Quantitative assessment of the sources of human salmonellosis attributable to pork

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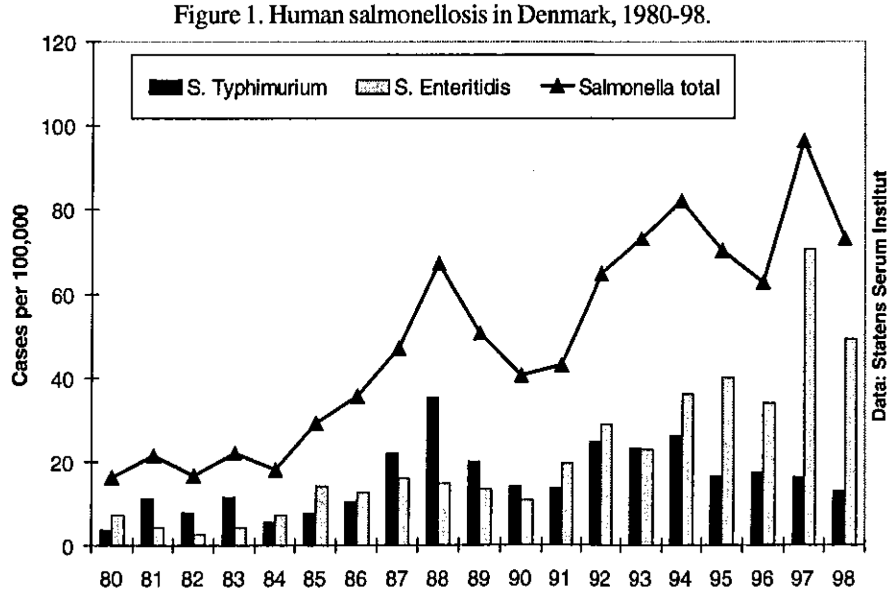
Introduction

Salmonellosis is the main cause of food-borne human gastroenteritis in Denmark. The annual incidence of registered cases increased throughout the 1980'ies reaching a maximum of 67.2 cases per 100,000 inhabitants in 1988 (Figure 1) (1). At that time, the most prevalent serotype encountered among humans was *S.* Typhimurium, which also occurred frequently in the broiler production, where 80-90% of the flocks was infected. As a consequence, a voluntary *Salmonella* control programme was implemented in the broiler production in 1989 (2). Further, the Danish food authority carried out a campaign informing the consumers about the correct handling and preparation of poultry and poultry products. These actions led to a decrease in the annual incidence of human salmonellosis.

In 1991, the incidence increased again reaching another maximum in 1994 (82.2 per 100,000). The dominant serotypes isolated from humans were *S.* Typhimurium and *S.* Enteritidis, where the latter was almost exclusively related to poultry. The occurrence of *S.* Typhimurium was now decreasing in broilers and the number of cattle herds infected with this serotype had remained constantly low through the last decade. In the pig production, however, an increase of clinical infections caused by *S.* Typhimurium and other serotypes was observed (3). In addition, an investigation of *S.* Typhimurium strains isolated from food-producing animals and humans in 1992 showed that the predominating *S.* Typhimurium phage type (DT) in pig herds was DT12, which also occurred most frequently in humans. In contrast, the phage types dominating in the broiler production (DT110, DT120, DT135 and DT193) only comprised a minor part of the types found in humans (1). Taken together, this suggested that pork played a major role as a source of human salmonellosis in Denmark.

A mandatory surveillance programme, monitoring the occurrence of Salmonella in pork and beef at the slaughterhouse level was initiated in 1993 (4). This was followed by the implementation of a serological surveillance and control programme in slaughter-pig herds in 1995 (5). As a result of these initiatives, the prevalence of *Salmonella* in pig herds and pork as well as the incidence in humans declined. The decrease in humans was solely due to a reduction of human cases caused by *S.* Typhimurium, whereas the number of cases caused by *S.* Enteritidis showed an increasing trend. In 1997, it was estimated that 10-15% of human infections, corresponding to 9.6-14.3 cases per 100,000, could be attributed to pork or pork products. S. Typhimurium DT12 was the predominant type found in humans comprising 58.8% of the isolates. This type was also dominating among isolates from red meat, but not from broilers (6). Since the prevalence of *Salmonella* in beef was lower than in pork and because Danes generally eat more pork, the majority of these cases was assumed to be related to pork.

Figure 1. Human salmonellosis in Denmark, 1980-98.



The annual incidence of human salmonellosis reached its highest level so far with 95 cases per 100,000 in 1997. The increase observed from 1995 to 1997 was exclusively due to an increase of infections with *S.* Enteritidis and the major source to these infections was presumed to be egg and egg products. In 1998, the human incidence decreased to 73.3 cases per 100,000. A revised control program launched in the table-egg production in March 1998 has undoubtedly contributed considerably to this reduction (7).

The purpose of this paper is to give an outline of the method we use for quantitatively assessing the sources of human salmonellosis in Denmark. The *Salmonella* statistics of 1998 will be used as an example.

Materials and Methods

The continuous monitoring of all major food animals and food of animal origin for *Salmonella* results in a collection of 5-10,000 *Salmonella* isolates per year. All isolates are serotyped and all isolates of *S.* Typhimurium and approximately 25% *S.* Enteritidis are phage typed. In addition, a selection of the strains is typed by molecular methods, primarily pulsed-field gel electrophoresis, and antibiogram typing. The latter method is applied to all *S.* Typhimurium isolates.

A comparison of *Salmonella* types isolated from animals and food with isolates from humans makes it possible to produce estimates of the number of human cases attributable to certain animal sources. It is prerequisite that some of the predominating *Salmonella* types are found almost exclusively in a single animal reservoir. Further, it is assumed that all human infections with these so-called "typical" types originate only from that particular source. Human infections caused by *Salmonella* types that are found in several reservoirs, may then be distributed in proportion to the occurrence of the typical types, assuming that the pathogenicity of the different *Salmonella* types is equal.

Consequently, the method requires the collection of representative *Salmonella* isolates from the stable-to-table chain followed by the application of discriminatory epidemiological typing methods. Finally, it is important that larger outbreaks are recognized and the sources identified. One reason is that unrecognized outbreaks caused by typical types will tend to overestimate the total number of infections originating from the reservoir harboring this type.

During the last decade, where this method has been applied to Danish monitoring data, the accuracy of the resulting estimates has been improved considerably. First of all, the *Salmonella* surveillance programs have been extended gradually resulting in more abundant data, which at the same time has become more representative. Further, the estimates have been more accurately assessed by also considering additional information. This includes information regarding the number of human infections acquired abroad, the annual amount of various foods consumed per capita, and the prevalence of *Salmonella* in imported food. The proportion of travel-associated cases is estimated from information given by the general practitioners when they fill out the forms that accompany fecal samples from patients to the diagnostic laboratory, whereas consumption figures are obtained from the national statistics. The importance of imported foodstuff is not sufficiently clarified. However, in June 1998, a continuous monitoring of imported pork, beef and poultry was initiated giving an overview of the prevalence and types of *Salmonella* in meat products of foreign origin.

The described method was also applied for the 1997 *Salmonella* statistics from three countries participating in an EU research project entitled "Salmonella in Pork" (SALINPORK) (8). In the Netherlands, data on *Salmonella* infections in man, animals and foods in 1997 was obtained from the National Institute of Public Health and the Environment (RIVM), who carries out surveillance and epidemiological studies of *Salmonella* infections, including the sero- and phage typing of *Salmonella* isolates. Data on the occurrence and distribution of *Salmonella* serotypes in Germany was taken from the annual report on zoonoses in 1997 (9), whereas information regarding the phage type distributions was kindly provided by the National Reference Centre for *Salmonella* and other Enterics (10). The Public Health Laboratory Service (PHLS) provided information regarding the annual incidence of salmonellosis in England and Wales in 1997, including the sero- and phage type distribution of isolates. Data on the distribution of sero- and phage types in the animal population was obtained from the annual *Salmonella* report published by the Central Veterinary Laboratory (11).

Results

In 1998, a total of 5,744 isolates of *Salmonella* from humans, animals and food was collected in Denmark. The serotype distribution of the isolates can be seen from Table 1, whereas the phage type distribution *S.* Typhimurium and *S.* Enteritidis is shown in Table 2.

Table 1. Serotype distribution of *Salmonella* from animals, meat at slaughterhouses and humans in Denmark, 1998.

Serotype	Humans	Pork	Beef	Broiler flocks	Layer flocks
S. Enteritidis	67.2	0.0	0.0	24.5	85.4
S. Typhimurium	17.5	54.0	10.5	18.2	6.8
S. Hadar	1.6	0.0	0.0	3.7	0.0
S. Manhattan	1.1	0.0	0.0	3.0	0.0
S. Infantis	0.9	17.2	0.0	17.5	4.9
S. Virchow	0.8	0.2	0.0	0.4	0.0
S. Agona	0.8	0.0	0.0	1.1	0.0
S. Derby	0.6	6.3	10.5	1.1	0.0
S. Newport	0.6	0.0	0.0	0.0	0.0
S. Java	0.5	0.0	0.0	0.0	0.0
S. Stanley	0.5	0.0	0.0	0.0	0.0
S. Braenderup	0.4	0.0	0.0	0.0	0.0
S. Bovismorbificans	0.4	0.0	0.0	0.0	0.0
S. Glostrup	0.3	0.0	0.0	0.0	0.0
S. Heidelberg	0.3	0.2	0.0	0.0	0.0
S. Saintpaul	0.3	0.0	0.0	0.0	0.0
S. Dublin	0.3	0.2	68.4	0.0	0.0
Others incl. not typable	6.1	21.9	10.5	30.5	2.9
Total	100	100	100	100	100
Number typed	3,880	448	19	269	103

Table 2. Phage type distribution (%) of *S.* Enteritidis and *S.* Typhimurium from humans, poultry and meat at slaughterhouses, 1998.

	S. Enteritidis		
Phage type	Humans	Layers	Broilers
8	39.9	59.1	30.6
6	28.9	22.7	48.0
4	11.0	4.6	14.3
1	4.1	1.1	2.0
21	4.1	6.8	0.0
6a	1.8	0.0	1.0
29	1.4	0.0	0.0
1b	0.9	0.0	0.0
21b	0.9	0.0	0.0
25	0.9	2.3	0.0
34	0.9	0.0	0.0
17	0.9	0.0	0.0
Others incl. not typable	4.1	3.4	4.1
Total	100	100	100
Number typed	218	88	98

	S. Typhimurium				
Phage type	Humans	Pork	Beef	Broilers	Layers
12	43.3	42.6	0.0	15.2	14.3
104	13.1	0.3	0.0	0.0	0.0
66	4.9	10.2	0.0	15.2	0.0
U288	3.8	1.5	0.0	0.0	0.0
135	3.6	1.2	0.0	2.2	14.3
193	3.4	5.7	0.0	2.2	28.6
17	2.1	7.2	0.0	6.5	0.0
120	1.7	1.5	0.0	2.1	0.0
302	1.7	0.6	0.0	0.0	0.0
10	1.1	3.6	0.0	0.0	0.0
107	0.8	1.8	0.0	0.0	0.0
177	0.8	1.2	0.0	4.4	0.0
110	0.6	0.9	50.0	34.8	14.3
41	0.4	0.0	0.0	4.4	28.6
15a	0.4	3.0	0.0	0.0	0.0
Others incl. not typable	18.4	18.6	50.0	13.0	0.0
Total	100	100	100	100	100
Number typed	474	333	2	46	7

Assessment of the proportion of travel-associated cases

Of the 3,880 registered cases of human salmonellosis 12-17% were estimated to be associated with travelling abroad (Figure 2). There was some serotype variation, though, since a lower percentage (approximately 7%) of *S.* Typhimurium infections was assumed to be travel-associated compared to the other serotypes.

Assessment of the sources of human S. Typhimurium infections in Denmark

Of the 678 registered cases of *S.* Typhimurium, around 630 were assumed to be infected in Denmark. The phage types dominating among humans were DT12, DT104, DT66, DTU288 and DT135 comprising 69% of the isolates (Table 2). In pork, beef and broilers, these phage types were found in 56%, 0% and 33% of the isolates, respectively. Phage type U288 was only found in pork (typical type) and pork was therefore assumed to be the single source of these infections. In broilers, the predominating phage type was DT110, which was found in 35% of the flocks infected with *S.* Typhimurium. This type was only isolated from 0.6% of the humans infected with *S.* Typhimurium suggesting that broilers are of minor importance as a source of human salmonellosis. Consequently, the majority (approximately 70%) of the *S.* Typhimurium infections acquired in Denmark was estimated to be attributed to pork. The remaining 30% were distributed among poultry (app. 10%) including imported poultry, Danish produced broilers (<1%) and unknown (app. 20%).

Assessment of the sources of human S. Enteritidis infections acquired in Denmark

In 1998, 2,607 infections with *S.* Enteritidis were registered. Of these, 2,209 were assumed to be acquired in Denmark. *S.* Enteritidis was almost exclusively related to layers and poultry (Table 1) and beef and pork was not considered as potential sources in 1998.

Sources of human infections caused by other serotypes

A total of 595 infections of other *Salmonella* serotypes were registered in 1998. Approximately 80% were assumed to be acquired in Denmark. In the primary production, *S.* Hadar and *S.* Agona were almost exclusively found in poultry, which were considered the main source of these infections. An outbreak of *S.* Manhattan associated with smoked fillet of pork (12) assigned almost all these infections to pork, whereas *S.* Infantis and *S.* Derby infections were distributed among pork and poultry (incl. broilers) by considering the previous identified typical types, the *Salmonella* prevalence in these products and the average amount of pork and poultry consumed. Finally, all *S.* Dublin infections were assumed to be caused by beef. A considerably part (58%) of the exotic *Salmonella* infections could not be associated with any specific source and were classed as unknown.

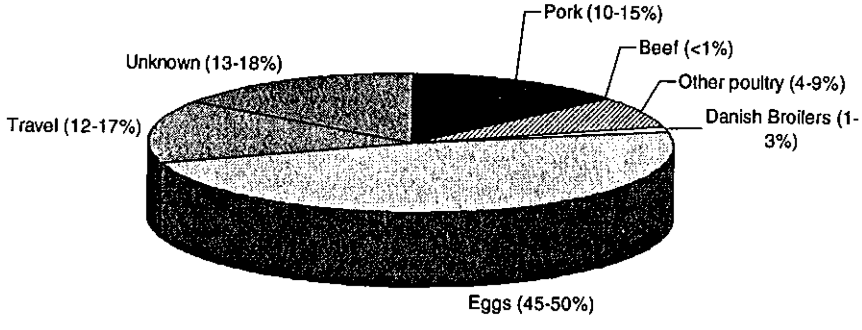
In summary, the estimated number of human cases of salmonellosis pr. 100,000 inhabitants that could be attributed to various sources in 1998, was as follows: eggs: 33-37; travel: 9-13; pork: 7-11; poultry: 3-7; Danish produced broilers: 1-2; beef: < 1. Approximately 603 cases (11.4 per. 100,000) could not be associated with any specific source. The distribution of sources is illustrated by percentages in Figure 2.

Assessment of the sources of human Salmonella infections in The Netherlands, Germany, and England and Wales in 1997

Using the same principles as described above, the relative importance of pork as a source of human salmonellosis was estimated for human cases in The Netherlands and Germany. Because information of the number of travel-associated cases was not available, it was assumed that the proportion of these cases per serotype was distributed as in Denmark in 1997.

—In The Netherlands, the RIVM registered 2,557 (17 per 100,000) cases of salmonellosis in 1997. *S.* Typhimurium comprised 786 cases of which 5% were assumed to be acquired abroad. Of the domestically acquired cases, 43% were assumed to originate from the consumption of pork. *S.* Enteritidis was almost exclusively found in poultry and poultry products, and pork was not considered a potential sources of

Figure 2. Quantitative assessment of sources of human salmonellosis in Denmark, 1998.



these infections. Finally, approximately 15% of the 687 cases caused by other serotypes were estimated to be attributable to pork. The dominating serotypes among the pork related cases were *S.* Infantis, *S.* Panama, *S.* Derby, and *S.* Livingstone. In total, pork was assumed to be responsible for 14-19% (2.4-3.2 cases per 100,000) of all *Salmonella* cases in 1997.

In 1997, 105,340 cases (128,4 per 100,000) of human salmonellosis were registered in Germany (9). *S.* Entertidis was the predominating serotype among humans and poultry, but was not found in pork. Consequently, the role of pork as a source of these infections was assumed to be negligible. Of the domestically acquired *S.* Typhimurium infections, approximately 65% were estimated to be attributable to pork. For the remaining serotypes this proportion was estimated to about 25%, among which *S.* Infantis, *S.* Bovismorbificans, *S.* Derby and *S.* Livingstone were dominating. In total, pork was assumed to be responsible for 18-23% (23-30 cases per 100,000) of all *Salmonella* cases in 1997.

The method was not applicable for England and Wales, since it was impossible to identify "typical" types within each reservoir, meaning that the prerequisite, that some of the predominating *Salmonella* types should be found almost exclusively in a single animal reservoir, was not fulfilled.

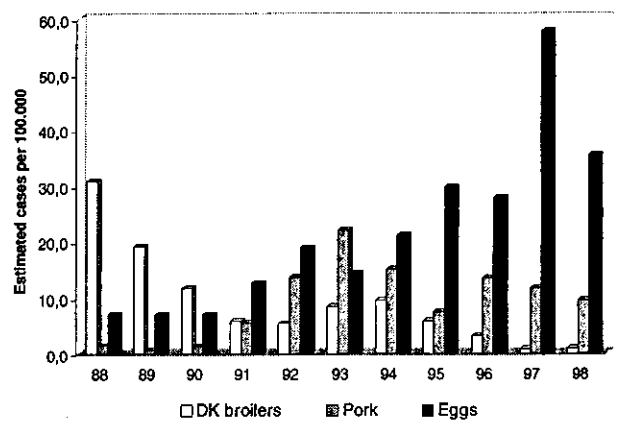
Discussion

Although the zoonotic *Salmonella* types can occur in almost all food-producing animals, there are often rather strong associations between certain types and a particular animal reservoir (1,3). In The Netherlands, the *Salmonella* types associated with pigs and pork over the past decade have accounted for on average 15% of identified cases of human salmonellosis. On the basis of this it is estimated, that 15% (5-25%) of all cases of human salmonellosis are attributable to pork (13). This corresponds well with our estimates of the proportion of pork related cases in The Netherlands. However, differences between reporting systems for human gastrointestinal diseases and variations in case-definitions makes it impossible to compare the estimated number of cases per 100,000 attributable to pork in Denmark, The Netherlands and Germany.

A weakness of this method is, that it will not give a detailed pathway of the *Salmonella* transmission and the identification of critical points of contamination and cross-contamination along the stable-to-table. Cases due to cross-contaminated products or person-to-person transmission will be classified according to the occurrence of the causal type in the primary production. However, depending on the purpose of estimating the number of human cases attributable to different sources, some would argue that this actually is an advantage. For instance, consider some cases of person-to-person transmission, where the index case was infected by consuming a meal of lets say pork contaminated with *Salmonella*. All cases infected by the index case are according to this method classed as infected from contaminated pork. This is quite acceptable, since preventing *Salmonella* in pork will also prevent the secondary cases.

We conclude, that even though the resulting risk estimates reflect a simplification, the method has proved to be a powerful tool in monitoring the main sources and dynamics in the occurrence of human salmonellosis in Denmark. Figure 3 shows that Denmark has experienced three waves of human salmonellosis, where the majority of cases was caused by three distinct sources: broilers in the late 80'ies, pork in the beginning of the 90'ies and eggs in the mid/late 90'ies. At each peak a new or revised control program was implemented and a reduction of human cases attributable to that particular source was observed.

Figure 3. Estimated major sources for human salmonellosis in Denmark, 1988-98.



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