Salmonella Serovars from Foodborne and Waterborne Diseases in Korea, 1998-2007: Total Isolates Decreasing Versus Rare Serovars Emerging

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INTRODUCTION

Salmonella has been a major foodborne and waterborne pathogen in Korea (1, 2). In 2006, Salmonella was a major foodborne bacterial pathogen in the United States, causing more deaths than any other foodborne pathogen (3). S. enterica Typhi, a causative agent of typhoid fever, has especially threatened Korean health (4). There were 1,921 deaths resulting in 17% mortality because of typhoid fever during 1945-1960 in Korea. Salmonellosis caused by non-typhoid Salmonella (NTS) gives rise to diarrhea, vomiting, abdominal pain, and enteric fever. Occasionally, systemic infection with bowel peroration, septicemia, and osteomyelitis are also caused by NTS (5-8).

At present, there are more than 2,500 Salmonella serovars in the world with new serovars emerging yearly. Salmonella serotyping is very important to the epidemiology study. Unquestionably, Salmonella serotyping is time-consuming and complex work for the serological identification of bacteria (9). To serotype Salmonella, lipopolysaccharide epitopes in bacterial membrane (O antigens) and flagella proteins (H antigens) should be identified with the respective antibodies. For the identification of S. enterica Typhi, additional antibody specific to capsular polysaccharides (Vi antigen) is essential. Until 1990, reference laboratories in Korea merely performed sero-grouping with only O antigen-specific and Vi-specific antibodies. Therefore, they were unable to complete serotyping of Salmonella due to expensive commercial antibodies and lack of interest in surveillance. Truly, at present, most poor or developing countries perform only sero-grouping of Salmonella because of the cost problem (10).

Korea National Institute of Health (KNIH) is the headquarters for the national surveillance of Salmonella in Korea. KNIH gathered Salmonella isolates and analyzed their epidemiological data from 17 Research Institutes of Health and Environment located in cities and provinces and 13 quarantine stations located in airports and harbors in Korea. The microbiologists in these 17 regional institutes covering all country and 13 quarantine stations isolated enteric bacteria from patients according to the standardized protocols distributed by KNIH. According to Korea’s Infectious Diseases Prevention Act which has been enforced since 1954, certain infectious diseases must be reported to government authorities. These diseases are classified into four classes according to the grade of danger and threat to public health. Among the infectious diseases caused by Salmonella species, typhoid fever and paratyphoid fever are classified as Class 1 notifiable infectious diseases, and NTS-causing diseases are classified as Class 4.

As Korea has developed, the desire for an advanced social hygiene system for the well-being of the people has increased.

Salmonella enterica has been one of the most widespread foodborne pathogens in Korea. Between 1998 and 2007, a total of 9,472 Salmonella isolates were identified from foodborne and waterborne illness patients. During that time, Korea was transitioning into a developed country in industry as well as in its hygiene system. Although the isolation number of total Salmonella including serovar Typhi has decreased since 1999, the isolation of rare Salmonella serovars has emerged. Three most prevalent serovars during 1998-2007 were S. enterica Typhi, S. enterica Enteritidis, and S. enterica Typhimurium. There were remarkable outbreaks caused by rare serovars such as S. enterica Othmanschen, S. enterica London and S. enterica Paratyphi A, and overseas traveler-associated infections caused by S. enterica Weltevreden and S. enterica Anatum. Salmonella serovars from overseas travelers made a diverse Salmonella serovar pool in Korea. This study is the first review of the status of the human Salmonella infection trend in a developing country during 1998-2007. Newly emerging rare Salmonella serovars should be traced and investigated to control new type pathogens in the developed world.

Key Words: Salmonella; Serovar; Foodborne Diseases
Transportation, water supply and drainage, medical and food-processing systems were developed. Between the 1990s and 2000s, Korea had one of the highest economic growths in the world. This rapid growth resulted in dramatic changes in lifestyles as well as in incidence of foodborne *Salmonella* (11).

In this review, three major *Salmonella* serovars, *S. enterica* Typhi, *S. enterica* Enteritidis, and *S. enterica* Typhimurium, and several remarkable outbreaks caused by rare *Salmonella* serovars in 1998-2007 are discussed. As complete serotyping of *Salmo- nella* was started and its security electronic database was constructed in national reference laboratories from 1998, this study is the first review of the status of the human *Salmonella* infections trend in Korea during 1998-2007. Finally, overseas-travel associated infection cases and the effects on serovar prevalence in Korea are also discussed.

**THREE PREVALENT SEROVARS**

From 1998 to 2007, *S. enterica* Typhi, *S. enterica* Enteritidis, and *S. enterica* Typhimurium were the most frequent *Salmonella* serovars in diarrhea patients and foodborne diseases in Korea (Fig. 1, Table 1) (12). From 422 to 2,252 culture-proven *Salmo- nella* infection cases and their isolates have been identified every year since 1998. The percentage of these 3 serovars among *Salmonella* isolates was over 70% almost every year from 1998 to 2007.

*S. enterica* Typhi is a causative pathogen of typhoid fever. Typhoid was an endemic enteric fever disease in Korea. It is not only systemic infection with high morbidity but also a common public health problem in Korea. The overall incidence of typhoid fever was 0.41 per 100,000 population from 1992 to 2000 (4). The number of *S. enterica* Typhi isolates was always among the three

![Incidence of four serovars, *S. enterica* Typhi, *S. enterica* Paratyphi A, *S. enterica* Typhimurium and *S. enterica* Enteritidis and nontyphoidal *Salmonella* serovars excluding *S. enterica* Typhi, 1998-2007.](http://jkms.org)

**Table 1. Top 15 Salmonella serovars from foodborne and waterborne diseases and their number of isolates, 1998-2007.**

| Year | Serovar          | Number of Isolates |
|------|------------------|--------------------|
| 1998 | Typhi            | 626                |
| 1999 | Typhi            | 1,334              |
| 2000 | Typhi            | 3,681              |
| 2001 | Typhi            | 3,157              |
| 2002 | Typhi            | 3,443              |
| 2003 | Typhi            | 4,454              |
| 2004 | Typhi            | 5,083              |
| 2005 | Typhi            | 5,257              |
| 2006 | Typhi            | 5,472              |
| 2007 | Typhi            | 5,734              |

| Year | Serovar          | Number of Isolates |
|------|------------------|--------------------|
| 1998 | Enteritidis      | 718                |
| 1999 | Enteritidis      | 1,147              |
| 2000 | Enteritidis      | 2,154              |
| 2001 | Enteritidis      | 2,115              |
| 2002 | Enteritidis      | 2,221              |
| 2003 | Enteritidis      | 2,261              |
| 2004 | Enteritidis      | 2,342              |
| 2005 | Enteritidis      | 2,405              |
| 2006 | Enteritidis      | 2,482              |
| 2007 | Enteritidis      | 2,571              |

| Year | Serovar          | Number of Isolates |
|------|------------------|--------------------|
| 1998 | Typhimurium      | 195                |
| 1999 | Typhimurium      | 427                |
| 2000 | Typhimurium      | 715                |
| 2001 | Typhimurium      | 864                |
| 2002 | Typhimurium      | 1,084              |
| 2003 | Typhimurium      | 1,211              |
| 2004 | Typhimurium      | 1,303              |
| 2005 | Typhimurium      | 1,367              |
| 2006 | Typhimurium      | 1,431              |
| 2007 | Typhimurium      | 1,496              |

| Year | Serovar          | Number of Isolates |
|------|------------------|--------------------|
| 1998 | Infantis         | 38                 |
| 1999 | Infantis         | 95                 |
| 2000 | Infantis         | 131                |
| 2001 | Infantis         | 190                |
| 2002 | Infantis         | 261                |
| 2003 | Infantis         | 317                |
| 2004 | Infantis         | 378                |
| 2005 | Infantis         | 407                |
| 2006 | Infantis         | 437                |
| 2007 | Infantis         | 464                |

| Year | Serovar          | Number of Isolates |
|------|------------------|--------------------|
| 1998 | Bareilly         | 8                  |
| 1999 | Bareilly         | 12                 |
| 2000 | Bareilly         | 15                 |
| 2001 | Bareilly         | 19                 |
| 2002 | Bareilly         | 25                 |
| 2003 | Bareilly         | 30                 |
| 2004 | Bareilly         | 35                 |
| 2005 | Bareilly         | 40                 |
| 2006 | Bareilly         | 45                 |
| 2007 | Bareilly         | 50                 |

| Year | Serovar          | Number of Isolates |
|------|------------------|--------------------|
| 1998 | Derby            | 11                 |
| 1999 | Derby            | 15                 |
| 2000 | Derby            | 19                 |
| 2001 | Derby            | 23                 |
| 2002 | Derby            | 28                 |
| 2003 | Derby            | 32                 |
| 2004 | Derby            | 36                 |
| 2005 | Derby            | 40                 |
| 2006 | Derby            | 45                 |
| 2007 | Derby            | 50                 |

| Year | Serovar          | Number of Isolates |
|------|------------------|--------------------|
| 1998 | Paratyphi A      | 7                  |
| 1999 | Paratyphi A      | 12                 |
| 2000 | Paratyphi A      | 17                 |
| 2001 | Paratyphi A      | 22                 |
| 2002 | Paratyphi A      | 27                 |
| 2003 | Paratyphi A      | 32                 |
| 2004 | Paratyphi A      | 37                 |
| 2005 | Paratyphi A      | 42                 |
| 2006 | Paratyphi A      | 47                 |
| 2007 | Paratyphi A      | 52                 |

| Year | Serovar          | Number of Isolates |
|------|------------------|--------------------|
| 1998 | Total            | 1,135              |
| 1999 | Total            | 1,390              |
| 2000 | Total            | 1,673              |
| 2001 | Total            | 1,897              |
| 2002 | Total            | 2,104              |
| 2003 | Total            | 2,215              |
| 2004 | Total            | 2,322              |
| 2005 | Total            | 2,433              |
| 2006 | Total            | 2,544              |
| 2007 | Total            | 2,655              |

Source: Reference (12).
most prevalent Salmonella serovars in Korea from 1998 to 2007, excluding 2002 (Table 1). In 2002, S. enterica Paratyphi A and S. enterica Braenderup were the third and fourth most prevalent respectively followed by S. enterica Typhi. During 1961-1963, S. enterica Typhi was the most dominant serovar in Daegu, Korea, showing 92.6% frequency (13).

Unlike other serovars, S. enterica Typhi infects only humans. Therefore, with good hygiene and control of healthy carriers, the incidence of typhoid fever could be decreased (4). Korea Ministry of Health has controlled typhoid fever as a Class 1 notifiable disease. KNH receives S. enterica Typhi isolates along with the epidemiological data from medical doctors according to the Infectious Diseases Prevention Act. Vi-passive hemagglutination for preliminary test and S. enterica Typhi isolation and identification are performed with the stool and the blood culture of the patients. In addition, quarantine stations obtain stool samples from overseas travelers who come from high-risk infectious diarrheal disease areas. Regional Health & Environment Institutes then isolate and identify the pathogens from the specimens (14).

The number of S. enterica Typhi isolates has decreased gradually since 1998. Busan and Gyeongsang-do, which face the East Sea, were higher incidence areas for typhoid fever than other Korea peninsulas (4). The integron-associated multidrug-resistant (MDR) S. enterica Typhi was first identified in Korea in 1999. The MDR isolates were resistant to six antimicrobial agents that were ampicillin, chloramphenicol, trimethoprim-sulfamethoxazole, streptomycin, tetracycline, and gentamicin. All the resistance determinants, aacA4b, catB8, aadA1, dfrA1, aac(6′)-Ila, and blaP2, were clustered in about a 50 kb plasmid (15). Even though most S. enterica Typhi strains are susceptible to antimicrobial agents, drugs for the treatment of typhoid fever should be chosen carefully (16). From the early 2000s, nalidixic acid resistant S. enterica Typhi isolates were also identified in Korea. The genetic relation by PFGE revealed that the nalidixic acid resistant S. enterica Typhi in Korea was closely linked to those in India, Nepal, and Bangladesh. Increased overseas travelers were one of the main reasons. From the 1990s in Korea, ciprofloxacin was recommended as the drug of choice for typhoid fever (4). Since then, ciprofloxacin has been used widely in other developing and developed countries. According to recent phage-typing study of S. enterica Typhi, major phage type transition from M1 and E1 to A occurred in Korea from 1992 to 2006 (17).

S. enterica Typhimurium is a zoonotic pathogen infecting domestic animals and causing salmonellosis in humans (18). S. enterica Typhimurium was one of the major foodborne pathogens in Korea during 1998-2007 (Table 1). It was a more serious problem because the frequency of MDR S. enterica Typhimurium human isolates increased yearly. MDR S. enterica Typhimurium definitive type (DT) 104 first emerged in Korea in 1997 (19). S. enterica Typhimurium DT104, which harbors SGI1, has been identified as a worldwide threat to human and animal health, and showed a high degree of clonality between isolates obtained from different countries (20-22). Over 50% of MDR S. enterica Typhimurium DT104 isolates during 1997-2007 in Korea were resistant to five antimicrobial agents that were ampicillin, chloramphenicol, streptomycin, sulfonamides, and tetracycline (ACSSuT) like the antibiogram of pandemic S. enterica Typhimurium DT104. During 1997-1998, there were five S. enterica Typhimurium DT104-associated foodborne-disease outbreaks nationwide (Table 2). Some S. enterica Typhimurium DT104 Korean isolates had an additional genetic arrangement of antibiotic resistance determinants with those of pandemic S. enterica Typhimurium DT104 (19). A PFGE experiment revealed that some S. enterica Typhimurium DT104 Korean isolates had an indistinguishable PFGE pattern with those of S. enterica Typhimurium DT104 isolates from American cattle (20). Even S. enterica Typhimurium swine isolates in Korea were resistant to at least 4 antimicrobial agents with class 1 integron. Among the isolates, DT104 was found by phage typing (23). Salmonella Genomic Island 1 (SGI1) in S. enterica Typhimurium DT104 chromosome harboring resistance determinants with two class 1 integrons was first identified only in S. enterica Typhimurium, but it has been discovered in other Salmonella serovars now (24), for example, S. enterica Derby (25), S. enterica Paratyphi B (26), and S. enterica Schlessehim (S. Kim, unpublished data). DT104 represented approximately 7% of phage types of all S. enterica Typhimurium human isolates. However, the major phage type was U302 (approximately 45%) (S. Kim, unpublished data).

S. enterica Enteritidis has been the most ubiquitous Salmonella serovar from diarrhea patients since 1998 in Korea. The percentage of S. enterica Enteritidis from all isolated Salmonella spp. from 1998 to 2007 was 47.5% (Table 1). This result indicated that half of the Salmonella isolates from humans were S. enterica Enteritidis. Similarly, S. enterica Enteritidis was the most prevalent serovar among the NTS serovars between 2000-2002 in the world (27). Main infection sources were poultry and eggs.

**Table 2. Selected Salmonella outbreaks in Korea mentioned in this review**

| Salmonella        | Year        | Source                                      | Region     | Number of patients | Reference          |
|-------------------|-------------|---------------------------------------------|------------|--------------------|--------------------|
| S. enterica Typhimurium DT104 | 1997-1998 | Pork and beef                             | Nation-wide | 41                 | Kim et al., 2009 (19) |
| S. enterica Enteritidis | 1999       | Boiled cockle and beef                      | Hamyang    | >200               | Kim et al., 1999 (30) |
| S. enterica London | 2000-2001  | Powdered milk                              | Nation-wide | >70                | Kim et al., 2003 (33) |
| S. enterica Paratyphi A | 2002      | Water                                      | Busan      | >200               | Kim et al., 2003 (40) |
| S. enterica Infantis | 2007      | Not found                                  | Jeollanam-do | >49                | S. Kim, unpublished data |
| S. enterica Ochtmarschen | 2007      | Eggs, squash, and seafood                  | Guri       | 72                 | Kim et al., 2007 (45) |
which were contaminated easily with *Salmonella enterica* Enteritidis. Molecular epidemiological and phage typing study with *Salmonella enterica* Enteritidis isolated from patients and chickens revealed that common phage types and PFGE patterns were found in both isolates. The common phage types in isolates from both patients and chickens were PT1 and PT21. Moreover, they were also the most predominant types among the isolates (28). The PFGE patterns of *Salmonella* Enteritidis isolates from various sources by using XbaI, SpeI, or NotI restriction enzyme were highly clonal and related (29).

In 1999, there were huge foodborne-disease outbreaks caused by *Salmonella enterica* Enteritidis in Korea (Table 2). The outbreaks which resulted in more than 200 inpatients and one death, occurred by consumption of *Salmonella* Enteritidis contaminated boiled cockle and beef (30). In addition to these outbreaks, there were many other outbreaks caused by *Salmonella* Enteritidis infections in 1999. Consequently, the number of *Salmonella* Enteritidis isolates showed the highest peak in 1999 (Fig. 1).

Most of *Salmonella* Enteritidis isolates from layers were not MDR (23). However, antimicrobial resistance rate of human isolates has been increasing yearly. Resistance to clinically important antimicrobial agents such as quinolone and cephalosporine has increased in *Salmonella* Enteritidis isolates. Resistance rate to nalidixic acid was 21.6% which was higher than that of *Salmonella* Typhimurium (12.1%) (31). PT1 was the most frequent phage type among nalidixic acid resistant isolates (31). Extended spectrum β-lactamase (ESBL)-producing *Salmonella enteritidis* isolates were found. The ESBL type was TEM-52 which spread clonally and horizontally in Korea (32).

**REMARKABLE OUTBREAKS BY RARE SEROVARS OF SALMONELLA**

There were many *Salmonella* outbreaks in Korea during 1998-2007. Here, I would like to introduce four outbreaks caused by rare *Salmonella* serovars that are worthy of note in *Salmonella* human infection history in Korea (Table 2). The rare *Salmonella* serovars mentioned in this review have not been spotlighted or ranked among the most widespread *Salmonella* serovars in Korea before 1998.

**S. enterica serovar Paratyphi A (antigenic formula: 1,2,12: a: [1,5])**

*Salmonella enterica* Paratyphi A was a rare serovar until 2001. Since 2002, *Salmonella* Paratyphi A has been ranked among the top 10 *Salmonella* serovars. *Salmonella* Paratyphi A is a causative pathogen for paratyphoid fever which is a Class 1 notifiable disease in Korea like typhoid fever (35). Outbreaks by this pathogen infection were not frequent in the world but were reported in India, Nepal, and Singapore (36-39). There was a big waterborne outbreak in Busan by *Salmonella* Paratyphi A infection in early 2002 (40). More than 200 people were hospitalized. Epidemiologists found that the water-supply system was contaminated with the bacteria. Most of the isolates were resistant to nalidixic acid. The resistance mechanism was due to the point mutation in the 83rd codon of gyrA gene as found by performing allele-specific PCR and restriction fragment length polymorphism (AS-PCR-RFLP). Recent studies showed that plasmid-mediated quinolone resistances were spreading to Enterobacteriaceae, suggesting that the mechanisms of resistance to quinolone or fluoroquinolone are developing in bacteria (41).

Some patients were not cured after being treated with ciprofloxacin because of decreased susceptibility to the antimicrobial agent (42). Currently, nalidixic acid resistant *Salmonella* Paratyphi A is very common in Korea as well as India and Mid-East Asia (42, 43).

**S. enterica serovar Infantis (antigenic formula: 6,7,14: r: 1,5)**

In fact, *Salmonella* Infantis was not a rare serovar but steady during 1998-2007 (Table 1). It was the fifth common serovar in 2002 worldwide and had been commonly isolated from farm animals and their feed in Europe nations (27). In 2007, there was a huge outbreak caused by *Salmonella* Infantis infection in Jeollanam-
do, a rural site in Korea (S Kim, unpublished data). Although 49 culture proven human cases were found from the outbreak, the source of contamination was not found. The antibiotic phenotype of the isolates was susceptible to 16 antimicrobial agents which were a standard set for antimicrobial test of enteric pathogens in Korean reference laboratories. Due to the outbreak, S. enterica Infantis ranked second most-frequent among serovars in 2007.

**S. enterica serovar Othmarschen (antigenic formula: 6,7,14: g,m[1]: -)**

Many foodborne outbreaks arise in schools, parties, companies, and other gathering places today (44). Mass catering is rising because of increased provision of meals in public; so many people are exposed to possible foodborne diseases. An outbreak in 2007 caused by *Salmonella enterica* Othmarschen was such a case. In a funeral service, more than 300 mourners were exposed to contaminated foods with the pathogen and among them 72 persons became ill. The characteristics of this salmonellosis were severe diarrhea, abdominal pain, and fever. Yellow or white watery diarrhea for about 5 days with maximum 50 incidents was a typical symptom. *S. enterica* Othmarschen was isolated from those patients, food handlers, and foods containing eggs, squash, and seafood. The PFGE patterns of the outbreak isolates were all identical and indistinguishable from that of an American *S. enterica* Othmarschen isolate (45). It was very interesting that the identical clones were found in geographically distant nations even though *S. enterica* Othmarschen is such a rare *Salmonella* serovar in the world (46). All these molecular epidemiological evidences could be harvested because of the PulseNet International activities.

**SALMONELLA SEROVARS FROM OVERSEAS TRAVELERS**

For the quarantine activity and early detection of contagious diseases in airports and harbors, quarantine stations carry out rectal swabs or stool sampling from overseas travelers and crew who show fever, or notify symptoms of diarrhea, abdominal pain, vomit in questionnaire sheets and come from Thailand, and other East South-Asian nations. The 3 serovars were mostly isolated from such travelers. *S. enterica* Weltevreden has been isolated from seafood at relatively high frequency in those nations. In Thailand, *S. enterica* Weltevreden had been the most prevalent serovar among *Salmonella* isolates from 1993 to 2002 (48). Recently, there were outbreaks caused by *S. enterica* Weltevreden infections in France (49), Norway, Denmark, and Finland (50). These phenomena indicated that contaminated food trades and infected travelers between nations seriously affects the health of people in a distance and a nation’s health defense (51).

**CONCLUSIONS**

In this study, I showed epidemiologic evidence that the incidences of *S. enterica* Typhi, *S. enterica* Enteritidis, and *S. enterica Typhimurium* decreased significantly in Korea during the last 10 yr since 1998. It was a very encouraging epidemiologic trend because these serovars have been health-threatening pathogens in Korea as well as the world. However, the rates of imported cases and outbreaks caused by rare serovars increased during that time. As reviewed in this study, I would like to stress that newly emerging rare *Salmonella* serovars should be traced and investigated to control new type pathogens in the developed world.

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