

Annual Report on Zoonoses in Denmark 2012



DTU Food National Food Institute

Annual Report on Zoonoses in Denmark 2012

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Introduction

In 2012, the number of human *Salmonella* cases increased slightly after several years of decrease. The increase can be explained by an increase in cases due to monophasic *S*. Typhimurium strains (*S*. 1,4,[5],12;i;-). Cases due to *S*. Enteritidis continued to decrease. A total of 11 *Salmonella* outbreaks were reported. Seven of these were caused by *S*. Typhimurium and the monophasic strains. Two outbreaks were caused by Danish beef and one by Danish pork. An outbreak due to *S*. Saintpaul in imported duck meat was also detected. Duck meat is an uncommon source of outbreaks in Denmark.

As in previous years, the *Salmonella* source account estimated that almost half of the human cases of salmonellosis could be attributed to travel. More than 75 % of the *S.* Enteritidis cases was acquired abroad, which is a small increase compared to previous years. The majority of the *S.* Typhimurium cases was still acquired in Denmark. For the sporadic cases not related to travel, Danish pork was estimated to be the most important source followed by Danish beef for which the estimated number of cases increased from 6 in 2011 to 85 in 2012. Two outbreaks contributed to the increase in cases from Danish beef. As in 2011, no cases were attributed to Danish broiler meat.

The number of human *Campylobacter* cases decreased by 8 % compared to the previous two years, and the prevalence in the flocks decreased by 19 %. *Campylobacter*, however, continues to be the most common zoonotic pathogen reported in Denmark contributing more than 50 % of all reported cases.

Campylobacter action plan

The new *Campylobacter* action plan 2013-2016 was drafted in 2012 and adopted in 2013. It covers initiatives in the broiler production at farm level as well as at the slaughterhouse, consumer information campaigns, and as a new element it covers sources and routes of transmission other than from the broiler production. Targets for *Campylobacter* in broiler flocks and in fresh broiler meat have been set. In broiler flocks the target is a 20% reduction in the prevalence by 2016 compared to the level in 2012. In fresh broiler meat target is defined as a reduction of the relative risk compared to the level in 2012 and depends on the prevalence as well as the concentration of *Campylobacter* in broiler meat.

The industry is free to choose the methods used to reach the targets, but the action plan suggests some options described in chapter 8.

Fruit and vegetables

Foodborne outbreaks caused by contaminated fruits and vegetables are an increasing problem globally and in recent years several large outbreaks have been described. Therefore, a number of risk management initiatives were taken in 2012 including implementation of mandatory heat treatment of frozen raspberries used as an ingredient in ready-to-eat dishes, guidelines for handling fruits and vegetables, guidelines for import of fruits and vegetables, and consumer and restaurant/ catering business campaigns.

In 2012, as in previous years, *Salmonella* was most often found in imported leafy greens and herbs. Salmonella has never been detected in products of Danish origin and detection of *Campylobacter* is rare.

In 2012, EU adopted four legal acts on sprouts which laid down the rules for traceability of sprouts and seeds, microbiological criteria for sprouts, approval of establishments for producing sprouts and certification requirements for imports into the EU of sprouts and seeds.

Special guarantees for *Salmonella* in the Danish table egg production

Denmark obtained special guarantees for *Salmonella* in table eggs July 1st 2012. By granting special guarantees to Denmark, the EU has acknowledged the effort made in Denmark for reducing the prevalence of *Salmonella* in table eggs. The special guarantees imply that eggs sold to Denmark should be followed by a specific certificate, and the flocks of origin must be tested negative for all *Salmonella* serotypes not only the serotypes covered by the EU legislation. Further, Danish eggs sold to other Nordic countries no longer need to be accompanied by specific certificates to facilitate trade.

The annual Report on Zoonoses presents a summary of the trends and sources of zoonotic infections in humans and animals, as well as the occurrence of zoonotic agents in food and feeding stuffs in Denmark in 2012. Greenland and the Faroe Islands are not represented. The report is based on data collected according to the Zoonoses Directive 2003/99/EC, supplemented by data obtained from national surveillance and control programmes as well as data from relevant research projects. Corrections to the data may occur after publication resulting in minor changes in the presentation of historical data in the following years report. The report is also available at www.food.dtu.dk.

Salmonella in pork at retail

To investigate the prevalence of Salmonella in pork at retail, six surveys have been conducted since 2001. Results from the four surveys showed a higher prevalence in butcher shops compared to supermarkets. There are a difference in the supply pattern between the two types of retail shops as butcher shops more often get the meat from small cutting plants than supermarkets. A survey showed that the Salmonella prevalence was significantly higher in small cutting plants compared to large cutting plants. Therefore, it could not be excluded that the higher Salmonella prevalence for butcher shops was a result of higher input of Salmonella from the supplying cutting plants. A survey therefore investigated the hygiene performance in butcher shops and supermarkets. Results from the supermarkets showed, that microbial contamination of processed pork could primarily be explained by the input via the raw meat, whereas results from the butcher shops showed that hygiene performance in the shop had an additional significant effect. To keep a low occurrence of Salmonella in pork at retail, focus on the hygiene practices in smaller cutting plants as well as in butcher shops is important. The project identified critical procedures and factors associated with hygiene performance, which have led to the preparation of tailored tools for the establishments to use in order to support a high level of hygiene.

Salmonella in the poultry production

In the table egg production, one breeding flock was positive for *Salmonella* in 2012. This is the first time in four years a positive flock has been reported. Only 0.8 % of the table egg layer flocks were found positive. In EU, a 2 % permanent target for *S*. Enteritidis and *S*. Typhimurium in table egg layer flocks is set. *Salmonella* in Danish layer flocks has been below the target for many years.

In the broiler production, no positive breeding flocks was reported in 2012. As regards broiler flocks, the *Salmonella*

prevalence has been low for more than a decade, and only 0.8 % of the flocks were positive in 2012. The permanent EU target of 1 % for *S*. Enteritidis and *S*. Typhimurium was reached by Denmark already in 2000. In 2012 the prevalence for these two serotypes was 0.2 %.

For breeding and fattening turkey flocks, the EU target of maximum 1 % flocks positive with *S*. Enteritidis and *S*. Typhimurium had to be reached by December 31st 2012. Denmark had no positive flocks in 2012.

The Danish EU-Presidency 2012

Denmark held the EU-Presidency from January 1st to July 1st 2012. During this period, the future meat inspection for pigs and the combat against antimicrobial resistance were dealt with.

At a conference on "Future meat inspection for pigs" steps towards a more risk based and flexible inspection were discussed including the four subjects: 1) no *Trichinella* testing of pigs from controlled housing conditions, 2) measures for reduction of *Salmonella* in pork, 3) omission of routine palpation and incision and, 4) that official inspection should focus on animal health, animal welfare and food safety only.

The Commission has included these conclusions in the proposals for a future regulation on meat inspection, and control of *Salmonella*.

A conference on "Combatting Antimicrobial Resistance -Time for a Joint Action" was held, and among other subjects the prudent use of antibiotics, critically important antimicrobials and improved data collection and monitoring of antimicrobial resistance were discussed. A very important statement from the conference was to keep a "One Health" perspective when fighting antimicrobial resistance. The conference was followed by the adaptation of a EU council Conclusion.



1. Trends and sources in human salmonellosis

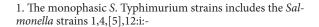
By Leonardo de Knegt (ledkn@food.dtu.dk) and Tine Hald

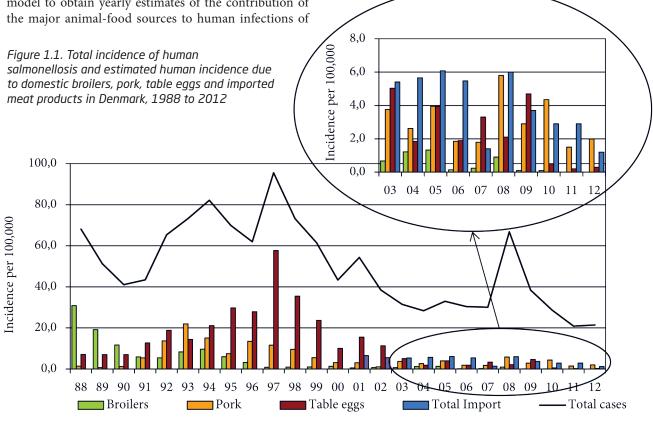
Salmonella is the second most frequently reported cause of bacterial foodborne infections in Denmark, with 1,198 cases reported in 2012, compared to 3,728 cases of campylobacteriosis [1]. Since the 1980s, different food-animals have been considered the main reservoir for *Salmonella* at different times. In the late 1980s, broilers were considered the main source, but with the implementation of targeted interventions, pigs and pork took over the role as the main source in the mid-1990s, and finally, laying hens in the end of decade [2,3]. Starting in the late 1990s, the total number of cases of salmonellosis in Denmark have been slowly decreasing, and eggs are again being replaced by pork as the most common implicated Danish vehicle of sporadic cases [1,2,4].

The relative contribution of different reservoirs to the total number of cases of salmonellosis is a dynamic process, which can be affected by intervention programmes aimed at specific animal reservoirs. Therefore, being able to identify the main causative food sources of *Salmonella* is believed to be a solid support to risk management decisions, allowing the evaluation of implemented interventions, as well as the need for new ones. For that purpose, the Danish Zoonosis Centre, National Food Institute, uses a source attribution model to obtain yearly estimates of the contribution of the major animal-food sources to human infections of

Salmonella. The principle of the method is to compare the number of human cases caused by different *Salmonella* sero- and phage types with the distribution of the same subtypes isolated from the various animal-food sources. Antimicrobial resistance profiles of *S.* Typhimurium isolates are also included to further distinguish between similar phage types found in animals, food and humans. Due to the growing importance of monophasic *S.* Typhimurium strains [1], sources of human infections associated with these strains will be discussed separately when describing *S.* Typhimurium cases.

The incidence of human salmonellosis in 2012 was 21.4 cases per 100,000, showing a slight increase from the 20.9 observed in 2011, but maintaining a lower incidence than observed during most of the last decade. Of these cases, 4.3/100,000 were caused by *S*. Entertitidis and 7.4/100,000 by *S*. Typhimurium (including 3.4/100,000 by the monophasic strains) (Appendix Table A2). The overall trend in human





Source: Danish Zoonosis Centre, National Food Institute

salmonellosis cases attributable to the major food-animal sources is presented in Figure 1.1.

Domestically produced pork was estimated to be the most important food source of salmonellosis in Denmark in 2012 with 8.0 % of all cases, which is a little higher than in 2011, where 7.4 % of all the cases were estimated to be caused by pork. In 2012, there was one foodborne outbreak due to pork which could explain some of the increase. The second most important source was domestic beef (7.0 %) which increased markedly compared to last year, where only 0.5 % of cases were attributed to this source. The increase was partly due to two outbreaks with monophasic *S*. Typhimurium, but there were also an increased number of sporadic cases especially due to *S*. Dublin. The estimated number of egg-related cases was slightly higher in 2012 compared to 2011, however it was still lower than in previous years. Since 2010, no outbreaks have been linked to table eggs.

Imported pork and imported beef both showed a considerable decrease in the estimated number of cases in 2012 compared to 2011. Imported pork decreased from 3.7 % of cases to 0.2 % and imported beef decreased from 2.8 % to 0.9 %. These decreases may be a result of risk-based control strategies, although a part can also be explained by the relative increase in importance of other sources, like domestic beef.

The number of cases attributed to imported ducks decreased from 2.4 % in 2011 to 1.9 % in 2012 (including 0.3% from an outbreak caused by *S*. Saintpaul) and the number of cases attributed to imported broilers was slightly lower in 2012 (1.8 %) than in 2011 (2.0 %) (Appendix Table A1). The total contribution of imported meat products decreased with almost a hundred cases when compared to 2011 (Table A1). It should be noted that part of the reduction may be explained by a decreasing number of samples taken from imported meat in the case-by-case control. Overall the number of samples was reduced by 30 % from 2011 to 2012.

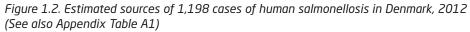
As in 2011 and 2010, cases estimated to have been acquired during international travels corresponded to nearly half (45%, 539 cases) of all Salmonella cases.

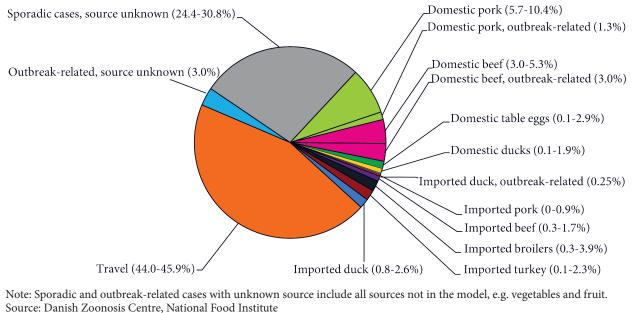
A total of 27.7% of reported sporadic *Salmonella* cases could not be attributed to any of the included food sources. These cases may be caused by imported or domestically produced fruits and vegetables (survey data presented in Table 5.1), food-animals not included in the national surveillance or by non-food sources of infection, such as direct contact with pet animals.

Of the 242 reported *S*. Enteritidis cases, 76.4 % was estimated to be related to international travel. Since 2010, there have been no *S*. Enteritidis outbreaks related to Danish sources.

A total of 415 S. Typhimurium cases were reported in 2012 which include 192 monophasic strains (46.3%). Cases of S. Typhimurium related to international travel were estimated to be 21.7%, of which approximately half of the cases were monophasic strains. There were 59 outbreak cases of S. Typhimurium and 51 cases (86.4%) were caused by monophasic strains. A combined outbreak of S. Typhimurium DT120 and monophasic S. Typhimurium DT193 which had domestic pork as the implicated source accounted for 15 of these cases, and two outbreaks of a monophasic strain which were related to Danish beef accounted for the remaining 36 cases.

In total, 91 *S.* Typhimurium cases (including 52 cases related to monophasic strains) were attributed to domestic products; 33.5 % were caused by strains susceptible to all tested antimicrobial agents and 66.5 % by strains resistant to one to three antimicrobial agents. No multi-resistant strains (resistant to four or more antimicrobial agents) were attributed to domestic foods in 2012, which is the first time during the last decade. As in 2011, all cases caused by *S.* Typhimurium strains resistant to quinolones were related to international travel (Figure 1.3). Among the 28 *S.* Typhimurium cases attributed to imported meat (including 13 cases infected with monophasic strains), no multi-resistant or quinolone-resistant cases were observed,





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and a large decrease from 71.8 % in 2011 to 41.6 % in 2012 was registered for resistant *S*. Typhimurium. In total, 87 *S*. Typhimurium infections was acquired abroad; 41.6 % were caused by resistant types, 14.4 % by multi-resistant types, 21.1 % by types resistant to quinolones, and 22.9 % by types susceptible to all tested antimicrobial agents.

Since 2010, the percentage of monophasic *S*. Typhimurium strains have increased from 21.1 % in 2011 to 46.3% in 2012. The increase is mainly to be found in domestic sources, as the percentage of cases of monophasic strains among *S*. Typhimurium attributed to domestic sources increased from 2.9 % in 2010 to 24.5 % in 2012. The estimated percentage of those strains in cases attributed to imported foods decreased from 20.1% in 2010 to 6.8% in 2012. Travel-related cases were comparable between years (36% in 2010 and 32% in 2012).

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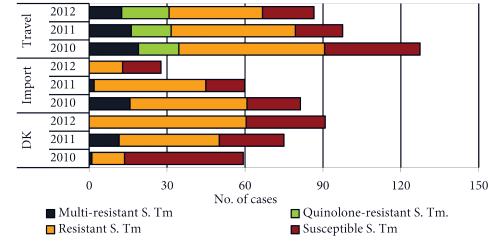


Figure 1.3. Estimated sources of antimicrobial resistant^o S. Typhimurium infections in humans, 2009-2012

a) Resistant: Resistant towards one to three antimicrobial agents; Multi-resistant: Resistant towards four or more antimicrobial agents. Source: Danish Zoonosis Centre, National Food Institute

Where do we acquire Salmonella infections?

By Birgitte Helwigh and Steen Ethelberg

In 2012, as in the previous years, Statens Serum Institut attempted to interview all registered *Salmonella* cases where no travel information was reported by the general practitioner. The patients were asked about the date of disease onset and whether they had travelled abroad within a seven-day period prior to disease onset. This information was complemented with information from general practitioners' reports. Travel information was obtained from a total of 67.9 % of the *Salmonella* cases in 2012. Compared to previous years, there was a decrease in the number of cases from whom it was possible to obtain information on travel. Among the cases with known travel history, 77.0 % of the *S.* Entertitidis cases, 25.2 % of the *S.* Typhimurium cases, 27.8 % of the monophasic *S.* 1,4,[5],12:i:- cases and 49.0 % of cases with other serotypes were infected abroad. The group of other serotypes comprises considerable variation in terms of serotypes between years (Table 1.1). The distribution pattern of travel-related and domestically acquired *Salmonella* infections (not including outbreak related cases) was comparable to previous years (Figure 1.4). Similar to 2011, almost 50 % of the travel-related *Salmonella* infections were acquired in Thailand, Turkey and Egypt in 2012.

In 2012, 11 outbreaks due to *Salmonella* with a total of 158 cases were reported. This is an increase compared to 2011. Seven of the outbreaks was due to *S*. Typhimurium or monophasic *S*. 1,4,[5],12:i:- (See chapter 6 for more information and Appendix Table A4). The total number of human salmonellosis cases increased slightly compared to last year mainly due to an increase in the number of monophasic *S*. 1,4,[5],12:i:- cases. The number of cases due to *S*. Entertitidis has decreased the last 10 years.

2012	12Number of patients (%)% patients infecteda Abroad Domestically2		2011	Number of patients (%)	-	nts infectedª Domestically	
Enteritidis	241 (20.1)	77.0	23.0	Enteritidis	293 (25.1)	71.7	28.3
Typhimurium	223 (18.6)	20.1	79.9	Typhimurium	245 (21.0)	19.5	80.5
Typhimurium (monophasic) ^ь			78.3	Typhimurium (monophasic) ^b	141 (12.1)	35.2	64.8
Dublin	50 (4.2)	0	100	Strathcona	43 (3.7)	0	100
Poona	28 (2.3)	26.7	73.3	Dublin	42 (3.6)	7.1	92.9
Stanley	28 (2.3)	68.4	31.6	Stanley	39 (3.3)	76.7	23.3
Infantis	25 (2.1)	36.4	63.6	Agona	22 (1.9)	50.0	50.0
Newport	24 (2.0)	62.5	37.5	Infantis	22 (1.9)	50.0	50.0
Virchow	21 (1.8)	81.3	18.8	Kentucky	17 (1.5)	100	0
Saintpaul	17 (1.4)	25.0	75.0	Virchow	17 (1.5)	76.9	23.1
Other serotypes	349 (29.1)	54.3	45.7	Other serotypes	285 (24.4)	54.5	45.5
Total	1,198 (100)	45.2	54.8	Total	1,166(100)	46.8	53.2

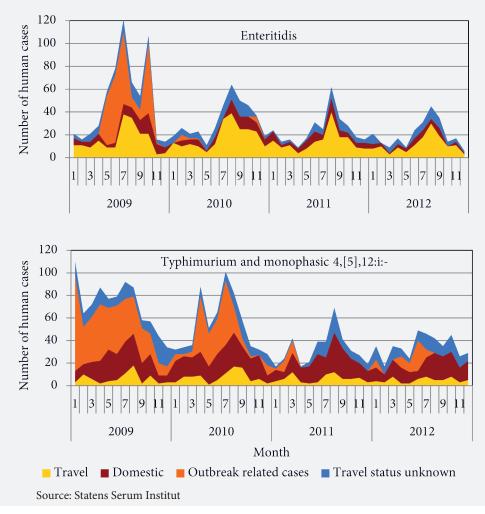
 Table 1.1. Top 10 Salmonella serotypes in humans and place of infection, 2011-2012

a) Patients with unknown travel information (32.1 % of all patients in 2012 and 22.4 % of all patients in 2011) were excluded from the percent calculations.

b) Typhimurium (monophasic) includes the Salmonella strains 1,4, [5], 12:i:-

Source: Statens Serum Institut

Figure 1.4. Monthly distribution of S. Enteritidis, S. Typhimurium, and monophasic S. 1.4.[5],12:i:- cases, 2009-2012



2. Ranking of food preferences based on age and gender of patients - a novel approach to outbreak investigations

By Anne Wingstrand (awin@food.dtu.dk), Sisse Fagt, Tue Christensen, Lone Jannok Porsbo and Tine Hald

In Denmark, age and gender of cases are part of the information which is available in the early phases of the investigation of foodborne outbreaks. The distribution of age and gender varies between outbreaks, and often diverges considerably from the demographic distribution in the Danish population registered by Statistics Denmark (Figure 2.1 and 2.2). From the Danish National Survey of Dietary Habits and Physical Activity 2003-2008 (DANSDA) [1], food consumption patterns are known to depend largely on age and gender.

Therefore, a study was initiated with the hypothesis that the age and gender distribution of a foodborne outbreak reflects a preference for the contaminated food item in the group of patients, and that the outbreak source may be found among the main food preferences of the patients. To our knowledge, no tool has so far been available for a structured, objective approach to this use of demographic information.

Information on the age and gender of outbreak cases was used to calculate and rank food preferences for a wide range of food items based on food intake data from DANSDA. Several approaches for calculation of food preferences have been evaluated on a selection of foodborne outbreaks with known source [2] and the most promising approach is presented below.

Food exposures in the Danish population

Quantitative food consumption data collected between 2005 and 2008 was obtained from DANSDA, comprising data from a total of 2,710 persons aged 4-75 years. The participants in the survey kept a food record for seven consecutive days using a pre-coded (semi-closed) questionnaire, and the recorded food intake for each participant was coded as exposure/non-exposure for 252 food items. Of these, 42 were excluded due to a negligible risk of foodborne infection (e.g. coffee, alcoholic beverages and dry bakery products). Five aggregated food groups each including several food items, were generated for calculation of the total exposure to beef, pork, eggs, sliced meat and tomatoes, respectively. Participants were stratified into 16 age and gender strata (Figure 2.1 and 2.2), and the proportion of participants in each stratum exposed to each food item was calculated (stratum specific exposures). The overall theoretical exposure in the Danish population aged 4-75 years was calculated for each food item based on the distribution of age and gender in the general population aged 4-75 years and the stratum specific exposure results from DANSDA.

Food exposures in selected foodborne outbreaks

Ten domestic foodborne outbreaks of bacterial gastrointestinal infection between 2007 and 2011 were selected for evaluation of the method. All with good evidence for the outbreak source. One outbreak (FUD 979 [5]) was analyzed as three outbreaks (FUD 979a, FUD 979b and FUD 979c) because the outbreak source changed during the course of the outbreak (Table 2.1). Thus a total of 12 outbreaks were analysed.

Only laboratory confirmed domestic cases aged 4-75 years were included in the study, as the DANSDA survey included participants aged 4-75 years. Cases identified as secondarily infected were excluded, and no larger suboutbreaks were identified. The selected outbreaks included two outbreaks caused by *Shigella sonnei* and eight outbreaks caused by *Salmonella* serovars.

For all outbreaks the proportion of cases in each of the 16 age and gender strata was calculated, and the overall theoretical food exposure was then calculated for each food item.

Indicator food items

For each outbreak, between 2 and 7 food items, which pointed directly or indirectly (e.g. mixed salad which could include the outbreak source) towards the outbreak source, were identified. For outbreak FUD 979c, which represented the "tail" of a larger outbreak preceded by two consecutive parts of the outbreak, FUD 979b+c with different sources, indicators for both sources were included.

FUD no.	Pathogen	Outbreak source	Year	No. cases 4-75 years / Total	% female	Mean age (4-75 years)
627 [3]	Shigella sonnei	Baby corn	2007	54/55	72	35.7
852	S. Typhimurium DT120	Smoked ham	2008	46/55	46	43.0
855 [4]	S. Typhimurium U288	Pork	2008	32/41	42	46.0
863	S. Typhimurium U312	Pork	2008-2009	25/36	52	48.7
891	S. Enteritidis FT8	Table eggs	2009	121/143	51	37.2
888	Shigella sonnei	Sugar peas	2009	8/8	100	31.3
996	S. Typhimurium DT120+DT7	Low-fat salami (pork and game meat)	2010	17/20	41	27.9
979 [5] 979a ^a 979b ^b 979c ^c	S. Typhimurium	Pork Teewurst Assumed pork and teewurst	2010	60/80 50/62 20/26	57 56 80	49.2 41.7 47.2
1067	<i>S</i> . Typhimurium DT120	Smoked pork ten- derloin	2011	20/22	70	54.4
1112	S. Strathcona	Datterino tomatoes	2011	31/43	55	33.9

Table 2.1. Ten foodborne outbreaks (12 including subsets) used for validation of food preference ranking

a) First 80 cases

b) Peak, week 27-31

c) "Tail", last 26 cases

Source: Central Outbreak Management Group and the Food- and Waterborne Outbreak Database (FUD)

Calculation and ranking of food preferences in the outbreaks

For each outbreak, the difference between the theoretical exposure to a food item in the outbreak and in the Danish population was calculated for each of the identified food items:

Diff(Exposure) = Exposure(Outbreak) - Exposure(DK population).

The preference in the outbreaks for each food item was then calculated as the ratio between the difference in exposure and the non-exposed part of the population:

Preference = Diff(Exposure) (1 - Exposure(DK population)).

For each outbreak, the food items, for which the exposure in the outbreak exceeded the exposure in the population, were ranked according to their preference (the higher preference, the lower rank number), and a preference profile including food items ranked in preference top-25 was created (examples in Table 2.2).

Results

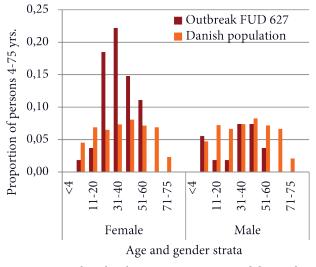
In the 12 outbreaks, the number of eligible cases varied between 8 and 121. The gender distribution ranged from 41 % females in FUD 996 (source: low-fat salami) to 100 % females in FUD 888 (source: sugar peas). Also the age distribution varied considerably. The mean age among cases ranged from 28 years in FUD 996 (source: low-fat salami) to 54 years in FUD 1067 (source: cold smoked pork tenderloin). Examples of age and gender distributions can be seen in Figure 2.1 and 2.2.

The top-25 food preference profiles of the outbreaks varied considerably. In FUD 627, where female and younger/ middle-aged cases were overrepresented (Figure 2.1), the profile was dominated by vegetables and fruits (Table 2.2). The outbreak source, baby corn, was not registered separately in DANSDA, but would most likely have been included in the group "Lettuce, incl. mixed salad and tomato salad", which was included in the top-25 preference list (Table 2.2). The meat preference in FUD 627 was chicken and the less specific "Casserole with meat".

In outbreak FUD 855, where male and elderly cases were overrepresented (Figure 2.2), a large part of the top-25 food preferences were food items typically associated with the traditional Danish lunch with open Danish sandwiches ("smørrebrød") (Table 2.2). The preferred meat was hamburger (beef) and pork including "other dishes with minced meat", which typically consists of beef and pork as well. Among the top-25 preferences were several indicator food items for pork, which was identified as the outbreak source.

For all outbreaks in this study, except FUD 979c, one or more food items indicating the outbreak source were

Figure 2.1. Age and gender distribution of 54 cases in an outbreak of Shigella sonnei from baby corn (FUD 627, 2008) compared to the distribution in the Danish population



Source: Central Outbreak Management Group and the Foodand Waterborne Outbreak Database (FUD) ranked in the top-25 food preference profile. The rank of the food items within top-25 did not give any further indication of the outbreak source.

Discussion and conclusion

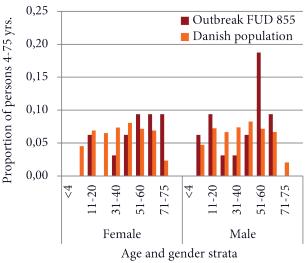
Systematic ranking of food preferences for cases in foodborne outbreaks based on the age and gender distributions of cases, is to our knowledge a new approach to outbreak investigation, which may be used in the early stages of an outbreak investigation if data on age and gender of cases are available. This type of investigation requires access to reasonably updated data on food intake in the population.

Results from this study indicate, that food preference profiling may be useful already at an early stage of an outbreak when only few cases are known. Further studies of food preferences in subsets of larger outbreaks could qualify the number of cases needed for the preference ranking, and the dynamics of age, gender and food preferences in long-lasting outbreaks. The method may be improved by adding information on season and geography to the calculations and by supplementing the background data from DANSDA with food intake data for small children and persons over 75 years of age. The method may provide decision support to guide and prioritize the investigation of foodborne outbreaks.

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Figure 2.2. Age and gender distribution of 32 cases in an outbreak of S. Typhimurium U288 from pork (FUD 855, 2008) compared to the distribution in the Danish population



Source: Central Outbreak Management Group and the Foodand Waterborne Outbreak Database (FUD) 2. Wingstrand A, Fagt S, Christensen T, Porsbo LJ and Hald T (2013). Køn og alder anvendt til indentifikation af kostpræferencer hos patienter i fødevarebårne udbrud. 1. Danske fødevarebårne udbrud med kendt kilde (In Danish). (In prep, electronic version will become available on www.food.dtu.dk)

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FUD 627, Shigella Sonnei, 54 cases Rank of food FUD 855, S.Typhimurium, 32 cases Rank of food (source: baby corn) preferences (source: pork) preferences Skimmed milk and buttermilk on Milk/cream for porridge/stewed fruit etc. 14 cereals Cheese, high-fat 5 Spreadable mixed butter products 20 Brie, camembert, blue cheese types etc. 11 Cheese as garniture/topping 17 Liver paté, paté etc. standard 12 Tomato/cucumber/pepper decoration 16 *Sliced meat. High-fat: roast pork, rolled 4 Chocolate on bread 23 meat, saveloy, rissole etc. Fish for Danish "smørrebrød", high-fat Pitabread etc. stuffed 25 2 (herring, salmon, eal etc) Carrots, incl. crudités 5 Eggs, whole 3 Tomato 6 Mayonnaise salads, standard 9 Cucumber 7 Tomato, "smørrebrød" 6 9 Pepper Tomato/Cucumber/Pepper, decoration 13 *Lettuce, incl. mixed salat and to-**Pickled** vegetables 7 1 mato salad Vegetable-mix 14 Avocado 24 Cabbage, all sorts 8 Apples 22 Pickled vegetables (not for bread) 15 Pears 13 Hamburger 17 Citrus fruits 18 Other dishes with minced meat 23 Bananas 3 *Pork, low-fat 25 Grapes 15 Fish, low-fat 20 Melon 21 Egg dishes incl. gratin 24 Raisins and other dried fruits 12 Potatoes, boiled 1 Chicken 19 Melted butter, fat dripping, cold butter/ 19 10 Casserole with meat margarine, garniture Dressing, unspecified, and cold sauce 8 21 Bearnaise sauce, sauce with roux, etc. Cake, unspecified (e.g. pie, apple cake) 11 Sauce, low-fat 16 Chocolate, incl. marzipan 4 Danish pastry, croissant etc. 10 Licorice, candy, jelly etc. 2 Cream buns 18 Aggregated food categories Agregated food categories 15 12 Tomatoes of various types *Pork total Eggs and buttermilk dish with raw eggs 7 *Sliced meat for "smørrebrød" low+high 2 fat Fruit Milk products Food items typical for Danish "Smørrebrød" (open sandwich) Meat Fast food Fish Vegetable Others

Table 2.2. Top-25 food preferences for two foodborne outbreaks, sorted by food type

Note: * and **bold**: Indicator food items for the outbreak source (baby corn for outbreak FUD 627 had no separate food category). Source: National Food Institute and Wingstrand et al., 2013

3. Risk factors for *Campylobacter* **colonisation in Danish broilers**

By Birgitte Borck Høg (bibo@food.dtu.dk) and Helle Mølgaard Sommer

Campylobacteriosis remains the most common cause of bacterial foodborne gastrointestinal illness in Denmark (Table A2) and broilers have been identified as the primary source [1, 2], though other sources also exist. Therefore, reducing the occurrence of *Campylobacter* in the broiler production is considered important for reducing the number of human cases. Risk factor studies have been carried out to identify the factors that significantly affect the risk of broilers becoming colonised with *Campylobacter*. In the following, results from two Danish studies are presented and discussed.

The first study (Study A) included the *Campylobacter* status for nearly 6,000 conventional broiler flocks from 244 farms and 43 explanatory variables. The *Campylobacter* data were obtained through the national surveillance programme, while the explanatory data were obtained through interviews with broiler farmers using a standardised questionnaire. The *Campylobacter* status of the flocks was followed over a 2-year period (1999-2000) and data were analysed by multivariate analysis using a generalised linear model. The unit of analysis was the broiler house, meaning that all results from a broiler house were aggregated, i.e. number of positive flocks out of the total number of flocks delivered over the 2-year period per broiler house [3].

The second study (Study B) included the Campylobacter status for approximately 8,000 conventional broiler flocks from 107 farms and 44 explanatory variables. This study was part of the EU financed "CamCon" project investigating novel approaches for controlling Campylobacter in the primary poultry production [4]. The explanatory data were collected using a standardised questionnaire filled out by the broiler farmers and the Campylobacter data were obtained through the national surveillance programme [5]. The questionnaire data were collected from December 2010 to January 2011 and Campylobacter data were collected from January 2010 through March 2012. These data were analysed using the same principles as in Study A, but the unit of analysis in this study was the broiler farm, meaning that all results from a broiler farm were aggregated into one prevalence measure, i.e. number of positive flocks out of the total number of flocks delivered over the 2-year period per farm.

Results

In Study A, the most important risk factors for a flock being *Campylobacter* positive were the age of the broiler house and quality of the rodent control procedures on the farm. Other important factors, but to a lesser extent, were age at introduction of whole wheat in the feed, age at slaughter, improper storage of whole wheat, number of chimneys on the broiler house, density of cattle farms in the surrounding area and having more than one broiler house on the farm.

Study B found the age of the broiler house, density of birds, level of biosecurity, length of downtime and type of drinkers to be associated with the risk of the broiler flocks becoming colonized by *Campylobacter*.

The statistically significant risk factors identified in the two studies with the associated p-values are presented in Table 3.1 and 3.2.

Discussion

While the two studies were designed differently, they shared many features. It was, therefore, of interest to compare outcomes of the two studies to identify differences and similarities as well as potential changes over time.

From 1999 to 2011, the Danish broiler industry has undergone some important changes. During this time, the number of broiler farms has decreased. Furthermore, in 2003 a strategy against Campylobacter was adopted (Annual Report, 2003), following which the Campylobacter flock prevalence in broiler flocks decreased. In 2008 an action plan was introduced, and testing of all flocks became mandatory from 2010 (Order no 1462 of 16/12/2009), see Appendix Table A33). A key element of this plan was to develop and implement a quality assurance programme¹ for broiler farmers, in order to increase the focus on biosecurity measures on broiler farms. Biosecurity measures include having anterooms divided in separate zones and/ or hygiene barriers at the house entrance; changing clothes and footwear and washing hands before entering the broiler house; using clean drinking water; rodent control and having vegetation free zones around the house.

Today, conventional broiler farms that produce broilers for the two main broiler companies in Denmark (98%

^{1.} Kvalitet i Kyllingeproduktionen (KIK)

Source of variation	p-value (type 3 test)	Effect on the prevalence	e	
Age of broiler house	< 0.001	Older houses	\rightarrow	Higher prevalence
Rodent control (improper)	< 0.001	1 Improper rodent control		Higher prevalence
Age of chickens at introduction to whole wheat	0.01	Older chickens	\rightarrow	Higher prevalence
Age of chickens at slaughter	0.01	Older chickens	\rightarrow	Higher prevalence
Storage of wheat	0.01	Inappropriate storage	\rightarrow	Higher prevalence
Number of chimneys	0.03	Many chimneys	\rightarrow	Higher prevalence
Two or more broiler houses	0.03	Several houses	\rightarrow	Higher prevalence
Density of cattle farms	0.04	Cattle farms close by	\rightarrow	Higher prevalence
No. of observations in the final model	469	·		

Table 3.1. Significant explanatory variables from Study A, 1999-2000.

Source: Sommer, HM et. al, 2013

Source of variation	p-value (type 3 test)	Effect on the prevalence	;	
Age of house	0.04	Older house	\rightarrow	Higher prevalence
Average stocking density	0.01	Lower density	\rightarrow	Higher prevalence
Own anteroom for each house	0.01	Not separate anteroom	\rightarrow	Higher prevalence
Bootdip at broilerhouse entrance	0.03	Bootdip present	\rightarrow	Higher prevalence
Average length of downtime	0.03	Long downtime	\rightarrow	Higher prevalence
Type of drinkers	0.04	Drinkers with cup	\rightarrow	Higher prevalence
No. of observations in the final model	93			

Table 3.2. Significant explanatory variables from Study B, 2010-2011.

Source: National Food Institute

of the Danish broiler production) comply with a quality scheme laid down by the broiler companies. These changes in management affected the responses to the questionnaire in Study B, i.e. all involved farms had agreed to comply with the industry's quality assurance programme and there was very little variation in the responses to the biosecurity questions [5].

Different questionnaires were used in the two studies. In Study A, data referred to the house level, while Study B referred mainly to farm level variables. It should also be taken into account that data were collected differently as questionnaire data collected through interviews (Study A) are likely to be more uniform than data filled out by farmers themselves (Study B), even when standardised questionnaires are used.

Both studies, identified the age of the broiler house as a significant risk factor. Thus, flocks raised in older houses were more often found to be colonised with *Campylobacter*. This may be explained by the fact that it is more difficult to implement and uphold modern biosecurity standards in older houses, e.g. older houses may be less sealed towards the surrounding environment and *Campylobacter* may, therefore, be more easily introduced via insects, rodents and birds etc. Also older houses may be more difficult to clean and disinfect properly.

In both studies, a number of factors related to the biosecurity on the farms were found to be significantly associated with occurrence of *Campylobacter*. In Study A, it was insufficient rodent control, a high number of chimneys on the houses and improper storage of whole wheat used as feed (i.e. not stored in a silo). A large number of chimneys provide easy access to the broiler house for flies and other insects, thereby increasing the risk of *Campylobacter* being introduced in the house. Improper storage of wheat is likely to be associated with birds and rodents that may contaminate the wheat with *Campylobacter* while it is in storage.

In Study B, the risk factors were lack of a separate anteroom, the presence of boot dips, the length of downtime between flocks (the number of days from a house is emp-

tied until it is restocked), and the type of drinkers used (nipples with or without cups or bells). It was interesting that boot dips were associated with a higher Campylobacter prevalence (Study B). Boot dips are typically placed at the entrance to the broiler houses and used to disinfect boots before entering the house and they are usually an alternative or supplement to changing boots. However, the effectiveness of the dips is highly dependent on whether or not they are maintained correctly. The fact that they can be associated with a higher Campylobacter prevalence may reflect that the boot dips are not maintained properly, leading to insufficient disinfection of footwear. It could also be speculated that the presence of the boot dips may lead to a lack of focus on the importance of the boots as vehicles of Campylobacter, e.g. the farmer may be less likely to change boots or physically clean the boots if boot dips are available.

Study B found a long downtime to be associated with a higher prevalence of Campylobacter among the broiler flocks. At first glance this seems somewhat odd, since a long down-time, in theory, allows the broiler house to dry completely, providing Campylobacter with the least favourable conditions for survival. However, farms with long downtimes also seemed to have less efficient rodent control and less formalised cleaning procedures, so it could also reflect the overall management on the farm, i.e. farms with a long downperiod had a less intensive production and perhaps not completely formalised and scheduled procedures for upholding a high level of biosecurity at all times. Interestingly, Study B also found decreasing density of birds to be a risk factor. We speculate that this result may be a reflection of an overall efficient management, with a high level of biosecurity on farms with a high stocking density, but this could not be elucidated by the available data.

Other factors associated with a higher *Campylobacter* prevalence (Study A) was the "density of cattle farms in the area", and "number of houses on the farm". Both of these factors reflect, that if there is a high density of potential carriers of *Campylobacter* in close proximity of the broiler house, there is an increased risk of the flocks being colonised. Age at slaughter was also found to be associated with a higher risk, and may simply reflect that the longer the broilers live, the more likely they are to be exposed to *Campylobacter* from the environment [3].

Overall, the results of both Study A and Study B point to the importance of having broiler houses where it is possible to maintain an overall high level of biosecurity. It seems especially important to properly control carriers of *Campylobacter* such as rodents. Study B also included information on whether or not the broiler houses had fly screens on the ventilation inlets to prevent insects from entering. However, only three farms had fly screens and the factor was not found significant, but the p-value was as low as 0.12. The importance of preventing carriers such as insects from entering the broiler houses has previously been demonstrated [6].

The data from Study B will later be re-analysed with comparable data from five more countries to investigate if different countries in Europe share the same risk factors for broiler flock colonization.

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Risk factors for Campylobacter colonisation in Danish broilers



4. *Salmonella* and indicator bacteria in pork: a comparison of butcher shops and supermarkets

By Tina Beck Hansen (tibha@food.dtu.dk), Anne Mette Bollerslev, Gudrun Sandø and Søren Aabo

To investigate the prevalence of *Salmonella* in pork at retail in Denmark, six surveys have been conducted since 2001. In 2001/2002, a survey investigating *Salmonella* and enterococci in minced pork was conducted (Table 4.2) and in 2002, a national survey on *Salmonella* in pork cuttings was carried out (Table 4.1). The aim was to estimate the *Salmonella* prevalence in meat from butcher shops and supermarkets. Approximately 50 % of all retail stores in Denmark were included in the surveys and three samples were collected in each shop. For each sample the meat supplier was identified.

In 2006, the national survey on *Salmonella* in pork cuttings was repeated for approx. 14 % of Danish retail stores (Table 4.1). It was found that the overall prevalence at retail had increased significantly from 2002 to 2006 and that the *Salmonella* prevalence in pork sampled in butcher shops and supermarkets was significantly different [1]. During the same period there was no increase in the prevalence of *Salmonella* positive carcasses at Danish slaughterhouses, suggesting that the increase most probably was related to changed hygiene performances either at the cutting plants or at the retailers themselves. Consequently, three follow-up surveys were conducted to investigate the possible causes. The occurrence of enterococci and Enterobacteriaceae is hypothesized to be negatively correlated to hygiene performance. These indicators were, therefore, used to supplement *Salmonella* detection in the samples in all three follow-up surveys.

The first survey was carried out in 2010 at 18 cutting plants with the objective to determine the prevalence of *Salmonella* in pork sampled from small and large cutting plants (Figure 4.1). The second survey was performed in 2010/2011 and was a repetition of the national surveys on *Salmonella* in pork cuttings carried out in 2002 and 2006 (Table 4.1). This time three pork cuttings were sampled from each of 134 butcher shops and 278 supermarkets. In the third survey conducted in 2011, 20 butcher shops and 20 supermarkets were selected for a detailed study combining microbiological swabs of equipment with sampling of pork from two processing lines producing minced pork and pork chops, respectively (Table 4.2, Figure 4.3 and 4.4). In all three surveys, *Salmonella* was detected in 25 g meat

		2002	2		200)6		2010/2011			
Type of retailer	Ν	%pos	[95% CI]	Ν	%pos	[95% CI]	Ν	%pos	[95% CI]		
Butcher shops	1,025	1.8	[1.0-2.8]	259	8.1	[5.1-12.0]	40	1.0	[0.3-2.5]		
Supermarkets	3,473	1.0	[0.7-1.4]	628	2.5	[1.5-4.1]	834	4 0.7	[0.3-1.6]		
Total	4,498	1.2	[0.9-1.5]	887	4.2	[2.9-5.7]	1,235	5 0.8	[0.4-1.5]		

Table 4.1. Detection of Salmonella in 25 g pork cuttings sampled from Danish butcher shops and supermarkets

Source: Danish Veterinary and Food Administration

		Minced meat							Cuttings			
	2001/2002			2011		2010/2011			2011			
Type of retailer	Ν	%pos	[95%CI]	Ν	%pos	[95%CI]	Ν	%pos	[95% CI]	Ν	%pos	[95% CI]
Butcher shops	434	36.9	[32-42]	72	38.9	[28-52]	401	10.2	[7.4-14]	68	16.2	[8.4-27]
Supermarkets	1,160	29.2	[27-32]	87	11.5	[5.7-20]	828	3.9	[2.7-5.4]	68	2.9	[0.4-10]
Source: Danish Va	torinory	and Eag	d Administ	ration								

Source: Danish Veterinary and Food Administration

samples from shoulder, middle or hind parts of the carcass. When pork samples were analysed for enterococci and Enterobacteriaceae, quantitative methods were applied. Swab samples from equipment were analysed qualitatively for *Salmonella* and quantitatively for enterococci and Enterobacteriaceae.

Salmonella prevalence at retail

The overall prevalence of *Salmonella* in fresh pork at retail increased significantly from 1.2 % in 2002 to 4.2 % in 2006 (Table 4.1). In 2010, the prevalence was, however, similar to in 2002, indicating that the findings in 2006 might be a peak rather than an increasing trend. In all three surveys, a higher prevalence was found for butcher shops compared to supermarkets. A higher occurrence of enterococci in minced pork was also found for butcher shops compared to supermarkets in the surveys from 2001/2002 and 2011 and for pork cuttings analysed in 2010/2011 and 2011 (Table 4.2).

These results suggest that poor hygiene in butcher shops resulted in a higher *Salmonella* prevalence compared to supermarkets. However, the supply patterns for butcher shops and supermarkets differed considerably [2,3]. Hence, it could not be excluded that the higher *Salmonella* prevalence for butcher shops was a result of higher input of *Salmonella* from the supplying cutting plants.

Salmonella input to retail from cutting plants

In 2010, the occurrence of *Salmonella* and enterococci was analysed in 759 samples from six large cutting plants and 770 samples from 12 small cutting plants (Figure 4.1) as well as for Enterobacteriaceae (results not presented). The *Salmonella* prevalence was significantly lower at the

large cutting plants (1.3 %) compared to the small cutting plants (3.9 %). The occurrence of Enterobacteriaceae and enterococci was also significantly lower for the large cutting plants, indicating differences in hygiene performance between large and small cutting plants. Results revealed large variations between cutting plants in the prevalence of *Salmonella* and occurrence of enterococci; particularly among the small cutting plants, where *Salmonella* prevalences from 0 % to 22 % and occurrences of enterococci from 0 % to 65 % were found.

Based on the surveys from 2002, 2006 and 2010/2011, 75 % of the pork sampled in the butcher shops were supplied by small cutting plants and 87 % of the pork sampled in the supermarkets were supplied by large cutting plants (results not presented). This information indicated that a higher input, particularly of *Salmonella* and enterococci, would be expected for butcher shops compared to supermarkets.

Hygiene performance at retail

In 2011, differences in hygiene performance on two processing lines at retail producing minced pork and pork chops, respectively, were investigated in a survey including 20 butcher shops and 20 supermarkets. Hygiene performance reflects itself in: 1) cross-contamination events, 2) microbial growth during handling, and 3) microbial growth during storage as a result of time and/or temperature abuse. Samples were analysed for presence of *Salmonella* and for the concentration of Enterobacteriaceae and enterococci.

As expected from the results presented in Figure 4.1, prevalence of *Salmonella* and occurrence as well as concentration of enterococci in the raw material was higher for butcher shops compared to supermarkets, which was not

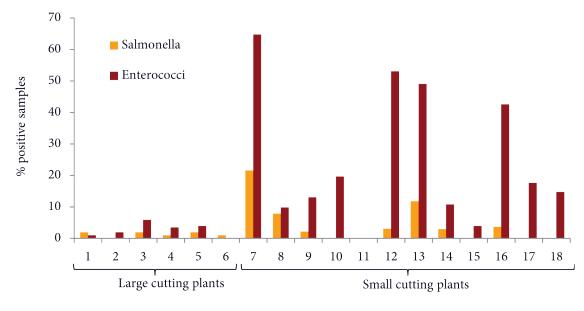


Figure 4.1. Salmonella prevalence (detection in 25 g samples) and occurrence of enterococci (\geq 100 cfu/g) in pork cuttings sampled from 18 Danish cutting plants in 2010.

the case for Enterobacteriaceae (results not presented). This identified Enterobacteriaceae as a good microbial indicator for detection of differences in hygiene performance as the influence of raw material could be excluded.

Pair-wise comparisons of Enterobacteriaceae counts in raw material and processed meat showed that crosscontamination might play a significant role in butcher shops, as low input raw material ended up as processed meat with high concentrations of Enterobacteriaceae (Figure 4.2). This was not the case for supermarkets where no significant difference was observed for Enterobacteriaceae counts in raw material and processed meat (Figure 4.2). However, cross-contamination events might be affected by the prevalence of the microorganism in question, meaning that different cross-contamination patterns might be observed for highly prevalent organisms in pork, e.g. Enterobacteriaceae, compared to low prevalent organisms, e.g. Salmonella. Compared to Salmonella, other Enterobacteriaceae are highly prevalent in pork, and, therefore, results for Enterobacteriaceae might not be directly applicable to cross-contamination mechanisms for Salmonella. Consequently, the low prevalent indicator enterococci might be more suitable for identification of cross-contamination risk from Salmonella during processing, such as chopping and grinding.

In butcher shops, a higher proportion (approx. 3-fold) of the enterococci negative samples became positive during processing compared to supermarkets. As the concentration of enterococci in raw material and processed meat was comparable, this difference probably resulted from the significantly higher occurrence of enterococci in raw material in butcher shops compared to supermarkets, leading to enhanced cross-contamination risk in the butcher shops. This was also observed as a 2- to 4-fold higher occurrence of enterococci on cutting boards and in meat grinders for the butcher shops compared to the supermarkets (results not presented).

Salmonella was only detected in one of the 232 swabs of equipment performed in the survey. This was from the meat grinder in a butcher shop. *Salmonella* was also detected in the raw material and the minced pork processed in the particular grinder.

Measured as concentration of Enterobacteriaceae on cutting boards, the risk of cross-contamination appeared to be higher in the shops handling meat at temperatures $\geq 18^{\circ}$ C (Figure 4.3). The combination of production temperatures $\geq 18^{\circ}$ C and a time period ≥ 4 hours since the cutting board was last cleaned resulted in significantly higher levels of Enterobacteriaceae (Figure 4.3). For samples collected at temperatures $\geq 20^{\circ}$ C, increased levels of Enterobacteriaceae were found already after 1-2 hours (results not presented).

The proportion of cutting boards with more than 100 enterococci per 400 cm² was also highest when \geq 4 hours had elapsed since the last cleaning of the cutting board in a production area with temperature \geq 18°C (results not presented). As the cutting boards harbouring the highest Enterobacteriaceae levels also harboured the highest enterococci levels, it indicated that microbial growth had occurred in meat residues present on these cutting boards.

These results pinpointed the microbial status of cutting

Figure 4.2. Paired Enterobacteriaceae counts (log cfu/g) in raw material and processed meat in Danish butcher shops and supermarkets.

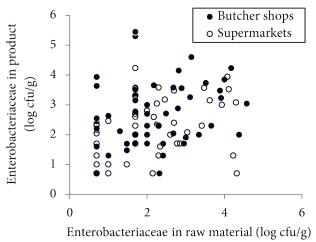
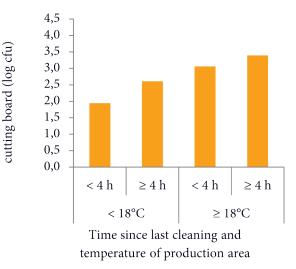


Figure 4.3. Concentration of Enterobacteriaceae on 400 cm² cutting board used for processing of raw pork in 38 Danish retailers in 2011.



Counts below the detection limit were included by assuming these to be 50 % of the detection limit.

Enterobacteriaceae counts below the detection limit were included by assuming these to be 50 % of the detection limit.

boards as an indicator of the hygiene performance in the retail stores, although the impact of cutting board hygiene was most pronounced for butcher shops. As shown in Figure 4.4a and 4.4b, the distribution of Enterobacteriaceae counts in samples used for production of minced pork shifted toward higher concentrations for butcher shops after processing on the cutting board and when analysing samples from the cutting boards, significantly higher levels of Enterobacteriaceae were found for butcher shops compared to supermarkets (Figure 4.4c).

Conclusion

The present surveys analysed the differences in Salmonella prevalence and other process hygiene indicators in pork sampled in butcher shops and supermarkets in Denmark during the past 10 years. Results from the supermarkets showed that microbial contamination of processed pork could primarily be explained by the input via the raw material, whereas results from the butcher shops showed that hygiene performance had an additional significant effect. To keep the presence of Salmonella in pork at retail low, these results suggest to focus on hygiene practices both in smaller cuttings plants and butcher shops. The industry has been involved in the project throughout the period, and has used the results constructively in their communication and dialogue with cutting plants and butcher shops. Furthermore, the project has identified critical procedures and factors associated with hygiene performance, which has led to the preparation of relevant tools for the establishments in order to support a high level of hygiene.

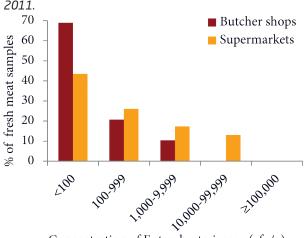
The projects were funded by the Danish Veterinary and Food Administration and carried out in cooperation with the National Food Institute with involvement of indstry stakeholders

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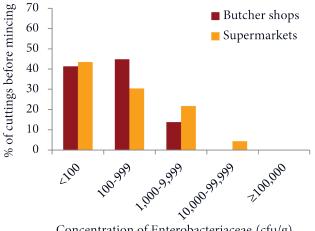
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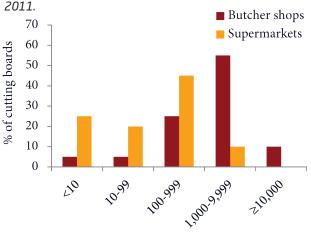
Concentration of Enterobacteriaceae (cfu/g)

Figure 4.4b. Distribution of Enterobacteriaceae after processing of pork cuttings used for minced pork in Danish butcher shops and supermarkets in 2011.



Concentration of Enterobacteriaceae (cfu/g)

Figure 4.4c. Distribution of Enterobacteriaceae on cutting boards from Danish butcher shops and supermarkets in



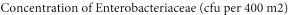


Figure 4.4a. Distribution of Enterobacteriaceae in raw material from Danish butcher shops and supermarkets in

5. Risk management initiatives against microbiological risk from fruits and vegetables

By Niels Ladefoged Nielsen (nln@fvst.dk)

Foodborne diseases caused by contaminated fruits and vegetables are an increasing problem globally and several large outbreaks have been described during the last few years – including the VTEC O104 outbreak in Germany and France in 2011 caused by contaminated fenugreek seeds / sprouts, and a large outbreak of norovirus in Germany in 2012 caused by frozen strawberries from China. In recent years, foodborne outbreaks caused by fruits and vegetables have been reported in Denmark – e.g. *Salmonella* in tomatoes and norovirus in raspberries (See Annual Report 2011 and 2010 for more details).

The increasing number of foodborne outbreaks caused by fruits and vegetables has led to a number of risk management initiatives described in a report by the Danish Veterinary and Food Administration (DVFA):

- The Order No 970 on Food Hygiene, September 28th 2012 has been revised introducing mandatory heat treatment of frozen raspberries used as an ingredient in dishes (e.g. deserts, cakes and smoothies) intended to be served without further heat treatment
- The DVFA Guideline on Hygiene has been revised introducing guidelines for handling fruits and vegetables – including washing, heat treatment of certain risk foods like baby corn and sugar snaps, and refrigerated storage of e.g. sprouts
- DVFA Guidelines regarding the import of fruits and vegetables have been introduced including the obligation for the food business operators to have procedures to ensure that suppliers fulfill a certain level of hygiene in production and distribution, e.g. as stated in the Guidelines from Codex Alimentarius or the Global GAP concept. These procedures must be described in their own check system
- Consumer information campaigns
 - * Television spots informing the consumers about the importance of thoroughly washing of fruits and vegetables in order to reduce the risk of food borne disease
 - * Free post cards in cafes and restaurants advising the consumers to boil frozen raspberries in order to avoid the risk of being infected by norovirus

- · Restaurant and catering business campaigns
 - * During 2012, the DVFA performed a campaign especially aiming at increasing the awareness of risk associated with fruits and vegetables and the correct handling of these in restaurants and in the catering business
 - * A control campaign related to the legislation on the import of fruits and vegetables is planned for 2013.

Since 2009, a centrally coordinated monitoring project on microbiological contamination of fruits and vegetables has been performed by the DVFA laboratories. In 2012, results of the monitoring program shows that especially imported leafy greens and herbs may pose a risk regarding exposure to *Salmonella* (Table 5.1). This is similar to findings from 2010 and 2011. *Salmonella* has never been detected in products of Danish origin. *Campylobacter* was detected in one batch of leafy greens from Denmark in 2012. In previous years, *Campylobacter* was mainly found in different imported products.

High levels of *E. coli* were detected in samples of baby corn, sugar snaps, leafy green, sprouts and herbs. This is similar to results from 2010 and 2011.

In 2012, fruits and berries were included in the monitoring project for the first time. Six batches of berries of EU origin, nine batches of berries produced outside the EU and 48 batches of apples and pears were tested. *Salmonella*, *Campylobacter* or high levels of *E. coli* were not detected in any of these batches.

For further details on the results of the study on for fruits and vegetables in 2012, please see Table 5.1.

	Saln	ionella	Campyl	obacter	<i>E. coli</i> (>	-100 cfu/g)
Type of sample	Ν	Pos	Ν	Pos	Ν	Pos ^j
Vegetables						
Baby corn	19	0	19	0	19	7
Cucumber	21	0	21	0	21	0
Pepper	28	0	28	0	28	0
Salad	48	3 ^e	48	1^{i}	48	5
Sprouts	18	0	18	0	18	2
Sugar peas	34	0	34	0	34	2
Tomato	41	0	41	0	41	0
Other vegetables ^c	22	0	22	0	22	0
Herbs						
Coriander	5	0	5	0	5	0
Lemon Grass	2	1^{f}	2	0	2	0
Parsley	13	0	13	0	13	3
Rosemary	2	0	2	0	2	0
Sage	2	0	2	0	2	0
Spearmint	7	0	7	0	7	3
Spring onions	17	1 ^g	17	0	17	1
Tarragon	3	0	3	0	3	1
Other herbs ^d	35	$1^{\rm h}$	35	0	35	1
Fruit and berries						
Apples	32	0	32	0	32	0
Pears	16	0	16	0	16	0
Berries	15	0	15	0	15	0
Other fruit and berries ^k	37	0	37	0	37	0
Total	417	6	417	1	417	25

Table 5.1. Pathogens in batches^a of ready-to-eat vegetables and herbs^b, 2012

a) Five samples per batch

b) Centrally coordinated study (see section 10.4 for description) to control and investigate Salmonella, Campylobacter and E. coli

in Danish and imported ready-to-eat vegetables, sprouts and herbs.

c) including cauliflower, cabbage, broccoli, beans, zucchini and onions

d) including chives, basil, thyme, dill, watercress, lovage, garlic, lemon balm, leafy celery, oregano and salicornia

e) S. Napoli in 2 batches of leafy green from Italy and S. Bongori 48:z35:- in one batch of leafy green from Germany

f) S. Infantis and S. Javiana in one batch from Thailand

g) S. 8,20:y:- in one batch from Thailand

h) S. Brunei in a leafy celery batch from Vietnam.

i) One batch of leafy green from Denmark

j) Batches with >100 cfu/g in one or more samplesk) includes plum, peach, persimmon fruit, nectarine, grapes, carom, cherries

Source: Danish Veterinary and Food Administration



6. Outbreaks of special interest

By Central outbreak management group

For the investigation of national foodborne outbreaks, outbreak control teams typically involve members from Statens Serum Institut, from the National Food Institute, Technical University of Denmark and from the Danish Veterinary and Food Administration. Local outbreaks are often handled by the Food Control Offices (FCO) with assistance from the Danish Health and Medicine Authorities (medical officers). The outbreak investigation procedures in Denmark are described in further detail in Chapter 8.2.

Investigators have access to the online Food- and Waterborne Outbreak Database (FUD) where notification and reporting of outbreaks take place. A table with an extract from this database listing the outbreaks that occurred in 2012 is shown in Appendix Table A4. Household outbreaks and clusters that could not be verified as common source outbreaks are not included in the table. As seen from the table, 82 outbreaks were reported to FUD in 2012. In total 2,203 cases were reported with a median of 25 per outbreak (range 2-200). Only five of the reported outbreaks were national outbreaks, which were slightly more than reported in 2011 (three national outbreaks). Half of all the outbreaks were reported as taking place in restaurants, hotels or canteens and half of all outbreaks occurred in the Copenhagen area. Norovirus was by far the pathogen associated with the most outbreaks (Figure 6.1); these outbreaks accounted for

more than half of all recorded outbreaks (45 outbreaks). No travel associated outbreaks were reported. A few selected outbreaks of special interest are described below.

Outbreak of VTEC and HUS

In September and October 2012, Denmark experienced an outbreak of Verocytotoxin-producing E. coli (VTEC) O157:H7 (FUD no. 1210) [1]. The toxin profile of the outbreak strain, eae, vtx1a and vtx2a, had rarely been seen in Denmark. A total of 14 cases from ten families were identified - two cases were found by screening healthy family members. Eight patients developed haemolytic uraemic syndrome (HUS). No deaths occurred among the patients. An epidemiological investigation pointed towards ground beef as the vehicle of infection. All cases had eaten ground beef prepared at home during the incubation period. In six of the ten households, the cases had eaten pan fried ground beef patties- a known vehicle for VTEC O157 outbreaks. In two of these six households, the pan fried ground beef patties were undercooked in the middle when eaten. The Danish Veterinary and Food Administration collected information on purchase dates and other labeling information on the ground beef consumed by five families and used this information to trace back the ground beef. The outcome of this investigation showed, that the meat used

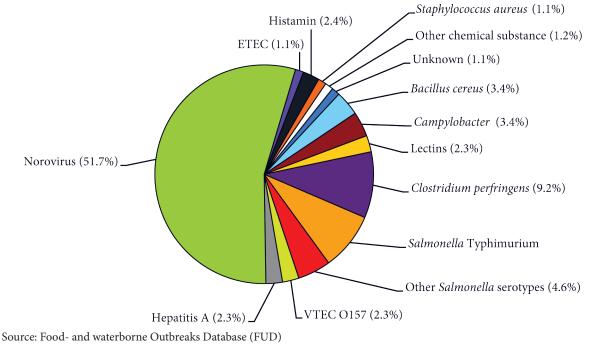


Figure 6.1. Aetiology of the 82 foodborne disease outbreaks reported with a causative agent in the Food- and waterborne Outbreak Database (FUD), 2012. Percentage of total outbreaks indicated in brackets

as raw material for the production of minced meat on two implicated establishments possibly came from animals slaughtered on either one of two possible slaughter dates on either one of two major slaughterhouses in Denmark. No meat samples were available from the slaughterhouses from the relevant slaughter dates and it was, therefore not possible to conclude from which date and slaughterhouse the contaminated meat came, nor was it possible to definitely exclude if contaminated meat from other sources than the establishments in question could be the source of the outbreak. Three samples of ground beef from the home of a patient were collected and analyzed – but none of them were found positive for VTEC.

This was the first VTEC outbreak in Denmark initially apprearing as a cluster of HUS cases. The outbreak could have been larger had it not been for the short shelf life (7 days) of ground beef sold chilled and packaged in modified atmosphere as well as the fact that this type of beef is generally thoroughly cooked in Denmark. This outbreak further underlines the importance of timely diagnosis and subtyping of VTEC.

Outbreaks with Salmonella

An increase in the number of outbreaks with *S*. Typhimurium and its monophasic variant (3 and 4 outbreaks respectively) was observed in 2012, as only two outbreaks with *S*. Typhimurium were reported in 2011. However four outbreaks with *S*. Typhimurium and two monophasic outbreaks were reported back in 2010. The largest outbreak in 2012 (FUD 1191) occurred in May-June and was caused by *S*. 4,5,12:i:- MLVA type 0006 with resistance pattern: Amp-Strep-Sulpha-Tet (R-ASSuT). A catering company in the Copenhagen area was found to have caused sub-outbreaks in a series of small companies to which it supplied lunches. Participants in a sports event also got ill after eating food provided by the same catering company. Epidemiological investigations indicated that cross contamination from marinated beef to other ready-to-eat dishes served at these locations had caused the illnesses. During the same period 24 cases infected with the same strain appeared throughout Denmark (FUD 1192). Case-interviews could not clarify the cause of disease; however the interview results indicated beef as the source of the infection.

Outbreaks with S. Enteritidis have become rare in the recent years as a result of the successful Salmonella control programs targeting the Danish laying hens and broilers and the EU harmonised Salmonella programmes in laying hens and broilers which include a ban on sellling fresh eggs from flocks positive with S. Enteritidis or S. Typhimurium.

Several small outbreaks with exotic *Salmonella* serotypes were reported including an outbreak with *S*. Bareilly with eight confirmed cases; three of the cases were identified by active case finding. Seven cases could be traced back to an unknown food source served at a specific restaurant in the city of Aarhus (FUD 1215). Another outbreak was caused by the rare *Salmonella* serotype *S*. Mikawasima and included 3 confirmed cases (FUD 1206). Cases were also reported from Norway and the Netherlands; however the source was not identified.

Outbreaks with norovirus

As in previous years, norovirus was by far the pathogen involved in most outbreaks (Figure 6.1). Laboratory confirmation of the diagnosis was not present for all outbreaks; some were classified as norovirus outbreaks based on the symptoms typical for norovirus (Kaplan criteria) [2]. However, investigators will often ask a small number of cases to submit a stool sample; and the number of Danish clinical laboratories offering PCR analysis for the presence of norovirus is steadily increasing. In 2012, the majority

Increase of *Yersinia enterocolitica*

Routine surveillance identified an increase in the number of *Yersinia enterocolitica* cases in Denmark from January to April 2012. An investigation was initiated to assess whether an outbreak was occurring or whether the observed increase in cases could be explained otherwise.

The number of *Y. enterocolitica* cases in 2012 was compared with the number of cases reported in previous years and analysis of geographical information and age distribution of cases was performed. Additionally, available biotype and serotype data were analyzed and PFGE was performed on a subset of the isolates. The presence of virulence genes among isolates was assessed by PCR.

The investigation indicated a 50-100 % increase in the number of cases per month in the first 4 months of 2012 compared to previous years. Typing showed that 72 % of all typed isolates was Biotype 1A and no unexpected virulence genes were found in these isolates. PFGE showed a wide variety in patterns among the tested isolates. Based on the available information, it was concluded that the *Y. enterocolitica* cases from January to April 2012 did not appear to result from a single source outbreak.

of the outbreaks were caused by infected food handlers contaminating the food when preparing it in restaurants, canteens or similar places.

In Denmark, waterborne outbreaks are quite rare and have occured on average 3-4 times per decade. However since 2007, four waterborne outbreaks have been registered in 2007, 2009, 2010 and 2012. In December, 2012, a waterborne norovirus outbreak took place in a town in the western part of Zealand, following contamination of the drinking water system supplying approx. 500 inhabitants (FUD 1233). A total of 18 cases of norovirus infection were laboratory confirmed to be genotype GII.4. Norovirus of the same genotype was also detected in water samples. There were an estimated 250 cases in total based on results from a questionnaire survey conducted by Statens Serum Institute. The study showed a dose-response relationship between intake of tap water and the risk of becoming ill. The cause of the contamination was identified as a leakage to an underground drinking water pipe running beneath a leaking sewage pipe - the contamination occurred following maintenance work on the water system, which meant that the pressure was temporarily lowered, whereby small amounts of sewage water could enter the drinking water system from the outside.

An outbreak of Cryptosporidium among veterinary students

In early spring 2012, illness was reported by several veterinary students, who had attended the same university course. The students suffered from diarrhea, abdominal pain and stomach cramps. In class, the students (divided in groups) had been working with calves that had diarrhea. It was suspected that the diarrhea was due to *Cryptosporidium* which is a very common infection in young calves, and also pathogenic for humans. Illness was also reported among veterinary students who had attended

another course with livestock animals including calves. One of the students had been hospitalized. University of Copenhagen contacted the National Food Institute and Statens Serum Institut in order to investigate the event. The students were encouraged to submit a stool sample, and six students were tested positive for Cryptosporidium parvum. An electronic questionnaire was circulated by email to the 106 students who had been attending the two courses. In total, 73 students filled in the questionnaire, and of these 24 had had symptoms of cryptosporidiosis. There had been no shared meals or drinks among the students and none had been drinking water during class. All had worn gloves and lab coats, but none had used face masks. The investigation showed that illness was not equally distributed among the groups of students and some of the infected students reported that cleaning of the classroom was conducted while they were still in the room. This was done by flushing the dirty surfaces with a water jet, which caused spray and aerosol formation in the room.

It was concluded that the *Cryptosporidium* outbreak was associated with contact to calves, but whether cleaning of the classroom while there were still students present and/or changing of clothes after class were contributing factors could not be determined. Following the outbreak, the cleaning procedure has been changed and students are now instructed to take hygienic precautions.

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7. International topics

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7.1 Control of zoonoses in animal populations 7.1.1 EU coordinated monitoring studies

Based on Zoonosis Directive 2003/99/EC and Regulation (EC) No 2160/2003, the Commission can initiate harmonised studies in order to generate comparable prevalence data from all Member States with the purpose of setting common EU targets for the reduction of the pathogens in question. So far, eight such baseline studies have been carried out concerning *Salmonella*, *Campylobacter*, *Listeria* and MRSA. The EU results have been published on the EFSA website (www.efsa.eu), except from the results of the baseline study on *Listeria*. The Danish results from these studies have been presented in Annual Reports 2005-2009.

7.1.2 EU harmonised *Salmonella* surveillance programmes

Based on the results of baseline studies in flocks of poultry, harmonised regulation on targets and surveillance in the poultry production has been laid down by the Commission.

In 2010, the EFSA published a scientific opinion [1] on monophasic *S*. Typhimurium-like strains in which it was concluded that the monophasic *Salmonella* strains with the antigenic formula *S*. 1,4,[5],12:i:- should be treated equal to *S*. Typhimurium. Subsequently EU regulations on target end criteria set on *Salmonella* have included these types as well.

According to Regulation (EC) No 584/2008, the EU target at 1% for breeding and fattening turkey flocks positive for S. Typhimurium or S. Enteritidis had to be reached at the end of 2012. This regulation has now been replaced by Regulation (EC) No 1190/2012 laying down a permanent target of maximum 1% positive breeding and fattening turkey flocks. The regulation enters into force in 2013. In Denmark, all turkey flocks were negative in 2012 (Appendix Table A11).

In breeding flocks of *Gallus gallus*, the target of 1 % positive adult flocks had to be reached by the end of 2009 according to Regulation (EC) No 1003/2005. The target was set for *S*. Typhimurium, *S*. Enteritidis, *S*. Hadar, *S*. Infantis and *S*. Virchow. This regulation has been replaced by Regulation (EC) No 200/2010 laying down a permanent target of maximum 1 % adult flocks positive for *S*. Typhimurium including the monophasic *S*. 1,4,[5],12:i:- strains, *S*. Enteritidis, *S*. Hadar, *S*. Infantis and *S*. Virchow. In the legislation no distinction is made between breeding flocks from the

table egg and broiler production lines. In Denmark, one pullet rearing flock from the table egg production was positive with *Salmonella* 4,5,12:i:- phage type DT120 in 2012 (Appendix Table A8 and A10).

The EU baseline study on table egg laying flocks carried out in 2004 showed large differences in the prevalence between Member States. Therefore, Member States specific targets were set either as an annual 10-40 % reduction of positive adult flocks dependent on the prevalence of adult flocks in the Member State the previous year or a maximum of 2 % adult flocks positive (Regulation (EC) No 1168/2006). The target was set for S. Typhimurium and S. Enteritidis and had to be reached by December 31st 2010. This regulation has been replaced by Regulation (EC) No 517/2011 laying down permanent targets for the reduction of Salmonella in laying flocks. The new regulation maintains the previous targets. For Denmark, the target is a maximum of 2 % adult flocks positive for S. Typhimurium including the monophasic S. 1,4,[5],12:i:strains and S. Enteritidis. The prevalence in Denmark has been lower than 2 % since 2004. In 2012, two flocks were positive for the target serotypes (S. Enteritidis and Salmonella 4,5,12:i:-) and one flock was positive for S. Infantis (Appendix table A8).

In broiler flocks of *Gallus gallus*, the target of maximum 1 % flocks positive for *S*. Typhimurium and *S*. Enteritidis had to be reached by December 31st 2011 according to Regulation (EC) No 646/2007. This regulation has been replaced by Regulation (EC) No 200/2012, which maintains the previous target at 1 % including the monophasic *S*. 1,4,[5],12:i:- strains. Denmark has had intensive *Salmonella* control programmes since the 90's and the target of 1 % was reached in 2000. In 2012, 0.2 % of broiler flocks was positive with *S*. Typhimurium including the monophasic *S*. 1,4,[5],12:i:- strains (Appendix Table A10).

7.2 Special guarantees for *Salmonella* in the Danish table egg production

Denmark obtained special guarantees for *Salmonella* in table eggs July 1st 2012. By granting special guarantees to Denmark, the EU has acknowledged the effort made in Denmark for reducing the prevalence of *Salmonella* in table eggs.

Denmark forwarded the application for special guarantees for *Salmonella* in table eggs to the EU according to article 8 (3) (b) of Regulation (EC) No 853/2004 laying down specific hygiene rules for food of animal origin in 2007. The Danish control programme for *Salmonella* in table eggs was discussed at several working group meetings in the EU and a guideline was developed setting up minimum requirements for a control programme to be regarded equivalent with the ones in Finland and Sweden. An EU expert group then evaluated the Danish control programme and concluded that the programme was equivalent to the programmes in place in Sweden and Finland.

Regulation (EC) No 427/2012 constitutes the legal basis for granting special guarantees to Denmark for table eggs. Special guarantees implies that eggs sold to Denmark should be followed by a specific certificate and the veterinary authorities in the country of origin must certify that the flocks of origin have been tested negative for all *Salmonella* serotypes, and not only the serotypes covered by the EU legislation (*S.* Enteritidis and *S.* Typhimurium including the monophasic *S.* 1,4,[5],12:i:- strains). Danish table eggs sold to the other Nordic countries no longer need to be accompanied by specific certificates to facilitate trade with Danish table eggs to other Nordic countries.

7.3 New legislation on sprouts

A major outbreak of verocytotoxin producing *Escherichia coli* (VTEC) O104 in Germany occurred in May 2011 (See Annual report 2011 for more details). As a follow up on this outbreak the EU Commission asked the European Food Safety Authorities (EFSA) to evaluate the risk related to sprouts and to provide guidance on possible additional risk management options. In November 2011, EFSA published a scientific opinion on the public health risk of VTEC and other pathogenic bacteria that may contaminate seeds and sprouted seeds.

The EU adopted the following four legal acts on sprouts and an accompanying Commission guidance document in 2012. The new regulations will apply from July 1st 2013:

• Regulation (EC) No 208/2013 on traceability requirements for sprouts and seeds intended for the production of sprouts

- Regulation (EC) No 209/2013 amending Regulation (EC) No 2073/2005 as regards microbiological criteria for sprouts and the sampling rules for poultry carcasses and fresh poultry meat
- Regulation (EC) No 210/2013 on the approval of establishments producing sprouts pursuant to Regulation (EC) No 852/2004 of the European Parliament
- Regulation (EC) No 211/2013 on certification requirements for imports into the EU of sprouts and seeds intended for the production of sprouts
- Guidance on Implementation of Certain Provisions of Regulation (EC) 852/2004 on Hygiene of Foods (as regards sprout production).

The key points in the new legislation are:

- A requirement of approval for establishments producing sprouts as these premises are regarded as primary production in the EU legislation
- A requirement for certificates for sprouts imported into Member States from third countries. The certificate should be used by the producers to attest the use of Good Agricultural Practice in primary production of seeds intended for sprouting
- Specific requirements to improve traceability for sprouted seeds in line with EFSA's recommendations.
- A new microbiological criterion for the absence of the six O-types of VTEC of major public health significance in sprouted seeds. Furthermore, a preliminary (pregermination) test for VTEC and *Salmonella* should be carried out on a representative sample from every consignment or batch of seeds used
- Guidance on best practice for the production of sprouts,
 e.g. storage of seeds, washing of seeds with potable water,
 the importance of keeping the products cold along the
 entire production chain, and hygiene of staff handling
 the seeds and sprouts.



7.4 Antimicrobial resistance

In November 2011, the European Commission published a five year action plan against the rising threats posed by antimicrobial resistance in which the Commission unveils 12 concrete action points. A substantial enforcement of actions are needed in order to reduce the use of antimicrobials, prevent further spread of resistance and preserve the ability to combat microbial infections in humans. EU-wide data published by the European Centre for Disease Prevention and Control (ECDC) on antibiotic resistance supports this need by showing that resistance to last-line antibiotics is increasing in Europe. Thus resistance to pathogens which frequently cause pneumonia and urinary tract infections in hospitals is increasing and is now established in several countries. In 2012, EFSA published two reports on the technical specifications on the harmonised monitoring and reporting of antimicrobial resistance in Salmonella, Campylobacter, indicator bacteria transmitted through food, and methicillin-resistant Staphylococcus aureus in food-producing animals and food [2,3]. In these reports proposals to improve the analysis and reporting of data in the EU Summary Reports on antimicrobial resistance in animals and food collected in the Member States have been described in detail. A working group on antimicrobial resistance with representatives from all Member States and the EU Commission are working on a proposal on harmonized monitoring of antimicrobial resistance in food and animals and a guidance document for prudent use of antimicrobials. The decision on harmonized monitoring is expected to be adopted in 2013 and enter into force in 2014.

7.5 The Danish EU Presidency 2012

Denmark held the EU-Presidency from January 1st to July 1st 2012; two important subjects dealt with were future meat inspections and antimicrobial resistance.

7.5.1 Conference on "Future Meat inspection for pigs"

A conference on "Future Meat Inspection for pigs" took place in February where authorities from most Member States participated as well as several third countries, the Commission and



representatives from the industry. With the overlying principle of protecting public health, animal health and animal welfare, a number of issues pertinent to the future meat inspection were discussed. An overall conclusion amongst the participants was that an evaluation of the current procedures was needed and that future meat inspections have to move towards a more risk based approach. During the conference, four issues were discussed, which also constitute the steps towards a more risk based and flexible inspection:

- No *Trichinella* testing of pigs from controlled housing conditions
- Focus on measures for a gradual reduction of *Salmonella* in pork
- Omission of routine palpation and incision when prerequisites are met
- Official inspection shall, besides animal health and animal welfare, focus on food safety only

The Commission has included these conclusions in the proposals on a future regulation on meat inspection and control of *Salmonella*.

7.5.2 Conference on "Combatting Antimicrobial Resistance - Time for Joint Action"

A conference on "Combating Antimicrobial Resistance – Time for Joint Action" was jointly organised in March by the Ministry of Health and the Ministry of Food, Agriculture and Fisheries. Experts from across the world were invited and the main issues of the conference were:

- Prudent use of antibiotics
- Critically important antimicrobials
- Improved data collection and monitoring of antibiotic use and resistance

The conference was followed by adoption of Council Conclusions (by the European Council). The conclusions call for intersectional collaboration, enhanced monitoring and a special focus on critically important antimicrobials, which is crucial in relation to maintaining options for treatment with antimicrobials. A very important statement in the conclusions was to keep a "One Health" approach when fighting antimicrobial resistance. A "One health" perspective is intended to strengthen the collaboration between the human and the veterinary field.

In line with the Council Conclusions, the Commission is now working on a proposal on harmonised monitoring for antimicrobial resistance in animals and meat in all member states.

7.6 The new Codex Alimentarius on microbiological criteria

The Codex Alimentarius Commission, established by the Food and Agriculture Organization of the United Nations (FAO) and the World Health Organization (WHO), has as its purpose to protect the health of consumers and ensuring fair practices in the food trade. In the field of food hygiene, the central standards are published as the "Codex



basic texts on food hygiene" [4]. These aim to promote the understanding of how rules and regulations on food hygiene are developed and should be applied internationally. Included are the definitive standards on Good Hygiene Practice (GHP), Hazard Analysis and Critical Control Points (HACCP), and the Principles for the establishment and application of microbiological criteria (MC) for foods.

The current Codex text on MC was adopted in 1997 and had major effect on how sampling and testing have been performed since. The EU regulation on MC (Regulation (EC) No 2073/2005 on microbiological criteria for foodstuffs) was based on this standard. Currently the standard is, however, under revision due to the recent emergence of improved scientific and industrial applications of MC. A new text "Proposed Draft Principles and Guidelines for the Establishment and Application of Microbiological Criteria Related to Foods" is expected to be approved by the Codex Alimentarius Commission in July 2013.

The work on the revision started in 2010 and involved several working groups. The revised text can be viewed as an extension of the document from 1997 and builds on the legislation that has been implemented as a consequence of the 1997 document. The new text includes the principles for applying MC's and the different purposes for using these. These purposes are illustrated by a list of practical examples, developed by sub-working groups comprising Codex member states all over the world. An example of a risk-based approach is included in the draft document, which is of special interest for Denmark. The idea is to take a sample, perform a quantitative analysis, and feed the result into a Quantitative Microbiological Risk Assessment model (QMRA) to achieve an estimate of the risk. The competent authorities may then use this risk estimate to decide if the batch is safe for human consumption. This approach is an entirely new way of establishing MC's. The principle has been developed taking into account the case-by-case risk assessments of raw meat that has been performed in Denmark since 2007.

Codex's work is based on reaching consensus between a large number of countries from all regions of the world. Therefore, the progress is relatively slow, but it is important and necessary work because the decisions made has a great and determining impact on international trade. In the Codex commission, all member countries speak with an equal voice, and arguments based on scientific evidence are listened to, making it possible for small countries to have an impact on the decisions made.

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8. Surveillance and control programmes

The collaboration between national and regional authorities, the industry and non-governmental organizations is presented in Figure 8.1. According to the Danish legislation, 41 infectious diseases are notifiable in Denmark. An overview of the notifiable and non-notifiable human and animal diseases presented in this report is provided in Appendix Table A30 and Table A31, respectively, including reference to the relevant legislation.

8.1 Surveillance of human disease

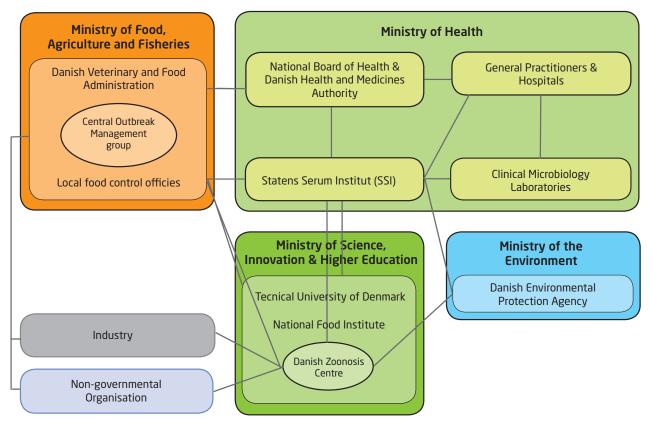
Information on human cases due to zoonotic pathogens presented in this report is reported to Statens Serum Institut through different channels depending on the disease:

• Notifiable through the laboratory surveillance system: Salmonella, Campylobacter, Yersinia, Verocytotoxinproducing E. coli (VTEC) and Listeria

- Individually notifiable zoonotic pathogens: *Chlamydia psittacci* (ornithosis), *Leptospira*, *Mycobacterium*, Bovine Spongieform Encephalopathy (BSE) prions (var. Creutzfeldt-Jakob Disease), Verocytotoxin-producing *E. coli* (VTEC) and *Lyssavirus* (rabies)
- Non-notifiable zoonotic pathogens: *Brucella*, *Cryptosporidium*, *Echinococcus*, *Toxoplasma* and *Trichinella*.

In Denmark, the physicians report individually notifiable zoonotic diseases to the Danish Health and Medicines Authority and the Department of Epidemiology at Statens Serum Institut. Positive cases diagnosed by a clinical microbiological laboratory are reported through the laboratory surveillance system to the Unit of Gastrointestinal Infections at Statens Serum Institut. Physicians send specimens from suspect cases to one of the clinical microbiology laboratories depending on county of residence of the requesting

Figure 8.1. Overview of the monitoring and outbreak investigation network for reporting infectious pathogens in humans, animals, foodstuffs and feedstuffs in Denmark, 2012



Source: Danish Zoonosis Centre, National Food Institute

physician. The laboratories must report positive results to Statens Serum Institut within one week. Furthermore, all *Salmonella* and VTEC isolates are sent to the reference laboratory at Statens Serum Institut for further sero- and genotyping. The *Salmonella* isolates are sent to the National Food Institute, Technical University of Denmark for phage typing (see Appendix Table A38 for more detailed information on typing methods). The results are recorded in the Register of Enteric Pathogens maintained by Statens Serum Institut. Positive cases are reported as episodes, i.e. each patient-infectious agent combination is only recorded once in any six-month period. Overviews of results from the Register of Enteric Pathogens are presented as follows:

- All laboratory confirmed human cases are presented in Appendix Table A2
- VTEC O-group distribution in humans is presented in Appendix Table A3
- The *Salmonella* sero- and phage type distributions are presented in Appendix Tables A5-A7.

8.2 Outbreaks of zoonotic gastrointestinal infections

In Denmark, local and regional foodborne outbreaks are typically investigated by the Local Food Control Offices in collaboration with the Danish health and medicine authorities, and the regional clinical microbiology laboratories. Larger regional and national outbreaks are investigated by Statens Serum Institut, the National Food Institute, and the Danish Veterinary and Food Administration in collaboration. These institutions may also aid in the investigation of local outbreaks. Representatives from these institutions meet regularly in the Central Outbreak Management Group to discuss surveillance results, compare the reported occurrence of zoonotic agents in animals, food and feedstuffs with that in humans, and coordinate the investigation of major outbreaks. The formal responsibility of investigating food- or waterborne outbreaks is currently divided between three ministries based on the outbreak source: the Ministry for Interior and Health for infectious diseases; the Ministry of Food, Agriculture and Fisheries for foodborne and animal related diseases; and the Ministry of the Environment (along with the municipalities) for waterborne diseases.

Outbreaks may be detected in various ways. Individuals who experience illness related to food intake in settings such as restaurants or work place cafeterias may report these incidents directly to the Local Food Control Office. General Practitioners and Hospitals are obliged to report all suspected water- and foodborne infections to the Danish health and medicine authorities , who then reports to Statens Serum Institut. Clusters of cases may also be noted in the laboratory or identified at Statens Serum Institut through the laboratory surveillance system of gastrointestinal bacterial infections or through subtyping of bacterial isolates from patients.

A list of verified outbreaks (not including household outbreaks) reported to the Food- and waterborne Outbreak Database (FUD) are presented in Appendix Table A4 and some of the more noTable outbreaks from 2012 are outlined in Chapter 6.

8.3 Surveillance and control of animals and animal products

Salmonella surveillance and control programmes for poultry, pigs and cattle are presented in Appendix Tables A32-A37. Sample analysis is performed at authorised private laboratories, the local food control offices, the National Food Institute, and the National Veterinary Institute. *Salmonella* isolates are forwarded to the National Food Institute for serotyping, some isolates are also phage- and genotyped as well as tested for antimicrobial resistance. An overview of the methods used for subtyping is presented in Appendix Table A38.

Overviews of results from surveillance and control of *Salmonella* are presented as follows:

- Results from the Table egg production are presented in Appendix Tables A5-A9
- Results from the broiler production are presented in Appendix Tables A5-A7 and A10
- Results from the duck and turkey productions are presented in Appendix Table A5-A6 and A11
- Results from the pig production are presented in Appendix Tables A5-A6, A15 and Figures A1-A3
- Results from the cattle production are presented in Appendix Tables A5, A16-A17 and Figure A4
- Results from the feeding stuff production are presented in Appendix Tables A20-A21
- Results from the rendering plants are presented in Appendix Table A22
- Results based on suspicion of diseases in pets, zoo animals and wild life are presented in Appendix Table A23-A24.

Overviews of results from monitoring of *Campylobacter* are presented as follows:

- Results from the broiler production are presented in Appendix Tables A12-A13
- Results based on suspicion of diseases in pets, zoo animals and wild life are presented in Appendix Table A23-A24
- Results on the relative distribution of *Campylobacter* species in broilers, pigs and cattle are presented in Appendix Tables A14 and A18

Pig and cattle carcasses are screened for *Mycobacterium* and *Echinococcus* during meat inspection at the slaughter-

house. Although Denmark is assigned as a region where the risk of *Trichinella* in domestic swine is negligible, all slaughter pigs are still examined for *Trichinella* at slaughter as well as wild boars, and horses slaughtered for human consumption. In addition, boars and bulls are tested for *Brucella* and bulls are tested for *Mycobacterium* at semen collection centres. All positive results for notifiable infectious diseases are reported to the Danish Veterinary and Food Administration. Results are presented in Appendix Table A15-A16.

Results from the surveillance for Bovine Spongiform Encephalopathy (BSE) in cattle, Transmissible Spongiform Encephalopathy (TSE) in sheep/goat are presented in Appendix Tables A25-A27.

Results from the monitoring of *Coxiella burnetii* (Q fever) in cattle are presented in Appendix Table A16.

Results based on suspicion of diseases with *Chlamydia psittacci*, *Cryptosporidium*, *Trichinella*, classical rabies and European Bat *Lyssavirus* in zoo animals, pets and wild life are presented in Appendix Table A23-A24.

8.4 Official testing of zoonotic pathogens in foodstuffs

In Denmark, control of zoonotic microorganisms in foodstuffs is mainly carried out as projects which are coordinated at the central level of the Danish Veterinary and Food Administration. Sampling and testing are carried out with the following purposes:

- To verify that food business operators comply with microbiological criteria laid down in the legislation
- To verify the microbiological safety of food for which no microbiological criteria are laid down at EU Community level
- To monitor the effect of established risk management procedures in order to evaluate if these provide the desired results or need to be reconsidered
- To generate data for the preparation of risk profiles and risk assessments to support microbial risk management
- To discover emerging problems with microbiological contaminants.

Appendix Table A28 provides information on the centrally coordinated studies conducted in 2012. Results for the following project are presented elsewhere in the report:

- Intensified control of *Salmonella* and *Campylobacter* in Danish and imported meat based on a case-by-case risk assessment (Appendix Table A19)
- Findings of *Campylobacter* in non-heat treated meat cuts from broilers (Appendix Table A13)
- Findings of *Listeria monocytogenes* in ready-to-eat products (Appendix Table A29)
- Findings of *Salmonella* in fresh and imported duck meat (Appendix Table A4-A5)
- Findings of *Salmonella*, *Campylobacter* and *E. coli* in ready-to-eat fruits, vegeTables and herbs (Chapter 5).

For further information consult the webpage of the Danish Veterinary and Food Administration, www.fvst.dk (in Danish).



Campylobacter action plan on broilers, food and environment 2013-2016

By Gudrun Sandø (gus@fvst.dk)

The previous action plan against *Campylobacter* in broilers covered the period 2008 – 2012. An evaluation carried out in collaboration between the National Food Institute and the Danish Veterinary and Food Administration concluded that initiatives directed against high levels of *Campylobacter* in imported broiler meat had had an effect, whereas initiatives directed at *Campylobacter* in the Danish production seemingly had not had the expected effect. The evaluation group recommended that the coming action plan should include setting of targets for reduction of *Campylobacter* in the broiler production, development and implementation of fly screens on relevant broiler houses, setting of process hygiene criteria in the production of fresh broiler meat, an enhanced focus on slaughter hygiene and initiatives to obtain more knowledge and data concerning alternative sources and routes of transmission. This evaluation together with other relevant information formed the background material for the new action plan.

The *Campylobacter* action plan 2013 - 2016 was developed during the autumn 2012 by the Danish Agriculture & Food Council, the Danish Broiler Association, the National Food institute, the National Veterinary Institute, and the Danish Veterinary and Food Administration. The work was finalised in spring 2013 and the plan was adopted with the overall aim to reduce the number of human *Campylobacter* cases.

The new action plan covers initiatives in the broiler production at farm level as well as at the slaughterhouse, information to consumers, and as a new element it also covers sources and routes of transmission other than from the broiler production. Targets have been set in the broiler production; The prevalence of positive flocks shall be reduced by 20 % by 2016 compared to the level in 2012. The target for fresh broiler meat at the slaughterhouses is formulated as a reduction of the relative risk compared to the level in 2012 and depends on the prevalence as well as the concentration of *Campylobacter* in fresh broiler meat. By the end of 2014 and 2016 the relative risk should be reduced by 25 % and 50 %, respectively.

The industry is free to choose the methods to obtain these targets. Nevertheless, the action plan describes initiatives where there has been a mutual agreement between stakeholders as well as a sound knowledge on effects. Some of these initiatives are:

- Implementation of a quality assurance programme¹ at the slaughterhouse laid down by the broiler industry where maximum limits on hygienic markers such as faecal leakage are defined by the slaughterhouse in order to improve the slaughter hygiene
- Initiatives on other sources such as fruit and vegetables
- A continued effort on Campylobacter in imported meat
- A continued effort on improving consumer awareness
- A research project on development and implementation of fly screens on relevant broiler houses should also be conducted.

The plan will be evaluated on a yearly basis. The report is available at the Danish Veterinary and Food Adminstrations webpage www.fvst.dk (in Danish).

^{1.} Kvalitet i Kyllingeproduktionen (KIK)

Appendix A

Trends and sources in human salmonellosis

Table A1. Estimated no. of reported human cases and percentage of cases per major food source, travel or outbreaks, 2010-2012

	2012		2011		2010	
Source	Estimated no. of reported cases (95 % credibility interval ^a)	Percen- tage of reported cases	Estimated no. of reported cases (95 % credibility interval ^a)	Percen- tage of reported cases	Estimated no. of reported cases (95 % credibility interval ^a)	Percen- tage of reported cases
Domestic pork	110 (84-139)	8.0	86 (44-131)	7.4	242 (238-283)	16.4
Domestic beef	85 (72-100)	7.1	6 (0-28)	0.5	12 (0-38)	0.7
Domestic table eggs	15 (1-35)	1.3	11 (2-22)	1.0	28 (18-41)	1.8
Domestic broilers	0	0	0	0	8 (4-14)	0.5
Domestic ducks	10 (1-23)	0.8	19 (4-37)	1.7	2 (0-7)	0.1
Imported pork	3 (0-10)	0.2	65 (28-104)	5.6	86 (59-115)	5.4
Imported beef	11 (4-20)	0.9	33 (10-48)	2.8	30 (4-51)	2.0
Imported broilers	21 (3-47)	1.8	24 (2-51)	2.0	5 (0-17)	0.2
Imported turkey	13 (1-28)	1.1	13 (1-38)	1.1	17 (2-37)	1.0
Imported duck	22 (13-34)	1.6	28 (8-54)	2.4	21 (10-37)	1.3
Travels	539 (527-550)	45.0	538 (531-546)	46.2	749 (740-758)	46.9
Unknown source	332 (293-369)	27.7	288 (252-330)	24.7	316 (275-354)	19.8
Outbreaks, unknown source	37	4.3	55 ^b	4.3	62	3.9
Total	1,198		1,166		1,598	

a) The model is based on a Bayesian framework which gives 95 % credibility intervals.

b) Five cases are known to have been caused by pork, but of unknown origin. In one outbreak with 43 cases the source was tomatoes imported from Italy.

Source: Danish Zoonosis Centre, National Food Institute

Appendix B

Human disease and outbreak data

	Incidence per 100,000 inhabitants		Re	eported no.	of cases		
Zoonotic pathogen	2012	2012	2011	2010	2009	2008	2007
Bacteria							
Brucella abortus/melitensis ^{a,d}	-	2	7	6	7	8	20
Campylobacter coli/jejuni ^ь	66.5	3,728	4,068	4,035	3,352	3,454	3,868
Chlamydia psittaci ^b	0.2	12	7	9	14	6	11
Leptospira spp. ^b	0.1	7	11	10	12	13	10
Listeria monocytogenes ^b	0.9	50	49	62	97	51	58
<i>Mycobacterium bovis</i> ^b	-	0	1	2	0	1	1
Salmonella total ^b	21.4	1,198	1,166	1,598	2,129	3,656	1,647
S. Enteritidis ^b	4.3	242	293	388	600	638	566
S. Typhimurium ^{b,c}	7.3	415	386	521	767	2,002	343
Other serotypes ^b	6.9	541	487	689	762	1,016	740
VTEC total ^b	3.4	190	224	184	165	159	161
O157	0.6	36	27	25	24	14	25
other or non-typeable	2.7	154	197	159	141	145	136
Yersinia enterocolitica ^b	5.2	291	224	192	238	330	270
Parasites							
Cryptosporidium spp. ^{a,d}	-	8	31	25	35	92	49
Echinococcus multilocularis ^{a,e}	-	7	4	1	0	0	3
Echinococcus granulosus ^{a,e}	-	20	31	10	11	5	9
Trichinella spp. ^{a,e}	-	0	0	0	0	0	1
Viruses							
Lyssavirus ^ь	-	0	0	0	0	0	0

Table A2. Zoonoses in humans, number of laboratory-confirmed cases, 2007-2012

a) Not notifiable hence the incidence cannot be calculated.

b) Notifiable.

c) S. Typhimurium and monophasic S. 1,4,[5],12:i:- strains

d) Data presented are from one laboratory (Statens Serum Institut) only, representing a proportion of the Danish population (approximately 1/3 in 2012). The proportion of the population represented varies from year to year, thus results from different years are not comparable. Testing for these pathogens is carried out only if specifically requested on the submission form. e) The cases were imported.

Source: Statens Serum Institut

O-group	Number of episodes	O-group	Number of episodes
0157	39	026	8
O145	19	O128	8
O146	17	O91	7
O117	11	O-rough	9
O103	11	Notification ^b	16
		Other O-groups or not-typed	1 63
Conti	nued in the next column	Total	208

Table A3. VTEC O-group distribution in humans^a, 2012

a) All O-groups that resulted in five or more episodes are listed.

b) The cases are reported through the notification system, isolates not available for analysis.

Source: Statens Serum Institut

Pathogen	No. of patients	Patients labora- tory confirmed	Setting	Source	FUD no.
Bacillus cereus	4		Restaurant	Rice (cooked)	117
Bacillus cereus	13	·	Hotel	Composite meal	1250
Bacillus cereus	26	•	Hotel	Broiler meat	1250
		•		Broiler meat	
Campylobacter	13	•	Canteen		1142
Campylobacter	11	5	Farm	Raw milk	1194
Campylobacter	7	3	Hotel	Broiler meat	1216
Clostridium perfringens	8	•	Restaurant	Broiler meat	1164
Clostridium perfringens	17	•	Restaurant	Composite meal	1162
Clostridium perfringens	8		Private party	Composite meal	117
Clostridium perfringens	18	•	Restaurant	Composite meal	1184
Clostridium perfringens	2	•	Restaurant	Composite meal	1207
Clostridium perfringens	9		Restaurant	Composite meal	122
Clostridium perfringens	70	•	Canteen	Composite meal	1227
Clostridium perfringens	43		Restaurant	Composite meal	1242
ETEC	4	1	Private party	Fresh herbs (imp)	1168
Hepatitis A virus	6	6	National	Unknown	1172
Hepatitis A virus	7	7	Regional	Unknown	1232
Histamin	2		Shop	Fish (canned tuna) (imp)	115
Histamin	5	•	Private party	Fish (canned tuna) (imp)	122
Lectines	8	·	Restaurant	Elderberries	122
Lectines	18	•	Restaurant	Dried beans (imp)	121
Norovirus	10	•	Private party	Composite meal	114
Norovirus	24	2	Restaurant	Oysters (imp)	115
Norovirus	10	1	Restaurant	Composite meal	115
Norovirus	9	3	Restaurant	Oysters (imp)	115
Norovirus	65	5	Restaurant	Unknown	1150
Norovirus	4	1	Canteen	Unknown	116
Norovirus	20	1	Institution	Buffet meal	1163
Norovirus	20 12	1	Restaurant	Composite meal	116
Norovirus	12	1	Canteen	Buffet meal	116
Norovirus	15	· 4			
		4	Shop	Composite meal Buffet meal	1169
Norovirus	65	·	School	Cake	117
Norovirus Norovirus	9 15	•	Private party	Buffet meal	1172 1170
		·	Private party	Buffet meal	
Norovirus	62 16	•	Restaurant		117
Norovirus	16	4	Restaurant	Buffet meal	117
Norovirus	21	•	Canteen	Buffet meal	118
Norovirus	25	•	Canteen	Buffet meal	118
Norovirus	19	•	Private party	Buffet meal	118
Norovirus	46	1	Restaurant	Unknown	118
Norovirus	12		Restaurant	Composite meal	118
Norovirus	70	•	Canteen	Composite meal	118
Norovirus	54	2	Restaurant	Composite meal	1190
Norovirus	12	•	School	Unknown	119
Norovirus	43	1	Private party	Composite meal	119
Norovirus	17	4	Canteen	Composite meal	120
Norovirus	4		Restaurant	Composite meal	120
	-	Continued on the		- I	

Table A4. Food- and waterborne disease outbreaks^a reported in the Food- and Waterborne Outbreak Database (FUD) (n=82), 2012

Pathogen	No. of	Patients labora-	Setting	Source	FUD
	patients	tory confirmed	Getting	Jource	no.
Norovirus	25		Canteen	Composite meal	1204
Norovirus	14	3	Private party	Buffet meal	1211
Norovirus	15		Private party	Buffet meal	1217
Norovirus	13		Restaurant	Buffet meal	1218
Norovirus	17		Canteen	Composite meal	1222
Norovirus	88	7	Canteen	Buffet meal	1223
Norovirus	61		Canteen	Buffet meal	1224
Norovirus	100		Canteen	Buffet meal	1225
Norovirus	10		Shop	Composite meal	1229
Norovirus	200		School	Unknown	1231
Norovirus	183	18	Other	Tap water	1233
Norovirus	38		School	Cake	1234
Norovirus	67		Restaurant	Composite meal	1235
Norovirus	30	3	Canteen	Composite meal	1236
Norovirus	15		Restaurant	Oysters (imp)	1237
Norovirus	15		Shop	Composite meal	1238
Norovirus	11		Restaurant	Unknown	1248
Norovirus	40		Hotel	Unknown	1253
Norovirus	9		Restaurant	Composite meal	1251
Other chemical agent	15		School	Composite meal	1147
S. Bareilly	11	8	Restaurant	Unknown	1215
S. Mikawasima	3	3	National	Unknown	1206
S. 4,5,12:i:-, MLVA0006	64	12	Canteens	Beef	1191
S. 4,5,12:i:-, MLVA0006	24	24	National	Beef	1192
S. 1,4,5,12:i:-, MLVA0126	9	4	Private party	Composite meal	1185
S. 4,5,12:i:-, MLVA0201	6	6	Restaurant	Buffet meal	1199
S. Saintpaul	3	3	Shop	Duck (imp)	1193
<i>S</i> . Typhimurium DT120, MLVA0007	15	15	Shop	Pork	1245
S. Typhimurium DT120, MLVA0995	7	7	Regional	Unknown	1186
S. Typhimurium DT120, MLVA0006	6	6	Regional	Unknown	1200
S. Poona	10	10	National	Unknown	1174
Staphylococcus aureus	68		Restaurant	Composite meal	1228
Unknown	11		Private party	Unknown	1247
VTEC O157	4	4	Regional	Unknown	1159
VTEC O157	14	12	National	Beef	1210
Total	2,203	192			

Table A4. Food- and waterborne disease outbreaks^a reported in the Food- and Waterborne Outbreak Database (FUD), 2012 (Continued from previous page)

Note: (imp)= imported product

a) In addition, three confirmed household outbreaks were registered (FUD 1208, 1209 and 1239). FUD 1208 involved two cases and was caused by histamin in imported canned tuna. FUD 1209 and 1239 both involved two cases each, and was caused by the presence of other chemical agents in imported pine kernels and kaki fruit, respectively.

Source: Food- and Waterborne Outbreak Database (FUD)

Appendix C

Monitoring and surveillance data

	Human	Pig ^a	Pork ^{b,g}	Beef ^b	Layer ^c	Broiler	Duck ^c		Impo	rted meat	(batch)	
		herds	batches	batches	flocks	flocks	flocks	Pork ^d	Beef ^d	Broiler ^d	Turkey ^d	Duck ^e
Serotype	N=1.198	N=198	N=137	N=13	N=3	N=29	N=62	N=23	N=2	N=18	N=5	N=41
Enteritidis	20.2	0.5	0	0	33.3	0	0	0	0	16.7	0	0
Typhimurium	18.6	15.2	18.2	0	0	17.2	4.8	26.1	0	0	20.0	24.4
Typhimurium	16.0	15.7	21.9	0	33.3	13.8	0	26.1	0	0	40.0	4.9
(Monophasic) ^f												
Dublin	4.2	0	0	23.1	0	0	0	0	50.0	0	0	0
Stanley	2.3	0	0	0	0	0	0	0	0	5.6	0	0
Poona	2.3	0	0	0	0	0	0	0	0	0	0	0
Infantis	2.1	1.0	6.6	0	33.3	6.9	0	0	0	27.8	0	0
Newport	2.0	0	0	0	0	0	32.3	0	0	16.7	0	2.4
Virchow	1.8	0	0	0	0	0	0	0	0	0	0	0
Saintpaul	1.4	0	0	0	0	0	0	0	50.0	0	0	2.4
Java	1.3	0	0	0	0	0	0	0	0	0	0	0
Braenderup	1.1	0	0	0	0	0	0	0	0	0	0	0
Corvallis	1.0	0	0	0	0	0	0	0	0	0	0	0
Kentucky	1.0	0	0	0	0	10.3	0	0	0	0	0	0
Derby	0.8	62.1	35.0	0	0	10.3	0	34.8	0	0	20.0	0
Others	21.4	4.0	8.0	0	0	41.4	59.7	8.7	0	33.3	20.0	61.0
Unknown	2.4	1.5	10.2	76.9	0	0	3.2	4.3	0	0	0	4.9
Total	100	100	100	100	100	100	100	100	100	100	100	100

a) Isolates collected from coecum samples taken randomly at slaughter.

b) Sampling of beef and pork carcasses at slaughterhouses according to surveillance programmes (Tables A36 and A37).

c) Sampling in production flocks prior to slaughter according to surveillance programmes (Tables A32-A34).

d) Case-by-case control of imported meat. For further information regarding case-by-case control programme, see Annual Report on Zoonoses in Denmark 2007.

e) Imported duck meat sampled at retail (centrally coordinated studies, Table A28).

f) Typhimurium (monophasic) includes the Salmonella strains 1,4,[5],12:i:-.

g) 15 of the serotypes isolated from pork were established using a DNA based molecular approach as the strains autoagglutinated (exhibited a rough phenotype) when typed by traditional slideagglutination.

Source: Danish Veterinary and Food Administration, Statens Serum Institut, and National Food Institute

	Human	Pork ^b	Layer ^c	Broiler	Duck ^c	Impo	orted meat (b	atch)
		batch	flocks	flocks	flocks	Pork ^d	Turkey ^d	Ducks ^e
Phagetype	N=415	N=55	N=1	N=9	N=3	N=12	N=3	N=12
120	26.3	18.2	100	0	55.6	33.3	0	0
193	22.7	20.0	0	0	0	25.0	0	0
RDNC	7.5	3.6	0	0	22.2	0	0	0
U323	4.1	0	0	0	0	0	33.3	0
104	3.4	0	0	0	0	0	0	0
U302	2.7	0	0	0	11.1	8.3	0	0
U310	2.4	0	0	0	0	0	0	0
8	1.9	0	0	0	0	0	0	83.3
U292	1.9	0	0	0	0	0	0	0
41	1.9	1.8	0	0	0	0	0	8.3
Other	14.0	20.0	0	0	11.1	25.0	0	8.3
Unknown	11.3	36.4	0	100	0	8.3	66.7	0
Total	100	100	100	100	100	100	100	100

Table A6. Top 10 (human) phage type distribution (%) of S. Typhimurium and the monophasic strains S. 1,4,[5],12:*i:*-from humans, animals and imported meat, 2012

a-e) See footnotes a-e in Table A5.

Source: Danish Veterinary and Food Administration, Statens Serum Institut, and National Food Institute

	Human	Layer ^c	Imported meat (batch)
		flocks	Broilers ^d
Phagetype	N=242	N=1	N=3
8	20.7	0	0
4	10.3	0	33.3
21	9.5	100	66.7
14B	8.3	0	0
1	5.8	0	0
6A	4.1	0	0
RDNC	3.7	0	0
2	2.9	0	0
3	2.9	0	0
11	2.5	0	0
Other	16.5	0	0
Unknown	12.8	0	0
Total	100	100	100

Table A7. Top 10 (human) phage type distribution (%) of S. Enteritidis from humans, animals and imported meat, 2012

a-e): See footnotes a-e in Table A5.

Source: Danish Veterinary and Food Administration, Statens Serum Institut, and National Food Institute

		ng period nt flocks)		period t flocks)	Pullet-rearing flocks Table egg layer flo		layer flocks	
	Ν	Positive	Ν	Positive	Ν	Positive	Ν	Positive
2003	24	0	15	0	367	4	611	10
2004	9	2	9	0	368	1	641	5
2005	16	0	9	0	355	6	655	7
2006	17	0	11	0	289	2	565	2
2007	11	0	12	0	326	0	510	5
2008	10	0	6	0	258	1	508	4
2009	13	0	6	0	253	0	454	8
2010	15	0	9	0	225	0	455	8
2011	8	0	9	0	195	0	410	2
2012	9	0	8	0	197	1^{b}	359	3°

a) See Tables A32 and A34 for description of the surveillance programmes.

b) One flock was positive with S. 4,5,12:i:- DT120

c) One flock positive with *S.* Enteritidis, one flock positive with *S.* 4,5,12::- and one flock positive with *S.* Infantis Source: Danish Agriculture and Food Council, and Danish Veterinary and Food Administration

Table AO Ossumes as a	£ C = = = .	the tells are lower.	fleater as the distance of	f production, 2003-2012
ιπηέρας υςτυπέρησε η	r Saimoneila <i>in</i> 1	the table ead laver	τιοςκς ςοιτέσι ον τνοέ ο	T DEDAILETION 2003-2017
Tuble 715. Occurrence o	j Sunnonena m	the tuble egg luyer	JIOCKS SOLICG Dy Lype O	

	De	ep litter	Fre	ee range	С	rganic	Ba	attery	
	Ν	Positive	Ν	Positive	Ν	Positive	Ν	Positive	
2003	191	2	71	2	173	1	167	9	
2004	214	0	72	2	175	1	177	2	
2005	217	3	70	0	178	0	175	4	
2006	185	0	62	0	164	2	148	0	
2007	155	2	56	0	146	2	146	1	
2008	151	0	61	2	145	1	135	1	
2009	133	1	78	0	130	4	110	3	
2010	117	0	45	2	136	1	157	5	
2011	109	0	40	0	130	1	131	1	
2012	101	0	37	1^{a}	136	1 ^b	85	1^{c}	

a) One flock positive with S. Enteritidis

b) One flock positive with *S.* 4,5,12:i:-

c) One flock positive with S. Infantis

Source: Danish Agriculture and Food Council, and Danish Veterinary and Food Administration

		Rearing period (parent flocks)		ılt period ent flocks)	Broi	ler flocks	Slaughterhouse (flocks/batches)		
	Ν	Positive	Ν	Positive	Ν	Positive	Ν	Positive	
2003	265	2	182°	4	4,414	77	1,552	77	
2004	275	1	155°	6	4,246	64	1,472	24	
2005	214	0	185°	0	4,034	87	1,174	27	
2006	190	0	282	5	3,621	71	875 ^d	17	
2007	152	0	258	3	3,703	60	884	10	
2008	146	0	293	2	3,845	43	518 ^e	3	
2009	140	0	225	4	3,767	35	375	3	
2010	126	0	200	5	3,773	43	346	1	
2011	114	0	213	0	3,795	47	306	0	
2012	123	0	183	0	3,342 ^f	27 ^g	368	0	

Table A10. Occurrence of Salmonella in the broiler production^o, 2003-2012

a) See Tables A32 and A33 for description of the surveillance programmes.

c) In 2003-2005, only one flock per house was registered per year although there may have been more than one flock in the house, however all flocks were sampled according to the surveillance programme.

d) From 2006, data cover only samples taken following the *Salmonella* programme. Verification samples taken once a week by producers of poultry meat approved to market *Salmonella*-free poultry meat are not included. Collection of verification samples started in the middle of 2005.

e) From 2008, all AM positive flocks are heat treated at slaughter. Sampling is now carried out as verification of the AM results of the negative flocks.

f) Data include 213 organic flocks.

g) S. 4,12:i:- (2), S. 4,5,12:i:- (1), S. Bareilly (2), S. Derby (3), S. Infantis (2), S. Kentucky (3), S. Montevideo (2), S. Typhimurium (4), S. Goverdhan (4).

Source: Danish Agriculture and Food Council, and Danish Veterinary and Food Administration

	Duck flocks			Turkey f	locks	
	Ν	% pos		Ν	% pos	
2006	266	80.5		11	0	
2007	-	-		13	0	
2008	68	64.7		10	10.0	
2009	85	63.5		15	0	
2010	108	56.5		24	4.2	
2011	95	58.1		38	2.6	
2012	96	49.0		23	0	

Table A11. Occurrence of Salmonella in turkey and duck flocks^o, 2006-2012

a) See Table A35 for description of the surveillance programmes. The two major turkey and duck slaughterhouses in Denmark closed down in 2004 and 2007, respectively. Therefore, most commercially reared duck and turkey flocks are transported abroad for slaughter.

Source: Danish Agriculture and Food Council

	Cloacal swa	bs at slaughter	Sock sam	oles at farm
	Ν	% pos	Ν	% pos
2004	5,157	27.0	-	-
2005	4,952	30.4	-	-
2006	4,522	30.8	-	-
2007	4,527	26.8	-	-
2008	4,950	26.3	-	-
2009	4,591	29.4	-	-
2010 ^a	-	-	3,132	16.5
2011	-	-	3,379	14.4
2012	-	-	3,376	11.6

Table A12. Occurrence of Ca	npylobacter <i>in broiler</i>	flocks, 2004-2012
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a) From 2010, results from broiler flocks are not comparable to results from previous years, as the sampling method changed from cloacal swabs at slaughter to boot swabs collected in the stable 7-10 days before slaughter according to Regulation No. 1469 of 15/12/2010 as ammened.

Source: Danish Agriculture and Food Council, Danish Veterinary and Food Administration, and National Veterinary Institute

Table A13. Occurrence of Campylobacter in non-heat treated broiler meat at slaughter and retail[®], 2012

		Chille	Fi	Frozen broiler meat (samples)						
	At slaughter At retail							At	retail	
	Denmark		Denmark		Import		Denmark		Import	
	Ν	% pos	Ν	% pos ^b	Ν	% pos ^b	Ν	% pos ^b	Ν	% pos ^b
2012	1,044	21.5	521	9.7	154	28.2	216	5.8	149	3.3

a) Centrally coordinated studies (see section 8.4 for describtion). Limit of quantification: 10 cfu/g.b) The prevalence is calculated as a mean of quarterly prevalences.

Source: National Food Institute

Table A14. Relative distribution of Campylobacter species (%) in broilers before slaughterab, 2003-2012

	Ν	C. jejuni	ejuni C. coli C. upsaliensi		NT/other
2003	113	92.9	6.2	0	0.9
2004	101	94.1	5.9	0	0
2005	109	90.8	0	2.8	6.4
2006	113	92.0	7.1	0.9	0
2007	111	91.9	0.9	5.4	1.8
2008	100	90.5	0	2.8	6.6
2009	105	89.0	11.0	0	0
2010	-	-	-	-	-
2011	46	95.7	4.3	0	-
2012	44	93.2	6.8	-	-

a) Samples were collected as part of the DANMAP programme and isolates were examined using conventional microbiological methods.

b) Since 2010, samples were only tested for *C. coli* and *C. jejuni* Source: National Food Institute

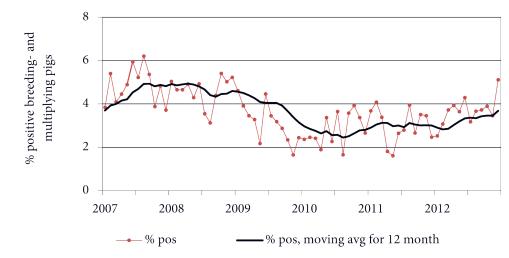
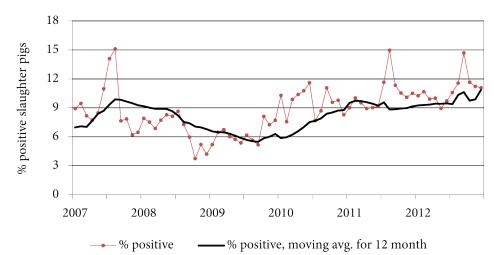


Figure A1. Serological surveillance of Salmonella in breeding and multiplying pigs^a based on monthly testing of blood samples, 2006-2012

a) For more information about the surveillance programme, see Table A37. Source: Danish Agriculture and Food Council

Figure A2. Serological surveillance of Salmonella in slaughter pigs^o, 2006-2012. Percentage of seropositive meat juice samples (first sample per herd per month)^b



a) For more information about the surveillance programme, see Table A37.b) The peak in late summer 2007, the very low level during 2008 and the peak in June 2012 were due to technical problems in the laboratory. Peaks in January 2010 and August 2011 were due to data transfer problems.Source: Danish Agriculture and Food Council

	Н	erds	Animals/	Animals/Samples		
Zoonotic pathogen	N	Pos	N	Pos	% pos	
At farm						
Brucella abortus ^a	-	-	38,617	0	0	
Leptospira ^b	62	2	157	4	2.5	
At slaughterhouse (slaughter pigs)						
Salmonella spp. ^{c,d}	7,181	414 ^e	-	-	-	
<i>Salmonella</i> spp. ^{c,f} (slaughtering >50 pigs/month)	-	-	18,655	-	1.2^{f}	
Salmonella spp. ^{c,g} (slaughtering 50 or less pigs/month)	-	-	480	-	0^{f}	
Salmonella spp. ^{c,h}			833	198	23.8	
Trichinella spp. ⁱ	-	-	18,883,606	0	-	
Mycobacterium bovis ⁱ	-	-	19,056,065	0	-	
Echinococcus granulosis/multilocularis ⁱ	-	-	19,056,065	0	-	

Table A15. Occurrence of zoonotic pathogens in pigs and pork in Denmark, 2012

a) Including samples from boars (examined at pre-entry, every 18 month, and prior to release from semen collection centres) (5,906 samples), samples collected in connection with export (32,606 samples), import (5 samples) or diagnostic samples (100 samples). 5-8 ml blood samples were analysed using either the SAT, RBT, CFT or ELISA methods.

b) Sampling is based on suspicion of leptospirosis due to increased abortions or other reproductive problems in a herd. Samples are investigated using immunoflourescence techniques.

c) See Table A37 for describtion of the Salmonella surveillance programme.

d) Data are from December 2012. Slaughter pig herds monitored using serological testing of meatjuice samples collected at slaughter.

e) Includes herds belonging to level 2 and 3 only.

f) Swab samples from four designated areas of the half-carcass were collected at the slaughterhouse after min. 12 h chilling. Sample size is $4x100 \text{ cm}^2$. Samples from five animals were pooled, except at slaughterhouses where 50 pigs or less were slaughtered per month, in which case samples were analysed individually.

g) When estimating the prevalence of *Salmonella*, both the loss of sensitivity and the probability of more than one sample being positive in each pool are taken into consideration. A conversion factor has been determined on the basis of comparative studies, as described in Annual Report 2001.

h) Coecum samples are randomly collected from slaughter pigs at slaughter.

i) Samples collected from slaughter pigs at slaughter were examined using the method described in Directive 2075/2005/EEC. In 2007, Denmark achieved official status as region with negligible risk of *Trichinella*, according to EU Regulation (EC) No 2075/2005. j) Slaughter pigs were examined by meat inspectors at slaughter.

Source: Danish Veterinary and Food Administration, National Veterinary Institute, and National Food Institute

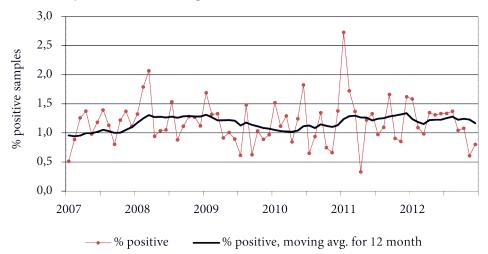


Figure A3. Salmonella in pork, monitored at slaughterhouses^a, 2006-2012

a) For more information about the surveillance programme, see Table A37. Source: Danish Veterinary and Food Administration

	He	rds	Animals/S	Animals/Samples	
Zoonotic pathogen	Ν	Pos	Ν	Pos	% pos
At farm					
Brucella abortusª	-	-	1,664	0	-
Mycobacterium bovis ^{b, c}	-	-	1,428	0	-
Coxiella burnetii	78 ^d	48	195°	34	17.4
At slaughterhouse					
<i>Salmonella</i> spp. ^f (slaughtering >50 cattle/month)	-	-	5,315	-	0,3 ^g
Salmonella spp. ^f (slaughtering 50 or less cattle/month)	-	-	379	-	0,8
Mycobacterium bovis ^{b, h}	-	-	495,700	0	-
VTEC O157 ⁱ	251	21	-	-	-
Echinococcusus granulosis/multilocularis ^h	-	-	495,700	0	-

Table A16. Occurrence of zoonotic pathogens in cattle and beef in Denmark, 2012

a) Denmark has been declared officially brucellosis free since 1979. The last outbreak was recorded in 1962. Including samples from bulls (examined at pre-entry, every year, and prior to release from semen collection centres), samples collected in connection with export, import or diagnostic samples. 5-8 ml blood samples were analysed using either the SAT, RBT, CFT or ELISA methods.

b) Denmark has been declared officially tuberculosis free since 1980. The last case of TB in cattle was diagnosed in 1988.
 c) Analysis using the tuberculin test. Including samples from bulls (examined at pre-entry, every year, and prior to release from

semen collection centres) and samples collected in connection with export

d) Bulk tank milk samples taken for diagnostic testing and analysed using an ELISA method.

e) Serum samples taken for diagnostic testing (139 samples, 33 pos), export (55 samples) and breeding (1 sample, 1 pos) and analysed using an ELISA method. An additional 6 samples from placenta was analysed using the FISH method, two sample was positive. f) See Table A36 for describtion of the surveillance programme. Swab samples from four designated areas of the half-carcass were collected at the slaughterhouse after min. 12 h chilling. Sample size is 4x100 cm². Samples from five animals were pooled, except at slaughterhouses where 50 cattle or less were slaughtered per month, in which case samples were analysed individually.

g) When estimating the prevalence of *Salmonella*, both the loss of sensitivity and the probability of more than one sample being positive in each pool are taken into consideration. A conversion factor has been determined on the basis of comparative studies, as described in Annual Report 2001.

h) Slaughtered cattle were examined by the meat inspectors at slaughter.

i) Caecal content are tested from one animal per herd, collected at slaughter (DANMAP programme). A 25 g faecal sample from one slaughter calf per herd is examined using overnight enrichment, immunomagnetic separation method and plating on CT-SMAC plates for O157.

Source: Danish Veterinary and Food Administration, Danish Agriculture and Food Council, National Veterinary Institute, and National Food Institute

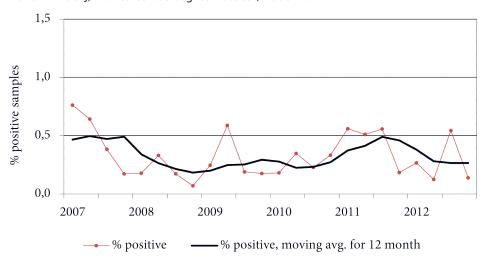


Figure A4. Salmonella in beef, monitored at slaughterhouses^a, 2006-2012

a) For more information about the surveillance programme, see Table A36. Source: Danish Veterinary and Food Administration

			Non- producii		Milk producing herds N % pos		
Salmonel	la Dublin l	evel	Ν	% pos			
Level 1	1a	On the basis of milk samples	598	4.0	3,399	91.3	
	1b	On the basis of blood samples	13,591	90.0	10	0.3	
	Total	Probably Salmonella Dublin free	14,189	94.0	3,409	91.5	
Level 2	2	Titer high in blood- or milk samples	39	0.3	62	1.6	
	2R	Titer high, official restrictions	361	2.4	248	6.7	
	2	Contact with herds in level 2 or 3	297	1.9	3	0.1	
	Total	Non Salmonella Dublin free	697	4.6	313	8.4	
Level 3	Total	Salmonellosis, official supervision	1	0.01	2	0.1	
Unknowi	1	Too few blood samples	208	1.4	0	0	
Total nun	nber of hei	rds sampled	15,095	100	3,724	100	

Table A17. Cattle herds in the S. Dublin surveillance programme^a, January 2012

a) See Table A36 for description of the surveillance programme. Source: Knowledge Centre for Agriculture, Cattle

			Pigs				Cattle	
	Ν	C. coli	C. jejuni	other/unknown	Ν	C. coli	C. jejuni	other/unknown
2003	242	-	-	100	56	-	-	100
2004	152	98.0	1.3	0.7	43	2.3	97.7	0
2005	158	97.5	2.5	0	31	0.0	100	0
2006	154	97.4	2.6	0	99	15.2	84.8	0
2007	205	97.6	2.4	0	93	4.3	95.7	0
2008 ^b	198	97.5	2.5	-	103	4.9	95.1	-
2009	160	85.6	14.4	-	110	2.7	97.3	-
2010	-	-	-	-	-	-	-	-
2011	168	96.4	3.6	-	123	8.1	91.9	-
2012	160	88.1	11.9	-	113	2.7	97.3	-

Table A18. Relative distribution of Campylobacter (%) in pig and cattle herds^a, 2003-2012

a) Samples were collected as part of the DANMAP programme. Caecal content was tested from one animal per herd. Data does not refelct the national prevalence of *Campylobacter* in pigs and cattle.
b) Since 2008, samples are only tested for *C. coli* and *C. jejuni*.

Source: National Food Institute

		Batches tested	No. of batches positive	No. of batches deemed unsafe based on a risk assessment	Batches deemed unsafe based on Microbiological criteria ^a	Mean prevalence in positive batches ^b	Mean relative human risk in positive batches ^c
Campylobacter	r						
Danish	Broiler	122	24	2	-	40.9%	2.5
Imported	Broiler	116	52	7	-	43.9%	3.7
Salmonella							
Danish	Beef	105	15	9	-	13.6%	150.8
	Pork	237	20	2	-	18.6%	4.6
	Broiler	96	0	0		-	-
Imported	Beef	101	3	1	-	28.4%	65.9
	Pork	230	20	0	-	9.6%	2.8
	Broiler	152	14	2	3	16.6%	24.7
	Turkey	49	5	0	1	17.3%	8.7

Table A19. Results from the intensified control of Salmonella and Campylobacter in fresh meat based on a case-by-case risk assessment, 2012

a) Regulation (EC) No 1086/2011

b) The *Salmonella* prevalence in each batch is based on the proportion of positive pooled samples (12 pools per batch) and number of subsamples per pool. Only results for batches subjected to risk assessment have been included.

c) Calculated as the risk relative to a batch of the same size with a mean prevalence (weighted average in Danish and imported meat) of *Campylobacter* or of a *Salmonella* type with an average impact to cause human infection.

Source: Danish Veterinary and Food Administration, and National Food Institute

Table A20. Feed business operators own sampling of Salmonella in compound feeds, feed processing and feed material
(batch-based data), 2009-2012

	20	012	20	011	2010	
	Ν	Positive	Ν	Positive	Ν	Positive
Feed processing plants (process control) ^a :						
Ordinary inspections - clean zone	7,105	11 ^d	7,359	10	7,963	12
Ordinary inspections - unclean zone	736	82 ^e	767	72	548	58
Compound feed, farm animals	316	0	386	0	390	0
Feed materials, farm animals ^b	1,369	25 ^f	1,849	60	1,285	49
Transport vehicles, clean zone/hygiene samples ^c	884	0	835	0	963	0
Transport vehicles, clean zone/hygiene samples ^c	259	0	273	0	224	1

a) Presence of *Salmonella* in compound feed is indirectly monitored by environmental samples collected during feed processing. b) Predominantly soy bean meal and rapeseed cake.

c) Samples from transport vehicles (hygiene samples) prior to loading of feed compounds.

d) S. Derby, S. Idikan, 3,10:d:-, 4,5,12:i,:-

e) Most of the samples were S. Putten from one factory. The rest were S. Derby, S. Havana, S. Infantis, S. Livingstone, S. Rissen.

f) S. Agona, S. Banana, S. Cerro, S. Cubana, S. Derby, S. Infantis, S. Livingstone, S. Mbandaka, S. Minnesota, S. Quakam, S. Rissen, S. Schwarzengrund, S. Senftenberg.

Source: Danish Veterinary and Food Administration and the feed business operators.

	2012		2011		2010		2009	
	Ν	Positive	Ν	Positive	Ν	Positive	Ν	Positive
Feed processing plants (process control) ^a :								
Ordinary inspections ^c	311	11^{d}	377	12	558	5	907	18
Feed materials, farm animals ^b	99	4^{e}	68	3	379	24	186	4

Table A21. Control of Salmonella in compound feeds, feed processing and feed material (batch-based data), 2008-2012

a) See footnote a) to Table A20. Companies are sampled one to four times per year.

b) See footnote b) to Table A20.

c) Primarily findings of *Salmonella* in the dirty zone.

d) S. Cubana, S. Infantis, S. Putten, S. Tennessee

e) S. Putten

Source: Danish Veterinary and Food Administration.

Table A22. Salmonella in three categories of meat and bone meal by-products not intended for human consumption^a, 2012

Category of		Own-ch	eck samples	Produ	ct samples
processing p	lant	Ν	Positive	Ν	Positive
1+2	By-products of this material cannot be used for feeding purposes	256	0	-	-
2	By-product of this material may be used for feed for fur animals	211	4	61	2
3	By-products from healthy animals slaughtered in a slaughterhouse. Products of these may be used for petfood ^b and for feed for fur animals	255	1	535	0
	Total	722	5	596	2

a) Regulation No. 1774 of 03/10/2002.

b) For cats and dogs. Only by-products from pigs are used in this petfood.

Source: Danish Veterinary and Food Administration

		Pet animals						Zoo animals			
	D	ogs	С	ats	Ot	thers		mals & tiles	Bir	ds	
Zoonotic pathogen	Ν	Pos	Ν	Pos	Ν	Pos	N	Pos	Ν	Pos	
Salmonella spp.	7 ^b	0	1°	-	0	-	16	0	2	0	
Campylobacter spp.	1	0	0	-	1	0	4	0	1	0	
Brucella canis/abortus ^f	22	0	0	-	0	-	68 ^g	0	0	-	
Chlamydia psittaci	0	-	0	-	3	0	0	-	3	0	
Cryptosporidium spp.	11	2	9	0	0	-	4^{d}	1^{e}	0	-	
Lyssavirus (classical)	1	0	2	0	0	-	0	-	0	-	
European Bat Lyssavirus	0	-	0	-	0	-	0	-	0	-	

Table A23. Occurrence of zoonotic pathogens in pets and zoo animals in Denmark^a, 2012

a) All samples are analysed based on suspicion of disease, except dogs and cats tested for *Salmonella* and does not reflect the country prevalence

b) 4 export samples, 3 import samples

c) Export sample

d) 4 monkeys

e) 1 chimpanzee

f) Results based on serological testing of blood samples

g) 3 alpaca, 5 bison, 12 camel, 20 deer, 3 elephant, 2 giraffe, 6 impala, 8 reindeer, 2 vikunja, 1 wild boar, 6 other

Source: National Veterinary Institute, and Danish Veterinary and Food Administration

Table A24. Occurrence of zoonotic	pathoaens in wild ar	nd farmed wildlife in	Denmark ^a , 2012

		Farmed	wildlife		Wildlife				
	Wil	d boar	Minks & cilla		Mar	nmals	Bir	·ds	
Zoonotic pathogen	Ν	Pos	Ν	Pos	Ν	Pos	Ν	Pos	
Salmonella spp.	0	-	48	21 ^b	17°	9 ^d	5	0	
Campylobacter spp.	0	-	46	3	0	-	0	-	
Chlamydia psittaci	0	-	0	-	0	-	52 ¹	13	
Cryptosporidium spp.	0	-	6	3	47 ^e	9^{f}	0	-	
Echinococcus multilocularis	0	-	0	-	315 ^g	$4^{\rm h}$	0	-	
Trichinella spp ^m	648	0	0	-	896 ⁱ	0	0	-	
Lyssavirus (classical)	0	-	0	-	4^j	0	0	-	
European Bat Lyssavirus	0	-	0	-	11^k	0	0	-	

a) All samples are analysed based on suspicion of disease and does not reflect the country prevalence, except for animals analysed for *Echinococcus multilocularis*. These animals are collected as part of survey.

b) 20 S. Dublin, 1 S. Typhimurium

c) 12 hedgehogs, 5 badgers

d) Hedgehogs

e) 14 racoon dog, 28 roe deer, 1 fallow deer, 1 red deer, 1 minke whale, 1 wolf, 1 hedgehog

f) 6 racoon dogs, 1 hedgehog, 2 roe deer

g) Results from a survey. 262 foxes, 3 badgers, 49 racoon dogs, 1 wolf

h) Foxes

i) 9 badgers, 1 marten, 111 raccon dogs, 1 wolf, 768 foxes, 6 racoons

j) 2 foxes, 1 marten, 1 wolf

k) Bats

l) Mallards, tested during investigation of an outbreak of Chlamydia psittaci

h) In 2007, Denmark achived official status as region with negligible risk of *Trichinella*, according to EU regulation (EC) No 2075/2005

Source: National Veterinary Institute, and Danish Veterinary and Food Administration

Type of surveillance	N^{b}	Positive
Active surveillance		
Healthy slaughtered animals (>48 months)	54,687	0
Risk categories:		
Emergency slaugthers (>48 months)	800	0
Slaughterhouse antemortem inspection revealed suspi-	0	0
cion or signs of disease (>48 months)		
Fallen stock (>48 months)	20,312	0
Animals from herds under restriction	0	0
Passive surveillance		
Animals suspected of having clinical BSE	1	0
Total	75,800	0

Table A25. The Bovine Spongiform Encephalopathy (BSE) surveillance programme^a for cattle, 2012

a) According to the EU Regulation (EC) 999/2001 as amended, Commission Decision 2009/719/EC as amended and Danish Order no. 499 of 26/05/2011 as amended.

b) Samples (brain stem material) are tested using a IDEXX technique or Prionics-Check PrioStrip. Confirmatory testing is carried out using Western blot (definitive diagnosis if positive case), else with histopathology or immunohistochemistry. Further confirmation on autolysed material is performed at the European Union TSE reference laboratory.

Source: National Veterinary Instistute, and Danish Veterinary and Food Administration

Type of Surveillance	N^b	Positive
Active surveillance		
Fallen stock (>18 months)	3,771	0
Animals from herds under restriction	0	0
Passive surveillance		
Animals suspected of having clinical TSE	0	0
Total	3,771	0

Table A26. The Transmissible Spongiform Encephalopathy (TSE) surveillance programme^a for sheep and goats, 2012

a) According to the EU Regulation (EC) 999/2001 as amended and Danish Order no. 1288 of 20/12/2011.

b) Samples (brain stem material) are tested using a IDEXX technique or Prionics-Check PrioStrip. Confirmatory testing is carried out using Western blot (definitive diagnosis if positive case), else with histopathology or immunohistochemistry. Further confirmation on autolysed material is performed at the European Union TSE reference laboratory.

Source: National Veterinary Institute, and Danish Veterinary and Food Administration

	Genotype	Sheep n=100
NSP 1	ARR/ARR	13.0
NSP2	ARR/AHQ	4.0
	ARR/ARQ	23.0
	ARR/ARH/Q	1.0
NSP 3 (ARQ/ARQ)	ARQ/ARQ	40.0
NSP 3 (Other)	AHQ/AHQ	0
	AHQ/ARQ	0
	ARH/ARH	2.0
	ARH/ARQ	0
	ARQ/ARH	1.0
	ARH/AHQ	1.0
	ARQ/AHQ	5.0
NSP4	ARR/VRQ	1.0
NSP5	ARQ/VRQ	9.0
	AHQ/VRQ	0
Total		100

Table A27. Distribution^a (%) of prion protein genotype of sheep randomly selected, 2012

a) The genotypes were grouped in the NSP classification system according to their different susceptibility: NSP 1: Genetically most resistant, NSP 2: Genetically resistant, NSP 3: Genetically little resistance, NSP 4: Genetically susceptible, and NSP 5: Genetically highly susceptible. Source: National Veterinary Institute

Table A28. Centrally coordinated studies conducted in	2012
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Title of project	No. of samples	Pathogen surveyed	Further information
DANMAP, antimicrobial resistance in Danish and imported broiler, beef and pork	1,000	Salmonella spp.,Campylobacter spp., Escherichia coli, Entero- coccus faesium, Enterococcus faecalis	Results are presented in the DANMAP Report 2012
MRSAª, ESC ^b in pigs at slaughter	1,600	Staphylococcus aureus, Escheri- chia coli	Results are being processed
<i>Campylobacter</i> spp. in fresh, chilled Danish broiler meat	1,300	Campylobacter spp.	Appendix Table A13
<i>Campylobacter</i> spp. in fresh, chil- led and frozen Danish and imported broiler meat	1,500	Campylobacter spp.	Appendix Table A13
<i>Salmonella</i> spp. in fresh imported duck meat	300	Salmonella spp.	Appendix Table A5-A7
Intensified control for <i>Salmonella</i> spp. and <i>Campylobater</i> in fresh Danish and imported meat	1,025°	Salmonella spp., Campylobacter spp.	Appendix Table A19
<i>Salmonella</i> spp antibiotic resistance in slaughter pigs	200	Salmonella spp.	Data are being processed
<i>Salmonella</i> spp. i table eggs - trade	150	Salmonella spp.	Results are publisched on DFVA website www.fvst.dk (in Danish)
Salmonella spp. in dried spices	500	Salmonella spp.	Data are being processed
Salmonella spp. and Escherichia coli in raw, frozen scallop from Greenland	50	Salmonella spp., Escherichia coli	Data are being processed
<i>Listeria monocytogenes, Salmonella spp., Escherichia coli,</i> staphyllococci in fish goods from Greenland	100	Salmonella spp., Listeria mo- nocytogenes, Escherichia coli, staphyllococci	Data are being processed
<i>Listeria monocytoges</i> in meat product - wholesale	650	Listeria monocytogenes	Appendix Table A29
Microbiological classification of mussel production areas in Denmark	100	Salmonella spp., Escherichia coli	Data are being processed
Pathogens in Danish and imported ready-to-eat vegetables	2,000	Salmonella spp., Campylobacter spp., Escherichia coli	Chapter 5
Water quality in food business - who- lesale	200	Total viable counts, coliforms and <i>Escherichia coli</i>	Data are being processed
<i>Listeria monocytoges</i> in meat product - at retail	100	Listeria monocytogenes	Appendix Table A29
<i>Listeria monocytogenes</i> - control of cleaning and desinfection - wholesale	450	Listeria monocytogenes	Results are publisched on DFVA website www.fvst.dk (in Danish)
Pathogens in salad bars	350	Salmonella spp., Listeria mono-	Date are being processed

Title of the project	No. of samples	Pathogen surveyed	Further information
<i>Salmonella</i> in herbs (Regulation (EC) No 669/2009)	100	Salmonella spp.	Data are being processed
Slaughter hygiene - cattle slagterhouses	400	Total viable counts, Salmonella spp., E. coli	Data are being processed
<i>Salmonella</i> and <i>E. coli</i> - meat prepara- tion - at retail	500	Salmonella spp., E. coli	Data are being processed
ESC ^b and <i>E. coli</i> in poultry meat	600	Escherichia coli	Data are being processed
MRSA ^a in milk producing cattle	500	Staphylococcus aureus	Data are being processed
ESC ^b in Danish slaughter poultry	400	Escherichia coli	Data are being processed
Official verification of microbiolo- gical criteria (Regulation (EC) No 2073/2005)	4,550	Total viable counts, Salmonella spp., Listeria monocytogenes, E. coli	Data are being processed

Table A28. Centrally coordinated studies conducted in 2012 (Continued from previous page)

a) MRSA: Methicillin-resistant *Staphylococcus aureus*.

b) ESC: Expanded-Spectrum Cephalosporin-Resistant Strains
 c) Batches

Source: Danish Veterinary and Food Administration, and National Food Institute

			amples qualitat			Samples analysed by a quantitative method			
		Batc	hesc	Sing	le samples	Batc	hesc	Single samples	
Food category	Sampling place	N	Pos	Ν	Pos	Ν	Pos	N	Pos
Cheese, RTE	At processing	43	1	4	0	27	0	0	-
	At retail	0	-	0	-	0	-	0	-
Milk and dairy products, RTE	At processing	51	0	4	0	54	0	0	-
	At retail	0	-	0	-	0	-	0	-
Products made from broiler meat, RTE	At processing	2	0	0	-	1	0	5	-
	At retail	0	-	0	-	0	-	0	-
Products made from other	At processing	2	0	0	-	2	0	0	-
poultry meat, RTE	At retail	0	-	0	-	0	-	0	-
Products made from pork,	At processing	68	4	0	-	51	0	0	-
RTE	At retail	0	-	0	-	37	0	2	0
Products made from beef,	At processing	11	1	0	-	9	0	2	0
RTE	At retail	0	-	0	-	10	0	0	-
Fruit, RTE	At processing	2	0	0	-	6	0	0	-
	At retail	0	-	0	-	0	-	9	0
Vegetables, RTE	At processing	11	0	0	-	9	0	0	0
	At retail	0	-	0	-	2	0	143	-
Fish and Fishery products,	At processing	14	3	0	-	21	0	0	-
RTE	At retail	0	-	0	-	0	-	0	-
Shellfish and products there	At processing	3	0	0	-	4	0	0	-
off, RTE	At retail	0	-	0	-	0	-	0	-
Other RTE products	At processing	71	9	4	0	36	1^d	0	-
	At retail	0	-	0	-	0	-	1	0

Table A29. Listeria monocytogenes in Danish produced ready-to-eat foods^a, 2012

a) Samples are collected by the local food control offices according to EU Regulation (EC) No 2073/2005.

a) samples are concerted by the local hold control onces according
b) *Listeria monocytogenes* present in a 25 g sample of the product.
c) 5 samples from each batch, analysed individually.
d) >100 cfu/g detected in one or more samples.
Source: Danish Veterinary and Food Administration

Appendix D

Monitoring and surveillance programmes

Patogen	Notifiable	Notification route
Bacteria		
Brucella spp.	no	-
Campylobacter spp.	1979 ^a	Laboratory ^b
Chlamydophila psittaci (Ornithosis)	1980 ^a	Physician ^c
Listeria monocytogenes	1993 ^a	Physician
<i>Leptospira</i> spp.	1980 ^a	Physician
Mycobacterium bovis/ tuberculosis	1905 ^a	Physician (and laboratory ^d)
Coxiella burnetii	no	-
Salmonella spp.	1979 ^a	Laboratory
VTEC	2000 ^a	Physician and laboratory
Yersinia enterocolitica	1979 ^a	Laboratory
Parasites		
Cryptosporidium spp.	no	_
Echinococcus multilocularis	no	-
Echinococcus granulosus	no	-
Toxoplasma gondii	no	-
Trichinella spp.	no	-
Viruses		
Lyssavirus (Rabies)	1964 ^a	Physician (via telephone)
Prions		
BSE/Creutzfeld Jacob	1997 ^a	Physician

Table A30. Overview of notifiable and non-notifiable human diseases presented in this report, 2012

a) Danish order no. 277 of 14/04/2000. Cases must be notified to Statens Serum Institut.

b) The regional microbiological laboratories report confirmed cases.

c) The physician report individually notifiable infections.

d) The laboratories voluntarily report confirmed cases.

Source: Statens Serum Institut

Patogen	Notifiable	EU legislation	Danish legislation
Bacteria			
Brucella spp.	1920 ^a		
Cattle	OBF in 1979 ^b	Decision 2003/467/EC	Order no 305 of 3/5 2000
Sheep and goats	ObmF in 1995 ^c	Decision 2003/467/EC	Order no. 739 of 21/8 2001
Pigs	No cases since 1999	Directive 2003/99/EC	Order no. 205 of 28/3 2008
Campylobacter spp.	no	-	-
<i>Chlamydophila psittaci</i> Birds and poultry	1920	-	Order no. 871 of 25/8 2011
Listeria monocytogenes	no	-	-
<i>Leptospira</i> spp. (only in production animals)	2003	-	Act no. 432 of 09/06/2004
Mycobacterium bovis/tuber-			
culosis	1920ª		
Cattle	OTF in 1980 ^d	Decision 2003/467/EC	Order no. 1417 of 11/12 2007
Coxiella burnetii	2005	-	Act no. 432 of 09/06/2004
Salmonella spp.	1993 ^e	-	
Cattle			Order no. 1723 of 22/12/2010
Swine			Order no. 1722 of 22/12/2010
Poultry			Order no. 1462 of 16/10/2009
VTEC	no	-	-
Yersinia enterocolitica	no	-	-
Parasites			
Cryptosporidium spp.	no	-	-
Echinococcus multilocularis	2004	Council Directive 64/433/EC	Act no. 432 of 09/06/2004
Echinococcus granulosus	1993	Council Directive 64/433/EC	Act no. 432 of 09/06/2004
Toxoplasma gondii	no	-	-
Trichinella spp.	1920ª	Regulation 2075/2005/EC	Order no. 412 of 28/05/2008
Viruses		-	
Lyssavirus (Rabies)	1920	-	Order no. 330 of 14/04/2011
Prions			
TSE			
Sheep and goats	yes	Regulation 999/2001/EC (as amended)	Order no. 1288 of 20/12/2011
BSE			
Cattle	yes	Regulation 999/2001/EC (as amended)	Order no. 499 of 26/05/2011 (as amended)

Table A31. Overview of notifiable and non-notifiable animal diseases presented in this report, 2012

a) Clinical cases, observations during the meat inspection at the slaughterhouse, positive blood samples or finding of agens are notifiable.

b) Officially Brucellosis Free (OBF) according to Council Directive 64/432/EC as amended and Commision Decision 2003/467/EC. No cases in cattle since 1962.

c) Officially *B. melitensis* Free (ObmF) according to Council Directive 91/68/EC and Commision Decision 2003/467/EC. Never detected in sheep or goat.

d) Officially Tuberculosis Free (OTF) according to Council Directive 64/432/EC as amended and Regulation (EC) No 1226/2002, and Commission Decision 2003/467/EC. No cases in cattle since 1988 or in deer since 1994.

e) Only clinical cases notifiable.

Source: Danish Veterinary and Food Administration

Time	Samples taken	Material	Material
Rearing flocks		Grandparent generation	Parent generation
Day-old ^{a,b}	Per delivery	5 transport crates from one delivery: crate liners (>1m ² in total) or swab samples (>1m ² in total). Analysed as one pool	5 transport crates from one delivery: crate liners (>1 m^2 in total) or swab samples (>1 m^2 in total). Analysed as one pool
1st & 2nd week ^{b, c}	Per unit	-	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample of 60g
4th week ^{a,c}	Per unit	5 pairs of boot swaps (analysed as two pooled samples), or 1 faeces sample consisting of 2x150g	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample of 60g
8th week ^{b,c}	Per unit	2 pairs of boot swabs (analysed as one pooled sample). Cage birds: 60x1g samples of fresh droppings. Analysed as one pool	2 pairs of boot swabs (analysed as one pooled sample). Cage birds: 60x1g samples of fresh droppings. Analysed as one pool
2 weeks prior to moving ^{a,d}	Per unit	5 pairs of boot swabs (analysed as two pooled samples), or 1 faeces sample consisting of 2x150g	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample of 60g
Adult flocks		Grandparent generation	Parent generation
Every two weeks ^{a,b} (Every 16th week) ^e	Per flock	Hatcher basket liners from 5 baskets (>1m ² in total) or 10g of broken egg- shells from each of 25 hatcher baskets (reduced to 25g sub-sample). Analy- sed as one pool	Hatcher basket liners from 5 baskets (>1m ² in total) or 10g of broken eggs- hells from each of 25 hatcher baskets (reduced to 25g sub-sample). Analysed as one pool
After each hatch ^b	Per hatch	Wet dust samples. Up to four hatchers of the same flock can be pooled	Wet dust samples. Up to four hatchers of the same flock can be pooled
Every week ^b	Per unit	-	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample of 60g
0-4 weeks after moving, 8-0 weeks before slaughter ^{b,d}	Per unit	5 pairs of boot swabs (analysed as two pooled samples), or 1 faeces sample consisting of 2x150g	5 pairs of boot swabs (analysed as two pooled samples), or 1 faeces sample consisting of 2x150g
After positive findings ^{b,d}	Per unit	5 pairs of boot swabs (analysed as two pooled samples), 2 dust samples (250 ml) and 5 birds (analysed for antimi- crobial substances)	5 pairs of boot swabs (analysed as two pooled samples), 2 dust samples (250 ml) and 5 birds (analysed for antimi- crobial substances)

Table A32. Salmonella surveillance programme for the rearing flocks and adult flocks of the grandparent and parent generation of the broiler and table egg production, 2012

a) Sampling requirements set out by Regulation (EC) No 2160/2003.

b) Samples collected by the food business operator.c) Sampling requirements set out by Order no 1463 of 16/12/2009.

d) Samples collected by the local food control offices.

e) When eggs from a flock exceed the capacity of one incubator, each incubator should be sampled as described. Source: Danish Veterinary and Food Administration

Time	Samples taken	Material
Salmonella		
15 - 21 days before slaughter ^{a,c,d}	Per flock	5 pairs of boot swabs. Analysed individually
7 - 10 days before slaughter ^{b,e}	Per flock	5 pairs of boot swabs. Analysed individually
After slaughter ^{b,c}	Per batch	300x1g neck skin, analysed in pools of max. 60 grams. Sampling size depends on whether the slaughterhouse slaughters only AM-negative flocks or AM-negative as well as AM-positive flocks
Campylobacter		

1 pair of boot swabs

Table A33. Salmonella and Campylobacter surveillance programme for the broiler flocks, 2012

a) Sampling requirements set out by Regulation (EC) 2160/2003.

b) Sampling requirements set out by Order no 1462 of 16/12/2009.

c) Samples collected by the food business operator.

7 - 10 days before slaughter^{b,e}

d) Once a year, one pair of socks is collected by the local food control offices.

e) Samples are collected by a representative of the slaughterhouse, laboratorium or the local food control offices.

Per flock

Source: Danish Veterinary and Food Administration

Table A34. Salmonella surveillance programme for the pullet-rearing, table egg layer and barnyard/hobby flocks in the table egg production, 2012

Time	Samples taken	Material
Pullet- rearing		
Day-old ^{a,d}	Per delivery	5 transport crates from one delivery: Crate liner (> 1 m ² in total) or swab samples (> 1 m ² in total) (Analysed as one pooled sample)
4 weeks old ^{b,d}	Per flock	5 pairs of boot swabs (analysed as two pooled samples) or 5 faeces samples of 60 gram
2 weeks before moving ^{a,c}	Per flock	5 pairs of boot swabs (analysed as two pooled samples) or 5x60 g faeces samples. 60 blood samples (serology)
Table egg layers (Production for certi	fied packing station	ons)
24 weeks old ^{a,c}	Per flock	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample consisting of 2x150 g. 250 ml (100 g) dust or 1 pair of boot swabs. 60 eggs ^b (serology)
Every 9 weeks ^{a,d,e}	Per flock	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample consisting of 2x150 g. 60 eggs ^b (serology)
After positive serological findings ^c	Per flock	5 pairs of boot swabs (analysed as two pooled samples) or 5x60 g faeces samples, and 60 hens
After positive findings of other serotypes than <i>S</i> . Enteritidis, <i>S</i> . Hadar, <i>S</i> . Infantis, <i>S</i> . Virchow or <i>S</i> . Typhimurium including the mo- nophasic strains <i>S</i> . 1,4,[5],12:i:- ^c	Per flock	5 pairs of boot swabs (analysed as two pooled samples) or 5 faeces samples consisting of 60 gram each
Barnyard and hobby flocks		
Every 18 weeks ^{b,d}	Per flock	Egg samples

b) Sampling requirements set out by Order no 1260 of 15/12/2008.

d) Samples collected by the food business operator.

e) According to Regulation (EC) 2160/2003 sample collection must be carried out every 15 weeks as a minimum.

c) Samples collected by the local food control offices.

Time	Samples taken	Material
Duck production		
Max. 21 days before slaughter ^{a,b}	Per flock	2 pairs of boot swabs. Analysed individually
Turkey production		
Max. 21 days before slaughter ^{c,d}	Per flock	2 pairs of boot swabs. Analysed individually

Table A35. Salmonella surveillance programmes for the duck and turkey flocks, 2012

a) Sampling requirements set out by Order no 1260 of 15/12/2008.

b) Samples collected by the food business operator.

c) Sampling requirements ser out by Regulation (EC) 584/2008.

d) Samples collected by the food business operator or the local food control offices.

Source: Danish Veterinary and Food Administration

No. of samples	Samples taken	Comment
Milk producing herds		
4 samples distributed over 18 months	Bulk tank samples	Calculation of herd level ^b
10 samples	Blood samples	If the owner wants a herd moved from
		level 2 to 1b
Non-milk producing herds		
1 sample every 180 days at slaughter ^c	Blood samples	Calculation of herd level $^{\rm b}$
4-8 samples	Blood samples	Consecutive negative samples required for level $1b^d$
Beef carcasses at the slaughterhou	ise	
5 samples daily, pooled into	Swab samples from 4 designated areas	Slaughterhouses slaughtering more than
one analysis	after 12 hours chilling (4x100cm ²)	200 cattle per day
5 samples per 200 slaughtered	Swab samples from 4 designated areas	Slaughterhouses slaughtering more than
cattle, pooled into one analysis	after 12 hours chilling (4x100cm ²)	200 cattle per month but 200 or less cattle per day
5 samples every 3 rd month,	Swab samples from 4 designated areas	Slaughterhouses slaughtering 50-200
pooled into one analysis	after 12 hours chilling (4x100cm ²)	cattle per month
1 sample every 3 rd month	Swab samples from 4 designated areas	Slaughterhouses slaughtering less than
	after 12 hours chilling (4x100cm ²)	50 cattle per month

Table A36. Salmonella surveillance programme^a for the cattle production, 2012

a) Order no. 143 of 22/2/2012 as ammended. In 2010, the programme for eradication of *Salmonella* Dublin from the Danish cattle production was intensified. This implies a new category of level 2 (level 2R) where the most contagious herds in this level are placed under official restrictions by the veterinary authorities.

b) Herd levels based on serological testing (blood and milk):

Level 1a: Milk producing-herd assumed free of infection (based on bulk tank samples),

Level 1b: Non-milk producing-herd or milk producing-herd assumed free of infection (based on blood samples),

Level 2: Herd not assumed free of infection,

Level 3: Herd infected,

Unknown level: insufficient number of blood samples have been taken from herd and no samples had antibody levels above the limit value.

c) No samples are taken, if the herd has been tested for *S*. Dublin within the last 180 days or 8 samples have been tested within the last 24 months.

d) Number of samples equals total number of animals in the herd minus 2 (max. 8 animals, min. 4 animals).

Source: Danish Agriculture and Food Council, and Danish Veterinary and Food Administration

Time	Samples taken	Purpose
Breeding and multiplier herds		
Every month	10 blood samples per epidemiological unit	Calculation of <i>Salmonella</i> -index based on the mean from the last three months with most weight to the result from the more recent months (1:3:6)
Max. twice per year	Herds with <i>Salmonella</i> -index 5 or above: Pen-faecal samples ^{b, d}	Clarify distribution ^c and type of infection in the herd
Sow herds		
When purchaser of piglets is assigned to level 2 or 3, max. twice per year	Pen-faecal samples	Clarify distribution ^c and type of infection in the herd, and clarify possible trans- mission from sow herds to slaughter pig herds
Herds positive with <i>S</i> . Typhimu- rium, <i>S</i> . Infantis and <i>S</i> . Derby are considered positive for the fol- lowing 5 years ^f	No samples are collected from the herd during the 5 year period when the herd is considered po- sitive, unless the herd is proven negative	Reduce repeated sampling in positive herds infected with a persistent serotype
Slaughter pigs, herds		
At slaughter	Meat juice, 60-100 samples per herd per year. Herds in RBOV ^{d, e} : one meat juice sample per month	Calculation of slaughter pig index based on the mean from the last three months with most weight to the result from the most recent month (1:1:3). Assigning herds to level 1-3 and assigning herds to risk-based surveillance (RBOV) ^e
Slaughter pigs, animals		
At slaughter	Coecum samples, 80 samples per month, 11 month per year	Random collection of samples for mo- nitoring of the distribution of serotypes and antimocrobial resistance.
Herds assigned to level 2 or 3, max. twice a year	Pen-faecal samples	Clarify distribution and type of infection in the herd
Pork carcasses at the slaughterhouse		
5 samples daily, pooled into one analysis	Swab samples from 4 designa- ted areas after 12 hours chilling (4x100cm ²)	Slaughterhouses slaughtering more than 200 pigs per day
5 samples per 200 slaughtered pig, pooled into one analysis	Swab samples from 4 designa- ted areas after 12 hours chilling (4x100cm ²)	Slaughterhouses slaughtering more than 200 pigs per month or 200 or less pigs per day
5 samples every 3 rd month, pooled into one analysis	Swab samples from 4 designa- ted areas after 12 hours chilling (4x100cm ²)	Slaughterhouses slaughtering more than 50 pigs per month or less than 200 pigs per month
1 sample every 3 rd month	Swab samples from 4 designa- ted areas after 12 hours chilling (4x100cm ²)	Slaughterhouses slaughtering less than 50 pigs per month

Table A37. Salmonella surveillance programme^a for the pig production, 2012

a) Sampling requirements set out by Order no. 1722 of 22/12/2010.

Source: Danish Veterinary and Food Administration

b) Herds with index above 10 have to pay a penalty for each pig sold.c) Pigs from herds in Level 3 must be slaughtered under special hygienic precautions.

d) The herd owner must inform buyers of breeding animals about the infection level and type of *Salmonella*. e) RBOV: risk-based surveillance where the sample size in herds with a SP-index of zero (no positive samples in the previous three months) are reduced to one sample per month. f) These serotypes are primarily spread by live trade, and are known to persist in herds.

Methods	Human	Food	Animal
Salmonella enterica			
Serotype	All	All	All
Phage type ^a	S. Typhimurium and S. Enteritidis	Few S. Typhimurium and S. Enteritidis	Few S. Typhimurium and S. Enteritidis
Antimicrobial resistance	S. Typhimurium, 50 % of <i>S</i> . Enteritidis, approx. 90 % of other serotypes	Almost all isolates	Almost all isolates
MLVA	S. Typhimurium and S. Enteritidis	S. Typhimurium (outbreak investigations), research	S. Typhimurium (outbreak investigations), research
PFGE	Outbreak investigations	Outbreak investigations	Outbreak investigations
Campylobacter coli/je	ejuni		
Antimicrobial resistance	Isolates from 3 districts for DANMAP surveillance	For DANMAP surveillance purposes and the case-by-case program	Only for DANMAP surveillance purposes
FlaA-SVR	Outbreak investigations	Outbreak investigations	None
MLST	Outbreaks investigaions, research	None	None
VTEC			
Serotype	All	None	All (O157)
Virulence profile	All	None	All (O157)
PFGE	O157	None	Outbreak investigations
Listeria			
Serogroup	All	None	None
MLVA	All	None	None
PFGE	All	All	All
Yersinia enterocolitico	1		
O-group	Isolates from one district	None	None

Table A38. Typing methods used in the surveillance of foodborne pathogens in Denmark, 2012

a) For the *Salmonella* source account and outbreak investigations. Source: Statens Serum Institut, and Danish Zoonosis Laboratory, National Food Institute

Appendix E

Population and slaughter data

Table A39. Human population, 2012

Age groups (years)	Males	Females	Total
0-4	160,671	152,285	312,956
5-14	340,049	324,591	664,640
15-24	366,867	350,982	717,849
25-44	709,414	701,385	1,410,799
45-64	749,899	746,684	1,496,583
65+	451,952	547,849	999,801
Total	2,778,852	2,823,776	5,602,628

Source: Statistics Denmark

Table A40. Number of herds/flocks, livestock and animals slaughtered, 2012

	Herds/flocks ^a	Livestock ^a (capacity)	Number slaughtered
Slaughter pigs (>27 kg)	7,507	6,612,144	19,056,065
Cattle	20,689	1,618,851	495,700
Broilers	298	20,700,921	100,231,000
Layers (excl. barnyard)	418	4,935,017	-
Turkeys	46	510,334	5,498
Sheep & lambs	7,632	156,029	80,128
Goats	3,487	23,953	2,241
Horses	-	-	1,673

a) March 2012.

Source: The Central Husbandry Register, Statistics Denmark, and Danish Veterinary and Food Administration

	No. of holdings	No. of houses/flocks	Livestock (capacity)
Rearing period (grandparent)	4	14	90,000
Adult period (grandparent)	4	7	80,000
Rearing period (parent)	15	90	130,000
Adult period (parent)	44	143	720,000
Hatcheries	5	-	-
Broilers	245	566	-

Table A41. Number of farms in the broiler production, 2012

Source: Danish Veterinary and Food Administration and Danish Agriculture and Food Council

	No. of holdings	No. of houses/flocks	Livestock (capacity)
Rearing period (parent)	5	6	20,000
Adult period (parent)	9	10	50,000
Hatcheries	5	-	-
Pullet-rearing	82	138	1,440,000
Layers (excl. Barnyard)	165	220	2,910,000

Table A42. Number of farms in the table egg production, 2012

Source: Danish Veterinary and Food Administration, and Danish Agriculture and Food Council

Table A43. Distribution of import, export and production of fresh and frozen meat and the production of table eggs in Denmark, 2008-2012^a. Data is presented in tons

		•					
	Year	Pork	Beef	Broiler meat ^a	Turkey meat	Duck meat ^c	Table eggs ^d
Import	2008	83,057	81,427	32,480	8,264	4,494	-
	2009	83,265	88,818	30,321	7,000	4,251	-
	2010	89,565	100,528	43,746	8,728	4,863	-
	2011	85,035	92,293	51,294 ^f	8,270	6,117	-
	2012	88,192	90,694	59,914 ^f	6,942	3,778	-
Export	2008	1,386,849	66,690	109,725	2,345	772	-
	2009	1,321,820	78,572	108,377	1,564	534	-
	2010	1,399,397	88,169	115,674	2,921	810	-
	2011	1,563,511	83,944	81,358 ^f	3,335	1,685	-
	2012	1,350,419	81,276	72,682 ^f	2,553	1,872	-
Danish	2008	1,602,648	149,744	157,543	49	37	67,900
production	2009	1,508,640	163,068	159,723	93	-	60,600
	2010	1,584,503	184,979	171,210	78	-	63,200
	2011	1,668,991	169,241	165,154	48	1,297	66,000
	2012	1,580,163	165,435	159,355	41	1,021	66,300
Consumption ^e	2008	298,857	164,481	80,298	5,968	3,722	-
	2009	270,084	173,314	81,667	5,529	3,717	-
	2010	274,671	197,338	99,283	8,728	4,053	-
	2011	190,515	177,590	-	4,983	3,390	66,000
	2012	317,936	174,853	-	6,969	2,927	66,300

a) Data from 2011 and 2012 are extracted on March 25^{th} 2013. Data from 2012 are preliminary and will be updated in the 2013 report.

b) Natural-marinated chicken is included.

c) Mixed products of ducks, geese and guinea fowl are not included.

d) Consumption of table eggs is assumed to be roughly the same as the production, since import and export of table eggs is minimal.

e) Consumption = Production + import - export.

f) Due to data problems, data might not reflect the true Danish import and export and thus the consumption has not been calculated Source: Statistics Denmark

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