboratory Standards guidelines (9). Based on the disk diffusion method, the strains were resistant to the penicillin and cephalosporin classes of antibiotics. Additionally, these isolates were resistant to gentamicin, tobramycin, chloramphenicol, and tetracycline and remained susceptible to quinolones (pefloxacin, norfloxacin, and nalidixic acid) and amikacin. The MIC of cefotaxime, ceftazidime, and aztreonam was determined by the agar dilution method (10) to be >128  $\mu$ g/ml. Based on the double-disk synergy test (6) performed with cefotaxime, ceftazidime, aztreonam, and amoxicillin-clavulanic acid, all strains showed synergy between amoxicillin-clavulanic acid and expandedspectrum cephalosporins.

Analytical isoelectric focusing (IEP) of crude extracts (8) demonstrated the production of SHV-12 (IEP = 8.2), an extended-spectrum lactamase responsible for high-level expanded-spectrum cephalosporin-hydrolyzing activity. SHV-specific PCR followed by sequencing (7) confirmed this result. This is the first report of SHV-12 in *Salmonella*.

Extended-spectrum beta-lactamases (ESBLs) are rarely associated with the genus *Salmonella*. Multiresistant *Salmonella* 

*enterica* serovar Typhimurium of definitive phage type (DT) 104 with chromosomal integration of the genes coding for resistance to ampicillin, chloramphenicol, streptomycin, sulfonamides, and tetracycline has been previously described (12). In Africa, the first *Salmonella* strains with ESBLs were identified in 1988 in Tunisia and belonged to serotype Wien (4). In Senegal, the first multiresistant *Salmonella* strains with ESBL activity belonged to serotype Kentucky and were identified only in a human sample (5).

All the strains seem to be epidemiologically related; however, an investigation has failed to prove any relationship between the animal and human isolates. Nosocomial transmission is not probable since isolation of the strains from each patient was done within the 48 h following admission to the hospital.

How has this multiresistant strain emerged? The extensive use of antimicrobial agents as feed additives for farm animals and in the control and treatment of Salmonella infection has been suggested as a predisposing factor in the evolution of multiresistant strains (2). In Senegal, antibiotics are used extensively in the poultry industry to reduce mortality and to increase the body weight of the animals. In most of these artisan or familial farms, there is a drastic absence of adherence to measures of good farming practice and good manufacturing practice during production and processing. Contrary to practices in developed countries, a decontamination step for eliminating pathogenic microorganisms from meat has not been considered in Senegal. Additionally, poultry products are served at street food stalls or in fast-food restaurants where food processors are not always aware of food safety rules. This is the first isolation of this Salmonella serotype from animals in Senegal. There is a potential threat for the transmission of this multiresistant, ESBL-producing strain via the food chain to humans.

## REFERENCES

- Bean, N. H., J. S. Goulding, C. Lao, and F. J. Angulo. 1996. Surveillance for foodborne disease outbreaks United States, 1988–1992. Morb. Mortal. Wkly. Rep. CDC Surveill. Summ. 45:1–65.
- 2. Dupont, H. L., and J. H. Steele. 1987. Use of antimicrobial agents in animal feeds: implications for human health. Rev. Infect. Dis. 9:447–460.
- Echeita, M. A., A. Aladuena, S. Cruchaga, and M. A. Usera. 1999. Emergence and spread of an Atypical *Salmonella enterica* subsp. *enterica* Serotype 4,5,12:i:- strain in Spain. J. Clin. Microbiol. 37:3425.
- Hammami, A., G. Arlet, S. Ben Redjeb, F. Grimont, A. Ben Hassen, A. Rekik, and A. Phillipon. 1991. Nosocomial outbreak of acute gastroenteritidis in a neonatal intensive care unit in Tunisia caused by multiply drug resistant *Salmonella* Wien producing SHV-2 beta-lactamase. Eur. J. Clin. Microbiol. Infect. Dis. 10:641–646.
- Hugard, L., and B. N'Doye. 1993. Isolement de Salmonella ser. Kentucky présentant une bêta-lactamase à spectre élargi: à propos d'un cas. Feuill. Biol. 195:79–80.
- Jarlier, V., M. H. Nicolas, G. Fournier, and A. Phillipon. 1988. Extended broad-spectrum beta-lactamases conferring transferable resistance to newer beta-lactam agents in *Enterobacteriaceae*. Hospital prevalence and susceptibility patterns. Rev. Infect. Dis. 10:867–878.
- Mabilat, C., and S. Goussard. 1993. PCR detection and identification of genes for extended-spectrum β-lactamases, p. 553–559. *In* D. H. Persing, T. F. Smith, F. C. Tenover, and T. J. White (ed.), Diagnostic molecular

microbiology: principles and applications. American Society for Microbiology, Washington, D.C.

- Matthew, M., A. M. Harris, M. J. Marshall, and G. Ross. 1975. The use of analytical isoelectric focusing for detection and identification of beta-lactamases. J. Gen. Microbiol. 88:169–178.
- National Committee for Clinical Laboratory Standards. 1993. Performance standards for antimicrobial disk susceptibility tests, 5th ed., vol. 13, no. 24. Approved standard M2–A5. National Committee for Clinical Laboratory Standards, Villanova, Pa.
- National Committee for Clinical Laboratory Standards. 1993. Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically, 3rd ed. Approved standard M7–A3. National Committee for Clinical Laboratory Standards, Villanova, Pa.
- Popoff, M. Y., and L. Le Minor. 1977. Antigenic formulas of the Salmonella serovars, 7th ed. WHO Collaborating Center for Reference and Research on Salmonella, Institut Pasteur, Paris, France.
- Threlfall, E. J., J. A. Prost, L. R. Ward, and B. Rowe. 1996. Increasing spectrum of resistance in multiresistant *Salmonella typhimurium*. Lancet 347:1053–1054.

Eric Cardinale ISRA/LNERV Dakar, Senegal

**Pierre Colbachini** Hopital Principal Dakar, Senegal

Jean David Perrier-Gros-Claude Amy Gassama Awa Aïdara-Kane\* Laboratoire de Bactériologie Expérimentale Institut Pasteur BP 220 Dakar, Senegal

\*Fax: 221 839 92 36 E-mail: aidarawa@pasteur.sn