Revista Brasileira de Ciência Avícola
ISSN: 1516-635X
revista@facta.org.br
Fundação APINCO de Ciência e Tecnologia Avícolas
Brasil

Brazilian Journal of Poultry Science
Revista Brasileira de Ciência Avícola

ISSN 1516-635X Jan - Mar 2010 / v.12 / n.1 / 01 - 11

Sources of Human Non-Typhoid Salmonellosis: A Review

Author(s)

Freitas Neto OC de[1]
Penha Filho RAC[2]
Barrow P[3]
Berchieri Junior A[4]

[1,2,4] Universidade Estadual Paulista "Julio de Mesquita Filho"- campus de Jaboticabal Departamento de Patologia Veterinária.
[3] The University of Nottingham School of Veterinary Medicine and Science

Mail Address

E-mail: oliveirocaetano@yahoo.com.br

Keywords

Antimicrobial resistance, beef, food, pork, poultry, *Salmonella* spp., vegetables,

Arrived: January/2010
Approved: March/2010

ABSTRACT

Salmonellosis is a worldwide disease caused by bacteria of the genus *Salmonella*. Currently, there are over 2,500 identified serovars of *Salmonella*. A reduced number of these serovars, about eighty, are implicated in most animals and human diseases. Most cases of salmonellosis in humans are associated with the consumption of contaminated food products such as beef, pork, poultry meat, eggs, vegetables, juices and other kind of foods. It may also be associated with the contact between humans and infected pet animals. Therefore, the chain of human salmonellosis is very complex and in most cases the origin of the infection is difficult to establish. The use of antimicrobial agents to treat and to prevent bacterial infections in humans and animals, as well as as growth promoters in animal production, has favoured the selection and transference of resistance genes between different bacteria, including *Salmonella* serovars. Many studies have confirmed the role of foods of animal origin as a source of multi drug-resistant *Salmonella* serovars. For this reason, continuous surveillance of these pathogens along the food chain together with the responsible use of antimicrobial agents is necessary.

BACKGROUND

The bacteria of the genus *Salmonella* are spread worldwide and are responsible for illnesses in human beings and animals. Currently, the genus is divided into two species, *Salmonella enterica* and *Salmonella bongori* (Popoff & Minor 2001). *S. enterica* is divided into six subspecies (*enterica*, *salamae*, *arizonae*, *diarizonae*, *houtenae* and *indica*), each of which has several serovars or serotypes (Table 1). Thus, a serotype is nominated in the following way: *Salmonella enterica* subspecies *enterica* serotype Typhimurium, which may be simplified as *Salmonella* Typhimurium. Nowadays, more than 2,500 serotypes are known and most of them (almost 1,500) belong to subspecies *enterica* (Porwollik *et al*., 2004; CDC, 2007c).

Table 1. Distribution of serovars by *Salmonella* species.

Species	***S. enterica***						***S. bongori***
Subspecies	*enterica*	*salamae*	*arizonae*	*diarizonae*	*houtenae*	*indica*	
Number of Serovars	1490	500	94	329	72	12	22

Salmonellosis is the disease caused by *Salmonella* serovars. A few serotypes are host-specific. *Salmonella* Pullorum and Gallinarum are responsible for pullorum disease and fowl typhoid in poultry, respectively, *S.* Typhi for typhoid fever in human beings, *S.* Dublin for disease in bovine, etc. However, the others are not host-specific and may infect several animal species including human beings. These serotypes are generally responsible for food-borne diseases and foods of animal origin are the main source. Historically, *S.* Typhimurium is the most common agent of human food-borne disease, although in the last few decades *S.* Enteritidis has become more common (Braden, 2006; Fernandes *et al*., 2006). Among all the 2,500 serovars, about eighty are most frequently involved in animal and human salmonellosis. *S.* Typhimurium and *S.* Enteritidis are the most common agents of disease in human beings and animals, but there is also increasing concern about the Infantis, Agona, Hadar, Heidelberg, and Virchow *Salmonella* serotypes.

Salmonellosis is one of the most important human food-borne diseases. Improvements in the quality of life as a result of hygienic conditions and sewage treatment have helped to prevent typhoid fever, and its occurrence is now frequently related to bad weather conditions like storms resulting in contamination of drinking water with sewage. The world population has increased dramatically, and to address the demand for food it has been necessary to increase production in large scale intensive farms. In these conditions, enteric pathogens like *Salmonella* serovars that have non-specific hosts, once introduced on farms may easily become disseminated among animals, resulting in persistence. Once this happens, it becomes almost impossible to eradicate the pathogens from farms and to eliminate them from products used as food or for preparing foods. To give an idea of the complexity of the chain of food-borne salmonellosis, at the beginning of the 1970s, many cases of human salmonellosis caused by *Salmonella* Agona in Europe and in the United States of America were associated with the ingestion of broiler meat from flocks fed with feed containing fish meal from Peru (Clark *et al*., 1973). Since the 1980s, worldwide outbreaks of human salmonellosis have been caused by *S.* Enteritidis present in eggs and contaminated broiler meal. In this case, breeders and laying hens were often thought to be infected by rodent feces and urine and transmitted the bacterium vertically, resulting in infected progeny. In addition, during the same period, outbreaks of human salmonellosis, resulting from ingestion of pork and beef meat contaminated with *Salmonella* serovars, have been reported in many countries (CDC, 1981; Spitalny *et al*., 1984; Delpech *et al*., 1998), with *S.* Typhimurium being the main serotype involved. Pork and beef were probably contaminated during the slaughter process followed by inadequate cooking.

Despite the strong link between food of animal origin and human salmonellosis, people may be infected by other ways, such as by cross contamination at home and commercial kitchens, through contact with other people, pets - especially dogs, cats and reptiles, as well as by ingestion of vegetables and fruits. This review attempts to provide information about different sources of *Salmonella* serovars which may be involved in human salmonellosis outbreaks. In addition, a basic knowledge about antimicrobial resistance in *Salmonella* serovars isolated from foods of animal origin is also presented since many drugs are used frequently in animal production.

Red meat and red-meat products as a source of Salmonella serovars for human beings

Red meat and red-meat products are recognized as an important source of human salmonellosis. Only in 2007, for instance, beef and pork (and products thereof) contaminated with *Salmonella* serovars were responsible for 800 cases of human salmonellosis involving 37 outbreaks in Europe (EFSA, 2009).

Additionally, in 2005, Hess *et al*. (2008) reported that lamb's liver was responsible for an outbreak of *S.* Typhimurium phage type 197 in Australia. In Germany, from 2001 to 2005, epidemiological studies, microbiological testing, and trace-back investigations showed that pork and pork products were involved in human salmonellosis outbreaks (Jansen *et al*., 2007). Moreover, in Italy, an outbreak of *S.* Typhimurium phage type DT 104A involving 63 cases was reported. In this study, a case-control study was conducted and data were analyzed by multivariate analysis. No food samples were available for testing; however, in northern Italy, two months prior to the outbreak, the veterinary surveillance system identified the first isolation of *S.* Typhimurium DT104A in a pig. Both human (patients) and pig isolates showed indistinguishable pulsed-field gel electrophoresis (PFGE) patterns. It was not possible to trace the pig after the sample collection. The results of the case-control study suggested that the consumption of pork salami was associated with this outbreak, underlining the importance of good manufacturing practices for ready-to-eat foods (Luzzi *et al*., 2007).

In Denmark an outbreak of human salmonellosis caused by *S.* Typhimurium DT 104 occurred between July and August 2005, and the comparison of strains from patients and food using PFGE, tandem repeat analysis (MLVA), and antibiogram tests showed that the source of the outbreak was imported beef served as carpaccio (Ethelberg *et al*., 2007). Some months later there was an increase in the number of *S.*

Typhimurium DT 104 cases in the Netherlands (Kivi *et al*., 2007). These occurrences were found to be epidemiologically associated with the disease in Denmark. A case-control study was conducted with 56 cases and 100 controls, and logistic regression analysis was used for analyzing the results (Ethelberg *et al*., 2007). From this investigation beef was considered the most probable vehicle of *S.* Typhimurium DT 104. Complementary assessment suggested that imported beef from a third European country had spread this *Salmonella* strain resulting in these outbreaks.

In the USA in 2004, an epidemiological investigation demonstrated that ground beef sold by a national chain of one supermarket was the source of food-borne salmonellosis due to *S.* Typhimurium in nine states. The PFGE patterns of *S.* Typhimurium isolates from patients and ground beef were found to be identical (CDC, 2006).

Many other reports involving human salmonellosis outbreaks associated with consumption of red meat have been recorded in the literature (CDC, 1995a; Davies *et al*., 1996; Roels *et al*., 1997; Shapiro *et al*., 1999; Haeghebaert *et al*., 2001). In most of cases, the disease was associated with the consumption of contaminated meat or as a result of incorrect or inadequate cooking.

Human non-typhoid salmonellosis caused by the ingestion of dairy products

Salmonella serovars are widespread in the environment and may contaminate a variety of food and food ingredients. Since dairy products are from animal origin, they may also be implicated in human food-borne salmonellosis. Usually milk and milk products are submitted to pasteurization, which kills *Salmonella* serovars. Milk-borne salmonellosis is therefore often related to the consumption of raw or inadequately pasteurized milk. However, *Salmonella* serovars may also contaminate dairy products after the pasteurization process.

S. Typhimurium caused an outbreak in Pennsylvania, USA, in 2007, through the consumption of raw milk. *S.* Typhimurium isolated from human and milk exhibited suspect identical PFGE patterns. Later, an epidemiological investigation confirmed that the source of *S.* Typhimurium was the raw milk and raw milk products from a dairy (CDC, 2007e). An outbreak of *S.* Newport in the USA in 2008 occurred predominantly among Hispanics in northeastern Illinois. *S.* Newport was recovered from patients and from a Mexican-style aged cheese made with unpasteurized milk (CDC, 2008c). The strains isolated from people and cheese had indistinguishable PFGE patterns. Another outbreak of human salmonellosis associated with cheese was described in Switzerland, Europe, where a nationwide outbreak of gastrointestinal disease caused by *S.* Stanley, a rare serovar in Europe, occurred from September 2006 to February 2007. *S.* Stanley strain was isolated from a local soft cheese and from 94% of affected people. The withdrawal of the cheese from the market resolved the problem (Pastore *et al*., 2008).

In view of the great number of human salmonellosis associated to the consumption of powdered milk products registered in the 1960's and 1970's, *Salmonella* control criteria were included in the Codex of Hygienic Practice for Foods for infants and children (Codex, 1979). Between 1985 and 2005 at least six outbreaks of salmonellosis, involving up to 250 infants, have been associated with powdered infant formula. For example, in 2005 in France, an outbreak affecting more than 100 infants was associated with powdered infant formula contaminated with *S.* Agona. Many of these outbreaks were identified because the *Salmonella* strains were unique in some way (e.g. a rare serovar). Another common feature of the outbreaks was the low level of *Salmonella* contamination detected in the implicated formula (*Salmonella* serovars may be missed in routine testing). These outbreaks likely represent only a small proportion of the actual number of *Salmonella* infections in infants that have been linked to powdered infant formula (Toyofuku *et al*., 2006; Cahill *et al*., 2008).

Salmonella serovars in foods from vegetable origin: a matter of increasing concern

In recent years, the importance of foods of vegetable origin as potential vehicles of gastrointestinal infection has been highlighted. During production, storage or even in retail outlets, *Salmonella* serovars may contaminate vegetables, fruits, juices, and, as they are usually consumed raw, they may also be implicated in human salmonellosis.

Unpasteurized orange juice was responsible for food-borne salmonellosis in 152 people in six states in the USA between May and July 2005 (Jain *et al*., 2009). Vojdani *et al*. (2008) reviewed fruit juice-associated outbreaks of illness reported to Centers for Disease Control and Prevention (CDC), in Atlanta, USA. From 1995 through 2005, 21 juice-associated outbreaks were reported to CDC; ten implicated apple juice or cider, eight were linked to orange juice, and three implicated other types of fruit juice. These outbreaks caused 1,366 illnesses, with a average of 21 cases per outbreak (range, 2 to 398 cases). Among the 13 outbreaks of known etiology, five were caused by *Salmonella* serovars.

Three cases of *S.* Thompson infection were registered by the Norwegian reference laboratory in November 2004. The results of the investigation indicated that the consumption of rucola lettuce and mixed salad were responsible for the outbreak (Nygård *et al*., 2008).

In addition, Little & Gillespie (2008) described general outbreaks of infectious intestinal disease linked to prepared salads reported in England and Wales from 1992 to 2006, as well as European outbreaks associated with prepared salads as a result of international trade. According to the authors, around 23% of these outbreaks were attributed to *Salmonella* serovars. Cross-contamination, infected food handler or inappropriate storage were the most commonly factors associated with vegetable contamination.

In 2002 tomatoes, grown and packed in Virginia state (USA), contaminated with *S.* Newport, caused illness in 510 patients in 26 other states (Kretsinger *et al*., 2003). Later, in July-November 2005, the same strain (confirmed by PFGE) caused illness in at least 72 patients in 16 states of the USA. A case-control study during the 2005 outbreak confirmed tomatoes as the source. The tomatoes were traced back to Virginia, where the *S.* Newport strain was isolated from pond water used to irrigate tomato fields, suggesting persistent contamination of the fields (Greene *et al*., 2008). In addition, during 2005-2006, three more outbreaks of *Salmonella* infections associated with eating tomatoes were detected in the USA and Canada. These outbreaks resulted in 387 culture-confirmed cases of salmonellosis, with isolation of *S.* Newport, *S.* Braenderup, and *S.* Typhimurium (CDC, 2007b).

Unicomb *et al*. (2005) described human salmonellosis outbreaks associated with sesame seed-based food. In November 2002, an outbreak of *S.* Montevideo occured in New South Wales, Australia. Infected persons were interviewed, and epidemiologically linked retail outlets inspected. Imported "tahini", a sesame seed-based food, was rapidly identified as the source of infection. A second outbreak was identified in Australia in June-July 2003 and another in New Zealand in August 2003. Of a total of 68 patients infected by *S.* Montevideo, 66 were contacted. Fifty-four (82%) reported consumption of sesame seed-based foods. Laboratory analyses demonstrated closely related PFGE patterns in the *S.* Montevideo isolates from human cases and sesame-based foods imported from two countries.

Although it is known that fresh salad vegetables, herbs, seeds and fruit may become contaminated from environmental sources, they have been associated only recently with food-borne diseases. Nowadays, the role of vegetables as vehicle for pathogens transmission has become clear and recurrent, demonstrating that major health problems can arise from consumption of contaminated vegetables if hygiene practices break down.

Pets as a source of Salmonella infection for human beings

Many pet animals can harbor and shed *Salmonella* serovars. The close contact between pets and people, especially children and immunosuppressed people, can result in human salmonellosis. Animals commonly infected with *Salmonella* serovars which may pose a risk to humans include amphibians, birds, cats, dogs, fish, guinea pigs, hamsters, horses, mice, rabbits, lizards, snakes, and turtles, although, according to literature reports, the last three (lizards, snakes, and turtles) are responsible for the majority of human salmonellosis outbreaks associated with pets (CDC, 2005; Bruins *et al*., 2006; Corrente *et al*., 2006; CDC, 2007f; Bertrand *et al*., 2008; CDC, 2008a).

The association of reptiles with human salmonellosis was first described in the 1950s (Boycott *et al*., 1953). Since then, reports have become more frequent. According to Mermin *et al*. (2004), approximately 1.4 million human cases of *Salmonella* infection occur each year in the USA and it has been estimated that 74,000 are the result of exposure to pet reptiles and amphibians. From 1994 to 1995, thirteen health state agencies in the USA reported to CDC human infection cases caused by uncommon *Salmonella* serovars, in which the patients had had direct or indirect contact with reptiles (snakes, lizards and turtles). In most cases, the same *Salmonella* serovar was isolated from the patient and from the reptile (CDC, 1995b). In 2005, at least six cases of human salmonellosis were associated with pet turtles in two states of the USA (CDC, 2005). Also in the USA, in October 2007, the North Carolina Division of Public Health notified to CDC human infections caused by *S.* Paratyphi B var. Java in several states. The results of the epidemiological and laboratory investigations showed that many of these infections occurred in young children and were associated with exposure to small turtles (CDC, 2008a).

A survey with the purpose of estimating the role of pet reptiles in human salmonellosis was done in Europe (Bertrand *et al*., 2008). In one study, in 2007-2008 seven human salmonellosis cases in Belgian were described which were related to contact with turtles and snakes. The same survey reported three reptile-associated cases of *Salmonella* infection in the past three years in France: two cases in 2005 and one, imported from China, in 2006. The patients were all young children, aged eight months, three years and four years, respectively. They were infected with a multi-resistant strain of *S.* Typhimurium. The first two cases had contact with, respectively, a snake and an iguana; in the third case an indirect link to a turtle (consumption of turtle soup) was found. At least 14 cases of human salmonellosis associated with reptile contact were identified in Ireland between 2005 and 2008, and 31 cases, also associated with reptiles, were reported in Germany during 2006-2008. Although infections in adults from contact with reptiles have been reported, in most cases infants less than one year old are affected.

In most countries, although cases of salmonellosis are reported within the national surveillance system, the source of infection is not routinely given, and the possible exposure to pet animals is usually only revealed in additional epidemiological investigations. Thus, the true number of cases due to direct or indirect contact with pet animals is likely underestimated.

Other sources of *Salmonella* for human beings

In some cases, salmonellosis may be associated with uncommon sources. It is frequently the result of factors including the ubiquitous distribution of *Salmonella* serovars, usage of contaminated ingredients, incorrect cleaning or disinfection of equipment in the food industry, and infected employees working in restaurant kitchens.

In the USA, an outbreak caused by a *Salmonella* strain with a specific PFGE pattern occurred during January 1- December 31, 2007. In this outbreak, a total of 401 cases from 41 states were identified and 32% of ill persons were hospitalized. A case-control study indicated that illness was associated with consumption of not-ready-to-eat pot pies. Later, the outbreak strain was isolated from 13 samples of unopened pot pies collected from the homes of patients (CDC, 2008b). Moreover, in November 2006, public health officials of CDC and state health departments detected an increase in the reported incidence of isolates of *S.* Tennessee. In a multi-state case-control study conducted during February 5-13, 2007, illness was demonstrated to be strongly associated with the consumption of two brands of peanut butter produced at the same process plant. A total of 628 persons infected with this outbreak strain of *S.* Tennessee had been reported from 47 states during 2006-2007 (CDC, 2007a). Later, on November 25, 2008, an epidemiological investigation began due to an increase of *S.* Typhimurium isolates that shared the same PFGE patters. Up to January 2009, 529 persons from 43 states from the USA and one person from Canada had been reported infected with this strain. A total of 116 patients were hospitalized, and the infection might have contributed to eight deaths. Sequential case-control studies indicated significant associations between illness and consumption of peanut butter, and specific brands of peanut butter biscuits. Epidemiological and laboratory findings indicated that peanut butter and peanut paste produced at one plant were the sources of the outbreak. These products were also ingredients in many foods produced and distributed by other companies. This outbreak highlights the complexities of "ingredient-driven" outbreaks and the importance of rapid outbreak detection and investigation (CDC, 2009).

On November 15, 2006, in the USA a case of salmonellosis in an employee of a poultry vaccine facility was notified. When a second case of salmonellosis in another employee at the same facility was reported on November 25, an outbreak investigation was initiated. Results of that investigation suggested that 21 employees of the facility became ill during a 1-month period from exposure to a strain of *S.* Enteritidis that was used in vaccine production. Infection was thought to have resulted from environmental contamination after the spill of a liquid containing a high concentration of *S.* Enteritidis. As a result, it was recommended that the facility improve its infection-control procedures to better protect workers. This outbreak highlights occupational risks that can be associated with the manufacture of veterinary biologicals derived from human pathogens (CDC, 2007d).

During October and November 2006, the Spanish National Reference Laboratory reported a series of isolations of *S.* Kottbus on the island of Gran Canaria. A matched case-control study was conducted and it showed that the ingestion of a brand of bottle water was related to outbreak. Microbiological and environmental analysis detected *S.* Kottbus in bottles randomly selected from markets, and also in the local factory where the water was bottled. A carrier pigeon loft was found near one water reservoir of the local factory and it could be verified that pigeons frequented this reservoir. Moreover, *Salmonella* spp. was detected in the pigeons, suggesting that they could be contaminating the water reservoir (Palmera-Suárez *et al*., 2007).

In 2002, an outbreak of *S.* Enteritidis phage-type 8 occurred at a convention center in Dallas, Texas, USA and continued for 6 weeks. From March to April, the hotel kitchen implicated catered for 41 multi-day conferences attended by 9,790 persons. A total of 617 illness reports from residents of 46 states were received. The epidemiological studies showed that sauces or items served with sauces were implicated in outbreak. Eleven food service employees, including one who prepared sauces, had stool cultures that yielded *S.* Enteritidis phage-type 8. Transmission ended with implementation of policies to screen food handlers and exclude those whose stool cultures yielded *S.* Enteritidis (Beatty *et al*., 2009).

In addition, in Germany, from October 2001 to March 2002 about 439 *S.* Oranienburg reports were registered. Simultaneously, an increase in this strain was noticed in other European countries. A case-control study was conducted, indicating that the consumption of chocolate from one company was associated with illness. Later, two brands from the same company were positive for *S.* Oranienburg. Isolates from humans and from chocolates had indistinguishable PFGE patters. No source or point of contamination was identified (Werber *et al*., 2005).

Antibiotic resistance in *Salmonella* serovars: A serious problem in public health

Since the start of the widespread use of antimicrobial agents by humans in the late 1940s, antibiotic resistance as a phenomenon has been observed in almost all bacterial species and against all drugs available. Antimicrobial resistance can increase the morbidity, mortality, and costs associated with disease. Moreover, it has social and economic consequences and requires strong scientific and public health efforts to improve the situation. The increase in the number of resistant and multiresistant (resistant to two and more antimicrobials) strains of bacteria has been recognized by the World Health Organization (WHO) and health authorities as one of the major problems in public health (Helmuth, 2001).

The use of antibiotics in food animal production systems has resulted in emergence of antibiotic resistant zoonotic bacteria that can be transmitted to humans through the food chain (Walsh & Fanning, 2008). Since the 1960s many studies have focused on this subject and the results are alarming. For instance, a rise in *S.* Typhimurium infection was observed in calves in United Kingdom during 1964-1966 following the adoption of the intensive farming methods (Anderson, 1968). A single phage type of *S.* Typhimurium, type 29, was incriminated as the major pathogen. Attempts to treat and control the disease with a range of antibiotics resulted in the acquisition of transferable multiple drug resistance by *S.* Typhimurium and the transmission of this strain, directly or indirectly, from bovines to man resulted in many human infections.

In Netherlands between 1972 and 1974, almost 50,000 *Salmonella* isolates from several sources (humans, animals, animal products, sewage, etc.) were tested for resistance to ampicillin, chloramphenicol, kanamycin, and tetracycline. In this study, the incidence of resistance to at least one of the above drugs ranged from 39.2% to 45.6%. Moreover, many multidrug-resistant strains of *S.* Typhimurium and *S.* Dublin were isolated from calves and cattle. In 1974, 64.4% of all strains of *S.* Typhimurium from these animals appeared to be resistant to ampicillin, tetracycline, chloramphenicol, and kanamycin, and 25.5% of *S.* Dublin were found to be resistant to chloramphenicol and tetracycline (Voogd *et al*., 1977).

Holmberg *et al*. (1984) assessed the origin of antimicrobial resistance in infections caused by *Salmonella* serovars in 52 outbreaks investigated by the CDC between 1971 and 1983. According to this study, in 38 outbreaks with identified source, food animals were involved in 11 (69%) of 16 resistant and 6 (46%) of 13 sensitive outbreak strains, *S.* Typhimurium being the main serovar isolated and beef and milk the main products involved. In addition, Cohen & Tauxe (1986) analyzed the occurrence of antimicrobial resistance in *Salmonella* serovars in USA between 1960 and 1980, concluding that the use of antibiotics in animal husbandry played a major role in the emergence or persistence of these resistant *Salmonella* strains.

In 1989 an outbreak of *S.* Typhimurium DT 193 resistant to four antibiotics (ampicillin, streptomycin, sulphonamides, and tetracyclines) occurred in northern

England, affecting 206 persons. An analytical study showed a significant association between illness and consumption of cold roast pork supplied by the butcher's shop. Later, *S.* Typhimurium DT 193 with the same antibiotic resistance pattern as the outbreak strain was isolated from samples of meat bought from the shop and from samples of pig feces taken from the farm supplying the shop. It was concluded that inadequate processing of infected pork meat at the shop may have contributed to this outbreak (Maguire *et al*., 1993).

Seyfarth *et al*. (1997) studied the frequency of antimicrobial resistance and epidemiological relatedness among 473 isolates of *S.* Typhimurium from human and veterinary sources in Denmark in 1993. In this study, the human strains were clinical isolates from patients with diarrhea (228 isolates) and animal strains were isolated from clinical or subclinical infections in cattle (48 isolates), pigs (99 isolates) and poultry (98 isolates). All strains were tested against 22 different antimicrobial agents used in both human and veterinary medicine with the disc diffusion method. Strains were also phage-typed and the plasmid content determined in all resistant strains. Of 228 human isolates tested, 19.3% of the strains were resistant to one or more antimicrobial agent compared with 10.4% of strains from cattle, 11.1% of strains from pigs and 9.2% of strains from poultry. Multiple resistance was found in 9.2% of the human strains, but in only two of the cattle isolates. Resistance in human strains was most common against tetracycline (13%), ampicillin (12%), sulphonamide (12%), streptomycin (10%), and chloramphenicol (8%). The resistance pattern differed somewhat in animal isolates. Poultry strains were usually resistant only to ampicillin, while pig and cattle isolates were most often resistant to sulphonamide, tetracycline and streptomycin. Typing of the strains showed that some animal strains and human strains were indistinguishable.

Moreover, Molla *et al*. (1999) assessed the antimicrobial resistance to 10 drugs in 39 *Salmonella* serovars isolated from raw minced beef and chicken (gizzard, liver, and heart) samples in Addis Ababa (Ethiopia). Thirty four isolates (87.2%) were resistant to one or more drugs. The antibiotics to which isolated strains were resistant included nitrofurantoin (48.7%), furazolidone (48.7%) and streptomycin (46.2%). Only four antimicrobials (gentamycin, kanamycin, rifampicin, and sulphamethoxazole-trimethoprim) were effective against all isolate*S.* About 80% of the *S.* Enteritidis strains showed multiple resistance to up to four antibiotics followed by *S.* Typhimurium (60.0%) and *S.* Dublin (33.3%).

In 1997, a *S.* Typhimurium DT104 strain resistant to five antimicrobials (ampicillin, chloramphenicol, streptomycin, sulfonamides, and tetracycline) affected 110 people in two outbreaks in USA. A case-control study and laboratory investigations indicated an unpasteurized Mexican-style cheese as the responsible for these outbreaks (Cody *et al*., 1999).

White *et al*. (2001) identified and characterized strains of *Salmonella* isolated from ground chicken, beef, turkey, and pork purchased at three supermarkets of Washington, USA. The isolates were characterized by serotyping, antimicrobial-susceptibility testing, and phage-typing. Additionally, PCR and DNA sequencing were used to identify resistance integrons and extended spectrum beta-lactamase genes. Of 200 meat samples, 41 (20 %) were contaminated with 13 different serovars. Eighty four percent of the isolates were resistant to at least one antibiotic, and 53% were resistant to at least three antibiotics. Sixteen percent of the isolates were resistant to ceftriaxone. Five isolates of *S.* Agona had resistance to 9 antibiotics, and the two isolates of *S.* Typhimurium DT208 were resistant to 12 antibiotics. Eighteen isolates, representing four serotypes, had integrons with genes conferring resistance to aminoglycosides, sulfonamides, trimethoprim, and beta-lactams.

In another study, Chen *et al*. (2004) analyzed 133 isolates of *Salmonella* serovars recovered from retail meats purchased in the USA and China, assessing the antimicrobial resistance of these strains. In this study, it was demonstrated that seventy-three (82%) of these *Salmonella* serovars isolates were resistant to at least one antimicrobial agent. Resistance to the following antibiotics was common among the USA isolates: tetracycline (68% of the isolates were resistant), streptomycin (61%), sulfamethoxazole (42%), and ampicillin (29%). Eight *Salmonella* isolates (6%) were resistant to ceftriaxone. Fourteen isolates (11%) from China were resistant to nalidixic acid and displayed decreased susceptibility to ciprofloxacin. Moreover, Miko *et al*. (2005) determined antimicrobial resistance of *Salmonella* serovars isolated from German foodstuffs. A total of 319 epidemiologically independent multidrug-resistant isolates comprising 25 different serovars were tested for their antimicrobial susceptibility by broth microdilution. *S.* Typhimurium was the prevalent serovar, and it was isolated from beef and pork meat. The results of this study shown that the most prevalent resistances found in the multidrug-resistant *Salmonella* serovars from foods were to streptomycin (94%), sulfamethoxazole (92%), tetracycline (81%), ampicillin (73%), spectinomycin (72%), chloramphenicol (48%), and trimethoprim (27%).

According to the USA Food and Drug Administration (FDA) (2006), in 2003, 22.5% of non-typhoid *Salmonella* isolates from humans were resistant to at least one antimicrobial agent and the most common multidrug-resistance phenotype reported was to ampicillin, chloramphenicol, streptomycin, sulfonamides, and tetracyclines, which was detected in 9.3% of isolates tested. While on the veterinary side, 44% of the *Salmonella* samples isolated from animal slaughter and veterinary diagnostic sources were resistant to at least one antimicrobial agent and the phenotype resistant to ampicillin, chloramphenicol, streptomycin, sulfonamides, and tetracyclines was also the most common multidrug resistance profile among veterinary isolates.

Between July 2004 and June 2005, a study was carried out in order to determine the prevalence and antimicrobial resistance of *Salmonella* serovars isolates from processed poultry in the USA. Four hundred eighty pre- and postchill whole broiler chicken carcasses were collected from a poultry processing plant. Water samples also were collected at the entrance and exit of the chiller. The proportions of pre- and postchill carcasses that were positive for *Salmonella* spp. were 88.4% and 84.1%, respectively. Ninety-two percent of water samples collected at the entrance of the chiller were positive for *Salmonella* spp., but all exit water samples were negative. Additionally, *Salmonella* spp. isolated were serotyped and tested for susceptibility to antimicrobials. Thirteen serotypes were identified; the most common were *S.* Kentucky (59.5%) and *S.* Typhimurium (17.8%). Three hundred thirty-nine (79.8%) of the isolates were resistant to at least one antimicrobial, and 53.4% were resistant to three or more antimicrobials. Resistance was most often observed to tetracycline (73.4% of isolates), ampicillin (52.9%), amoxicillin-clavulanic acid (52%), ceftiofur (51.7%), streptomycin (35.2%), and sulfisoxazole (21.8%). These results indicated that a large number of the isolates in poultry processing plants are frequently resistant to commonly used antimicrobials (Parveen *et al*., 2007).

Valdezate *et al*. (2007) conducted a study in order to determine the distribution and antimicrobial susceptibility of *Salmonella* serovars isolated in 2002 from food in 16 Spanish regions. Fifty one serovars were detected; the most common were *S.* Enteritidis and *S.* Typhimurium, followed by a group of 49 different serovars. The main sources of *Salmonella* serovars were eggs and their derivatives (21.6% of strains), poultry and related products (16.6%), and seafood (16.3%). Resistance rates for *S.* Enteritidis, *S.* Typhimurium, and the group of 49 serovars were, respectively, 8.3, 69.8, and 13.9% for ampicillin, 3.1, 52.8, and 59% for streptomycin, 40.6, 22.6, and 10.4% for nalidixic acid, 15.6, 71.7, and 31.1% for tetracycline, 7.3, 18.8, and 9.5% for trimethoprim-sulfamethoxazole, 0, 50.9, and 4.3% for chloramphenicol, and 6.2, 71.7, and 17.4% for multiple (at least four) antimicrobials. All the strains remained susceptible to other beta-lactams and fluoroquinolones.

From September 2003 to February 2004, Zewdu & Cornelius (2009) assessed antimicrobial resistance of 98 isolates of *Salmonella* serovars recovered from food in Addis Ababa, Ethiopia. The results of this study revealed that 32 *Salmonella* isolates were resistant to one or more of the 24 antimicrobials tested. The most common resistance was to streptomycin (75%), ampicillin (59.4%), tetracycline (46.9%), spectinomycin (40.6%), and sulfisoxazole (40.6%). The level of antimicrobial resistance was significantly higher for isolates from chicken carcass (62.1%) and pork isolates (22.7%).

In face of this public health problem, it is recommended a careful prescription of antimicrobial agents during veterinary practice, regardless of the purpose of this prescription (therapy, prophylaxis, and growth enhancement). This will increase the efficient use of antimicrobial agents after microbiological identification of the causative pathogen. Moreover, it is important to highlight that the use of any antimicrobial agent should not substitute good hygiene pratices and, wherever possible, alternative management methods should be sought and used. In addition, veterinarians should respect and follow the sanitary legislation concerning the use of antimicrobial agents in their country.

Concluding remarks

Members of the genus *Salmonella* are major enteric pathogens of human beings and animals. The increase of the human population and food production increases the potential for dissemination of these ubiquitous microorganisms. The high incidence of human non-typhoid salmonellosis demonstrates the potential harm of the presence of *Salmonella* in food and contact animals to people. In addition to disease caused by

Salmonella serovars, antibiotic resistance can be transferred by other microorganisms that are agents of human disease similar to those involved in hospital-acquired infections. Fortunately, we can learn from undesirable episodes of life. The outbreaks of food-borne *S.* Enteritidis disease related to chickens from 1980's trigged a great deal of discussion related to its control. As a result, in the understanding of the epidemiology of the infection in commercial birds improved. Thus, monitoring program could also be improved resulting in more efficient control programs in the food chain. In addition, nowadays there is an increasing worldwide preoccupation with the quality of the food; many countries have banned the use of antimicrobial agents in the feed and they are encouraging farmers to use non-harmful substances. All measures taken over animal husbandry may minimize the presence of *Salmonella* in food. However, the contribution of the consumers is also necessary by adopting the basic concepts of hygiene during food preparation and storing.

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