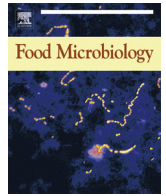




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Salmonella epidemiology: A whirlwind of change

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ABSTRACT

The field of infectious disease epidemiology for *Salmonella* and other enteric pathogens is undergoing some of the most profound changes since the time of Kauffman and White. Rapid advances in “big data” technologies such as genomics and metagenomics are making it possible to monitor and control salmonellosis in new and exciting ways. Epidemiological methods are becoming increasingly robust through the routine use of standardized hypothesis-generating questionnaires, iterative open-ended interviewing, informational trace-backs and new modeling techniques for describing the attribution of disease to food sources. In addition, *Salmonella* epidemiology is facing important challenges and new opportunities due to the rapid adoption of culture independent diagnostic test panels by clinical laboratories. Where is this unprecedented wave of change taking us? This chapter will examine emerging trends in *Salmonella* epidemiology, and take a peek into the not-so-distant future.

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1. Background

Diarrheal illness remains a significant cause of morbidity and mortality worldwide. The World Health Organization (WHO) estimates that approximately 1.9 billion people worldwide become ill with diarrhea each year, and 715,000 die. According to these estimates, approximately one third of these infections are transmitted through food. A higher proportion of *Salmonella* cases (52% of non-typhoidal and 37% of typhoidal *Salmonella* cases) than other enteric pathogens are thought to be foodborne, most of which are potentially preventable once vehicles are identified. *Salmonella* is

responsible for 180 million, or 9% of the diarrheal illnesses that occur globally each year. Among recognized etiologies, the number of estimated *Salmonella enterica* illnesses (180 million) is exceeded in frequency by norovirus (685 million), enterotoxigenic *E. coli* (241 million), *Shigella* spp. (190 million), and *Giardia* spp (183 million). However, a disproportionate number of deaths (298 thousand, or 41% of all diarrheal disease-associated deaths) can be attributed to *Salmonella*. Causative serotypes are not evenly distributed around the world. For example, non-typhoidal *Salmonella* (NTS) is more common in Africa, while typhoidal *Salmonella* (e.g. serotypes Typhi and Paratyphi A) are more common in Southeast Asia (WHO, 2016).

The epidemiology of *Salmonella* and other foodborne disease agents has been evolving for decades due to broad trends in food production and consumption, such as increasing industrialization

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and globalization of the food supply, growing demand for “ready-to-eat” and raw or lightly cooked foods, aging populations and higher proportion of immunologically compromised individuals, and use of antibiotics in food animal production (Tauxe et al., 2010, Newell et al., 2010, Quested et al., 2010).

The tools for analyzing trends in foodborne disease and detecting outbreaks have been continuously improving, such as PulseNet for detection of disease clusters that may represent possible outbreaks of *Salmonella* infections (Gerner-Smith et al., 2006). Use of new epidemiological methods have informed policy developed by regulatory agencies, and stimulated safety improvements both by the regulatory agencies and by the food industry. PulseNet, for example, has aided outbreak investigations that uncovered previously unrecognized problems in a wide variety of food commodities, such as various fruits, vine vegetables, leafy green vegetables, spices, tree nuts, poultry, beef, sprouts, flour, and ready-to-eat foods. Other types of trends, such as increased infections by reptiles and amphibian-related *Salmonella* serotypes, reflecting increased adoption of these animals as pets, can be detected through this type of laboratory-based surveillance (Gambino-Shirley et al., 2016; Centers for Disease, 2011). PulseNet has facilitated recognition of outbreaks dispersed over wide areas, which is the type of outbreak pattern that would be expected from commercially distributed food. Although multi-state and multi-national outbreaks represent only 3% of recognized foodborne disease outbreaks in the U.S., they account for 11% of hospitalizations and 56% of attributable deaths (CDC, 2015a). Consequently, rapidly identifying the source of these outbreaks and preventing further illnesses can have substantial impact. A 2016 cost-benefit analysis estimated that the PulseNet USA prevents 266,522 illnesses due to *Salmonella*, Shiga toxin-producing *Escherichia coli* (STEC), and *Listeria monocytogenes* each year, averting approximately \$507,000,000 in medical costs and lost productivity each year (Scharff et al., 2016). Advances have also been made in obtaining case exposure information, cluster and trace-back investigation, which have the potential for greatly increasing the sensitivity of outbreak detection and investigation (Besser, 2013). For example, routine acquisition of detailed exposure information and cluster follow-up for *L. monocytogenes* cases was pioneered by the Institut de Veille Sanitaire (InVS, now Santé Publique France) in France in 1999 (Goulet et al., 2008). In the U.S., the use of informational trace-back methods were used to rapidly test hypotheses in outbreak investigations (Miller et al., 2012).

Despite these advances, rates of salmonellosis have remained stubbornly stable or increased over time. Rates of *Salmonella* enteric fever increased in Australia from 1985 to 2010 from 4 to 7 cases per million person years (Commons et al., 2012). In the U.S., rates of salmonellosis in the FoodNet surveillance network remained essentially flat from 2000 to 2015 (Henao et al., 2015), as did the proportion of antibiotic resistance infections (CDC, 2015b). Nevertheless, the 2016 PulseNet cost benefit analysis (Scharff et al., 2016) provided evidence that surveillance and outbreak detection activities had an impact on salmonellosis rates in the U.S. Fig. A1 in the appendix of that publication highlights the relative rates of salmonellosis in U.S. states as a function of their PulseNet activities. In the states where PulseNet was more effectively utilized (“top 10 adopters”), rates of salmonellosis declined, whereas in states where PulseNet was less effectively used (“bottom 10 adopters”) rates increased. These data suggest the rates of salmonellosis in the U.S. would have increased from 2000 to 2015 in the absence of these surveillance and investigation activities. In other words, the relatively flat rates during this time period likely represents successful interventions triggered by PulseNet investigations.

Among the common enteric illnesses, *Salmonella* accounts for a

higher proportion of deaths, is largely preventable, and is one of the few etiologies for which testing is widely available throughout the world. In addition, steady progress has been made over the last 20 years in surveillance and investigation in the areas of microbiology, case information, cluster follow-up and product trace-back. By the early 2010's, the stage was set for rapid change as new technologies began to emerge.

2. Drivers of rapid change

Technical advances in molecular biology are driving major changes in public health laboratory practice, which will in turn require significant modification and expansion of epidemiology methods. The two technologies most impacting change in *Salmonella* epidemiology are nucleic acid sequencing and information technologies. A number of new epidemiological and trace-back methods are also being developed.

Nucleic acid sequencing technology has been advancing at a much faster pace than Moore's Law, which predicts the doubling of integrated chip processing power approximately every two years, with a commensurate drop in cost (NHGRI, 2016; Wikipedia, 2016). The relatively low cost of whole genome sequencing (WGS) made possible its prospective use in the detection and investigation of enteric disease outbreaks, including those caused by *Salmonella*. The low cost of sequencing should soon make it possible to use metagenomics (i.e. sequencing of all nucleic acids in an environment) for a wide range of studies in *Salmonella* epidemiology that were formerly difficult or impossible to conduct. Both WGS and metagenomics involve the processing, transmission, and storage of vast amounts of data, and successful implementation of these technologies will require comparable innovation in software analysis tools.

3. Whole genome sequencing in *Salmonella* surveillance

In the 2010's, WGS began migrating from research labs into real-time public health surveillance activities around the globe. In collaboration with scientists in France, England, and Belgium, the U.S. began real-time WGS on *Listeria monocytogenes* isolates from all human cases of listeriosis and from all food and environmental isolates obtained as part of regulatory monitoring in 2013. This was conducted in parallel with the standard subtyping method, pulse-field gel electrophoresis (PFGE). Comparing listeriosis clusters and outbreaks in the year preceding WGS implementation to year 2 of the project, the total number of clusters detected in the United States increased from 14 to 21 (50%), the number of outbreak solved (including outbreaks due to previously unrecognized vehicles) increased from 2 to 9 (350%), and the median number of cases per cluster or outbreak decreased from 6 to 3 (–50%) (Jackson et al., 2016). These observations are consistent with the hypothesis that the increased resolution and phylogenetic meaningfulness of WGS should make it possible to detect and solve more outbreaks, including smaller outbreaks that would not have been detectable or solvable using earlier methods. It accomplishes this by (1) increasing the specificity of the case definition, thereby reducing the number misclassified cases in clusters; (2) improving the sensitivity of the case definition by inclusion of true cases in clusters that would have otherwise been excluded, and (3) increasing the strength of hypotheses derived from clustering of patient and food/environmental isolates.

Although the epidemiology of listeriosis has important differences from that of salmonellosis, it seems likely that widespread adoption of WGS could similarly impact the epidemiology of *Salmonella*. Listeriosis is a relatively uncommon disease, with the global burden estimated to be 23,150 cases per year (Maertens de

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