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Salmonella, Enteric Fevers, and Salmonelloses

SALMONELLA bacteria are a cause of diarrhea, and less commonly enteric fever, throughout the world. They are distinct from most of the other major bacterial and viral agents of diarrhea (see chapters 11, 13, 16, and 17) in that, with the exception of the typhoid and paratyphoid bacteria, they commonly infect many species of mammals, birds, reptiles, and other animals. Human infections are frequently associated with contact with animal feces or ingestion of contaminated animal products.

Description of Pathogens and Diseases

Salmonella bacteria, and the infections they cause, are well described in a voluminous medical and veterinary literature. Only a brief summary is given here.

Identification

Salmonellosis is any infection with bacteria of the genus *Salmonella*. In man, most of the many serotypes of *Salmonella* give rise to a transient intestinal infection manifested as acute gastroenteritis with diarrhea and abdominal cramps. Some of the bacteria may transiently be found in the blood, and there may be fever, nausea, and vomiting as additional symptoms. But in infections with some serotypes, particularly *S. typhi*[1] (the causative organism of typhoid fever), *S. paratyphi* A, and *S. paratyphi* B (also designated *S. schottmuelleri*), the bacteria invade the tissues and produce a septicemia with a high temperature rather than diarrhea. This is known as enteric fever. Other salmonellae may sometimes give rise to a disease resembling typhoid, and a third syndrome—again associated mainly with particular serotypes, but especially in people with impaired resistance to infection—includes pyogenic lesions of internal organs. The salmonellae involved include *S. paratyphi* C (also designated *S. hirschfeldii*), *S. enteritidis* var. *chaco*, *S. sendai*, *S. dublin*, *S. typhimurium*, and *S. cholerae-suis*. Most other serotypes predominantly give rise to acute gastroenteritis, but many *Salmonella* infections are symptomless.

For convenience in this chapter, the septicemic syndrome seen most typically in typhoid will be referred to as enteric fever, and the primarily gastrointestinal pattern of infection as salmonellosis.

Diagnosis in cases of gastroenteritis is by isolation of the bacteria from feces or rectal swab, but in the first week of enteric fever isolation of the bacteria is more likely from blood culture, and only later are the organisms regularly found in the feces. In enteric fever there is commonly a rise in agglutinating antibodies during the course of infection. This is the basis of the Widal reaction as a method of diagnosis in the absence of bacteriological facilities. A positive reaction should be based on a fourfold or higher rise in the titer of antibodies, since past exposure or vaccination may have led to preexisting antibodies, but even then the test is unreliable because as many as half of all cases may fail to demonstrate a fourfold rise in antibody titer.

Occurrence

Salmonellae are found world-wide but the pattern of common serotypes varies considerably from region to region. Some serotypes, for instance *S. typhi* and *S. typhimurium*, are found in many parts of the world, whereas others have a very localized occurrence. In all countries a few serotypes are responsible for most reported human and animal infections, and many other serotypes are found rarely. In many developed countries *S. typhi* is now rare, and most of the cases that do occur are contracted elsewhere.

1. See the subsection "Infectious agents", below, for a note on taxonomic nomenclature.

Figure 15-1. Salmonella enteritidis *under scanning electronmicroscopy*. Colonization of the small intestine of a rat. Scale bar = 3 micrometers. (Photo: C. D. Garland, Department of Agricultural Science, University of Tasmania, Hobart, Australia)

Infectious agents

The genus *Salmonella* is a member of the family Enterobacteriaceae and is distinguished from other so-called genera of Enterobacteriaceae primarily on biochemical criteria. For example, unlike most *E. coli* (chapter 13), *Salmonella* strains typically do not ferment lactose. Salmonellae are nonsporulating, Gram-negative, motile, noncapsulate rods (0.5 by 2–4 micrometers) with peritrichous flagella (see figure 15-1). They are facultative anaerobes.

In 1934 it was agreed that each new antigenically distinct isolate of *Salmonella* should be assigned a specific Linnaean epithet, often according to the place at which it was first isolated. This led, however, to an unhelpful proliferation of names and, at the time of writing, there are around 2,000 named serotypes. Not all the serotypes listed in the 8th edition of *Bergy's Manual* (1974) have been given names, and the practice has been discontinued.

The genus *Salmonella* can be sub-divided on biochemical grounds into four subgenera; only subgenus I is common in human disease. Subgenera I, II, and IV include many named serotypes, whereas subgenus III contains only the named species. *S. arizonae*, although many unnamed serotypes have been described (table 15-1).

In an attempt to simplify the situation, Edwards and Ewing (1972) proposed that only three species of *Salmonella* should be recognized: *S. typhi* (one serotype), *S. cholerae-suis* (one serotype), and *S. enteritidis* (around 2,000 serotypes). Following this schema, all serotypes except *S. typhi* and *S. cholerae-suis* should be designated *S. enteritidis* serotype ——— (table 15-1). *S. arizonae* was considered to be a separate genus (*Arizona*) rather than a subgenus of *Salmonella*. A similar reform was proposed by Cowan and Steel (1974), save that *S.* [illegible] was the name given to the aggregate species. This move towards a reduced number of species is reflected in the approved list of bacterial names (Skerman, McGowan and Sneath 1980): *S. cholerae-suis* and *S. typhi* retain their specific status; *S. typhimurium* is retained because of the frequency of its occurrence; *S. enteritidis* is the aggregate species; and *S. arizonae* consists of members of subgenus III.

In this book we have adhered to the system of nomenclature described in the 8th edition of *Bergy's Manual*, except that we have continued to use the names *S. paratyphi* B and *S. paratyphi* C, since they are more familiar to medical microbiologists.

Reservoirs

Salmonellae are primarily pathogens of animals, which provide important reservoirs for the infections of man other than enteric fever. Person-to-person transmission also occurs, and the relative importance

Table 15-1. *Variations in the nomenclature of some important types of* Salmonella

Bergy's Manual (8th ed., 1974)	Edwards and Ewing (1972)
Subgenus I (numerous named serotypes)	
S. typhi	*S. typhi*
S. cholerae-suis	*S. cholerae-suis*
S. enteritidis	*S. enteritidis* serotype *enteritidis*
S. [illegible]	*S. enteritidis* serotype [illegible]
S. hirschfeldii (*paratyphi* C)	*S. enteritidis* serotype *paratyphi* C
S. paratyphi A	*S. enteritidis* serotype *paratyphi* A
S. schottmuelleri (*paratyphi* B)	*S. enteritidis* serotype *paratyphi* B
S. typhimurium	*S. enteritidis* serotype *typhimurium*
Subgenus II (numerous named serotypes)	
S. salamae	*S. enteritidis* serotype *dar es salaam*
Subgenus III (numerous unnamed serotypes)	
S. arizonae	*Arizona hinshawii*
Subgenus IV (numerous named serotypes)	
S. houtenae	*S. enteritidis* serotype *houten*

of the human and nonhuman reservoirs depends upon the dietary, agricultural, and sanitary situation in a particular community.

A few *Salmonella* serotypes are almost species specific. Most importantly, *S. typhi* is a pathogen of man, and therefore the source of infection is the human case or carrier. *S. paratyphi* A, B, and C and *S. sendai* also have their reservoirs primarily in man, although infections of other animals have been reported.

Salmonellae are commonly isolated from poultry, especially chickens, turkeys, and ducks (Goyal and Singh 1970), and from livestock such as pigs, cows, sheep, and horses (Baker 1970). The prevalence of infection among these poultry and farm animals is especially high under some systems of intensive indoor farming or where contaminated foodstuffs (especially fish meal and bone meal) are used (Al-Hindawi and Taha 1979; Joint Working Party 1965; Lee 1974). In the UK and the USA between 15 and 50 percent of dressed poultry in retail stores may be contaminated by *Salmonella*. Other domestic animals and pets (for instance, cats, dogs, mice, guinea pigs, and hamsters) are frequently infected. Both rats and mice that live in close proximity to human communities may be infected. Their carrier rate may sometimes rise above 10 percent, which is in contrast to the very low prevalence observed in surveys of other wild mammals that do not appear to constitute a reservoir of infection for man or domestic animals (Jones and Twigg 1976).

Gulls, pigeons, and doves have been implicated as major reservoirs of salmonellae, but other wild birds are more rarely infected. Jones, Smith and Watson (1978) isolated salmonellae from 62 percent of samples of gull droppings from a large gullery in northwestern England, whereas Plant (1978) isolated *Salmonella* from only 0.17 percent (1/599) of wild birds examined at two sewage treatment works in southeastern England. Hussong and others (1979) failed to isolate *Salmonella* from forty-four migratory waterfowl wintering at Chesapeake Bay (USA). The infection of gulls may be due to their habit of scavenging for food at rubbish tips and sewage treatment works. Gulls have been implicated in the contamination of water reservoirs by salmonellae, and in the spread of salmonellae from refuse tips to cattle on nearby pastures (Williams and others 1977).

Many species of reptiles and other cold-blooded animals are carriers of *Salmonella* (Goyal and Singh 1970). This is of epidemiological importance in cases where these animals have close contact with man. Particular concern has been expressed over the high prevalence of *Salmonella* excretion among pet turtles, terrapins, tortoises, frogs, and snails (Bartlett and Trust 1976; Bartlett, Trust and Lior 1977; Lamm and others 1972). It has been suggested that the decrease in human infection with *S. java*, *S. litchfield*, and *S. urbana* in Canada since 1975 has been due to the government prohibition of the importation of turtles, which were popular as pets (D'Aoust and Lior 1978). In 1975 the interstate transport of pet turtles became illegal in the USA (MMWR 1975). Among the arthropods, fleas, ticks, lice, cockroaches, and houseflies may harbor salmonellae.

Nonhuman isolations of *Salmonella* in England and Wales during 1968–74 were reviewed by Sojka and others (1977). Of a total of 23,609 incidents of *Salmonella* infection, 86 percent were in cattle, 7.4 percent in poultry and other birds, 2.9 percent in sheep, 2.4 percent in pigs, and 1.3 percent in other species, especially horses, dogs, mink, guinea pigs, and cats. These figures do not reflect the relative incidence of salmonellosis among various species but rather the commercial importance of certain farm animals and the policies of veterinary laboratories. Although 153 serotypes were isolated, 88 percent of incidents were due to *S. dublin* and *S. typhimurium*. Other common serotypes, in decreasing order of importance, were *S. cholerae-suis* (almost entirely restricted to pigs), *S. abortus-ovis* (entirely restricted to sheep), *S. newport*, *S. agona*, *S. virchow*, *S. anatum*, *S. enteritidis*, and *S. montevideo*.

Transmission

The transmission of typhoid fever was partly elucidated before the bacteriological era by William Budd (1856, 1873). He showed that it was spread by contagion, that infective material was excreted in the feces, and that the disease could be spread by the tainted hands of those who waited on the sick or by contamination of water and milk.

Salmonella transmission takes place when the infected feces of man or animal are ingested by a susceptible person. In the case of typhoid fever, transmission is only from human feces or urine to mouth. This fecal-oral (or occasionally urinary-oral) transmission may be direct where personal cleanliness is poor, or it may be via contaminated food or water. Large numbers of bacteria are excreted, and high infective doses are required to infect most persons and animals.

Cases of salmonellosis or enteric fever may excrete up to 10^{10} *Salmonella* per gram of feces. Unlike other bacterial enteric infections, asymptomatic carriers may also excrete very high concentrations. Thomson (1954, 1955) studied *Salmonella* excretion by cases and

carriers. Twenty cases of salmonellosis excreted 2.5×10^5–1×10^9 *Salmonella* per gram of feces, and 6 *Salmonella* carriers excreted 1×10^4–5×10^7. Seven cases of paratyphoid B excreted 1×10^4–5×10^9 *S. paratyphi* B per gram, and 12 paratyphoid B carriers (known duration 1–20 years) excreted 5×10^5–1.2×10^{10} *S. paratyphi* B per gram. Half of the paratyphoid B carriers excreted more *Salmonella* per gram than *E. coli*. Eight typhoid carriers (known duration 3–12 years) excreted 5×10^5–4.5×10^7 *S. typhi* per gram. Merselis and others (1964) found that thirteen typhoid carriers (known duration 4–41 years) excreted 10^4–10^{11} *S. typhi* per gram of feces.

The infective dose for typhoid and other salmonellae is high in healthy adults. In a major series of studies, McCullough and Eisele (1951*a*, 1951*b*, 1953*a*, 1953*b*) fed eggnog containing various doses of *S. anatum*, *S. bareilly*, *S. derby*, *S. meleagridis*, *S. newport*, and *S. pullorum* to healthy adult prisoners. The doses needed to produce clinical symptoms in about half of the volunteers were between 9×10^5 for *S. anatum* and 4×10^9 for *S. pullorum*. No clinical disease was produced with less than 1.3×10^5 organisms, but infection and a temporary carrier state was established with only 1.2×10^4 *S. anatum*.

Hornick and others (1970) reported a median infective dose (ID_{50}) of *S. typhi* for healthy adult male volunteers of 10^7 organisms in 30 milliliters of milk. No disease was produced in any of 14 volunteers by 10^3 organisms. In subsequent tests, five different *S. typhi* strains were fed to volunteers at a dose of 10^7 and induced disease in between 21 and 56 percent and infection or disease in between 60 and 93 percent. As with all ID_{50} data, it must be remembered that the attack rate in most common source outbreaks is very much less than 50 percent.

Although the ID_{50}s for salmonellae are high, some individuals may be infected and made ill by much smaller doses. D'Aoust and Pivnick (1976) reported outbreaks caused by 15,000 *S. cubana* in carmine dye capsules, by 1,000 *Salmonella* in ice-cream, and by chocolates containing only 100 *S. eastbourne* per gram. In children with abnormal gastrointestinal tracts due to cystic fibrosis, Lipson (1976) reported infection with *S. schwarzengrund* from probably <100 bacteria.

The infective dose of *Salmonella* for healthy cattle is high, although young calves can be readily infected, and sometimes killed, by a dose of only 10^4 *S. typhimurium* (Deans Rankin and Taylor 1966; Robinson and Loken 1968). Hall and Jones (1978) detected no infection in four cows each fed 1 liter of raw sludge per day containing 10^2–10^5 *Salmonella* per liter. In another group of cows fed 1 liter of sterilized sludge per day to which 10^5 *S. dublin* per liter were added, three out of four were infected but none developed diarrhea. The authors pointed out that these exposures (10^5 *Salmonella* per day) are higher than would be expected under normal farming conditions in which cattle graze on pasture spread with sludge. However, some cattle, like some humans, may be infected by fewer organisms if they are for any reason especially susceptible. Aitken and others (1976) showed that cows were more susceptible to *S. dublin* if they were also infected by the liver fluke, *Fasciola hepatica* (see chapter 27).

Although fecal-oral transmission of salmonellae is the norm for man and animals, the organism may be spread by other routes. Garg and Sharma (1979) isolated salmonellae from the nasal passages of 2 calves and 2 piglets out of 395 young farm animals in India. Crozier and Woodward (1962) found that chimpanzees could be given typhoid fever by the respiratory route with a dose of 10^8 *S. typhi*, compared with a dose of 10^{11} organisms needed for successful oral infection. Darlow, Bale and Carter (1961) found that 57 percent (34 of 60) of mice inhaling 5×10^4 *S. typhimurium* died in 4 weeks, and all except 1.7 percent (1 of 60) were infected. By contrast, when mice ingested the same dose only 1.7 percent (1 of 60) died in 4 weeks, and 75 percent (45 of 60) were not infected. Morse, Myhrom and Greenwood (1976) cited studies from the USSR showing that *Salmonella* infection of sheep and cows by aerosols was possible with 27 percent of the dose required for infection by the oral route. Moore (1957) reported that the ID_{50} of *S. enteritidis* for guinea pigs was 10^9 when fed by mouth and only 10^2 when placed on the eye (see also Duguid, Darekar and Wheater 1976). Thus, although fecal-oral transmission is the norm, fecal-nasal, fecal-ocular, and nasal-nasal routes are also theoretical possibilities (see also Wray and Sojka 1977).

Incubation period

In typhoid fever the incubation period is usually between 5 and 21 days, whereas that of paratyphoid tends to be less. Typically, the first symptom to appear is fever due to bacteremia; only later, usually in the second and third weeks of the illness, do bacteria invade the intestine from the blood stream in increasing numbers to be detected in fecal specimens. The typical symptoms of enteric fever may be preceded by acute gastroenteritis. Most other salmonelloses have considerably shorter incubation periods, generally between 8 and 36 hours, typically followed by headache, nausea, vomiting, abdominal pain, fever,

and diarrhea. Incubation periods are inversely proportional to infecting dose.

Period of communicability

In typhoid and other enteric fevers, the *Salmonella* species responsible appears in the excreta late in the first week of illness or thereafter. During the first few weeks of convalescence the proportion of patients excreting typhoid bacilli falls, until about 3 months after the onset of illness less than 10 percent of clinical cases continue to excrete *S. typhi* in their feces. Two to five percent may become chronic carriers and excrete the pathogen for over 1 year. A small proportion remain carriers for 10, 20, 30 years or a lifetime. As noted above, chronic typhoid carriers may excrete 10^4–10^{11} *S. typhi* per gram of feces and therefore constitute a major source of infection within a community. The gall bladder is the organ that typically remains infected, and gall stones may predispose towards the carrier state.

Some typhoid and paratyphoid cases pass *Salmonella* in their urine. Chronic urinary carriers of typhoid and paratyphoid are, however, much less common than fecal carriers, except in countries where urinary schistosomiasis (see chapter 32) is common. In such areas many urinary carriers of enteric fever bacilli are found, and the prevalence may be as high as 3 percent of typhoid convalescents. Hathout and others (1966) found that, among forty-nine males in Egypt with *Schistosoma haematobium* infection who contracted typhoid or paratyphoid A, 47 percent were still excreting *Salmonella* in their urine 1 year after the diagnosis of enteric fever. (Among those with *Schistosoma mansoni* infection, typhoid and some other *Salmonella* infections give rise to severe illness over many months.)

In other salmonelloses, the bacterium appears in the feces concurrently with the diarrhea and for a few days to a few weeks thereafter. A temporary carrier state for around 2 months occurs in 5–10 percent of convalescents, but chronic carriers (over 1 year) are rare (less than 1 percent of cases).

In many poor communities the incidence of *Salmonella* infection is high, and many infections do not produce disease. Therefore, at any time 1–5 percent of the healthy population will be excreting *Salmonella* bacteria. Infection prevalences are higher among workers who come into daily contact with potentially infected people, animals, or animal products. The carrier rates for salmonellae among healthy adults in Baghdad (Iraq) were 1.9 percent of random persons, 2.9 percent of buffalo owners, 3.2 percent of hospital personnel, 4.2 percent of slaughterhouse workers, 6.2 percent of poultry processers, and 8.6 percent of food handlers (Al-Ani and Saadallah 1979). No salmonellae were isolated from the urine of 905 people. Al-Dulaimy and Al-Allaf (1979) reported a 1.2 percent (23 of 2,000) prevalence of *Salmonella* excretion in Mosul (Iraq). Becerril, Bessudo and González Cortés (1979) found that 13 percent (110 of 850) of food handlers, and 5 percent (73 of 1,527) of the general population, were *Salmonella* carriers in Mexico City (Mexico). Various studies in South Africa showed *Salmonella* excretion in 2–6 percent of hospitalized children 0–2 years old without diarrhea and in 0–12 percent of healthy schoolchildren (Koornhof and others 1979).

In contrast, Gordon and others (1961) reported an average prevalence of *Salmonella* excretion among about 10,000 healthy children under 10 years old in highland and lowland villages in Guatemala of only 0.2 percent. Other data showed the *Salmonella* was not an important cause of diarrhea in that age group in those villages (Pierce and others 1961). Similarly, studies in the USA in the 1950s showed that *Salmonella* was rarely excreted by healthy children from lower socioeconomic groups. Among preschool children in Kentucky the prevalence of *Salmonella* excretion was 0.2 percent, whereas that for *Shigella* was 3.1 percent (Schliessmann and others 1958), and among children under 10 years old in California the prevalence of *Salmonella* excretion was 0.4 percent, whereas 4.6 percent excreted *Shigella* (Watt and others 1953).

Resistance

For the enteric fevers and the salmonelloses, individuals who have gastric hypochlorhydria, who are sick or who are on antibiotic therapy are especially susceptible. Indeed, *Salmonella* infections frequently select debilitated individuals, and hospital outbreaks are often especially serious.

An attack of typhoid fever confers some immunity to reinfection, usually life-long, but second and third attacks have been reported. Although antibodies to several *Salmonella* antigens are readily detectable in serum, they correlate poorly with resistance. In some areas where typhoid is endemic, about 5 percent of cases occur in those under 5 years old and perhaps 4 percent are fatal. About 45 percent of cases occur in the 5–19 age group, and fatalities range from 2 to 6 percent. Thereafter, from ages 20 to above 60 years, the incidence rate falls progressively, but the case fatality rate increases to over 30 percent in cases over 60 years old. It may be inferred that many in the community become immune during childhood and adolescent

exposure, though proof is lacking. The outcome of challenge depends also on dose size and the virulence of the strain of *S. typhi*.

Among the other salmonelloses, attack rates are highest in the age group under 5 years and especially high in infants under 1 year. Significant immunity is not conferred; in any case, the next challenge is likely to be from a different serotype.

Epidemiology

The salmonelloses have a markedly seasonal incidence, peaking in the warmest months along with all diarrheal diseases. This may be in the warm-wet season, as in Lesotho and India, or in the warm-dry season, as in such countries as the UK, El Salvador, Guatemala, and the USA. In those countries where it is endemic, typhoid may peak at the same time or a few weeks later. A late summer or autumn high incidence of typhoid, for example, follows the summer peak salmonellosis incidence in Lesotho, India, and Tunisia.

Although salmonelloses constitute an important cause of diarrhea, especially foodborne diarrhea, in many developed countries and are also of major veterinary importance in some areas, *Salmonella* are responsible for only a very small proportion of infant and childhood diarrhea in some developing countries. In these latter places *Salmonella* are completely overshadowed as a cause of childhood diarrhea by enterotoxigenic *E. coli* (chapter 13), rotavirus (chapter 11), and sometimes also *Shigella* (chapter 16) and *Campylobacter* (chapter 12). For example, *Salmonella* was associated with only 1 percent of childhood (1–5 years) diarrhea in Guatemala (Pierce and others 1961), 0–1.7 percent of childhood diarrhea in Panama (Kourany and Vasquez 1969), less than 1 percent of childhood (under 5 years) diarrhea in Bangladesh (Black and others 1979), and 1.4 percent of diarrhea cases among young children in a Gambian village (Barrell and Rowland 1979). In contrast, studies on both black and white children (0–2 years) in South Africa showed *Salmonella* excretion in between 6 and 17 percent of diarrhea cases and 2–6 percent of healthy controls (Koornhof and others 1979). This contrast might suggest that *Salmonella* diarrhea becomes more prominent as economic conditions improve, since the South African children studied were probably more urban and wealthier than the Guatemalan or Bangladeshi groups. However, this economic distinction falls down in the case of Panama, and it was shown in the 1940s and 1950s that *Salmonella* were not a major cause of childhood diarrhea in California (Watt and others 1953), Kentucky (Schliessmann and others 1958), and Texas (Watt and Lindsay 1948) in the USA. It may be concluded that there are major unexplained differences in the prominence of *Salmonella* diarrheas in children (as there are with all individual agents of diarrhea), but that in many poor communities salmonellae do not make a major contribution to overall pediatric diarrheal morbidity and mortality.

Typhoid is commonly endemic in Africa and is especially prominent in some upland areas. Characteristic features of endemic highland typhoid are illustrated by the situation in Lesotho described by Feachem and others (1978). At one major hospital (St. Joseph's Hospital, Roma) during 1971–74, typhoid accounted for 1 percent of all attendances (inpatients and outpatients) and 5 percent of all admissions (inpatients). The highest age-specific incidence occurred in the 15–24 age group. The fatality rate among all hospitalized typhoid cases was about 6 percent, whereas in patients over 35 years it was 13 percent. Typhoid in Lesotho had a pronounced seasonal pattern, with major outbreaks occurring every 2 or 3 years during March–April; temperature and rainfall peaked during November–March, and all diarrhea reportings peaked during December–February.

The maximum typhoid incidence among the 10–30 age group is typical of endemic typhoid and has been reported from, for instance, Antigua (Uttley 1960), Dominica (Grell 1979), India (Mathur and Sharma 1971), Jamaica (Miller, Grant and Irvine 1961), and Tunisia (Miled, Zribi and Ben Rachid 1973). Ashcroft (1962) reported a peak incidence of typhoid in Guyana during 1956–60 among slightly younger children (5–14 years) and suggested that the age of peak incidence might rise as sanitation improved and children were less frequently immunized by mild or asymptomatic infections.

Typhoid fever in the UK has been reviewed (Public Health Laboratory Service Standing Sub-Committee on the Bacteriological Examination of Water Supplies 1978). A major waterborne outbreak (341 cases and 43 deaths) occurred in Croydon in 1937 (Holden 1939). Between 1941 and 1970, several small outbreaks were traced to the ingestion of well or stream waters contaminated by chronic typhoid carriers. Other outbreaks were caused by contaminated milk, oysters, and imported corned beef. A major typhoid outbreak in Aberdeen (Scotland) in 1964, with 507 cases and 3 deaths, was caused by a single tin of contaminated corned beef from South America. The meat was sliced and sold at a supermarket, and about 50 people were infected from this corned beef. The remainder were

infected by eating other cold meats that had been sliced by the same machine in the same supermarket (Howie 1968; Walker 1965). Since about 1955 the annual incidence of typhoid in Britain has been very low. During 1975 and 1976, 419 cases were reported in England, Wales, and Northern Ireland, and 85 percent of these were infected abroad. By contrast, only 33 percent of 1,418 *S. typhi* infections in the USA during 1967–72 were associated with foreign travel, although there was a markedly rising trend in this percentage (17 percent in 1967, 46 percent in 1972) (Rice, Baine and Gangarosa 1977). Over this period, indigenous cases of *S. typhi* infection were mainly between 5 and 30 years old, cases associated with travel were mainly in those aged 10–30 years, and known typhoid carriers were predominantly elderly women.

Human salmonelloses in the USA during 1963–67 were reviewed by Aserkoff, Schroeder and Brachman (1970). About 20,000 cases were bacteriologically confirmed and reported annually. The annual peaks occurred in July–October, and the age-specific incidence was by far the highest in those under 4 years old. The reported incidence in Hawaii was nearly 10 times the national average in each year, and this was attributed to the use of pig intestines in soups and stews by certain ethnic groups. There were 180 epidemics, comprising 16,772 cases, during 1963–67 , and 156 of these were due to a common vehicle of infection—especially eggs, egg products, and turkey. Less commonly chicken, beef, pork, milk, pet chickens, pet ducks, and pet turtles were implicated. The largest number of epidemics were domestic, but the largest epidemics were those associated with banquets and schools. During the period *S. typhimurium* was by far the most commonly identified serotype, constituting 30 percent of all human isolations and 17 percent of all nonhuman isolations. Poultry and poultry products accounted for half of all nonhuman isolations of *Salmonella*. Some serotypes exhibited a considerable degree of species specificity: *S. pullorum* and *S. gallinarum* in fowl, *S. abortus-ovis* in sheep, and, to a lesser degree of specificity, *S. cholerae-suis* in pigs and *S. dublin* in cattle.

Food-borne outbreaks of typhoid and other salmonelloses are commonly reported. Sometimes they are due to contaminated raw materials, such as poultry, tinned meat, or eggs, while in other cases the food is contaminated by a food handler. Carrier rates for *Salmonella* are typically higher among food handlers than among the general population and there is a close relationship between contaminated food and infected food handlers which places the consumer of the food at risk.

"Food poisoning" of bacterial origin may be due to several different organisms, including *Staphylococcus aureus*, *Clostridium botulinum* and *Cl. perfringens*, and *Vibrio parahaemolyticus*. The bacteria multiply in the contaminated food and may produce exotoxins in the case of *Staphylococcus* and *Clostridium*, when symptoms result from ingestion of the preformed toxin. In infective food poisoning the gastrointestinal symptoms follow proliferation of the bacteria in the human intestine. When the pattern of food preparation provides opportunities for salmonellae to multiply, infection is naturally more likely.

In developed countries the primary vehicles of foodborne salmonelloses depend on dietary habits and particularly on the main source of animal protein. Poultry and egg products are the most frequently implicated vehicles in many countries. In developing countries, where less animal protein is eaten and animals are not typically raised by "factory farming" methods, the contamination of food by infected food handlers may be more important than the ingestion of naturally contaminated animal products.

Food contamination is believed to be an important factor contributing to the high prevalences of *Salmonella* excretion among African schoolchildren in South Africa (table 15-2). Surveys of food in Pretoria and Soweto showed that 48 percent of tripe samples, 29 percent of intestines, 40 percent of pork sausage, 64 percent of minced beef samples, 20 percent of chicken carcasses, and 16 percent of biltong samples (dried meat eaten raw) were contaminated by salmonellae (Prior and Badenhorst 1974; Richardson, Burnett and Koorhof 1968). Bokkenheuser and Richardson (1959) found that 4.3 percent (67 of 1,565) of food handlers employed at nineteen gold mines in the Transvaal were infected by *Salmonella*.

An outbreak of sixty-nine typhoid cases after a party in Cape Town (South Africa) in 1978 was traced to the main caterer who had prepared and stored chickens for the party under grossly unhygienic conditions (Popkiss 1980). The main caterer was found to excrete *S. typhi* of the same phage type as those isolated from the cases. On May 5–7, 1977, 545 university students at Trujillo (Peru) were hospitalized with acute gastrointestinal symptoms due to *S. thompson* (Gunn and Loarte 1979). The outbreak was traced to the university dining hall, and 93 percent of the students who regularly ate there became ill. The implicated vehicle was sardine-mayonnaise salad. Barrell and Rowland (1979) found that *Salmonella* were associated with 1.4 percent of diarrhea cases among young children in a Gambian village and that 4.7 percent of infant food samples were contaminated by salmonellae.

Waterborne outbreaks have been chiefly associated

with *S. typhi* and much less frequently with *S. paratyphi* or other *Salmonella* serotypes. The evidence implicating a water supply is usually circumstantial, and the causative organism is rarely isolated because the pollution that gives rise to an epidemic is often temporary or intermittent. Waterborne outbreaks due to gross contamination are usually characterized by an explosive onset. The majority of cases develop over a period of a few days, and these may be followed by a secondary crop of contact cases. Sometimes the outbreaks may happen as a series of scattered cases occurring over a considerable period of time, and this may be due to a lower and intermittent contamination of the supply.

Waterborne disease outbreaks in the USA during 1971–73 have been reviewed (Hughes and others 1975; Merson and others 1974). Typhoid fever was the cause of 2.5 percent (217 of 8,537) of known cases and 4.2 percent (3 of 71) of known outbreaks, while other salmonelloses were the cause of 0.04 percent (3 of 8,537) of cases and 1.4 percent (1 of 71) of outbreaks. Large, and probably waterborne, outbreaks of diarrhea due to *S. typhimurium* occurred in Riverside (California) in 1965 (Greenberg and Ongerth 1966) and in Suffolk County (New York State) in 1976 (Zaki and others 1979).

Feldman and others (1974) described an outbreak of typhoid (225 cases and no deaths) at a migrant farm labor camp in Florida (USA) in 1973. A case-control study linked the infection to the camp water supply, which was pumped from two wells. The water supply was chlorinated but with inadequate contact time during peak water demand, and the chlorinator had a history of malfunction. High coliform counts were obtained from the camp water on several occasions, and it was discovered that surface drainage and sewage might gain access to the wells. It was estimated that the average infecting dose in the outbreak was 10^3–10^5 *S. typhi* and that such doses could have been administered by the water supply if a case or carrier with heavily contaminated stools (say 10^{11} *S. typhi* per gram) had defecated in the pump house, around the well, or into the water storage tank.

In the notorious typhoid outbreak in Zermatt (Switzerland) in March 1963 (Bernard 1965) there were 437 cases and 3 deaths. Some evidence implicated the inadequately chlorinated water supply, which may have become contaminated by a carrier working on the catchment or by leakage of sewage into the water system.

Mendis and others (1976) described a very persistent outbreak of diarrhea due to *S. bareilly* in the maternity hospital in Colombo (Sri Lanka). Initial person-to-person spread led to the contamination of the hospital water system as a result of the suction of wastewater into water mains. *S. bareilly* became established in the water system and appeared to grow in the interior of taps and on the walls of water tanks at the water level. The contaminated water spread the infection throughout the hospital during 1968–69 so that. during January–March 1969, 48 percent (20 of 42) of hospital staff were infected. The rectification of certain plumbing defects, and the chlorination of the mains and tanks, led to the disappearance of *S. bareilly* from the water system, and the outbreak subsided.

A report from Trinidad (West Indies) described forty-eight cases of acute diarrhea among eighty-eight children and adults attending a church camp (Koplan and others 1978). *S. arechevalata* (a rare serotype) was isolated from cases, asymptomatic persons, two food items (a fish dish and stewed peas that had been prepared with roof-collected rain water), and from roof-collected rain water. The roof used for rainwater catchment was overhung by trees in which mocking birds, wrens, and doves nested and rested, and the roof was covered with dried and fresh bird feces. This may be the only account of a salmonellosis outbreak due to contaminated roof-collected rain water, but small outbreaks, in individual households, having roofs contaminated by bird droppings, may be commonplace where such rain water is a widely used water source.

The complex interactions possible between human and nonhuman reservoirs, and waterborne and foodborne transmission are illustrated by an outbreak of *S. paratyphi* B infection in five cows and ninety people in North Yorkshire (England) in 1970. The probable chain of events was that a chronic human carrier contaminated a stream, which infected a herd of cows, which infected farm workers—one of whom contaminated the water supply serving several villages (George and others 1972; Harbourne and others 1972). The failure of two types of sewage treatment to remove *S. paratyphi* B from sewage is also suggested by this study. The original chronic carrier contaminated the stream by way of sewage effluent from a "conventional" treatment plant (probably trickling filters), and the farm worker contaminated the village water supply by way of septic tank effluent from his cottage.

There is extensive evidence of direct person-to-person spread of salmonelloses and enteric fevers under conditions of poor hygiene, in crowded institutions, or simply in a normal family setting. Budd (1856) described an outbreak of typhoid at a military school in France in 1826 and reported that among twenty-nine cases nursed at home, eight were known to

have transmitted the disease to persons attending them. Rosenstein (1967) studied the family contacts of twenty-eight sporadic index cases of *Salmonella* diarrhea in Rhode Island (USA). Thirty-five percent (42 of 121) of family contacts were found to be infected by *Salmonella* of the same serotype as the index case, and 55 percent (23 of 42) of infected contacts had diarrhea. Sixty-one percent (17 of 28) of the families of the index cases had at least one infected contact. Among all family contacts, children were more likely to be infected and, if infected, were more likely to be ill. Baine and others (1973) reviewed *Salmonella* outbreaks in institutions in the USA between 1963 and 1972. Person-to-person transmission (as opposed to common vehicle outbreaks associated with contaminated food, water, or pharmaceutical products) was the major mode of transmisson in nurseries and pediatric wards and was also important in outbreaks among hospitalized adults. Person-to-person spread was implicated in 61 percent (46 of 76) of institutional outbreaks in which the mode of transmission was elucidated.

S. anatum could be recovered from artificially contaminated fingertips for over 180 minutes when the initial inoculation was 530 organisms per fingertip, and for 90 minutes when the initial inoculum was only 36 organisms (Pether and Gilbert 1971). In the same study, 10^6 *S. anatum* per fingertip were not removed completely by hand washing (a 15-second wash with soap and running warm water and drying on paper towels), and unwashed contaminated fingertips could readily contaminate corned beef and cooked ham.

Control Measures

The control of enteric fevers, which are primarily infections of man, has been achieved in many wealthy countries. The control of other salmonelloses has proved impossible due to their large and widespread animal reservoirs.

Individual

The widespread resistance of *Salmonella* species, including *S. typhi*, to many antibiotics is a serious problem brought about by excessive curative and prophylactic use of antibiotics in both human and veterinary medicine. Antibiotic therapy in man is not beneficial in uncomplicated *Salmonella* diarrhea, since it does not accelerate recovery and may prolong the period of *Salmonella* excretion during convalescence. Chemotherapy, usually with chloramphenicol, ampicillin, or cotrimoxazole, is required for the treatment of enteric fevers. Prophylactic use of antibiotics for man and farm animals (a very common practice, although now illegal in some countries) is generally condemned. Antibiotic resistance in *Salmonella* is usually caused by transmissible plasmids conferring multiple antibiotic resistance. Thus, misuse of a single antibiotic can give rise to multiresistant strains. Some major epidemics of enteric fever and salmonellosis, caused by multiresistant *Salmonella* strains (especially of *S. typhi*, *S. typhimurium*, and *S. wien*), have occurred recently in man and animals.

Typhoid fever is the only disease considered in this book, except poliomyelitis, for which there is a widely used and moderately effective vaccine, and vaccination can play a role in typhoid control. Inactivated whole-cell vaccines against typhoid are given in two intradermal or intramuscular doses, with an interval of 2–4 weeks, and provide 70–85 percent protection for a period of 3–4 years. To maintain immunity at a high level, revaccination is required every 2–3 years. It has been common practice to use a TAB vaccine, which combines *S. typhi* (T) with *S. paratyphi* A and B (AB) organisms and purports to protect against all three enteric fevers. The use of TAB vaccines is, however, officially discouraged (WHO 1979) because the addition of the *S. paratyphi* organisms requires a reduction in the number of *S. typhi* present below the 10^9 organisms necessary to produce significant antityphoid immunity. It is therefore recommended that antityphoid vaccine alone should be used. A new live oral typhoid vaccine has been tested in Egypt and is currently (1982) undergoing further field trials in Chile.

Carrier surveillance and control

Eradication of typhoid from the community requires the prevention or cure of the carrier state. No uniformly successful method of curing this condition has yet been devised, although a high proportion of both typhoid and paratyphoid fecal carriers can be freed from their infection by surgical removal of the gall bladder and concomitant ampicillin therapy. The names of all chronic carriers should be registered with the local health authority, occupational restrictions imposed, and careful instructions given on strict personal hygiene to avoid infecting others. Besides being prevented from handling food for others, carriers should not be employed on or admitted to water works.

Outbreaks of enteric fever can seldom be traced to the known chronic carriers, and it is the unknown carrier that is the main danger. It is valuable to

question new employees in the catering or water supply industries to establish whether they have previously suffered from enteric fever and to examine them by standard serological and cultural procedures. These tests should be repeated at regular intervals and on individuals returning to work after any enteric infection. Excreters of pathogenic organisms identified by these procedures should be legally precluded from working in food handling and water supply until it can be demonstrated that they are completely cured.

Environmental

Wide-ranging economic and sanitary changes in Europe and North America over the last century, combined with methods to prevent, detect and cure typhoid carriers, have caused a very great reduction in the incidence of the enteric fevers and have certainly removed them from the list of major infectious diseases. Whether simple water and sanitary improvements applied to poor communities in developing countries can have a measurable impact upon typhoid and paratyphoid incidence remains uncertain. It is probable that a combination of improved water supply, adequate excreta disposal facilities, strenuous health education programs, and national systems for identifying, controlling and treating carriers are required.

The control of salmonelloses will inevitably prove much more difficult because of the widespread animal reservoir. In the developed countries salmonelloses may have decreased since the last century (although this is not known), but they remain a major medical and veterinary problem. The trends in diet and farming practice that accompany economic development will tend to increase the salmonellosis problem. Improved water supply and excreta disposal, coupled with tighter control of the catering industry and improved food hygiene at home, will have a beneficial effect.

Cvjetanović, Grab and Uemera (1971, 1978) constructed a mathematical model of endemic typhoid to predict the impact of immunization and latrine construction. The values fed into the model were those corresponding to the demographic, medical, and economic situation in Western Samoa. As with their cholera model (see chapter 17), Cvjetanović and his coworkers overestimated the impact, and greatly underestimated the costs, of latrine construction. They excluded all labor and material costs, which were met by the householders, and deemed the cost of a latrine to be only the costs borne by the government in the provision of supervision and technical assistance. This government input was valued at US$3.15 per latrine or US$0.50 per capita served. The value of benefits was also underestimated, however, since it was taken to be the costs of hospital treatment and lost wages for typhoid only and thus excluded the cost of death (future working days forgone) and the many other health benefits that might result from latrine construction. The model showed immunization at 5-year intervals to be greatly more cost effective than was the case for cholera (chapter 17), because of the greater effectiveness (80 percent assumed) of the typhoid vaccine and its longer period of protection (5 years assumed). The costs of five-yearly vaccinations of the population (assuming 75 percent coverage), and latrine provision for the entire population over a 10-year period, were found to be very similar, and the two strategies were predicted to have a similar effect on typhoid incidence—reducing it from around 7.2 cases to 2.5 cases per 10,000 people per year over a 30-year period. However, if five-yearly vaccinations were discontinued, incidence would increase sharply—whereas the 10-year latrine program, and a low level follow up program to cope with population increase, would continue to bring the incidence down. With vaccination alone, cumulative benefits would exceed cumulative costs after 10 years; with latrine construction alone this point would be reached after 20 years, whereas with both vaccination and latrine construction benefits would not exceed costs until after 25 years. Cvjetanović and his colleagues concluded, by using both the model and actual data on typhoid incidence in England and Wales and the USA, that continuing improvements in sanitary and economic conditions would lead eventually to the eradication of typhoid (except for imported cases) without recourse to mass vaccination. Models of this type are useful in focussing attention on the dynamics of a disease and the potential impact of various interventions. The cost-benefit analyses could be greatly improved by reconsidering some of the epidemiological variables, especially the effect of sanitation on transmission, and by incorporating more rigorous economic costing and discounting techniques.

Studies in Lesotho (Feachem and others 1978) suggested that the periodic summer typhoid outbreaks, and the sporadic cases between outbreaks, were not associated with rainfall or increased water pollution and that villages with improved water supplies providing water of good quality were not protected against these outbreaks. It was also shown that the temporal and spatial patterns of two typhoid outbreaks studied in detail were not suggestive of waterborne transmission. It was concluded that much typhoid transmission in Lesotho was nonwaterborne

and that wide ranging improvements in waste disposal, cleanliness, food hygiene, and water supply were necessary to reduce typhoid incidence. Identical conclusions were reached for all diarrheal diseases in Lesotho, an unknown proportion of which are due to *Salmonella* infections.

It is illuminating to review a series of studies on salmonellosis and shigellosis among healthy African schoolchildren in the Transvaal (Republic of South Africa; Bokkenheuser and Richardson 1960; Richardson and Bokkenheuser 1963; Richardson and Koornhof 1965; Richardson, Koornof and Hayden-Smith 1966; and Richardson and others 1968). The schools took children from very different economic backgrounds and were, in rising order of economic standard and urbanization, located at Tlaseng, Komatipoort, Witkoppen, and Soweto. The results are summarized in table 15-2. The water supply and sanitation facilities were very poor, except at Tlaseng in 1966 after a new water supply had been constructed and at Soweto where high standards of water supply and excreta disposal prevailed. The authors of these studies started out believing that waterborne transmission of *Salmonella* and *Shigella* was important and ended up concluding that it was not. After the first Tlaseng study, Bokkenheuser and Richardson (1960) wrote that "water supply was probably implicated in the conveyance of the infections". After the Witkoppen study, Richardson and Bokkenheuser (1963) wrote that "the poor quality of the water, particularly that drawn from surface wells, made it highly probable that drinking water was involved in the transmission of the infections". After the Komatipoort study, Richardson and Koornhof (1965) wrote that "the suspicion that the water supply is a factor in the transmission of salmonellosis is strengthened by the results of the present survey". Then came the shock of finding prevalences in Soweto as high as, or higher than, those reported from relatively impoverished rural areas with poor sanitation. Richardson, Koornhof and Hayden-Smith (1966) then wrote that "although it is reasonable to assume that water contamination plays some part in the transmission of these organisms, it does not appear to be the most important factor", and again that "water supplied to each house by the Johannesburg municipality was of good quality, yet it did not affect the incidence of salmonellosis and shigellosis". The complete turnabout came after the resurvey of Tlaseng in 1966, when Richardson and others (1968) concluded that "in the environment of the Bantu children the provision of high quality community water as the only sanitary measure was without effect on the prevalence of intestinal *Salmonella* and *Shigella* infections".

Other work in South Africa by the same authors has strongly implicated *Salmonella* contamination of meat and meat products in the transmission of salmonellosis (Prior and Badenhorst 1974; Richardson, Burnett and Koornhof 1968). The contention is indirectly supported by parasitological data, obtained during the 1966 survey at Tlaseng, showing that 16 percent of children had *Taenia* infections (compared with only 3 percent having *Ascaris*, 2 percent having *Trichuris*, and 2 percent having *Necator*), data that indicate the common consumption of undercooked meat.

Occurrence and Survival in the Environment

Because of their very extensive nonhuman reservoir, salmonellae can be isolated frequently from a wide variety of environmental samples. Before the role of *E. coli* and *Campylobacter* in human diarrhea was recognized in the 1970s, the bacterial enteric pathogens of prime interest were the salmonellae, the shigellae, and *Vibrio cholerae*. Of these it is the salmonellae that are by far the most common in environmental samples, and so a great deal of work was done on the occurrence and survival of salmonellae in water and soils. Indeed, *Salmonella* became the favorite pathogen of the sanitary engineers and environmental microbiologists; whenever data on more than harmless indicator organisms were required, salmonellae would be studied. This situation has generated a large literature, second only to that on fecal indicators in the environment (see chapter 13), and only a small portion of it can be reviewed here. In the absence of specific information, it may generally be assumed that the survival, and the factors affecting survival, of *Salmonella* in the environment is similar to that described for fecal coliforms and *E. coli* in chapter 13.

In surface water

Salmonellae will be found in surface waters wherever there are animal populations. Those workers who have failed to find them have probably not looked hard enough. This involves sampling large volumes of water, or using suspended swabs to sample flowing water for several days. The occurrence and survival of salmonellae in surface waters has been reviewed by Wray and Sojka (1977).

Salmonellae are found in rural streams receiving run-off from agricultural land and discharges of domestic and other effluents from villages and rural industries. Harbourne (1977) and Harbourne, Thomas

and Luery (1978) readily isolated salmonellae from swabs suspended in streams in North Yorkshire (England). Smith, Jones and Watson (1978) sampled rivers and streams running through agricultural land in Cheshire and Lancashire (England). Salmonellae were detected at 71 percent (10 of 14) of the sites investigated. Sewage effluent discharges were believed to be the major source of the salmonellae, but farms, a dairy, an abattoir, and gulls were also implicated. In one stream the concentration of salmonellae was 1,100 per 100 milliliters at an effluent discharge point and 23 per 100 milliliters at a point 0.5 kilometer downstream.

Davis (1979) studied stream water quality in a rural area north of Houston (Texas, USA). During low flow, salmonellae concentrations were up to 5,800 per 100 milliliters, and during storms the maximum concentration was 2,500 per 100 milliliters. Salmonellae were sometimes detected when fecal coliforms were absent. Dondero and others (1977) isolated salmonellae from 39 percent of 322 swabs suspended in six streams in central New York state (USA). Cherry and others (1972) found that 44 percent of samples collected from unpolluted mountain streams ("judged to be completely free from human, domestic animal and industrial pollution") in Georgia (USA) contained salmonellae. One "unpolluted" stream acquired salmonellae within 100 meters of its source. The authors suggest that some salmonellae may have an aquatic reservoir and that they may be "superior to coliforms as indexes of water quality". These suggestions, apart from being contradictory, have not been supported by subsequent work.

Drainage from farms raising animals by intensive methods are responsible for major contributions of salmonellae to rural streams (see, for instance, Miner, Fina and Piatt 1967). The *Salmonella* serotypes found in a particular water may reflect the special character of the wastes contaminating that water. *S. agona*, a serotype associated with fish meal used as a poultry and animal feed, was found in all water samples from a creek receiving poultry wastes in Georgia (USA; Cook, Champion and Ahearn 1974).

Salmonellae can travel considerable distances in streams and rivers, especially at low water temperatures. Spino (1966) showed that *S. saintpaul* and *S. thompson* traveled for at least 120 kilometers under the frozen surface of the Red River of the North from their source at the Fargo-Moorhead (North Dakota-Minnesota, USA) effluent discharge sites.

Salmonellae are also found in urban streams and stormwater run-off, although in the absence of waste discharges the concentrations may be lower than found in rural catchments. Geldreich and others (1968) isolated 4,500 *S. thompson* per 100 milliliters from a sample of urban stormwater in Cincinnati (Ohio, USA). Olivieri, Kawata and Krusé (1978) reported that salmonellae were nearly always present in samples of urban stream water and stormwater in Baltimore (Maryland, USA) and that geometric mean concentrations at various sites were between 6 and 140 per 100 milliliters.

The detection of salmonellae in waters with very low fecal indicator counts is not uncommon. Dutka and Bell (1973) studied the St. Lawrence river (Canada) and detected salmonellae in 24 percent (17 of 72) of samples containing less than 9 fecal coliforms per 100 milliliters. Samples that contained more than 1,000 fecal coliforms per 100 milliliters were 86 percent (6 of 7) positive for salmonellae. Geldreich and Bordner (1971) reviewed a variety of stream pollution data from the USA and found that salmonellae had been detected in 54 percent of samples containing 1–1,000 fecal coliforms per 100 milliliters and in 96 percent of samples containing more than 1,000 fecal coliforms per 100 milliliters.

Many attempts have been made to relate the presence of salmonellae in surface waters to the concentrations of fecal coliforms or other fecal indicator bacteria (see chapter 13). Although in general the chances of isolating salmonellae increase as water pollution (and therefore indicator densities) increase, there is no precise or generally applicable relationship. Indeed, when analyzing data from a particular location several workers have found that *Salmonella* concentrations are among the most difficult bacterial pathogens to predict on the basis of indicator concentrations (Davis 1979; Olivieri, Kawata and Krusé 1978). Gallagher and Spino (1968) reviewed water pollution data from several rivers in the USA and failed to show any fecal coliform density below which *Salmonella* isolation would be particularly unlikely. Smith, Twedt and Flanigan (1973) isolated salmonellae from the Huron and Saline rivers (Michigan, USA) at sites where fecal coliform concentrations were low and found that, probably due to technical difficulties in the laboratory, "the probability of *Salmonella* isolation decreased as the fecal coliform concentration increased".

Salmonellae are more likely to be found in bottom sediments than in the overlying waters. Hendricks (1971*a*) isolated salmonellae from 0.6 percent (1 of 195) of water samples and from 4.6 percent (9 of 195) of bottom sediment samples in a stretch of river (Georgia, USA) below a sewage effluent outfall. Van Donsel and Geldreich (1971) collected simultaneous sediment and water samples at a variety of lake and river sites. Forty-six percent of sediment samples, and only 8 percent of

water samples, contained salmonellae. The highest *Salmonella* concentration in sediment was 790 per 100 milliliters. Salmonellae were isolated from 19 percent of sediment samples when the fecal coliform concentration in the overlying water was between 1 and 200 per 100 milliliters, from 50 percent when between 201 and 2,000, and from 80 percent when the fecal coliform concentration in the water was over 2,000 per 100 milliliters.

The survival of salmonellae, especially *S. typhi*, in water has been investigated by several researchers over the past 80 years (see the appendixes to Feachem and others 1980). McFeters and others (1974) studied the survival of several species of enteric bacteria in membrane chambers suspended in well water at 9–13°C. Approximate t_{90} values over 2 days were 80 hours for *S. paratyphi* A and *S. typhimurium*, 32 hours for *S. typhi*, and 19 hours for *S. paratyphi* B. Geldreich and others (1968) derived t_{90} values over 14 days for *S. typhimurium* in storm water of 240 hours at 10°C and 160 hours at 20°C. At both temperatures the dieoff of *S. typhimurium* was similar to that of fecal coliforms and considerably more rapid than *Str. faecalis*.

Dutka and Kwan (1980) studied the death of *S. thompson* in membrane chambers suspended in Lake Ontario and Hamilton Bay (Canada) when water temperatures were 17–19°C. Over the first 3 days, t_{90} values were 19–180 hours, and over the entire 28 days of the experiments average t_{90} values were 122–224 hours. Death rates were considerably greater near the surface than at greater depth and were higher in less polluted water (Lake Ontario) than in more polluted water (Hamilton Bay). *S. thompson* dieoff was similar to *Str. faecalis* and faster than *E. coli*.

The range of death rates reported is large and there is considerable interserotype, and probably also intraserotype, variation in ability to survive in water. It has been suggested that these differences in survival may partly explain why some serotypes are common and others are very rare in human and animal infection (Enkiri and Alford 1971). Few comparative studies on different serotypes in water under identical conditions have been conducted, but the data of McFeters and others (1974) showed that *S. typhimurium*, a very common serotype, survived for longer than *S. typhi* and *S. paratyphi* B.

Overall survival times reported range from 1 to over 100 days, with typical t_{90} values in the range of 20–200 hours. These results are similar to the fecal coliform t_{90} data reviewed in chapter 13, and most comparative studies have found *Salmonella* survival to be similar to that of fecal coliforms (see, for instance, Smith, Twedt and Flanigan 1973). The factors affecting *Salmonella* survival are the same as those affecting fecal coliforms. Survival is greatly prolonged at lower temperatures and somewhat prolonged in darkness. Experimental data show that survival is prolonged in more polluted waters, but many experiments are done in biologically inactive polluted waters (autoclaved or filtered), or in chambers suspended in polluted waters, where the salmonellae are not exposed to predation and competition. Experiments in New Zealand, however, showed that *S. typhimurium* and *S. bovis-morbificans* survived for under 2 weeks (t_{90} <56 hours) in clean water and for 12–16 weeks (t_{90} = 340–450 hours) in unsterilized water containing 5 percent (weight per volume) sheep feces (Tannock and Smith 1971).

Growth of salmonellae in polluted water is possible but unlikely, although concentration and perhaps growth in bottom sediments is more likely. Hendricks (1972) showed that *S. senftenberg* grew in autoclaved river water collected downstream of a sewage effluent outfall at 30°C. No growth occurred at 20°C or 5°C or in autoclaved river water collected upstream from the same outfall (see also Hendricks 1971*b* and Hendricks and Morrison 1967).

Viewed with hindsight, the interest in low levels of salmonellae in surface waters, and their relationship to the concentrations of indicator bacteria, seems excessive. From the viewpoint of human public health, the greater importance of other bacterial agents of diarrhea, the frequent transmission of salmonelloses by contaminated animal products, and the high infective dose for salmonellae make the presence of low concentrations in surface water a matter of only minor concern in developed countries. From a veterinary viewpoint, it has been suggested that the contamination of rural streams by salmonellae may help to maintain infection among farm animals (Smith, Jones and Watson 1978). There are other more likely routes of transmission among farm animals, although stream pollution has undoubtedly contributed to some outbreak (George and others 1972; Williams 1975; Wray and Sojka 1977). In developing countries, surface water contamination by salmonellae must be seen in the light of the probable minor importance of *Salmonella* in pediatric diarrhea and of the many alternate transmission routes for salmonellae in areas of poverty and poor hygiene (see table 15-2 and the discussion of salmonellosis in South Africa and typhoid in Lesotho, above).

In groundwater

Salmonellae are unlikely to be present in groundwater unless the water table is very shallow or fissured strata allow the direct flow of surface waters into an aquifer. Salmonellae have not been isolated from dual

Table 15-2. *Period and point prevalences of* Salmonella *and* Shigella *excretion by black schoolchildren in the Transvaal, Republic of South Africa*

Site	*Year*	*No. of children*	*Age of children (years)*	*No. of surveys per year*	*Water supply (E. coli concentrations are per 100 milliliters of water)*	*Excreta disposal facilities*	*1 year period prevalence[a] (percent)*		*Mean point prevalence (percent)*		*Source*
							Salmonella *excretion*	Shigella *excretion*	Salmonella *excretion*	Shigella *excretion*	
Tlaseng school, rural Western Transvaal	1958–59	75	6–16	7	Polluted shallow wells (2–900 *E. coli*)	None[b]	36[c]	25[c]	6.3[d]	4.2	Bokkenheuser and Richardson (1960)
	1966	92	7–16	4	Borehole with reticulation (0 *E. coli*)	Pit latrines[b]	13[c]	20[c]	3.8	5.5	Richardson and others (1968)
School near Komatipoort on Mozambique border in rural Eastern Transvaal Lowveld	1964	99	7 17	2	Piped water (0 *E. coli*) River water (50–250 *E. coli*) and salmonellae)	Pit latrines[b]	6	3	3.5	2.0	Richardson and Koornhof (1965)

School at Witkoppen, a periurban area 25 kilometers from Johannesburg	1961	75	7–18	8	Protected well with hand-pump (0 *E. coli*) Borehole with motor pump (0–4 *E. coli*) Borehole with wind pump (0–>4 *E. coli*) Open well (>4 *E. coli*)	Pit latrines[b]	30	2.7	5.8	0.3	Richardson and Bokkenheuser (1963)
Two schools in Soweto, a large African city near Johannesburg	1964	55	7–18	8	Chlorinated piped water (0 *E. coli*)	Waterborne sewerage	36	9	5.9	1.1	Richardson, Koornhof and Hayden-Smith (1966)

a. The recorded annual period prevalence of an acute and readily transmissible infection, such as salmonellosis, rises markedly as the number of surveys per year increases. Therefore, the figures for Komatipoort, for instance, are not comparable with those from Witkoppen.

b. This information is not given in the original source but in the summary by Koornhof and others (1979).

c. For the reasons stated in note a, it is not possible to compare the period prevalences from 1958–59 (seven surveys) with those from 1966 (four surveys).

d. This figure is elevated due to an outbreak of *S. mobeni* infection that coincided with the survey in December 1958.

purpose wells in Israel used for groundwater recharge and abstraction (Goldshmid 1974). Salmonellae will be found in many open wells due to the drainage of contaminated surface waters down unprotected well shafts.

In drinking water

It is uncommon to isolate salmonellae from piped water supplies, whether treated or untreated, and their presence suggests a serious fault in the design or maintenance of the system. Raman and others (1979) found that 9 percent (3 of 33) of tap water samples in Aurangabad (Maharashtra, India) contained salmonellae due to leakage of polluted water into the distribution system.

S. typhimurium was isolated from 7 percent (5 of 74) of water samples from taps and reservoirs of the water supply system of Riverside (California, USA) during the major outbreak of salmonellosis in May–June 1965 (Boring, Martin and Elliot 1971). One composite sample examined quantitatively revealed 1.7 *S. typhimurium* and 0.14 *E. coli* per 100 milliliters. All *S. typhimurium* isolates examined were of phage type 10, as were the clinical isolates during the outbreak. The water sources were deep wells, and the water was delivered unchlorinated.

Schubert and Scheiber (1979) reported that salmonellae could frequently be isolated from piped drinking water in Togo, even in the absence of *E. coli* and coliforms. This was attributed to faulty wellheads and broken reservoir covers allowing access to rainbow lizards (*Agama agama*), which were often found to excrete salmonellae and sometimes few or no *E. coli* and coliforms. Keeping small animals out of water systems, and thus reducing the risk of contamination by salmonellae, can prove extremely difficult, especially in arid areas.

A large proportion of the population of the developing countries drink untreated and unprotected surface waters or open well waters that are often heavily polluted by feces (table 13-1). Salmonellae may be expected in these drinking water sources, although few data on their presence exist. Salmonellae will be far more common than shigellae (chapter 16) in unprotected water sources because, unlike shigellae, they are excreted by some pigs and by donkeys, goats, cattle, camels, rats, dogs, and other village animals whose excreta commonly pollute streams, ponds, and open wells. Gracey and others (1979) isolated salmonellae from 48 percent of water samples and from 63 percent of sediment samples collected from a river and canals in Jakarta (Indonesia) used as drinking water sources by the poorer inhabitants of the city.

The contamination of clean drinking water by *E. coli* while the water is stored in the home is well documented (chapter 13). No similar data exist for salmonellae, although such pollution may be expected and has been shown for clean water used by infected turkeys (Gauger and Greaves 1946).

In seawater

Salmonellae are found in estuarine and marine environments where there is fecal contamination from domestic or agricultural wastewater discharges or where there are large populations of water birds. The occurrence and survival of salmonellae in seawater have been reviewed by Buttiaux (1962).

Colwell and Kaper (1978) reported that the ratios of salmonellae to fecal coliforms in Chesapeake Bay (USA) ranged from 1:100 to 1:1000 and that there were up to 240 salmonellae per 100 milliliters in Baltimore Harbor (Maryland, USA). Salmonellae could not be isolated from Chesapeake Bay in the winter, only during April–November. In contrast, Carney, Carty and Colwell (1975) failed to isolate salmonellae in an extensive survey of microbiological pollution in a subestuary of Chesapeake Bay, despite occasionally elevated concentrations of fecal coliforms (up to 5,400 per 100 milliliters)

Goyal, Gerba and Melnick (1977, 1978) studied the occurrence of fecal indicator bacteria and salmonellae in the waters (salinities 1.0–2.2 percent) and bottom sediment of canals bordering Galveston Bay (Texas, USA). The canal networks are largely man-made, and holiday homes are located along their banks. Domestic wastes are discharged into the canals through septic tanks or small treatment plants, and the canals are greatly used for bathing, boating, skiing, and diving. Salmonellae were isolated from 47 percent (17 of 36) of sediment samples but from only 3 percent (1 of 36) of water samples. Salmonellae concentrations in sediment were between 0 and 150 per 100 milliliters.

The limited information on *Salmonella* survival in seawater indicates that it may be only slightly less than survival in fresh water. Since *E. coli* survival is very much shorter in seawater than in fresh water (chapter 13), it follows that *Salmonella* are more persistent in marine environments than are *E. coli*. Therefore *E. coli* are poor indicators of salmonellae, as they are of enteroviruses, in marine environments (see Petrilli and others 1979).

Jamieson, Madri and Claus (1976) added 1.5×10^7 *S. typhi* to samples of sterilized seawater, adjusted to salinities of 0.5, 2, and 3.5 percent, and stored them at 4, 25, and 37°C. Survival was inversely proportional to salinity and temperature. Maximum survival was for 7

days (t_{90} = 23 hours) at 4°C and 0.5 percent salinity, and minimum survival was for 5 days (t_{90} = 17 hours) at 37°C and 3.5 percent salinity. Survival of *E. coli* was shorter than that of *S. typhi* at all temperatures and salinities. Vasconcelos and Swartz (1976) compared the survival of *S. heidelberg* and *E. coli* in sterilized seawater at 14.5°C. After 6 days, the *E. coli* concentration had declined by 6 log units (t_{90} = 24 hours), whereas the concentration of *S. heidelberg* was reduced by only 1.5 log units (t_{90} = 6 hours).

In feces and night soil

In developing countries, pooled human feces are likely to contain *Salmonella* in areas where asymptomatic excretion of the organism is fairly common (see above). Similarly, pooled animal excreta are also likely to contain *Salmonella*. Screening night soil for *S. typhi* is potentially useful in epidemiological investigations of typhoid, and screening farm wastes for specific *Salmonella* serotypes can assist livestock and poultry hygiene.

Jordan (1926) studied the bacterial content of feces from typhoid patients and carriers (Chicago, USA); the feces were stored at room temperature in sealed cans. *S. typhi* survived for between 3 and 52 days. Desiccation was not a factor contributing to death, since the moisture content of the feces stored in this manner was still over 80 percent after 77 days. No multiplication of *S. typhi* in stored feces was observed.

Tannock and Smith (1972) reported that *S. typhimurium* survived for 6–18 weeks (t_{90} = 150–450 hours) in sheep feces outdoors in New Zealand. Survival was longer on shaded than exposed sites and longer in summer than winter (presumably because of the bactericidal effect of freeze-thaw cycles in winter).

Berkowitz, Kraft and Finstein (1974) studied the survival of various *Salmonella* serotypes (*typhimurium*, *saintpaul*, *thompson*, and *infantis*) inoculated into samples of wet poultry excreta (80 percent water) and stored at various temperatures. Initial concentrations were 1.6×10^5–2.4×10^6 *Salmonella* per gram. Overall persistence was usually less than 1 month; t_{90} values (averaged over the first 3 log units' decline) averaged 184 hours at 9–12°C, 112 hours at 18–20°C, and 40 hours at 30°C. Growth occurred before decline in sixteen of twenty-three tests, with concentrations of *Salmonella* rising to a maximum of 1.4 log units above initial values. No clear pattern of differential survival among the four serotypes emerged. When samples were allowed to dry for 2 days (12 percent water after 2 days) t_{90} values became 21 hours at 20°C. However, the organisms that survived the 2 days drying then survived for considerably longer than in the undried samples, with overall survival times being 148 days at 11°C. It may be concluded that storing undried excreta is more effective in eliminating *Salmonella* than promoting initial rapid drying.

These and other studies (listed in the appendixes of Feachem and others 1980) suggest t_{90} values of 40–100 hours for *Salmonella* in feces or night soil in tropical climates (20–30°C).

In sewage

The monitoring of sewage for salmonellae in general, or for *S. typhi* in particular, is of practical public health value in epidemiological surveillance, investigating outbreaks of salmonellosis or typhoid, and, especially, in tracing typhoid carriers. The favored technique is the Moore's swab, an absorbant pad suspended in the flow for 2–3 days. If a pad of calcium alginate wool is used, it can be completely dissolved in sodium hexametaphosphate on return to the laboratory, thereby liberating all the entrapped bacteria. For more rapid results, a wipe swab can be used—the sewer wall is wiped and the swab immediately returned to the laboratory (Bokkenheuser 1964; Gell and others 1945; Harvey and Phillips 1955; Kelly, Clark and Coleman 1955; Moore, Perry and Chard 1952; Robinson 1958; Shearer and others 1959). Alternatively, various filtration methods have been devised for detecting salmonellae in large volumes of sewage collected at a single time. Hirn (1980) suggested that monitoring the effluent of food-processing plants for salmonellae was a useful contribution to the microbiological quality control of the processed food.

Reported concentrations of salmonellae in sewage vary considerably. Concentrations per 100 milliliters of 7–250 in India (Phirke 1974), 2–41 in South Africa (Grabow and Nupen 1972), 500 in Baltimore (Maryland, USA; Olivieri, Kawata and Kruse 1978), 8,000 in Houston (Texas, USA; Davis 1979), up to 2.3 in Finland (Hirn 1980), up to 7,240 in northwest England (Jones 1977), and 670 in Holland (Kampelmacher and van Noorle Jansen 1970) have been reported.

Several workers have recorded concentrations of salmonellae in sewage that are difficult to explain by analyzing known inputs from patients, carriers, farm wastes, or food-processing effluents. One such study in Holland concluded that salmonellae must be multiplying in the sewerage system (Kampelmacher and van Noorle Jansen 1976).

The concentration of salmonellae in sewage may fluctuate on a regular basis. McCoy (1977) reported that raw sewage in Hull (England) contained most

salmonellae (median concentration of 150–400 per 100 milliliters) during July–September and least during January–March. Yaziz and Lloyd (1979) studied the hourly fluctuations of *Salmonella* concentrations in raw sewage at Guildford (England). Peak concentrations occurred at 0900–1000 hours, 2 hours before the peak flow of sewage at midday. Samples of raw sewage collected during the morning peak over a 9-month period contained 20– > 1,800 (median 130) *Salmonella* per 100 milliliters at Guildford and 11–1,600 (median 170) *Salmonella* per 100 milliliters at Woking, a nearby town. *Salmonella* concentrations were higher in the cooler months than in the summer.

During the 1930s, a survey of sewage in Bandung (Java, Indonesia)—which then had a population of 200,000, of whom 40,000 were sewered—demonstrated the presence of typhoid bacilli in 62 out of 80 samples (Mom and Schaeffer 1940); *S. typhi* concentrations varied from less than 1,000 to 45,000 per 100 milliliters, with an average of 5,000 per 100 milliliters. Despite individual accounts of high levels of *Salmonella* in sewage, reported concentrations from developing countries are typically lower than from developed countries. This may reflect the greater input of effluents from food-processing plants in developed countries or it may be an artifact caused by differences in laboratory technique. Daniel and Lloyd (1980) reported geometric mean concentrations of salmonellae in two refugee camps in Bangladesh of only 7.1 and 7.7 per 100 milliliters, and they noted that these results could have been caused by the difficulty of selectively isolating salmonellae from sewage with a very high solids content (17,000 milligrams per liter).

There are few studies on *Salmonella* survival in sewage (see the appendixes of Feachem and others 1980), and most attention has focussed instead on survival in sludges and slurries (see below). t_{90} values of 77–108 hours may be computed from the data of Green and Beard (1938) on *S. typhi* in raw sewage at 7–20°C. *S. tennessee* inoculated at 10^9 per 100 milliters into strong Jerusalem (Israel) sewage (BOD 800–1200 milligrams per liter), survived for 22 days (t_{90} = 60 hours) in outdoor storage tanks in summer (Bergner-Rabinowitz 1956). Gallagher and Spino (1968) reported the survival of *S. typhimurium* and fecal coliforms in various process waters and effluents at two sugar beet factories in the USA. t_{90} values for *S. typhimurium* were generally over 72 hours and under 168 hours, and those for fecal coliforms were similar. In the absence of other information, it is reasonable to assume that *Salmonella* survival in sewage is similar to that of fecal coliforms (chapter 13), with t_{90} values in warm climates of 20–100 hours.

In sludge and slurry

The common practice of applying sludge and slurry[2] to arable and pasture land has stimulated interest in the occurrence and survival of salmonellae in these materials. Sludges from sewage treatment works will almost always contain salmonellae. Reported cencentrations vary greatly and may fluctuate seasonally. In England, concentrations of salmonellae per 100 milliliters of raw sludge have been reported as around 70 (median value), with 7 percent of samples containing over 2,400 near Hull (McCoy 1977, 1979), 40–11,000 in Yorkshire (Fennell 1977), and 4,000–23,000 in the northwest (Jones, F. 1977). In Switzerland, over 90 percent of raw sludge samples contained *Salmonella* with maximum and mean concentrations of 10^6 and 10^4 per 100 milliliters respectively (Hess and Breer 1975; Obrist 1979).

Pike (1981) reviewed the data on salmonellae in sewage sludges in England and Wales. Geometric mean counts per 100 milliliters of raw sludge reported from various regions were between 8 and 1,400. Salmonellae were more numerous and more frequently isolated from sludge at treatment works serving communities of 10,000–100,000 people than at works serving larger or smaller communities. Common *Salmonella* serotypes in sludge were those (particularly, *agona*, *typhimurium*, *heidelberg*, *virchow*, *anatum*, and *hadar*) that infect a wide range of animals and man and not those host-adapted serotypes (such as *dublin* in cattle, *abortus-ovis* in sheep, *cholerae-suis* in pigs, and *gallinarum* and *pullorum* in poultry) that are a major cause of salmonelloses in farm animals in the UK. Interestingly, of the ten most common serotypes in sludge, six were among the ten most common serotypes in human infections and only four were among the ten most common isolates from farm animal infections. This suggests that sludge and its disposal on land do not play a major role in transmitting salmonellae among farm animals in England and Wales. Hall and Jones (1978) examined eight raw sludges from sewage treatment plants in England and found between 34 and 11,000 *Salmonella* per 100 milliliters. The most commonly isolated serotypes were, in decreasing order of frequency of isolation: *newhaw*, *heidelberg*, *saintpaul*,

2. In this context, "slurry" refers to the mixture of animal feces, urine, water, and sometimes some straw or other bedding produced at farms. This material may be rich in salmonellae; it has a solids content similar to that of sewage works sludge (1–10 percent), and it presents disposal problems very similar to those of sludge produced at a sewage works receiving primarily domestic, rather than industrial, sewage. The major distinction is between a sludge of primarily human origin and a slurry that contains the wastes of farm animals.

typhimurium, *paratyphi* B, *oranienberg*, and *kaapstad*. Only one of these serotypes, *typhimurium*, is a prominent cause of farm animal infection in Britain.

The frequency and concentration of salmonellae in animal slurries are typically lower than those in sewage works sludges. The lower frequency is partly due to the size of the contributing population; the herd of animals on a typical farm is much smaller than the number of people in a typical town. Jones and Mathews (1975) isolated *Salmonella* from 11 percent (20 of 187) of cattle slurry samples taken randomly throughout England and Wales; concentrations were up to 180 per 100 milliliters. Jones, Bew and Gammack (1975) isolated *Salmonella* from only 3 percent (2 of 63) of dairy factory sludge samples in England. Jones and others (1976) isolated *Salmonella* from 22 percent (12 of 54) of pig slurries in southern England at concentrations up to 2×10^5 per 100 milliliters. Kraft and others (1969) isolated *Salmonella* from the wastes of 50 percent (18 of 36) of poultry farms in New Jersey (USA) at concentrations of up to 3.4×10^6 per 100 grams. Monitoring animal slurries for salmonellae can assist the detection and control of infection in the herds (Jones and Hall 1975).

The literature on *Salmonella* survival in sludges and slurries is extensive (see the appendixes of Feachem and others 1980). Jones (1978), reviewing European literature, reports the survival of salmonellae in slurry as being between 13 and 286 days. Strauch (1978) reported the survival of various *Salmonella* serotypes in animal waste slurries of up to about 1 year at 8°C and up to about 6 months at 17°C. At both temperatures and in four different slurries, *S. enteritidis* and *S. cairo* consistently survived for longer than *S. gallinarum*, *S. typhimurium*, or *S. paratyphi* B. Braga (1964) found that *S. cholerae-suis* survived for longer than *S. typhimurium*, *S. typhi*, or *S. enteritidis* in sewage sludges at 6 and 28°C. Initial concentrations were 6×10^5 per 100 milliliters, and survival times ranged from 20–38 days at 6°C and 4–9 days at 28°C. Findlay (1972) recorded the survival of *S. dublin* in cattle slurry in northeastern England for 31–33 weeks in winter and 18–19 weeks in summer. Ekesbo (1979) found that *S. dublin* and *S. zanzibar* survived for at least 13 weeks in cattle slurry (8–9 percent solids) and settled cattle slurry (5–6 percent solids) at 5°C.

Jones (1976) experimented with various *Salmonella* serotypes inoculated into cattle slurry from a dairy. When *S. dublin* was inoculated at 10^6 per milliliter into slurry (4.7 percent solids), t_{90} values were 528 hours at 5°C and 10°C, 228 hours at 20°C, and 52 hours at 30°C. Overall survival times were 132 days at 5°C and 10°C, 57 days at 29°C, and 13 days at 30°C. Survival increased as the solids content of the slurry was increased from 1 to 5 percent, but did not increase further at solids contents above 5 percent. When the survivals of eight strains (four serotypes) were compared (at 10°C and 5.5 percent solids), overall survival times ranged from 90–140 days and appeared to be related to strain not to serotype. There was no evidence of multiplication. Jones suggested that the storage of slurry for 1 month prior to spreading on pasture, followed by a further month during which the pasture is not grazed, would be a minimal treatment regime for protecting cattle from *Salmonella* infection by this route.

Salmonellae can multiply vigorously in sterilized sludge or slurry, but under natural conditions they are strongly inhibited by the activity of other microflora (Findlay 1973; Jones, Smith and Bew 1977).

In soil

Salmonellae are likely to be found in soils that have been treated with sludges, slurries, or effluents. Reported survival times in several countries are: *S. dublin* for up to 12 weeks in the autumn in southern England (Taylor and Burrows 1971); *S. dublin* for 24 weeks in winter and 13 weeks in summer in northeast England (Findlay 1972); *S. typhimurium* for more than 35 weeks in England (Mair and Ross 1960); *S. typhimurium* for 4–10 weeks in New Zealand (Tannock and Smith 1972); *S. typhi* for up to 17 weeks during the rainy season in California (USA; Beard 1940); and *S. typhi* for 5–19 days in Michigan (USA; Mallman and Litsky 1951).

The long reported survival times represent residual contamination by a very small proportion of the large original inoculum of bacteria. Delage (1961) found that 10^6 *S. abortus-ovis* in 1 gram of soil were reduced by 99.9 percent after 50 days but were still detectable for over 300 days.

Watson (1980) applied digested sludge, containing 25–30 *Salmonella* per 100 milliliters, to cabbage fields at a rate of 70 cubic meters per hectare. Survival times, during spring and summer in northern England (temperatures 3–32°C), were 42–49 days, with t_{90} values of 336–528 hours. Chandler and Craven (1978) also reported about 8 weeks survival for *S. typhimurium* on dry soil in Australia.

Zibilske and Weaver (1978) studied the effect of soil type, moisture, and temperature on the survival of *S. typhimurium* applied to soil in cattle slurry or saline. Survival was inversely proportional to temperature but was not obviously related to soil type or delivery medium. At 22°C survival times ranged from 3 days (t_{90} = 17 hours) to 84 days (t_{90} = 483 hours).

Dazzo, Smith and Hubbell (1973) studied the survival of *S. enteritidis* in fine sand (soil moisture 10 percent), which was returned to the laboratory following different regimes of cattle slurry application. Samples were stored in darkness at 22°C. Death rate was inversely related to the rate at which the sand had received slurry. When no slurry was applied, overall survival was 8 weeks (from an initial concentration of 10^6 *Salmonella* per gram of soil), and the t_{90} was 254 hours. At the maximum rate of slurry application tested (508 cubic meters per hectare per week), the t_{90} was 363 hours, and over 100 *Salmonella* per gram of soil remained after 8 weeks. The effect of slurry application in enhancing *Salmonella* survival in fine sand was not due to moisture content, since this was controlled at 10 percent throughout each experiment.

Bergner-Rabinowitz (1956) studied the survival of *S. tennessee* seeded into sewage (at about 10^8 per 100 milliliters) and applied to soil of low organic content near Jerusalem (Israel). During the winter (air temperature 2–21°C), at the soil surface (soil moisture 8–39 percent) overall survival times averaged 46 days, whereas at a depth of 100 millimeters (soil moisture 19–30 percent) survival was for 70 days. Coliforms were still detectable in low concentrations after 74 days. During the summer (temperature not given) survival at the surface (soil moisture 4–23 percent) was 15 days, and at 150 millimeters depth (soil moisture 17–29 percent) 11 days. These experiments were all on uncultivated plots using a single application of sewage seeded with *S. tennessee*. In subsequent experiments in summer, *S. tennessee* in sewage was applied to plots growing sunflowers, and the plots were reirrigated with sewage not containing *Salmonella* every 6–8 days (in line with normal practice in Israel). Survival at the surface (soil moisture 4–31 percent) was increased to 23 days, and at a depth of 150 millimeters (soil moisture 18–30 percent) to 37 days.

These and other studies show that salmonellae can survive in soil for periods of many months when conditions are ideal. The factors most affecting survival are temperature, exposure to sunlight, and the moisture content, pH, and the organic content of the soil (Gerba, Wallis and Melnick 1975; Rudolfs, Falk and Ragotzkie 1950). In hot and sunny climates, maximum survival times may be around 2 months, with almost complete elimination after 2 weeks.

On pasture

The role of sludge and slurry application to pasture in transmitting *Salmonella* infections remains controversial. Considerable evidence (reviewed by Williams 1975, 1979) has been accumulated in West Germany, Holland, and Switzerland (Hess and Breer 1975; Obrist 1979) to suggest that the use of inadequately treated sludges and slurries on pasture is a major factor in spreading bovine *Salmonella* infections. Some countries, Switzerland among them, have banned the use of unpasteurized sludge on pasture in summer (Williams 1979). In the UK, a general association between sludge application to pasture and salmonellosis in grazing animals has not been demonstrated. Wray and Sojka (1977) reviewed the subject and mention that, in addition to documented outbreaks in which pasture contamination has been implicated (for instance, Jack and Hepper 1969), the salmonellosis peak in cattle in many European countries occurs in late summer when the cattle are grazing and that the incidence declines markedly when cattle are brought indoors for the winter. This is scarcely evidence; the human salmonellosis peak occurs at the same time, and people neither graze contaminated pasture nor contract most of their infection from contaminated beef and milk. Further evidence against the importance of pasture contamination in cattle salmonellosis is the high infective dose of *Salmonella* (see the subsection on transmission, above, and the studies of Taylor 1973 and Taylor and Burrows 1971).

The interim guidelines produced by the Commission of the European Communities (Kelly 1978) recommended that, ideally, slurry should be applied only to arable land. If applied to pasture, it should receive a minimum of 60 days storage before spreading, and there should be a further interval of 30 days before grazing. Only healthy adult animals should be grazed on treated pasture.

Hess and Breer (1978) reported that salmonellae on grass treated with sludge could survive for up to 16 months in Switzerland. Most reported survival times are much shorter than this. In New Zealand, Josland (1951) reported survival of *S. typhimurium* on pasture of 12–24 weeks, and Tannock and Smith (1971) reported 6–14 weeks for *S. typhimurium* and 2–8 weeks for *S. bovis-morbificans*.

Taylor and Burrows (1971) applied cattle slurry (0.2 percent solids) containing 10^9 *S. dublin* per 100 milliliters to pasture during September–December in southern England. *S. dublin* survived for 11 days on grass 75–150 millimeters above ground, 18 days on grass at ground level, and 12 weeks in the soil. In similar experiments, with slurry impregnated with 10^8 *E. coli* per 100 milliliter, *E. coli* survival was only 3 days at 75–100 millimeters, 7 days at ground level, and 11 days in soil. Calves, given a choice of contaminated and

uncontaminated pasture, avoided the contaminated pasture for 2 days but then grazed it. When twelve calves were grazed on pasture that had been treated with slurry containing 10^8 avirulent *S. dublin* per milliliter, eight showed evidence of infection. None of six calves was infected when grazed on pasture treated with slurry containing 10^5 avirulent *S. dublin* per 100 milliliters.

Salmonella survival on pasture is considerably shorter than in soil and is shorter high up the blades of grass than near the ground. Heat, desiccation, and sunlight are all lethal factors, and survival on pasture in the tropics is unlikely to exceed 10 days.

On crops

Several outbreaks of salmonellosis and typhoid have been linked to the contamination of vegetables and fruit by sewage or sludge (Geldreich and Bordner 1971). Gayler and others (1955) linked an outbreak of gastroenteritis due to *S. miami* with sliced watermelon and showed that the act of cutting a dirty watermelon could contaminate the inner fruit and that multiplication of *S. miami* in the melon could then occur. Sixty-eight percent of lettuce and 72 percent of fennel samples marketed in Bari (southern Italy) were contaminated with salmonellae (Ercolani 1976). Twenty-two percent of vegetables purchased in Holland contained salmonellae, and contamination was especially prevalent on tropical imports (Tamminga, Beumer and Kampelmacher 1978).

In Colorado (USA) 100 percent of settled sewage samples contained salmonellae (concentrations 43–360 per 100 milliliters), and 63 percent of irrigation water samples from streams receiving raw or treated sewage contained salmonellae (concentrations 1–360 per 100 milliliters) (Dunlop and Wang 1961). Only 1 out of 97 samples of irrigated turnips, cabbage, spinach, endive, and lettuce contained *Salmonella*. This was attributed to the inadequate laboratory methods, which could only detect high levels of contamination, and to the use of furrow irrigation on sandy soil in a dry climate, which minimized the survival of salmonellae.

Salmonella survival data (see the appendixes of Feachem and others 1980), reviewed by Geldreich and Bordner (1971), indicate up to 53 days on root crops, up to 40 days on leafy vegetables, up to 5 days on berries, and over 2 days on orchard crops. Two early studies of *S. typhi* on radish and lettuce showed survival for 10 days in sunny sites and for 31 days in the shade (Creel 1912) and for 21–37 days (Melick 1917). The latter study also showed that *S. typhi* became attached to leaves lying on contaminated soil and were not readily removed by washing. Lovett and Francis (1976) concluded that fecal coliforms were good indicators of *Salmonella* survival on vegetables, but that coliforms and fecal streptococci were not.

The use of waters containing sewage effluents to irrigate crops is commonplace in developing countries and will increase as growing urban populations produce more wastewater and create an increasing market for intensively cultivated crops. The risks of crop contamination are reduced by adopting drip, furrow, or subsurface irrigation methods instead of using spray or flood irrigation. A further safeguard is to discontinue irrigation or fertilization with fecal materials 2 weeks prior to harvesting; the survival of salmonellae on crop surfaces will be very much reduced by heat, sunlight, and low humidity. If these precautions are taken *Salmonella* survival on crops is unlikely to exceed 10 days in hot and arid climates.

In fish and shellfish

Fish and shellfish from unpolluted waters, such as the open sea or mountain streams, do not contain salmonellae. Fish and shellfish living in waters polluted by waste discharges are commonly found to harbor *Salmonella* (Buttiaux 1962). Salmonellae are not known to cause disease in fish or shellfish, but they do cause temporary infection when the fish or shellfish are residing in waters containing salmonellae. Fish or shellfish can be decontaminated by placing them in clean water, but salmonellae seem to be eliminated rather more slowly than enteroviruses or *E. coli*.

Fish caught in deep-sea areas are free of *Salmonella* but may become contaminated prior to sale. Likely means of contamination are from ice made from polluted water, from storage in contaminated boxes or baskets, and from handling by infected packers or process workers. When salmonellae are isolated from fish intestines, contamination in polluted water is indicated; when salmonellae are isolated only from external surfaces, contamination during transport and handling is more probable. The contamination of fish meal used as a high protein food for poultry and pigs has been of great importance in the epidemiology of salmonelloses in Europe in recent years.

Geldreich and Clark (1966) studied the survival of various enteric bacteria in the sterilized intestinal contents of fish. In carp intestinal contents (pH = 7.3) at 10°C, *S. typhi*, *S. typhimurium*, *Shigella flexneri*, and fecal coliforms declined, whereas fecal streptococci grew slowly. At 20°C, fecal streptococci grew rapidly, fecal coliforms and *S. typhimurium* grew slowly, and *S. typhi* and *Sh. flexneri* declined. In bluegill intestinal

material (pH = 8.0) at 20°C, all species of bacteria tested grew rapidly except *S. typhi*.

In order to clarify the risks of salmonellosis associated with fish culture in sewage effluents, Heuschmann-Brunner (1974) experimented with carp and tench kept in water heavily contaminated by *S. enteritidis* and *S. typhimurium*. A few hours residence in heavily polluted water caused infection, and *Salmonella* spread rapidly along blood and lymph vessels throughout the body, including the musculature. Salmonellae were found most often, and for the longest time, in the digestive tract. At 9–12°C, *Salmonella* infection persisted in the tench gut for 60 days and in the carp gut for 68 days. In warmer water, infection persisted for longer—the reverse of what occurs with depurating shellfish, which cleanse themselves faster in warmer water because their rate of filter feeding increases. Lesions in the gut were produced by massive intralymphatic injection of salmonellae. Salmonellae isolated from fish several weeks after exposure were still pathogenic to mice. A great deal more research is required on the uptake and elimination of *Salmonella* by fish grown in sewage in developing and tropical countries.

Shellfish are often harvested from estuaries where polluted or potentially polluted waters flow into the sea. Salmonellae may be concentrated in the flesh of filter-feeding molluscs in the same manner as enteroviruses (chapter 9) and *E. coli* (chapter 13). Salmonellae are frequently isolated from shellfish harvested from contaminated waters and have given rise to major and many minor outbreaks of salmonelloses and enteric fevers (Buttiaux 1962). Depuration of shellfish, by placing them in clean water, seems to be less effective in removing *Salmonella* than in removing enteroviruses (chapter 9) and *E. coli* (chapter 13).

Janssen (1974) took oysters (*Crassostrea virginica*) from the Chesapeake Bay (USA) and kept them in an aquarium with salinity of 1.5 percent and water temperature of 20°C. Oysters were exposed to artificial seawater containing 2×10^7 *S. typhimurium* per 100 milliliters for 48 hours and then kept in clean water continually decontaminated by ultraviolet light. Oysters accumulated *S. typhimurium* up to a concentration of 2.8×10^4 per oyster and still contained 170 per oyster after 42 days in sterilized water. In other experiments in which the depuration water was only intermittently sterilized by ultraviolet light, oysters excreted *S. typhimurium* for 14 days and after 49 days still contained 6,000 per mollusc. These rates of *Salmonella* elimination by oysters in clean water are far slower than the reported rates for enteroviruses and *E. coli* elimination (DiGirolamo, Liston and Matches 1975; Hedstrom and Lycke 1964; Hoff and Becker 1969; Mitchell and others 1966).

Slanetz, Bartley and Stanley (1968) studied salmonellae in water and oysters in an estuary at Portsmouth (New Hampshire, USA). Water salinities were 1.1–2.5 percent and temperatures were 8–26°C. Salmonellae were readily isolated from water in which the coliform count was below the limit recommended for shellfish-growing waters (70 per 100 milliliters) and were on two occasions isolated from shellfish that met the coliform standard (less than 230 per 100 grams). On three occasions salmonellae were isolated from estuarine waters and shellfish containing no fecal coliforms per 100 milliliters and per 100 grams, respectively. Jegathesan and others (1976) studied 38 shellfish bought at markets in Malaysia. Three species were included: cockles (*Anadura granosa*) cultivated on muddy shores of the Malaysian west coast and mussels (*Modiolus senhaussi* and *M. metcalfi*) harvested from muddy and sandy shores, respectively. Cockles are commonly eaten half-boiled, whereas mussels are normally fried or baked. Two specimens contained *Salmonella*

In the air

Salmonellae are likely to be aerosolized and dispersed from flush toilets, spray irrigation devices, and activated sludge plants in the same manner as *E. coli* (see chapter 13). Comparatively few data on airborne *Salmonella* are available because they have been seldom studied and because they are present sporadically or in low concentrations in feces and sewage and so are far more difficult to detect in the air than *E. coli*.

Newson (1972) added 10^{10}–10^{11} *S. typhimurium* to toilet bowls and flushed, producing an average aerosol of 132 *Salmonella* per cubic meter of air. The survival ability of *Salmonella* in splashes produced by toilet flushing was tested by studying 0.1 milliliter droplets of water and feces, containing 10^8 *S. typhimurium*, on a laboratory bench. *Salmonella* survived for 12 days in both aqueous and fecal droplets, compared with 2–11 days for *E. coli* and 5 days for *Shigella sonnei* in parallel experiments. Katzenelson, Teltch and Shuval (1977) detected *S. infantis* 60 meters downwind from spray irrigators delivering raw sewage in Israel (see also Katzenelson and Teltch 1976). Hickey and Reist (1975) and Pereira and Benjaminson (1975) failed to detect *Salmonella* downwind of activated sludge tanks and grit chambers in the USA.

Inactivation by Sewage Treatment Processes

The literature on *Salmonella* in sewage treatment plants is limited, both in coverage of the various technologies and in the quality of the experimental procedures. Some studies are reviewed below, and others are listed in the appendixes of Feachem and others (1980). Unless otherwise stated, it may be assumed that removal of *Salmonella* during sewage treatment processes is similar in nature and degree to removal of *E. coli* (chapter 13).

By primary and secondary sedimentation

Mom and Schaeffer (1940) recorded that an Imhoff tank at Bandung (Indonesia) reduced an influent *S. typhi* concentration of 5,100 per 100 milliliters to an effluent concentration of 800 per 100 milliliters (an 84 percent reduction). Data on the removal of salmonellae by primary and secondary sedimentation at two treatment plants in southern England are given in tables 15-3 and 15-4, below, and are discussed in the section below on conventional treatment. Further literature is cited in the appendixes of Feachem and others (1980).

Although the number of studies reported is small, it may be concluded that *Salmonella* removal during sedimentation is similar to fecal coliform removal (chapter 13) and that salmonellae become concentrated in the sludge, as do all excreted viruses and bacteria undergoing sedimentation.

By septic tanks

Salmonellae, like all enteric pathogens, are not normal residents of the healthy gut and are found only sporadically, and in widely varying concentrations, in sewage treatment systems serving individual buildings or small clusters of buildings. Surprisingly, a survey of seven septic tanks in the USA detected *Salmonella* in 15 percent (4 of 27) of effluent samples even though the average number of people per septic tank was only 3.9 (Small Scale Waste Management Project 1978).

Green and Beard (1938) simulated a septic tank in the laboratory and found that *S. typhi* in the supernatant liquor at 15–21°C declined at a t_{90} rate of 52 hours. Howard, Lloyd and Webber (1975) installed an Oxfam sanitation unit (two large flexible septic tanks in series) in the yard of the cholera hospital in Dacca (Bangladesh). At a mean retention time of 8 days (4 days in each tank), and sewage temperatures of 22–32°C, the 2,900 *Salmonella* per 100 milliliters of influent were reduced by 98.8 percent (t_{90} = 100 hours).

From the data on survival in sewage reviewed above and in the appendixes of Feachem and others (1980), it may be concluded that *Salmonella* removal in septic tanks is unlikely to exceed 95 percent and will be very much less in an overloaded or sludge-filled unit.

By conventional treatment

Perhaps the most detailed study on *Salmonella* removal in conventional treatment plants is that on the trickling filter plant at Woking and the activated sludge plant at Guildford (both in the county of Surrey in southern England) reported by Yaziz and Lloyd (1979). The results are summarized in tables 15-3 and 15-4. Whereas the efficiencies of the two primary sedimentation systems are similar (79 percent removal at Woking, 73 percent at Guildford), the performance of the activated sludge plant is very much better in terms of *Salmonella* removal than that of the trickling filter plant. This difference results in a total removal of only 93 percent (1.2 log units) of *Salmonella* at Woking compared with 99.86 percent (2.9 log units) at Guildford. Judging from the extremely good BOD_5 removal performance of the two plants, they were in good operating order and were not overloaded. It is likely that the *Salmonella* removal rates reported are close to the maximum achievable by full scale plants employing conventional trickling filter and activated sludge processes. Many plants that are poorly designed or maintained, or that are overloaded, will achieve considerably lower removal rates.

Studies on four treatment plants in Holland showed overall *Salmonella* removals of 90 percent, 94 percent and 79 percent in three trickling filter plants and 99.4 percent in an activated sludge plant (Kampelmacher, Fonds and van Noorle Jansen 1977 and Kampelmacher and van Noorle Jansen 1970). Early laboratory studies on *S. typhi* showed a 90–99 percent reduction after 6 hours aeration in activated sludge and 95–99 percent reduction by trickling filters (Green and Beard 1938). Green and Beard (1938), like other early workers, were impressed by removal efficiencies of 95 percent and wrote that conventional treatment "may be expected to reduce greatly typhoid organisms present in sewage, and are, therefore, effective barriers for the protection of public health." This view, although widely held, was and is erroneous. It is only more recently that researchers have emphasized that, with high influent concentrations, 90–99 percent removal rates are poor and that, in any case, most of those *Salmonella* removed from the liquor are

Table 15-3. Salmonella *removal at the Woking trickling filter plant, UK*

	Salmonella *per 100 milliliters*			*Percentage reduction*		
Sewage and process	*Minimum*	*Maximum*	*Median*	*Minimum*	*Maximum*	*Mean*
Raw sewage[a]	11	1600	170			
Primary sedimentation[b]				29	99.00	79[c]
Settled sewage	1	160	20			
Trickling filters				−92.3[d]	91.7	68
Trickling filters plus secondary sedimentation				86	99.6	92.3
Final effluent[e]	0	250	3			
Total plant				64	100	93.0

Source: Adapted from Yaziz and Lloyd (1979).
a. BOD_5 = 194 milligrams per liter. Total flow = 7,600–12,100 cubic meters per day.
b. 6–7 hours' detention.
c. *Salmonella* removal correlated with suspended solids removals (r = 0.82).
d. Increase of 92.3 percent.
e. BOD_5 = 7 milligrams per liter.

concentrated in the sludge, which then presents its own treatment and disposal problems.

Kabler (1959) reviewed several studies on *S. typhi* removal by conventional treatment. These, plus other studies mentioned above and in the appendixes of Feachem and others (1980), indicate removal by trickling filter plants of 75–95 percent and by activated sludge plants of 90–99.9 percent. In other words, *Salmonella* removal by these processes is similar to the removal of fecal coliforms (chapter 13).

By oxidation ditch

Will, Diesch and Pomeroy (1973) studied *S. typhimurium* in a 1:10 scale model oxidation ditch treating cattle slurry (0.5–1.0 percent solids). Maximum survival times were 17 days under simulated summer conditions (20°C) and 47 days under simulated winter conditions (2°C). When oxidation ditch effluent was held in a settling chamber, *S. typhimurium* survived for 66 days in the liquid layer and 87 days in the sludge layer at 2–3°C.

Oxidation ditches are commonly used in Holland to treat the sewage of small communities, and two were studied by Kampelmacher and van Noorle Jansen (1973). *Salmonella* concentrations per 100 milliliters ranged from 23 to 2,400 in the influent and from 0 to 350 in the effluent. *Salmonella* reductions of 90–99 percent were recorded. Kampelmacher and van Noorle Jansen (1971) also studied an oxidation ditch treating pig wastes in Holland. Concentrations of *Salmonella* per

Table 15-4. Salmonella *removal at the Guildford activated sludge plant, UK*

	Salmonella *per 100 milliliters*			*Percentage removal*		
Sewage and process	*Minimum*	*Maximum*	*Median*	*Minimum*	*Maximum*	*Mean*
Raw sewage[a]	20	>1,800	130			
Primary sedimentation[b]				35	96.9	73[c]
Settled sewage	7	250	35			
Activated sludge[d] plus secondary sedimentation				93.6	100	98.7
Final effluent[e]	0	1.7	0.1			
Total plant				98.7	100	99.86

Source: Adapted from Yaziz and Lloyd (1979).
a. BOD_5 = 259 milligrams per liter. Total flow = 7,600–22,800 cubic meters per day.
b. 6–7 hours' detention.
c. *Salmonella* removal correlated with suspended solids removal (r = 0.65).
d. 6–9 hours' detention in the activated sludge tanks.
e. BOD_5 = 8 milligrams per liter.

100 milliliters were 33–1,600 in the influent, 3–75 in the ditch, and 1–12 in the effluent.

By waste stabilization ponds

Joshi, Parhad and Rao (1972) studied bacterial removal in a series of three ponds, with total retention time of 7 days, near Nagpur (India). *Salmonella* concentrations in the influent were 4–540 per 100 milliliters, and none was detected in the effluent. The reductions of coliforms, *E. coli*, and fecal streptococci in the same ponds were in the range 99–99.9999 percent. In a subsequent study (Joshi, Parhad and Rao 1973) on two ponds with 12 days total retention in Nagpur, *Salmonella* were present in the influent (3–100 per 100 milliliters) and in all effluent samples (qualitative determinations only). Reductions of indicator bacteria were only 43–98 percent. The striking difference in performance between the two sets of ponds was attributed to short-circuiting and poorly designed interpond connections in the second set of ponds. Two ponds in series in Lima (Peru), with total retention of 37 days and temperatures of 18–27°C, yielded salmonellae (mainly *paratyphi* B, *derby* and *newport*) in all influent and effluent samples (Yánez 1980). This must reflect poor pond design and major short-circuiting.

As far as is known, the processes affecting salmonellae removal in ponds are the same as those determining the removal of indicator bacteria (chapter 13). One study suggested that *Salmonella* death rates in ponds were similar to those of *E. coli* (Davis and Gloyna 1972). More commonly it has been found that *Salmonella* reduction is significantly less than that of coliforms, *E. coli*, or fecal streptococci in the same ponds (table 13-3; Coetzee and Fourie 1965; Walker, Carbonnelle and Leclerc 1977). The removal rate for salmonellae in ponds is very temperature dependent: higher removal is obtained in summer than in winter (for instance, Slanetz and others 1970), and ponds in the tropics remove salmonellae more effectively than those in temperate climates.

By tertiary treatment

The realization that salmonellae, like all excreted viruses and bacteria, may be present in moderately high concentrations in the secondary effluents of conventional sewage treatment plants has stimulated some research into *Salmonella* removal by tertiary processes. In the absence of other information it may be assumed that *Salmonella* behave like fecal coliforms during tertiary treatment (chapter 13).

LAGOONING. Lagooning of secondary effluents will remove salmonellae if retention times are long enough. Removal will be greatly enhanced at warmer temperatures. The secondary effluent from an activated sludge plant in London (England) was held in three lagoons in series with a total retention time of 17 days (Metropolitan Water Board 1963–64). The lagoon influent contained 0.2–29 salmonellae per 100 milliliters, while the effluent contained 0–0.2 per 100 milliliters.

DISINFECTION. Brezenski, Russomanno and DeFalco (1965) studied the effluents and receiving waters of four sewage treatment plants discharging into Raritan Bay (New York–New Jersey, USA). The treatment plants incorporated primary treatment (sedimentation) and chlorination (0.25–2.5 milligrams per liter of combined chlorine residual). When effluents were chlorinated, no salmonellae were isolated from the effluents or receiving waters. When chlorination was suspended for 1 week, salmonellae were recovered from two of the effluents and from the receiving waters.

Kampelmacher, Fonds and van Noorle Jansen (1977) studied three sewage treatment plants in Holland discharging effluents into lakes used for recreation during the summer. Plant 1 had trickling filters followed by chlorination (6 milligrams per liter chlorine added); plant 2 had trickling filters, aeration, $FeCl_3$ addition for phosphate removal, and chlorination (2 milligrams per liter chlorine added); and plant 3 had activated sludge and chlorination (2 milligrams per liter of chlorine added). Reductions of *Salmonella* by biological treatment (influent compared with effluent prior to chlorination) were 94 percent in plant 1, 79 percent in plant 2, and 99.4 percent in plant 3. *Salmonella* concentrations per 100 milliliters were $2->10^4$ in influents, $0–10^4$ before chlorination, $0–10^3$ after chlorination, and $0–10^3$ in the receiving lake. Ninety-two percent (68 of 74) of samples contained *Salmonella* before chlorination, whereas only 17 percent (9 of 52) were positive afterward.

Schiemann, Brodsky and Ciebin (1978) compared three methods of disinfection of an activated sludge plant effluent in Ontario (Canada) with respect to their ability to remove wild *Salmonella*. *Salmonella* were recovered from 89 percent of primary effluent samples, 73 percent of secondary effluent samples, 13 percent of chlorine dioxide treated effluent samples, 8 percent of ozonated effluent samples, and 0 percent of chlorinated effluent samples.

Oliver and Carey (1976) reviewed data showing that *S. typhi* was inactivated by chlorine and ultraviolet light to the same degree as *E. coli* but was considerably

more resistant to ozone. This finding was not supported by Burleson, Murray and Pollard (1975), who found that *S. typhimurium* and *E. coli* had a similar response to ozone, both in saline and secondary effluent.

LAND TREATMENT. Near Phoenix (Arizona, USA) secondary effluent from an activated sludge plant was treated by intermittent flooding onto soil basins (Gilbert and others 1976). Secondary effluent contained, on average, 21 *Salmonella* per 100 milliliters, and no *Salmonella* were detected from ground water 9 meters under the site. The *Salmonella* had been removed by percolation of wastewater through 1 meter of fine loamy sand and 8 meters of sand and gravel.

Inactivation by Night Soil and Sludge Treatment Processes

The realization that 70–99 percent of salmonellae entering a sewage treatment plant are concentrated in the sludge, and the widespread use of sludge in agriculture, have stimulated an increasing amount of research on the fate of salmonellae during sludge treatment. More research is urgently needed on salmonellae in simple sludge treatment processes (storing, drying, and composting) in warm climates and on the fate of salmonellae in night soil treatment and disposal systems in developing countries.

By pit latrines

Galvagno and Calderini (1908) reported *S. typhi* survival in pit latrines for 15–30 days. Survival may be estimated from reports on survival in feces, night soil, sludge, and slurry (see above and the appendixes of Feachem and others 1980).

By storage

Sludges stored for long periods will normally be free from *Salmonella*. In Yorkshire (England) lagooned raw sludges over 1 year old were negative or gave low counts commensurate with possible recontamination (Fennell 1977). Jones, P. (1977) found *Salmonella* in 50 percent of lagooned sludge samples less than 2 years old, but in no samples more than 2 years old. Jones (1975) reported that *S. dublin* underwent no loss of virulence when stored in cattle slurry for 1 month at 10°C.

By anaerobic digestion

Pike (1981) reviewed data on *Salmonella* removal by sludge digestion at various sewage treatment plants in England and Wales. Reductions in *Salmonella* concentrations were between 16 and 98 percent. Two mesophilic digesters in Yorkshire (England), with mean retention times of 30 days, were receiving on average 5,900 and 2,500 *Salmonella* per 100 milliliters and putting out on average 3,600 and 37 *Salmonella* per 100 milliliters—thus achieving very different removal efficiencies of 39 and 98.5 percent, respectively (Fennell 1977). Stokes and others (1945) found that *S. typhimurium* could be detected in sludge after 45 days anaerobic digestion at 26°C.

Obrist (1979) reported that approximately 70 percent of the 2 million cubic meters of municipal sewage sludge produced annually in Switzerland is applied to land, and of this 52 percent is applied to pasture and forage crops. Raw sludge contained *Salmonella* in 91 percent of samples, with maximum and mean concentrations being 10^6 and 10^4 per 100 milliliters respectively. Digested sludge contained *Salmonella* in 81 percent of samples, with maximum and mean concentrations being 10^5 and 10^2 per 100 milliliters respectively.

Dudley and others (1980) studied the bacterial content of digested sludges from three sewage treatment plants in the southern USA. *Salmonella* concentrations were 200–2,400 per 100 milliliters. Cooke, Thackston and Malaney (1978) studied *Salmonella* removal by three anaerobic mesophilic digesters at sewage treatment plants near Nashville (Tennessee, USA). Digester 1 had a mean retention time of 9 days, operated at 34°C, and reduced *Salmonella* concentration by 98 percent. Digester 2 had a retention time of 50 days, operated at 37°C, and reduced *Salmonella* concentrations by 99.4 percent. Digester 3 had a retention time of 38 days, operated at 36°C, and removed 99.9 percent of salmonellae.

Mom and Schaeffer (1940) studied sludge digestion in an Imhoff tank in Bandung (Indonesia). Raw sludge contained 0–2,400 *S. typhi* per 100 milliliters; after 30 days digestion at 27°C, the concentration was 0–2,200 per 100 milliliters.

Clearly, mesophilic digestion produces a sludge that may still retain a considerable population of *Salmonella*. Thermophilic anaerobic digestion at around 50°C will certainly eliminate *Salmonella* if operated as a batch process with a retention time of 5 days or more. Under continuous feed *Salmonella* elimination can never be guaranteed, although concentrations in the digested sludge should be very low.

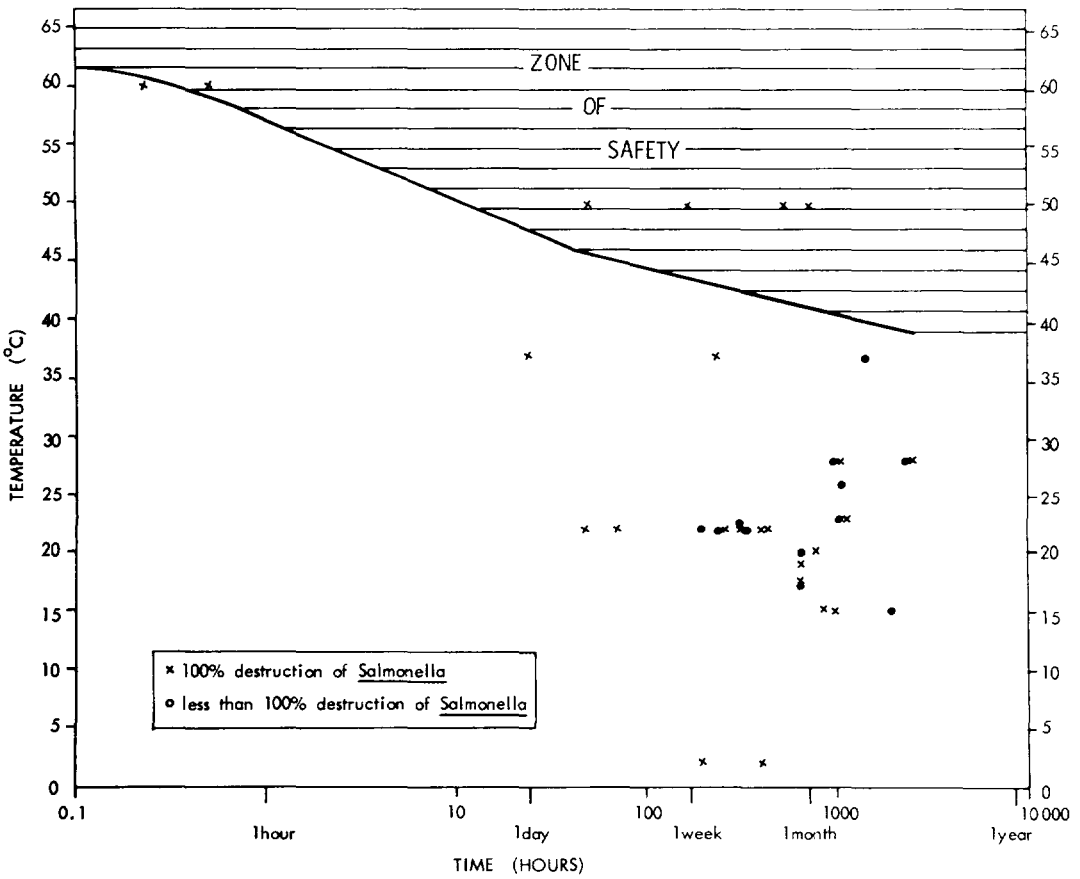

Figure 15-2. *The influence of time and temperature on salmonellae.* The points plotted are the results of experiments done under widely differing conditions. The line drawn represents a conservative upper boundary for death

By aerobic digestion

Thermophilic aerobic digestion (wet composting) is similar to composting in its effect upon enteric bacteria. If temperatures are maintained at the required level for the required time (figure 15-2) throughout the sludge mass, total destruction of *Salmonella* will occur. Smith, Young and Dean (1975) reported reductions in *Salmonella* of 99.8 percent in one aerobic digester (temperature 56°C, retention 4 days) and 100 percent in another (temperature 49°C, retention 4 days).

Aeration of stored slurry is used on some farms to promote bacterial dieoff. Willinger and Thiemann (1978) found that *S. typhimurium* survived for 6–12 days in continuously aerated cattle slurry in Austria. Average ambient temperature was 3–24°C, whereas the temperature in the aerated slurry was 29–35°C. *E. coli* survival in the same experiments was 9–12 days.

By drying

Pike (1981) reported that raw and aerobically digested sludges on drying beds in England maintained their levels of *Salmonella* contamination for up to 85 days. Stokes and others (1945) inoculated sludge (5.8 percent solids) with 2.5×10^9 *S. paratyphi* B per 100 milliliters and found that they were detectable after 27 days (solids content 36 percent), but not after 41 days (solids content 43 percent), on sludge drying beds in England during the summer. In similar experiments conducted during December–June with *S. typhimurium*, the bacteria could be detected after 180 days on the drying beds, by which time the solids content had risen to 86 percent.

By heating

All high temperature processes, such as pasteurization, wet oxidation, and the Porteous process, will eliminate *Salmonella* from sludge. Pasteurization is commonly used in Switzerland to treat digested or aerobically stabilized sludges prior to land application (Obrist 1979). Sludge is heated to 70°C for 30 minutes by steam injection. Hess and Breer (1975) reported that pasteurized sludges from five treatment plants

contained less than 10 Enterobacteriaceae per gram in 98–100 percent of samples.

In many situations, and especially in developing countries, the more practical and appropriate technologies for harnessing heat to destroy pathogens in sludge are aerobic thermophilic digestion (see above) and composting (see below). The time-temperature requirements for the destruction of salmonellae are shown in figure 15-2, and it may be seen that 1 hour at 60°C, 1 day at 50°C, and 1 week at 45°C are lethal combinations.

By composting

In laboratory experiments on composting poultry excreta, *S. typhimurium* were eliminated after 19 hours of composting during which the temperature rose to 64°C at hour 10 (Platz 1978). Salmonellae were more readily eliminated by composting when inoculated into the compost than when held in sealed glass ampoules within the compost, and this indicates that factors other than heat were contributing to bacterial death. In both cases *Salmonella* could not be detected after temperatures had risen to about 62°C.

Wiley and Westerberg (1969) determined that *S. newport* in nutrient broth was destroyed in 40 minutes at 60°C and in 30 minutes at 65°C. Dewatered primary sludge, containing 2.4×10^6 *S. newport* per gram, was then treated for 5 days in a continuously mixed, forced air, laboratory composter containing 1.1 cubic meters of sludge. The temperature within the composter was 60–76°C. *S. newport* was not detectable in the sludge after 25 hours of composting.

Savage, Chase and MacMillan (1973) experimented with various regimes for composting pig wastes (a combination of uneaten garbage and pig feces) in New Jersey (USA). In a windrow comprising 36 tons of pig waste that was turned twice per week, *Salmonella* concentrations rose from day 0 (temperature within windrow 36°C) to day 40 (temperature 48°C) and then declined to negligible levels by day 187 (temperature 68°C). In another windrow, comprising 36 tons of pig waste plus 1.4 tons of straw, turned 20 times per week, temperatures rose to 72°C within 10 days and remained above 60°C for 20 days. Coliforms, and presumably also salmonellae, were eliminated within 14 days.

The effect on enteroviruses and coliforms of the composting experiments at Beltsville (Maryland, USA) are described in chapters 9 and 13. In windrows of sludge and woodchips turned daily, salmonellae grew at first but were eliminated after 14 days. In piles of sludge and woodchips subjected to forced aeration, salmonellae again increased initially but were undetectable after 10 days (Burge, Cramer and Epstein 1978; Kawata, Cramer and Burge 1977). The importance of management and process control are illustrated by the poor *Salmonella* removal properties of a digested sludge plus sawdust composting plant at El Paso (Texas, USA) reported by Reeves (1959).

Samples of compost from Vietnamese double-vault composting toilets (retention times 6–7 weeks) have been found not to contain *Salmonella* (Nimpuno, personal communication). These results must be treated with caution, however, because they may result from studies of toilets operating under controlled, experimental conditions.

To eliminate salmonellae, all parts of the composting mass have to be brought to a warm enough temperature for a long enough time (figure 15-2). This generally requires the presence of a carbon source (refuse, straw, or woodchips), careful moisture control, and a supply of oxygen throughout the mass provided by turning or forced aeration.

By coagulation and vacuum filtration

Sludge coagulation followed by vacuum filtration is commonly used to dewater sludges in some developed countries. Kampelmacher and van Noorle Jansen (1972) studied three treatment plants in Holland; two plants added lime and ferrous sulfate prior to vacuum filtration, whereas the third used lime and ferric chloride. The solids contents were 4–10 percent and 25–30 percent, respectively, before and after dewatering. At the three plants raw sludge contained salmonellae in 59, 65, and 100 percent of samples, whereas dewatered sludge contained salmonellae in only 5, 5, and 14 percent of samples. The bactericidal properties of this process are due to the addition of coagulants and especially to their action of raising the pH.

By lime treatment

Any sludge treatment process involving the addition of lime is likely to produce a sludge free from salmonellae. Pike (1981) reviewed sludge treatment data for England and Wales and found that lime treatment was highly effective in removing *Salmonella*.

By irradiation

Experiments in the Federal Republic of Germany showed that 10^2–10^3 wild *Salmonella* in 100 milliliters of raw sludge could be reduced to zero by the

application of 3 kilogray (Lessel and Suess 1978). Hess and Breer (1975) concluded that a dose of 3 kilogray has an effect on Enterobacteriaceae in sludge similar to heating to 70°C for 30 minutes. *Salmonella* in sludge and in composted sludge (60 percent solids) were reduced by approximately 1 log unit for each 300 gray of ionizing radiation applied (Brandon, Burge and Enkiri 1977). Similarly, White (1979) reported inactivation of coliforms at a rate of 1 log unit per 200 gray at 20°C and inactivation of *Salmonella* at a rate of 1 log unit per 300 gray at 23°C.

It is clear that the radiation doses necessary to eliminate *Salmonella* are similar to those required for other Gram-negative enteric bacteria (Osborn and Hattingh 1978) and are about one-tenth of the doses required to inactivate enteroviruses (2.5–5 kilogray for a 1 log unit reduction—see chapter 9). Therefore, ionizing radiation treatment designed to inactivate viruses will certainly eliminate *Salmonella*.

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