### Technical University of Denmark



### **Annual Report on Zoonoses in Denmark 2016**

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# Annual Report on Zoonoses in Denmark 2016



**DTU Food**National Food Institute

### **Annual Report on Zoonoses in Denmark 2016**

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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 2016. 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 year's report. The report is also available at www.food.dtu.dk.

### Introduction

Campylobacter continues to be the most common food borne pathogen in Denmark with 4,677 cases in 2016. This is an increase compared to the previous year, but it is not possible to determine if the reason is due to the changes in the reporting system, changes in the diagnostic practice as changes in the analytical methods are being introduced or a true increase in number of cases. In 2016, there were three foodborne outbreaks caused by Campylobacter, two outbreaks with a few cases each and one outbreak due to imported duck meat caused 103 cases.

A total of 1,074 human cases of salmonellosis were reported in 2016, which is an increase compared to 2015 with only 925 human cases, but still lower than the previous years. *S.* Enteritidis and *S.* Typhimurium including monophasic strains continue to be the most common serovars found humans with an incidence of 4.3/100,000 inhabitants and 5.6/100,000, respectively. Both in humans and several animal and food sources the monophasic strains of *S.* Typhimurium was more common than the genuine *S.* Typhimurium.

In animals, *S*. Enteritidis was only reported from imported broiler meat and duck meat. Whereas *S*. Typhimurium including the monophasic strains were reported from several sources (Danish pigs and pork, broilers, layers and duck, and imported pork and duck meat).

The model behind the Salmonella source account links the number of human salmonellosis cases caused by different Salmonella subtypes to specific food and animal reservoirs. In 2016, Danish produced pork was estimated to be the most important food source of human salmonellosis (approximately 64 cases). This finding is in line with observations from 2012-2014, but an increase from last year, where imported pork was the most important food source. The increase in cases attributed to Danish produced pork can partly be explained by an outbreak related to this specific food source comprising 16 cases. The second most important source was estimated to be imported broiler meat (~43 cases) followed by imported pork (~40 cases). Further, around 22 cases were attributed to Danish produced table eggs in 2016, which is in contrast to last year where no cases were attributed to table eggs for the first time since the source account model has been applied.

In total, 49 foodborne outbreaks were reported in 2016, with a total of 1,825 registered cases of which 234 were confirmed in the laboratory. This is an increase compared to 2015, but the second lowest number of outbreaks reported since the introduction of the cen-

tral registration of outbreaks in the end of 2005. The slight increase is mainly seen in outbreaks caused by *Salmonella* and verocytotoxin-producing *E. coli* (VTEC), while outbreaks caused by other agents are stable in numbers. In total, 12 foodborne *Salmonella* outbreaks were reported compared to three in 2015. International collaboration was needed for several outbreaks including two travel-related outbreaks.

As in previous years Novovirus was the most frequent cause of foodborne outbreaks (18 outbreaks), and in total, 1,178 persons were affected by Novovirus outbreaks.

### Burden of foodborne disease in Denmark

Estimating the burden of foodborne diseases is important in order to be able to rank diseases in Denmark according to their overall health impact in the population, and ultimately to inform risk management strategies in the area of food and health.

Toxoplasmosis is a common foodborne parasitic disease caused by *Toxoplasma gondii* and it is ranked the third most important cause of foodborne disease in Europe although only few cases are registered annually. In Denmark, the estimated disease burden and public health impact of congenital toxoplasmosis (CT) (toxoplasmosis caused by transplacental transmission of *T. gondii*) was higher than the registered number of cases, demonstrating that CT is under-diagnosed or under-reported in Denmark. CT may be a severe and life-long disease, and knowledge about its public health impact is important for guiding public health policy at the national level.

Yersinia enterocolitica is, as most other microbiological foodborne disease, believed to be under-reported and the estimation of burden of disease showed that for each reported human infection approximately 11 people were estimated to be infected. This led to a total estimate of 1,860 cases of yersiniosis.

#### Vector-borne zoonoses

The first systematic survey of urban ticks and urban tick-borne pathogens was conducted in Copenhagen City and the northern and southern suburbs in 2016. The density of ticks is much lower in the urban areas compared to areas populated with roe deer, which is believed to be the main cause of high tick densities. Different species of *Borrelia* were found in ticks from seven of eight examined sites and at relatively high infection rates. This suggests that these urban tick vectors pose a very real risk of zoonotic infections.

## 1. Trends and sources in human salmonellosis

### By Nanna Sophia Mucha Munck (nsmm@food.dtu.dk) and Tine Hald

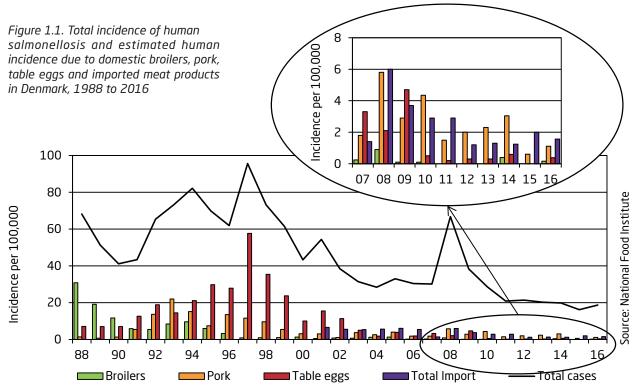
In 2016, a total of 1,074 human cases of salmonellosis were reported in Denmark. This corresponds to an incidence of 18.8 cases per 100,000 inhabitants, which is slightly higher than last year, where the lowest incidence since the 1980'ies was observed (figure 1.1). The incidence of *S.* Enteritidis was 4.3 per 100,000 inhabitants this year continuing the decreasing trend seen since 2013. The incidence of *S.* Typhimurium was 5.6 per 100,000 inhabitants in 2016, which is an increase compared to last year. The increase may be explained by the occurrence of several outbreaks of *S.* Typhimurium compared to last year, where only one *S.* Typhimurium related outbreak with six cases was detected¹.

The share of reported *S.* Typhimurium, the monophasic variant, increased from 50% (1.9 per 100,000) of total reported *S.* Typhimurium cases in 2015 to 64% (3.3 per 100,000) of the total reported *S.* Typhimurium cases this year. This predominance of monophasic *S.* Typhimurium strains was observed for the first time in 2014<sup>2</sup>.

The attribution of human salmonellosis cases to different sources is based on a mathematical model that links the

number of human salmonellosis cases caused by different *Salmonella* subtypes to specific food and animal reservoirs. The model is based on phenotyping and molecular analysis of *Salmonella* isolates and compares the number of human cases caused by different *Salmonella* subtypes with the distribution of the same subtypes isolated from various animal-food sources. Input data is obtained from the national *Salmonella* surveillance of food-producing animals and food of animal origin including imported meat, and the reported cases of human salmonellosis. Results of the source account model support risk management decisions, allowing for evaluation of implemented interventions as well as for the need for new initiatives.

Salmonella subtypes are defined by serotyping (all isolates), Multiple Locus Variable Tandem Repeat Analysis, MLVA-typing (S. Enteritidis and S. Typhimurium including its monophasic variants), and resistance profiling (S. Typhimurium including its monophasic variants). MLVA profile for S. Enteritidis is defined by the number of repetitions observed in five independent loci, namely SE1, SE5, SE2, SE9 and SE3. For S. Typhimurium and its monophasic variants the five loci are



Source: National Food Institute, Technical University of Denmark

STTR9, STTR5, STTR6, STTR10 and STTR3. For source attribution purposes, the complete scheme was used for *S*. Enteritidis, but the level of discrimination of the original scheme was reduced for *S*. Typhimurium and its monophasic variants to include only three loci, namely STTR9|STTR10|STTR3³. Implication of this decision on the source account results are discussed below, especially regarding Danish produced table eggs. In the source account model, monophasic strains of *S*. Typhimurium (S. 1,4,[5],12:i:-) are separated from classical *S*. Typhimurium strains to better identify any epidemiological changes of these types.

#### Salmonella source account 2016

The overall trend in human salmonellosis cases attributable to the major food-animal sources is presented in figure 1.1. Danish produced pork was assessed to be the most important food source of human salmonellosis (64 cases)(Figure 1.2 and Table A1). This finding is in line with observations from 2012-2014, but in contrast to findings from last year, where imported pork took the lead. The increase in cases attributed to Danish produced pork can partly be explained by an outbreak related to this specific food source comprising 16 cases (FUD 1521).

In 2016, 22 cases were attributed to Danish produced table eggs, which is in contrast to last year where no cases were attributed to table eggs for the first time since the source account model has been applied. Important to note is the wide credibility interval related to the mean estimate found this year (1-55 cases). The attribution is based on three positive samples identified in three different flocks of different sizes. The serotypes responsible for the cases were *S*. Typhimurium monophasic variant and *S*. Derby. In addition, the monophasic variant found in one of the flocks did not match

any human cases. Differences were found in the full MLVA profile and the resistance profile differed. Consequently, the prevalence for Danish table eggs was adjusted according to these observations omitting the positive flock from which the strain had no human cases matching. For the two other layer flocks, the strains found matched human cases, but these strains were also found in Danish and imported pork, which explains the wide credibility interval found for table eggs. From 2017, the *Salmonella* source account will be based on whole-genome sequencing data, which will provide us with a better tool to distinguish between isolates that are identical with the currently applied subtyping methods.

In contrast to last year, positive samples from Danish produced broiler meat were found in 2016 and consequently 9 cases were attributed to this source. Data from Danish produced ducks was available this year, compared to 2014 and 2015, and 5 cases were attributed to this source. No difference was observed in cases attributed to Danish produced beef in 2016 (17 cases) compared to 2015 (Figure 1.2 and Table A1).

Imported sources were responsible for 1.6 reported cases per 100,000 inhabitants in total. The most important source was imported chicken (43 cases), followed by imported pork (40 cases), imported duck (4 cases) and imported turkey (3 cases) (Figure 1.2 and Table A1). No positive samples were detected in imported beef and consequently no cases were attributed.

The source attribution model estimates travel related cases from the reported travel cases and also takes cases without travel information into account by adding a proportion of these to the travel related cases (Table 1.1 does not include cases without travel information which is why the numbers differ). In 2016, more than half of all reported

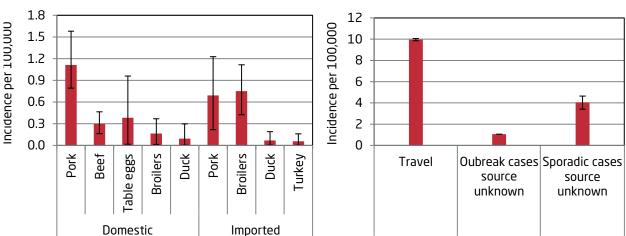


Figure 1.2. Estimated sources of 1,074 cases of human salmonellosis in Denmark, 2016. Incidences per 100,000 inhabitants and 95% credibility intervals are shown (See also Appendix Table A1)

Note: Domestic pork includes 16 outbreak cases. Sporadic and outbreak-related cases with unknown source include all sources not in the model. E.g. one outbreak with four cases was caused by contact to snakes fed with mice and is included in the 'outbreak cases, source unknown' category. Source: National Food Institute, Technical University of Denmark.

cases was estimated to be travel related (573 cases) which was an increase compared to the number of estimated travellers in 2015 (522 cases). Of the 246 reported *S*. Enteritidis cases, 71% was estimated to be travel related, which is lower than the share estimated in the previous two years (78.0% in 2015 and 77.9% in 2014). Less than a third of the 320 reported *S*. Typhimurium cases was estimated to be related to travel (30.9%), which is a small decrease compared to last year (33.8% in 2015). Sporadic *Salmonella* cases that could not be attributed to any of the sources included in the model were allocated to unknown sources. This year, a total of 21.7% of cases could not be attributed, which is in line with previous years.

Sporadic and outbreak cases allocated to unknown sources may be associated with exposure to foods not included in the national surveillance programs, or by nonfood sources such as direct contact with pet animals or person-to-person transmission. This year for instance, an outbreak was caused by contact to snakes fed with mice (FUD 1532). Thus, the related cases were included in the model as outbreak cases with source unknown.

### Antibiotic resistance in *S.* Typhimurium

The panel of antibacterial agents used to compose the resistance profiles was identical to the panel used last year, where Cefotaxime and Ceftazidime were introduced, replacing Ceftiofur.

Resistance information was available for 46 of the *S*. Typhimurium cases attributed to Danish produced food products. The vast majority (37 cases) was caused by resistant strains, while nine cases were caused by susceptible strains. This represents an increase in both resistant and susceptible strains compared to last year (Figure 1.3).

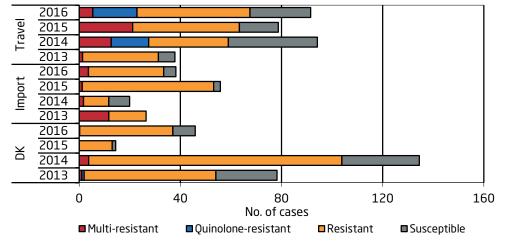
Resistant strains also dominated among cases attributed to imported food sources (30 cases out of 38 cases with available resistance information). Although the number of cases was lower than last year (52 reported cases in 2015), it was still higher compared to the years 2012-2014. An increase in reported cases caused by both multi-resistant strains and susceptible strains was observed compared to last year (Figure 1.3).

In total, 92 travel-associated *S*. Typhimurium cases had available resistance information. Among these resistant strains also dominated (45 cases). A reduction in multi-resistant strains was observed compared to last year, but was overtaken by an increase in quinolone-resistance strains of which the vast majority is also multi-resistant. Quinolone resistance in human strains was not observed in 2015.

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Figure 1.3. Distribution of antimicrobial resistance<sup>a</sup> in S. Typhimurium, including S. 1,4,[5],12:i:-, from human infections attributed to domestic or imported food sources, or travel in the Salmonella source account, 2013-2016



a) Resistant: Resistant towards one to three antimicrobial agents: Multi-resistant: Resistant towards four or more antimicrobial agents. Antimicrobials in the resistance profile for the Salmonella source account were: ampicillin, chloramphenicol, sulphamethoxazole, tetracycline, trimethoprim, ciprofloxacin, gentamicin, nalidixic acid, cefotaxime and ceftazidine.

Source: National Food Institute, Technical University of Denmark.

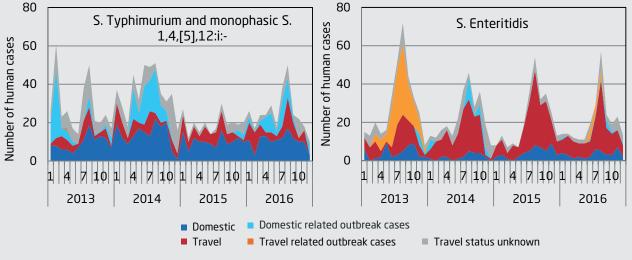
In 2016, as in the previous years, Statens Serum Institut attempted to interview all registered *Salmonella* cases where no travel information was reported by the general practitioner. The patients were asked about the date of disease onset and whether they had travelled abroad within a seven-day period prior to disease onset. This information was complemented with information from general practitioners' reports. Travel information was obtained from a total of 81.3% of the *Salmonella* cases in 2016. Among the cases with known travel history, 55.1% were infected abroad (Table 1.1). However, the proportion of travel-related cases varied greatly between the different serotypes, hence 78.2% of the *S.* Enteritidis cases, 33.0% of the *S.* Typhimurium cases, 30.6% of the monophasic *S.* 1,4,[5],12:i:- cases and 57.9% of cases with other serotypes were infected abroad. Similar to previous years, the majority of travel-related cases in 2016 travelled to Thailand (19.3%), Turkey (12.7%), and Spain (5.2%). Two travel-related outbreaks in patients returning from Greece and Turkey were identified (see chapter 2).

Table 1.1. Top 10 Salmonella serotypes in humans and information about travel abroad, 2015-2016

2016	Number of patients (%)	% of patier Abroad <sup>b</sup>	ntsª infected Domestically	2015	Number of patients (%)	% of pati Abroad <sup>b</sup>	ents <sup>a</sup> infected Domestically
Enteritidis	246 (22.9)	78.2	21.8	Enteritidis	258 (27.9)	78.2	21.8
1,4,[5],12:i:-	192 (17.9)	30.6	69.4	1,4,[5],12:i:-	117 (12.6)	34.3	65.7
Typhimurium	108 (10.1)	33.0	67.0	Typhimurium	116 (12.5)	32.7	67.3
Stanley	42 (3.9)	81.1	18.9	Newport	32 (3.5)	50.0	50.0
Newport	29 (2.7)	52.2	47.8	Oranienburg	24 (2.6)	25.0	75.0
Java	27 (2.5)	76.2	23.8	Infantis	21 (2.3)	53.3	46.7
Infantis	23 (2.1)	57.9	42.1	Stanley	21 (2.3)	80.0	20.0
Dublin	20 (1.9)	9.1	90.9	Dublin	19 (2.1)	30.0	70.0
Kentucky	19 (1.8)	94.1	5.9	0:4,5,12; H:b:-	17 (1.8)	54.5	45.5
Saintpaul	19 (1.8)	69.2	30.8	Java	16 (1.7)	71.4	28.6
Other serotypes	349 (32.5)	53.2	46.8	Other serotypes	284 (30.7)	57.8	42.2
Total	1,074 (100)	55.1	44.9	Total	925 (100)	56.6	43.4

a) Patients with unknown travel information (18.7% of all patients in 2016 and 17.3% in 2015) were excluded from the percent calculations.

Figure 1.4. Monthly distribution of S. Enteritidis and S. Typhimurium incl. monophasic S. 1,4,[5],12:i:- cases, 2012-2016



Source: Statens Serum Institut.

b) Infected abroad is defined as travel abroad in a seven-day period prior to disease onset.

Source: Statens Serum Institut.

### 2. Food- and waterborne outbreaks

### By the Central Outbreak Management Group

Food- and waterborne outbreaks in Denmark are reported in the Food- and waterborne Outbreak Database (FUD). Outbreaks that occurred in 2016 are presented in Appendix Table A4. Figure 2.1 shows the relative distribution of these outbreaks by the different causative agents. Household outbreaks and clusters that could not be verified as common source outbreaks are not included. The outbreak investigation procedures in Denmark are described in further details in Chapter 8.

In total, 49 foodborne outbreaks were reported to FUD in 2016 (Appendix Table A4). This is the second lowest number of outbreaks since the introduction of the central registration of outbreaks in FUD in the end of 2005. The slight increase is mainly seen in outbreaks caused by *Salmonella* and verocytotoxin-producing E. coli (VTEC) while outbreaks caused by other agents are stable in numbers. This could be explained by increased use of whole-genome sequencing (WGS).

In total, the number of persons affected by foodborne outbreaks was 1,825, with a median of 13 persons per outbreak (range 2 - 412). The outbreaks were mainly regional or local outbreaks (70%). Eleven outbreaks were considered national outbreaks and one was part of an international

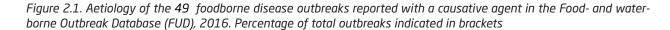
outbreak. The largest outbreak involving 412 persons was a national norovirus (NoV) outbreak caused by *Lollo Bionda* lettuce (FUD1500), see description below in section 2.1

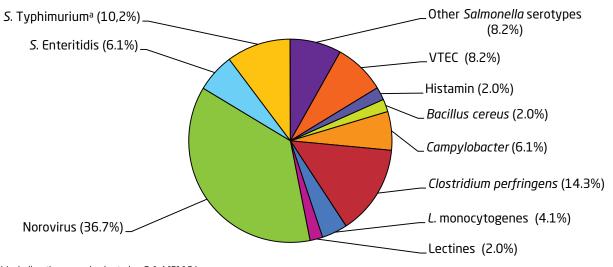
In 2016, *Clostridium perfringens* was associated with seven foodborne outbreaks affecting a total of 353 people compared to 11, 7, and 16 outbreaks caused by this agent in 2015, 2014, and 2013 respectively.

When dividing the outbreaks into reported settings, the most frequent setting was restaurants (18%) with 9 outbreaks affecting 309 people (mean: 34 people per outbreak). Outbreaks taking place in workplace canteens and catering (11 outbreaks) also affected a high number of people (821 people) and affected on average 75 persons per outbreak. Composite meals (10 outbreaks) and buffet meals (8 outbreaks) combined were the most frequently reported sources of outbreaks in 2016 and most often these outbreaks were associated with NoV or *C. perfringens* (Appendix Table A4).

### Norovirus outbreaks

As in previous years NoV was the most frequent cause of foodborne outbreaks (18 outbreaks), and in total, 1.178 persons were affected by NoV outbreaks. The transmission





a) Including the monophasic strains *S.* 1,4,[5],12:i:-. Source: Food- and waterborne Outbreak Database (FUD).

routes for NoV causing foodborne outbreaks were multiple. In Table 2.1 a breakdown of the number of outbreaks and the number of people affected per route of transmission for 2015-16 are shown. The most common way of infection with NoV in 2016 was contamination from ill or healthy carrier among kitchen staff. In 2016, this way of infection constituted 44% of the NoV outbreaks. The number of outbreaks caused by ill guest attending a buffet was at the same level in 2016 compared to 2015 however; the number of persons affected was more than doubled compared to 2015. This way of contamination deserves continued focus.

The largest NoV outbreak started in early April 2016, when an unusual high number of point-source outbreaks of gastrointestinal disease were reported to the regional Food Control Offices (FUD1500). A national investigation was initiated to investigate whether the outbreaks were related, which pathogen(s) were involved and whether there was a common food source causing the outbreaks. Outbreaks were individually investigated. Two analytical studies in a seminar and a high school were performed. Patient stool samples were collected and analysed; positive stool samples were sequenced. A total of 23 linked point-source outbreaks with 412 persons reported ill with diarrhea and/or vomiting occurred over the course of one week. Fresh green coral lettuce (Lollo Bionda lettuce) produced in France was found to have caused the large series of linked NoV outbreaks. In a cohort study including 234 participants a dish containing green lettuce was associated with illness. NoV of Genogroup I (GI) was detected in samples from 28 patients comprising eight of the outbreaks. Sequencing showed GI.P2-GI.2. NoV GI was detected in one of 20 examined lettuce heads1.

In November, another NoV outbreak occurred in a large company in the Copenhagen area (FUD1536). A questionnaire was distributed to approximately 1300 employees to identify possible sources of infection and stool samples were collected from cases. The descriptive and analytical analysis of the responses from the questionnaire identified that 284 persons had been ill and that having eaten in the canteen on October 31, 2016 was associated with a higher risk of being ill (risk ratio 4.97, [95% CI; 3.62-6.84]). No specific food item was found that could explain the outbreak fully. In total, 14 stool samples were all positive for NoV GII, 12 could be subtyped to GII.Pe-GII.4 Sydney 2012. It was concluded that one or more food items sold in the canteen on October 31, 2016 were responsible for the outbreak and recommendations on hygiene for both kitchen staff and guests in the canteen was given.

In October, a conference center in Copenhagen experienced an outbreak with NoV and a questionnaire survey was conducted (FUD1538). In all, 103 persons were ill with diarrhea and/or vomiting and seven were confirmed with a positive NoV stool sample (Nov GII.P16-GII.2 and NoV nd-Gii.2) Samples were from cases in two separate groups of guests and from one member of the kitchen staff. It was concluded that the outbreak most likely were caused by transmission from the ill kitchen staff via the food served to participants in five different events at the conference center.

### Outbreaks with Salmonella

In 2016, there was a marked increase in the number of outbreaks caused by *Salmonella*. In all, twelve *Salmonella* outbreaks were reported to FUD compared to three in 2015. Two outbreaks were related to travel and six outbreaks were

Table 2.1. Norovirus outbreaks per route of transmission based on number of cases or number of outbreaks, 2014-2016

	2016		2015		2014	
Transmission route/source	No. of outbreaks	No. of persons ill	No. of outbreaks	No. of per- sons ill	No. of outbreaks	No. of per- sons ill
Ill kitchen staff or healthy carrier of virus among kitchen staff	6	258	5	153	11	507
Kitchen staff tending to ill persons at home before entering the kitchen	2	40	4	96	6	729
III person/guest attending a buffet	4	355	4	108	2	43
Seafood (oysters)	3	92	1	22	3	40
Frozen raspberries/strawberries	0	0	1	9	2	20
Leafy greens / lettuce	3	433	0	0	0	0
Water	0	0	1	142	0	0
Total	18	1,178	16	530	24	1,339

Source: Food- and waterborne Outbreak Database (FUD).

small with less than ten cases and the source was not found despite interviews with cases.

In July 2016 a regional *Salmonella* 0:4,5,12;H:i:-MLVA0126 outbreak in Jutland was registered (FUD1521). Seven cases were connected to the same town fair in Mid Jutland. The source of these seven cases and one sporadic case was most likely Danish pork meat from a local slaughterhouse. The hypothesis was that the pork was insufficiently cooked and that cross-contamination from the meat to ready to eat dishes (salads and cooked meat) could have taken place. The sporadic cases with the same MLVA type is probably also due to fresh pork meat - however, no link to the same slaughterhouse could be established.

April 8, the Swedish health authorities reported a cluster of *Salmonella* 0:4,5,12; H:i:- with a specific MLVA profile (MLVA0338). April 21, the Swedish authorities declared that salami sticks from a Danish company produced in Poland was the suspected source – a product also sold in Denmark. The product was recalled from consumers in both Sweden and Denmark voluntarily by the Danish company on April 22. In Denmark, the first cases were registered April 25 (FUD1504). In all, 12 Danish cases were linked to this outbreak and further confirmed by WGS typing- they became ill between March 20 and May 11. Eight were female and four male and they were aged 3 months to 65 years. Nine were children under five years old. Interviews showed that 10/11 (one was not interviewed) had consumed salami sticks. The last case was a three months old baby for whom

secondary transmission was suspected. Salmonella was eventually detected by the Swedish authorities in samples of three opened packages and one unopened package of the product. Based on the Swedish investigation and the clear link for the majority of the Danish cases it was concluded that the salami sticks were the source of this outbreak.

### Other outbreaks of interest

In June, a large outbreak of gastroenteritidis was seen caused by a lunch catering company serving food for approximately 1,400 persons from 38 different companies (FUD1513). An electronic questionnaire survey was conducted and showed that 103/393 persons from 19 different companies were ill. Campylobacter jejuni was found in 19 of 25 patient samples. WGS of six of the samples showed a genetic cluster (ST48). Duck meat was suspected to be the source - sampling at the wholesaler of the same lots of duck meat delivered to the catering company showed Campylobacter in duck meat, but with a differed WGS profile. However, it is possible that more than one strain could be present in the duck meat at the same point in time. The explanation of this outbreak was that there most likely was a cross contamination from the fresh duck meat into ready-to-eat meat dishes. The handling of the fresh duck meat took place at adjacent tables to the tables where meat that had already been cooked took place.

Another *Campylobacter* cluster based on WGS was observed with patients from May-June 2016. In all, eight

### Pet snakes pose a risk of Salmonella infection

By Luise Müller (lum@ssi.dk)

A persistent outbreak of *Salmonella* Enteritidis phage type (PT) 8 infection, characterised by MLVA type 2-10-8-5-2, were reported in September 2015 by the United Kingdom. By whole-genome-sequencing (WGS) analysis, it was discovered that the outbreak had been ongoing in the United Kingdom since at least 2011. The British outbreak investigation team found that illness was associated with exposure to pet reptiles, in particular corn snakes, and feeder mice. The feeder mice were imported into the United Kingdom from a rodent farm in Lithuania. The European Centre for Disease Prevention and Control (ECDC) initiated a multi-national collaboration in order to clarify whether other countries in addition to the UK had been affected by this outbreak (1).

In Denmark, the same Salmonella Enteritidis MLVA type 2-10-8-5-2 was identified in 12 patients from 2014-2016. Three of these clustered by WGS with the British outbreak – however, it was not possible to establish an epidemiological link for two cases where interviews were possible, since they reported no contact to snakes. Another Danish cluster was however, revealed by WGS with four cases from January to December 2016 (FUD1532). Interviews showed that three patients reported contact to snakes (a boa constrictor, a corn snake and a kingpython) all fed with live mice. Comparison with British isolates showed that this type had been found in another cluster of 11 patients in United Kingdom – however, no interview data were available.

### Reference

1. European Centre for Disease Prevention and Control. Multi-country outbreak of *Salmonella* Enteritidis PT8 infection, MLVA type 2-10-8-5-2, associated with handling of feeder mice – 1 December 2016. Stockholm: ECDC; 2016.

cases living on the island of Bornholm. Interviews did not reveal any common event or connection between the cases. A non-confirmed source was babyleafs/lettuce because 4/5 interviewed reported consumption of this and three specifically mentioned lettuce in a bag ready to eat. In the weeks prior to the investigation a lettuce/babyleaf mix with rucola (Napolitana mix) had been recalled from consumers due to a finding of *Campylobacter* in the product – unfortunately no subtyping of this product was possible hence it is not known whether this batch or an adjacent batch of Napoliana Mix could have been the source of this outbreak.

The source of a long-lasting outbreak of listeriosis was found in 2016. The cluster was discovered by whole-genome sequencing already in 2015 with two cases in 2015 and one in 2014. In 2016, a Swedish patient was reported with the same type after consumption of a Danish cold cut ("rullepølse") made from pork. In 2016, additionally three cases were reported in Denmark. Interviews were possible for three of the Danish cases, who had all consumed "rullepølse". The Swedish authorities found the same strain of *Listeria* in the open package of "rullepølse" at the patient's home. This lead to an extensive investigation at the Danish producer with findings of the same strain in the environment and a withdrawal of several products.

In 2016, VTEC was increasingly subtyped by WGS. This revealed four outbreaks where the source was not found for three of them. One outbreak in August-September was a cluster of VTEC 0157:H7, vtx1a, vtx2c, ST11 in

Mid-Jutland. Six cases were reported with this type - four male and two female aged 15 - 41 years. The suspected source was beef used as kebab in take away restaurants. Cattle are a well-known reservoir for VTEC and there are no restrictions on presence of the pathogen in fresh beef, since heat treatment is sufficient to kill the bacteria. Trace back investigation could not point out a specific supplier of the kebab or a specific lot of meat. In June, a VTEC outbreak was seen in a day care center in Jutland (FUD1520). Within the same week, five children became ill. Two cases were diagnosed with VTEC 0121:H19, vtx2a, ST655 and one of them developed hemolytic uremic syndrome. There was no suspicion about a common food source consumed in the day care center, but the children had had several common risk exposures during the incubation period including playing in an open sand box, visiting farms with animals, and offered raw milk during a visit to a farm.

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Multi-national Salmonella outbreak linked to eggs from Poland

By Luise Müller (lum@ssi.dk)

From 1 May 2016 to 24 February 2017, 14 European countries have reported 218 confirmed *Salmonella* Enteritidis cases belonging to two distinct whole-genome sequencing (WGS) clusters (UK cluster 1.2.3.18.359.360.% and UK cluster 1.2.3.175.175.%), and 252 probable cases sharing the *S.* Enteritidis MLVA profiles 2-9-7-3-2 or 2-9-6-3-2. Outbreak cases, both confirmed and probable, have been reported by Belgium, Croatia, Denmark, Finland, France, Greece, Hungary, Italy, Luxembourg, the Netherlands, Norway, Slovenia, Sweden and the United Kingdom. The outbreak peaked at the end of September 2016. Available evidence from epidemiological, microbiological, environmental and tracing investigations identified eggs originating from three Polish packing centers as the vehicle of infection in this outbreak<sup>2</sup>.

In Denmark, six cases were identified to match the international case definition with the same distinct WGS profiles from June to October 2016 (FUD1531). Two of the cases were travel-related to Austria and Hungary. Four cases were infected in Denmark and interview showed that they had been visiting the same restaurant on Funen. It was not possible to link the food at the restaurant to eggs or poultry meat of Polish origin via the trace-back investigation; however, it is possible that eggs, egg products or poultry meat of Polish origin could have been sold on the Danish marked and even served at the restaurant as these products are widely distributed and often through more establishments before ending up being served or used in private households.

### 3. Burden of foodborne diseases in Denmark

### 3.1 Estimating the burden of foodborne disease

#### By Sara Pires (smpi@food.dtu.dk)

At the National Food Institute and under the Metrix Project<sup>a</sup>, we are developing methods to estimate the burden of a range of foodborne diseases caused by microbial agents, chemical hazards and diet-associated risk factors. These estimates will then be used to rank diseases in Denmark according to their overall health impact in the population, and ultimately to inform risk management strategies in the area of food and health.

To be able to compare diseases with different causes, incidence and symptoms, we apply a harmonized health metric that assesses the impact of diseases in terms of incidence, severity, duration, and mortality.

### Total incidence of disease by foodborne pathogens

Infections by foodborne and other zoonotic pathogens are notified to public health surveillance. Nonetheless, it is widely recognized that reported cases represent only the "tip of the iceberg", and that for many pathogens the true incidence of disease in the population is largely unknown. The gap between the true number of cases caused by contaminated foods and what is captured by public health surveillance systems can be easily explained: for a case to be identified, the ill person must seek medical care; the doctor must request a sample; the causative pathogen must be identified at a laboratory; and the results must be reported to public health officials. Any failure in this process leads to underdiagnosis and under-reporting.

To estimate the total incidence of a selected number of foodborne pathogens in Denmark, we derive multiplication factors that translate the probability that a patient seeks care, is diagnosed with infection by a specific foodborne pathogen, and the case is reported.

### Disability Adjusted Life Years (DALYs)

The disability adjusted life years is the most common metric used to estimate the burden of diseases. It accounts for the years lost due to decreased quality of life and/or premature death caused by a given disease or condition, at the individual or population level.

DALYs are the sum of years lived with disability (YLD), and the years of life lost due to premature death caused by

a) The Metrix Project is funded by the National Food Authorities and aims at performing integrated analyses of the health risks and benefits of food, nutrients, and food diets, as well as ranking foodborne diseases on the basis of their health and economic burden in Denmark.

the disease (YLL), aggregating morbidity, severity, duration and mortality.

### How are these estimates useful?

Burden of disease studies quantify the health impact of diseases in a population by integrating information on the incidence, mortality and disability caused by all potential harmful health effects of these diseases. Therefore, they provide the scientific evidence necessary to allow policymakers to rank different foodborne diseases and thus prioritize interventions to reduce their public health, as well as economic burden. We will expand our efforts to estimate the burden of other foodborne diseases in Denmark, thereby providing a more complete picture of the public health burden. The next step will be to integrate these estimates with economic analyses and calculate the total costs of foodborne illnesses in Denmark.

This chapter presents an update from the articles published in Chapter 3 of the Annual Report on Zoonoses in Denmark 2015, where we presented the burden of disease of *Salmonella*, *Campylobacter* and VTEC infections in Denmark in 2013-2015.

### 3.2 Disease burden of congenital toxoplasmosis in Denmark

### By Joanna Nissen, Christen Rune Stensvold, Henrik V. Nielsen and Sara M. Pires (smpi@food.dtu.dk)

Toxoplasmosis is a common foodborne parasitic disease caused by *Toxoplasma gondii*. More than one million cases of foodborne infection by this parasite are estimated to occur in Europe annually, ranking it the third most important cause of foodborne disease in the region<sup>1</sup>.

Congenital toxoplasmosis (CT) is caused by transplacental transmission of *T. gondii*. CT might lead to miscarriage (foetal loss before gestational age 22), stillbirth (foetal loss after gestational age 22), or sequelae in the child; however, the majority of congenitally-infected children have no apparent symptoms or clinical signs at birth.

In a recent study<sup>2</sup>, we used regional and nationwide data (see the box on page 17) to estimate the disease burden of CT in Denmark in 2014 in terms of incidence, occurrence of sequelae, mortality, and burden of disease as expressed by disability-adjusted life years (DALYs). The following health outcomes of CT were included in the model: sequelae manifesting in the first year of life (chorioretinitis, intracranial

calcification, hydrocephalus, central nervous system (CNS) abnormalities, and neonatal death), sequelae appearing after the first year of life (chorioretinitis), and still birth.

### Incidence and DALY estimation of congenital toxoplasmosis

We estimated that in 2014, a total of 105 pregnant women acquired a primary *T. gondii* infection leading to 0.8 CT-related stillbirth, and that 14 children were born with CT. Of the 14 children born with CT, 3 children were estimated to be born with CT-related sequelae, 3 children were estimated to be asymptomatic in the first year of life but will develop chorioretinitis before the age of 12, and 8 children will be asymptomatic until the age of 12 (Table 3.1). Among the children born with CT-related sequelae, intracranial calcification and chorioretinitis were estimated to be the most frequent outcomes, hydrocephalus and CNS abnormalities were estimated to be less frequent and neonatal deaths were estimated to be even more infrequent. In the model, we did not account for co-morbidity (Table 3.2).

The disease burden of CT was estimated to be 123b DALYs in 2014 (Table 3.1). In total, 80 years of life were lost due to premature mortality (YLLs), where CT-related foetal loss resulted in 78 DALYs and neonatal death resulted in 2 DALYs. A total of 42b years of life were lost due to disability (YLDs), where hydrocephalus and CNS abnormalities caused 13 DALYs and 14 DALYs, respectively; the remaining sequelae caused a lower disease burden (Table 3.2).

Our findings are in agreement with but slightly higher than the findings by Torgerson and Mastroiacovo (2013). They estimated 13 annually cases of CT resulting in 82 DALYs in Denmark<sup>3</sup>. Both investigtions estimated the annual disease burden of CT to be substantially lower than the an-

nual disease burden caused by salmonellosis (379 DALYs) and campylobacteriosis (1,586 DALYs)<sup>4</sup>. It is important to emphasize that our estimates do not included the health impact of acquired toxoplasmosis (i.e., post-natal infection) and that this still needs to be addressed.

The estimated disease burden of CT in Denmark is much lower compared to a Dutch estimate where CT caused 2,251 DALYs annually between 2007 and 2011<sup>5</sup>, causing 13.4 DALYs per 100,000 inhabitants. Our results showed a disease burden of 2.2 DALYs per 100,000 inhabitants. The difference between the two countries can largely be explained by the lower CT incidence in Denmark. The apparent CT incidence of 1.9 per 10,000 live-born children in Denmark was captured through The Danish National Neonatal Screening Programme for Congenital Toxoplasmosis (DNNSP)<sup>6</sup> (see box on page 17), a study which included >98% of all newborns in Denmark over a period of 8.5 years. Comparing the incidence estimates obtained using equivalent methods, the CT incidence in Denmark was almost 11 times lower than in The Netherlands<sup>7</sup>, two times lower than in Poland [8], three times lower than estimated by WHO-FERG for countries with low child mortality status, including EU<sup>9</sup> and a little higher than in Ireland<sup>7</sup>.

#### Under-ascertainment of CT

During the DNNSP, 6-19 children were annually diagnosed with CT. Ever since the DNNSP was discontinued in 2007, 0-5 children have annually been registered with CT (2008-2014) (Table 3.1). Our model estimated that 14 children were born with CT in 2014, and that three of these children had clinical signs of CT at birth. Nevertheless, no cases of CT were registered in 2014 and consequently, even symptomatic CT cases are missed. Moreover, comparing our estimated incidence with the registered number of CT cases indicates that for each CT case reported, at least five other CT cases could be expected to have occurred. These data

Table 3.1. Number of estimated (1) and registered (2) cases of congenital toxoplasmosis (CT) in Denmark in the period of 2008-2014

Year	2008	2009	2010	2011	2012	2013	2014
Live-births in DK <sup>a</sup>	65,038	62,818	63,411	58,998	57,916	55,873	56,870
1. Total estimated no. of CT cases	16	16	16	15	15	14	14
No. of cases with sequelae in the first year of life	4	4	4	3	3	3	3
No. of cases with no sequelae in the first year of life but who will have developed chorioretinitis until the age of 12	3	3	3	3	3	3	3
No. of cases who will not have developed sequelae until the age of 12	9	9	9	8	8	8	8
2. No. of registered cases of CT <sup>b</sup>	5°	1	3	0	1	1	0

a) Statistics Denmark, accessed on 1 June 2016 (http://www.danmarksstatistik.dk/en/)

Source: Adapted from Reference2.

b) Manually calculation of the sum of the health outcomes in Table 3.2 does not equal 123 DALYs and 42 YLDs. This is because there is not accounted for decimals for each health outcome.

b) The Danish National Registry of Patients, accessed on 23 June 2016.

c) One of these cases was registered in 2009.

indicate that CT is currently under-diagnosed or underreported and that public health surveillance data present an incomplete picture of the impact of the disease in the country. However, the Danish Health Authority is expected to include CT in the national laboratory-based infectious disease surveillance programme in the near future.

### Conclusions and future work

In Denmark, the estimated disease burden and public health impact of CT are lower than what has been estimated for other European countries, emphasizing the need for country-specific studies. Moreover, our findings demonstrate that without a surveillance programme, CT is under-diagnosed or under-reported in Denmark. CT may be a severe and life-long disease, and knowledge about its public health impact is important to guiding public health policy at the national level. The National Food Institute is now joining efforts with Nordic partners to estimate the most important sources and transmission routes of *T. gondii* infections, which will be crucial to identifying and prioritizing the most effective interventions.

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Table 3.2. Estimated incidence and disease burden of congenital toxoplasmosis (CT) in Denmark, 2014

		Estimated cases in 2	2014			
	Reported cases 1999-2002 <sup>a</sup>	Cases per 1,000 live births (Median and 95% Ul <sup>b</sup> )	Total cases (Median and 95% UI <sup>b</sup> )	Duration years <sup>c</sup>	Disability weight (Mean and 95% UI <sup>b</sup> )	Total DALYs <sup>d</sup> (Median and 95% UI <sup>b</sup> )
Foetal loss ≥ 22 weeks of gestation	-	-	0.84 (0.6-1)	92	1	78 (64-94)
Symptomatic in the	first year of li	fe				
Chorioretinitis	7	0.03 (0.01-0.06)	2 (1-3)	81	0.031 (0.019-0.049)	5 (3-8)
Intracranial calcification	10	0.04 (0.02-0.07)	3 (1-4)	81	0.01 <sup>g</sup>	3 (2-4)
Hydrocephalus	1	0.01 (0.001-0.02)	0.40 (0-1)	81	0.36 (0.16-0.56)	13 (4-29)
CNS abnormalities	1	0.01 (0.001-0.02)	0.40 (0-1)	81	0.36 <sup>g</sup>	14 (5-27)
Neonatal death <sup>5</sup>	0	0.0004 (0.0001-0.001)	0.02 (0-0.03)	92	1	2 (1-3)
Asymptomatic in th	e first year of	life				
Chorioretinitis later in life (Follow up to 12-3 years) <sup>f</sup>	6	0.05 (0.02-0.1)	3 (1-5)	69	0.031 (0.019-0.049)	7 (4-11)
Total	-	-	-	-	-	123 (100-148)

a) Number of CT-cases reported from the initial four years (1999-2002) of DNNSP

b) UI=uncertainty interval.

c) Duration of all health outcomes is life-long. Life expectancy data retrieved from Statistics Denmark (accessed on May 17, 2016)

d) DALYs, disability-adjusted life years. Calculated as median incidence x duration x disability weight

e) Minimum data from Denmark (zero neonatal deaths reported in initial four years [1999-2002] of DNNSP [10]); most likely and high value adapted from Havelaar et al. (2007)<sup>11</sup>, giving an interval of 0.7% (0-1.2)

f) Chorioretinitis later in life, based on follow-up observations of The Danish Neonatal Feasibility Study.

g) No uncertainty interval available.

Source: Adapted from Referense2.

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### 3.3 Disease burden of yersiniosis in Denmark

### By Joana Pessoa (joapes@food.dtu.dk), Steen Ethelberg and Sara M. Pires

Yersiniosis is a bacterial disease caused by Yersinia enterocolitica and Y. pseudotuberculosis. In the European Union, most cases are caused by Y. enterocolitica, where it has been the third most reported zoonosis for the past four years. Denmark has reported 8 and 10 cases per 100,000 population in 2014 and 2015, respectively, which makes it the country with the second highest notification rate in the region<sup>1</sup>. It should be noted, however, that a considerable proportion of these reported cases belong to the non-pathogenic biotype 1A.

The epidemiology of *Y. enterocolitica* is not completely known, but pigs are considered the main reservoir of human pathogenic strains<sup>2</sup>. Other sources have been identified, such as a wide range of animals, animal-derived food products, vegetables and water sources<sup>3</sup>.

The most common symptom of yersiniosis is gastroenteritis with self-limiting diarrhea associated with mild fever and abdominal pain. However, infection can also be limited to the right fossa iliaca, causing terminal ileitis or mesenteric lymphadenitis, which can lead to symptoms that can be confused with those of acute appendicitis (pseudo-appendicitis syndrome)<sup>3</sup>. Yersiniosis can also cause post-infection sequelae such as reactive arthritis (ReA), erythema nodosum and irritable bowel syndrome (IBS)<sup>4</sup>.

Even though yersiniosis is a mandatory notifiable disease, like other foodborne pathogens it is likely to be

The burden of disease caused by Toxoplasma gondii.

By Joanna Nissen and Sara M. Pires

### Danish Neonatal Feasibility Study (DNFS)

The DNFS was conducted from June 1992 to August 1996, where pregnant women from five counties in Denmark who gave birth to live children, representing one third of all deliveries in the country, were offered screening at delivery for primary *Toxoplasma gondii* infection acquired during pregnancy<sup>12</sup>. Guthrie cards of newborns were analysed for *T. gondii*-specific IgM and IgG antibodies and if found positive the mother's first-trimester serum sample (routinely taken for syphilis testing at weeks 8 to 12 of gestation) was thawed and analysed for *T. gondii*-specific IgG antibodies. When CT was suspected, confirmatory diagnostic serology was performed on blood samples from mother and child within 6 weeks after birth<sup>12</sup>. Of 89,873 children sampled during the DNFS, 27 children were diagnosed with CT, corresponding to an apparent incidence of 3.0 per 10,000 live-born children. Follow-up was performed for 26 of the 27 children for late development of chorioretinitis with a maximum follow-up time of 12 years (3.9-12.3 years, median age 10 years).

### The Danish National Neonatal Screening Programme for Congenital Toxoplasmosis (DNNSP)

The DNSSP was conducted from January 1999 to July 2007 and included >98% of all newborns in Denmark<sup>10,13</sup>. Guthrie cards of newborns were analysed for *T. gondii*-specific IgM and IgA antibodies. When CT was suspected, confirmatory diagnostic serology was performed on blood samples from mother and child. Of 560,000 children born during the DNNSP, 105 children were diagnosed with CT, corresponding to an apparent incidence of 1.9 per 10,000 live-born children. The 55 children born with CT in the first four years of the DNNSP were followed for three years for developmental and clinical outcomes<sup>10</sup> and this data were used to estimate sequelae in the first year of life.

underdiagnosed and underreported to public health surveillance. We estimated the total incidence of yersiniosis in Denmark by correcting for underreporting and the burden of disease by applying the DALY metric.

### Estimating the incidence of yersiniosis

To estimate the total incidence of yersiniosis in Denmark, we estimated a multiplication factor that corrects the reported number of cases for under-diagnoses and under-reporting, using non pathogen-specific and *Yersinia*-specific parameters informed by data collected through a population-based telephone survey conducted in 2009<sup>5</sup>, by literature review, and by expert elicitation (Table 3.3). As biotype 1A is considered non-pathogenic, cases caused by this type were excluded. We combined the parameters in different steps, re-constructing the surveillance pyramid, and used a probabilistic model with 20,000 iterations to account for uncertainty. All modelling steps were performed in R 3.3.2 (R Core Team, 2016).

#### **Health-outcomes and DALY calculations**

All the potential outcomes for yersiniosis were identified by a literature review, as were the probabilities of their occurrence given infection (Figure 3.2). Due to lack of data, we have not considered pseudo-appendicitis syndrome and erythema nodosum in our model. In consistency with models developed for other foodborne pathogens, we have also not considered sepsis<sup>6</sup>.

To calculate the total DALYs associated with yersiniosis, we estimated the incidence of each health outcome and adopted the disability weights (DW) from the Global Burden of Disease study<sup>7</sup> and from the Dutch Burden of Disease study<sup>8</sup>. For these calculations we used the DALY Calculator, an interface developed in R<sup>9</sup>.

### Burden of disease of yersiniosis

For each of the 172 *Yersinia* pathogenic biotypes cases reported in 2016, we estimated that around 11 people had

yersiniosis in Denmark, leading to an estimated 1,860 cases. While in the female population the incidence of yersiniosis was substantially higher in children under 5 and decreased with age groups, in males the distribution of cases in the population was less marked (Figure 3.3).

We estimated that the overall burden of yersiniosis in Denmark was of 88 DALYS, of which 63% were years lived with disability (YLD) and 37% years of life lost due to premature mortality (YLL). The health outcome leading to the highest burden was gastroenteritis (GE) which caused 38 DALYs, most of it due to the death of two cases (Table 3.4).

#### Conclusion

The estimated annual disease burden of yersiniosis in Denmark was low when compared to other foodborne pathogens. Even though our estimates point to a higher incidence in young children, the oldest age groups bear the highest burden due to mortality, and because the symptoms of disease in children are often less severe.

These are the first estimates of the burden of disease of yersiniosis in Denmark. Still, the assumptions made to address knowledge gaps and overall data limitations led to considerable uncertainty around our estimates, and results should be interpreted with care.

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Long-term sequelae

Death

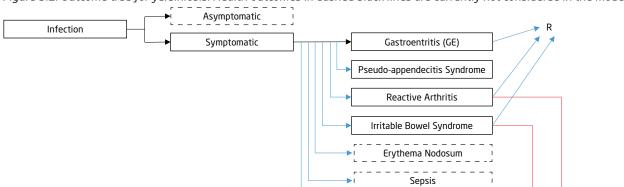
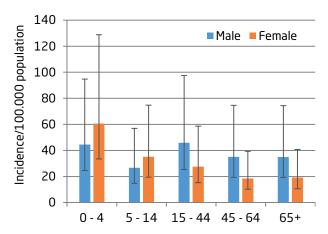


Figure 3.1. Outcome tree for yersiniosis. Health outcomes in dashed black lines are currently not considered in the model

Figure 3.2. Estimated incidence of yersiniosis per 100,000 population by age group and gender, 2016



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Table 3.3. General and Yersinia-specific parameters used to estimate the true incidence of yersiniosis

	Description	Reference
General Parameters  Yersinia-specific	Probability of seeking medical care	Müller et al. (2012)⁵
	Probability of submitting a stool sample for analysis	Müller et al. (2012) <sup>5</sup> ; Pires, (2014) <sup>6</sup>
	Probability of reporting a positive laboratory result	MiBaª
Yersinia-specific	Probability of testing for Yersinia in sample	S. Ethelberg, PCb
parameters	Sensitivity of laboratory analysis	E. Moller Nielsen, PCb
	Proportion of bloody diarrhea in cases	Schiellerup et al. (2008)10
	Proportion of hospitalized cases	Helms et al. (2006) <sup>11</sup>
	Proportion of biotype 1A	S. Ethelberg, PC <sup>b</sup>

<sup>1.</sup> MiBa: The Danish Microbiological Database

Table 3.4. Estimated total DALYs, YLD and YLL associated with the different health outcomes of yersiniosis in Denmark, 2016

2010								
	Gastroenteritis		Reactive Arthritis		Irritable b drome	Irritable bowel syn- drome		6 CF)
	Median	(95% CF)	Median	(95% CF)	Median	(95% CF)	Median	(95% CF)
DALY	38	(35-44)	22	(20-24)	28	(27-29)	88	(83-93)
YLD	5	(2-11)	22	(20-24)	28	(27-29)	55	(50-61)
YLL	33	-	0	-	0	-	33	-
Deaths	2	-	0	-	0	-	2	-
Cases	1,831	(1,505-2,217)	393	(356-434)	132	(127-136)	2,356	(2,026-2,739)

<sup>2.</sup> PC: Personal communication

### 4. Vectorborne zoonoses

### By René Bødker (rebo@vet.dtu.dk), Erika Vrbová and Kirstine Klitgaard Schou

The National Veterinary Institute, Technical University of Denmark monitors vectors and vector borne diseases in Denmark on behalf of the Danish Veterinary and Food Administration. The Veterinary Institute is responsible for the national weekly surveillance of mosquito and Culicoides vectors and for quantifying and mapping ticks and tickborne pathogens. The surveillance is focused on endemic vectors but also screens for exotic vectors.

The Nile Fever mosquito, Culex modestus, was previously discovered by the surveillance program around a single pond in the suburban area of Greve, 20 kilometers south of Copenhagen (see in Annual Report 2014). Since then, this important vector for the zoonotic West Nile virus practically vanished from the site (see Annual Report 2015). But here in 2016 it was rediscovered in large numbers around three ponds on the salt marches of Vest Amager, a protected natural wetland just outside Copenhagen close to the airport and approximately 15 kilometers from Greve across the brackish Bay of Køge (Figure 4.1). In August and September, it was the dominating man biting vector, during daytime. The unusually warm autumn in 2016 allowed intensive biting rates of this vector all September. The Vest Amager habitat is somewhat similar to the recently colonized north Kent marshes in the UK1. It is likely that Vest Amager is the main breeding site and that it is from there the mosquito occasionally colonize the Greve area.

In 2016, Culex modestus was also discovered for the first time by the Swedish Veterinary Institute at a coastal location in Sweden situated at the same latitude as Vest Amager. Man-biting Culex pipiens biotype molestus were reported from southern Sweden in 2016<sup>2</sup>. The same year, hybrids between this biotype and the bird biting Culex pipiens were reported from Linköbing area, 250 kilometers further north in Sweden<sup>3</sup>. In the 1920s, man-biting Culex pipiens were observed in Copenhagen and it is likely that they are still present in Denmark<sup>4</sup>. Culex modestus and Culex pipiens molestus bite both humans and birds and can therefore function as bridge vectors which can transfer virus from wild birds to humans and other mammals. The main zoonotic threats from man biting Culex mosquitoes are presently the West Nile virus and the Usutu virus, two flaviviruses which both are expanding their northern range in Europe. In 2016,

the first large outbreak of Usutu virus was reported in wild birds from Holland, less than 300 kilometers from the Danish border. Also, several outbreaks of West Nile virus have been reported in Austria. The finding of *Culex modestus* in high numbers very close to densely populated areas in Eastern Denmark is important when assessing the risk of zoonotic outbreaks of West Nile and Usutu virus in Denmark and the rest of southern Scandinavia.

In 2016, the first survey of urban ticks and urban tick-borne pathogens was conducted in Copenhagen City and the northern and southern suburbs of Copenhagen. These areas were all free from roe deer which in Denmark is considered to be the main driver of increasing tick densities<sup>5</sup>. This is the first time urban parks, cemeteries, and recreational areas have been systematically screened in Denmark. More than 600 ticks belonging to Ixodes ricinus were collected from 10 of the 27 sites. The majority was larvae and only 35 were nymphs and 14 were adults of which 8 were females. In Copenhagen City, we found a single co-infection with Borrelia afzelii and B. spielmanii in a nymph while eight adults and seven other nymphs were negative. In the northern suburbs, we found Borrelia in 4 out of 5 sites and identified either B. afzelii, B. valaisiana, B. miyamotoi, B. garinii, B. spielmanii or B. burgdorferi sensu stricto in four of 21 nymphs and in seven of nine adults. In the southern suburbs, we found Borrelia at both examined sites with either B. valaisiana or B. miyamotoi in two of nine 9 nymphs and in a single adult.

The densities of nymphs and adult ticks were much lower in the urban areas compared to nearby areas populated with roe deer. However, the infections rates with Borrelia species were surprisingly high in the urban ticks. Because the urban areas have relatively more human visitors per area, compared to forest areas, and because there are few other suitable large hosts than man and pets, the few urban nymphs and female adults may have a relatively high chance of getting in contact with a human host. The observed high infection rates in the urban nymphs and adult ticks suggest that these urban vectors carry a very real risk of zoonotic infections. Urban private gardens were not examined in this survey, but it is likely that urban gardens with park like vegetation also harbors tick populations some of which may carry Borrelia bacteria.





Figure 4.1. Man biting Culex modestus 8th September 2016 at Vest Amager recreational area just outside Copenhagen (Photo: Anders Lindström, SVA).

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### 5. Human psittacosis

By Charlotte Kjelsø (jel@ssi.dk), Søren Anker Uldum, Pernille Dahl Nielsen and Birgitte Beck Jørgensen

Psittacosis or parrot fever, is caused by *Chlamydophila psittaci*. The bacterium can withstand desiccation, and human infection occurs mostly by inhalation of aerosols, feces, or secretions from infected birds, but also by mouth-to-beak contact, or by contact with plumage and tissues of infected birds. Person-to-person transmission occurs rarely, if ever. The diagnosis can be made early in the disease process in the detection of *C. psittaci* using PCR technique on secretions from the lower respiratory tract. In addition, it is possible to detect antibodies in blood samples, but there are 2-3 weeks or longer before the assays become positive. In Denmark samples from humans are not cultured routinely for *C. psittaci*. Psittacosis can be treated with antibiotics.

The incubation period is usually between 7 and 14 days, but both shorter and longer periods are seen. The disease is in most cases mild and mostly appears with flu-like symptoms such as fever, muscle pain and headaches, but some patients develop a severe pneumonia. The disease is usually diagnosed in middle-aged men, who are predominantly infected in Denmark.

In the years 2011-2016, there have been an increasing number of human psittacosis cases infected in Denmark (TAble 5.1). In 2016 an outbreak of six human cases was registered after participating in an agricultural show with several bird species, amongst others parakeets, and parrots and pheasants. The Danish Veterinary and Food Administration, The National Veterinary Institute, The Danish Patient Safety Authority and Statens Serum Institut worked together to investigate the outbreak and prevent further infection. Persons with symptoms that had been at the county fair were recommended to consult a doctor to be tested for psittacosis. Just as physicians in the area were informed to pay attention in connection with the diagnosis and treatment of patients with atypical chest infections. There were subsequently diagnosed sick birds in a family who attended the agricultural show. In addition, in 2016 an outbreak of three human cases with contact to the same pet store with sick birds was detected. Both an employee and bird buyers were infected. The outbreak was handled in cooperation between the Danish Veterinary and Food Administration, The National Veterinary Institute, the Danish Patient Safety Authority, and Statens Serum Institut.

Upon investigation of the background of a relatively high number of human psittacosis in 2015 were among

others found an accumulation of eight cases infected through contact with ducks and mixed birds, including pigeons and chickens from breeders on Funen and Jutland from May to July. The Danish Veterinary and Food Administration took measures in relation to tracking and treating or culling birds from the infected herds.

In 2014 several smaller outbreaks were registered among people who had captured, put out or moved ducklings to new habitats with later hunting in mind. The ducklings were transported in the cabin of ordinary cars without shielded transport spaces.

Similarly, in 2014 a small outbreak with epidemiologically link between people who had bought parakeets from the same dealer was registered. The bird-dealer could not be confirmed as the source of the outbreak.

Clusters revealed in 2013 and 2012 were not possible to link epidemiologically apart from the fact that the majority of all recorded human cases of psittacosis were from Southern Denmark. This could however indicate a lack of attention to diagnostics and notification of psittacosis in humans in the rest of the country. An outbreak in Southern Sweden among people who had cleaned feeders in private gardens in Skåne and Kronoberg gave rise to an alert in Denmark. No increased incidence of *C. psittaci* in wild birds or Danish spread of human psittacosis was registered in Denmark in relation to the Swedish outbreak. The Danish Veterinary and Food Administration launched in 2011 a survey of the prevalence of Chlamydophila psittaci in parrots and parrotlike birds in pet shops. In 2012 the survey was extended to include suppliers to bird shops. They found a statistically significant increase in the number of positive samples in 2012 compared to 2011 but also that bird shops that were found psittacosis-positive in 2011 had no infected birds 2012. This suggests that control measures, with either killing or treating flocks of birds and subsequent cleaning and disinfection, works, and that infection does not persist but evolves through new-introduction of C. psittaci. An investigation of bird providers showed that especially being both a bird supplier and a pet retailer poses great risk of psittacosis. This underlines the importance of the veterinary quarantine rules of a minimum 30 days interval between introduction of birds to existing flocks.

All samples from the birds during the outbreaks and surveys from 2011-2016 were analyzed at The National Veterinary Institute at the Technical University of Denmark.

Ornithosis primarily affects parrot birds, pigeons and ducks. Although some serotypes seem more common in some species than in others, contamination between different bird species occurs, and from different bird species (both wild birds and poultry) to humans. Not all infected birds show clinical symptoms and in several outbreaks among ducklings, it is seen that there was also contact with pigeons.

Signs of infection in the animals include floats (secretions) from the eyes and nose, diarrhea and low body weight. The person responsible for a bird or poultry herd should if psittacosis (chlamydiosis) is suspected, immediately call a veterinarian. The veterinarian should contact the DVFA if the suspicion maintains after examination. The birds are then subject to isolation and movement restrictions and diagnostic samples should be sent to a laboratory approved by DVFA. If the samples are positive the birds are treated or killed and cleaning and disinfection are performed before movement restrictions are lifted. Animals with contact to sick birds are traced and will also be subject to movement restrictions and tested. Everyone who sells or buys birds or poultry has to keep records of seller/buyer information including addresses. This information is very important when it comes to the tracing of animals and humans who has had contact to

diseased birds. cf. Legislation on avian chlamydiosis:

Order no. 871 of 25.7.2011 on avian chlamydioses (Psittacosis). The 1st of July 2017 a new order on chlamysiosis comes into force, though there will be no significant changes for the public; The disease will still be notifiable on suspicion, and The DVFA will use the necessary measures in a risk based way to control the disease.

For more information about psittacosis in humans see SSI website at

http://www.ssi.dk/Service/Sygdomsleksikon/O/Ornitose.aspx . For more information about psittacosis in birds / poultry see DVFA website at

https://www.foedevarestyrelsen.dk/Leksikon/Sider/Ornitose-(Clamydiose).aspx.

Table 5.1. Registered human psittacosis in Denmark, 2011-2016

Year	Denmark	Abroad
2011	6	1
2012	10	2
2013	11	1
2014	15	1
2015	25	0
2016	23	1
Total	90	6





### 6. International topics

By Gudrun Sandø (gus@fvst.dk)

### **EU** targets

Harmonised regulation on targets and surveillance in the poultry production has been laid down by the Commission. An overview is presented in Appendix Table A30.

According to Regulation (EC) No 1190/2012, the EU target for *Salmonella* in breeding and fattening turkey flocks is 1% positive for *S.* Typhimurium or *S.* Enteritidis. In Denmark, no turkey flocks were positive with *S.* Typhimurium or *S.* Enteritidis in 2016 (Appendix Table A12).

In breeding flocks of *Gallus gallus*, Regulation (EC) No 200/2010 lays down a 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, three breeding flocks from the broiler production were positive with *S.* Stourbridge, *S.* Give and *S.* Derby in 2016 (Appendix Table A9 and A11).

Regulation (EC) No 517/2011 lays down targets for the reduction of *Salmonella* in laying flocks. The targets are Member States specific and are 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. 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 below 2% since 2004. In 2016, two flocks were found positive with *S.* 1,4,[5],12:i:- (Appendix table A9).

In broiler flocks of *Gallus gallus*, Regulation (EC) No 200/2012 lays down a target at a maximum of 1% flocks positive for *S*. Enteritidis and *S*. Typhimurium 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 2016, 0.4 % of broiler flocks was positive with *S*. Typhimurium including the monophasic *S*. 1,4,[5],12:i:- strains (Appendix Table A11).

## 7. Surveillance and control programmes

The collaboration on zoonoses between national and regional authorities, the industry and non-governmental organizations in Denmark is presented in Figure 7.1. According to the Danish legislation, 41 infectious diseases are clinically notifiable in Denmark. An overview of the notifiable and non-notifiable human and animal diseases, presented in this report, is provided in Appendix Table A31 and Table A32, respectively, including reference to the relevant legislation.

#### 7.1 Surveillance of human disease

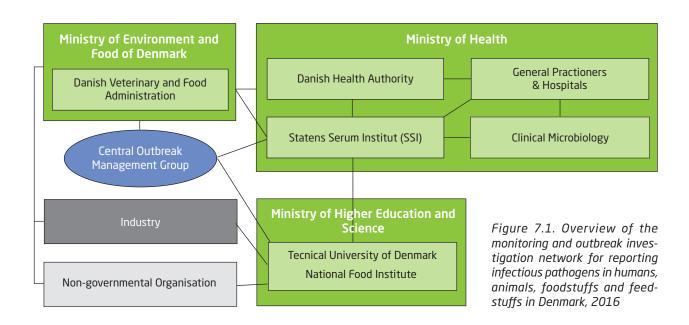
Information on human cases due to zoonotic pathogens presented in this report is reported to Statens Serum Institut through different channels depending on the disease:

- Notifiable through the laboratory surveillance system: Salmonella, Campylobacter, Yersinia, Verotoxin-producing E. coli (VTEC) and Listeria.
- Individually notifiable zoonotic pathogens: Chlamydia psittacci (ornithosis), Leptospira (Weils disease), Mycobacterium, Bovine Spongform Encephalopathy (BSE) prions (var. Creutzfeldt-Jakob Disease), Verotoxinproducing E. coli (VTEC) and Lyssavirus (rabies).
- Non-notifiable zoonotic pathogens: Brucella.

In Denmark, the physicians report individually notifiable zoonotic diseases to the Danish Health Authority and the

Department of Infectious Disease Epidemiology at Statens Serum Institut. Physicians send specimens from suspected cases to one of the clinical microbiology laboratories depending on the geographical region. 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. 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 results are recorded in the Register of Enteric Pathogens maintained by Statens Serum Institut. 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 serovar and MLVA distribution is presented in Appendix Table A6-A8.



### 7.2 Outbreaks of zoonotic gastrointestinal infections

In Denmark, local and regional foodborne outbreaks are typically investigated by the Food Control Officesa in collaboration with the Public Health Medical Officers at the Danish Patient Safety Authority, and the regional clinical microbiology laboratories. Larger regional and national outbreaks are investigated by Statens Serum Institut, the National Food Institute, Technical University of Denmark 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 outbreaks. The formal responsibility of investigating foodor waterborne outbreaks is currently divided between two ministries based on the outbreak source: the Ministry of Health for infectious diseases; the Ministry of Environment and Food for foodborne and animal related diseases, and for waterborne diseases. The latter are investigated in collaboration with the municipalities.

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 Food Control Office. General practitioners and hospitals are obliged to report all suspected water- and foodborne infections to the Danish Patient Safety Authority and 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 outbreaks from 2016 are outlined in Chapter 2.

### 7.3 Surveillance and control of animals and animal products

In Denmark, action plans and programs on zoonoses have been in place for more than 25 years. The first plan targeted *Salmonella* in the broiler production and was developed as a response to an increase in the number of human cases related to eating chicken meat. Since then, plans have been developed for *Salmonella* in pigs and pork, *Salmonella* in

layers (eggs), *Campylobacter* in broilers and *S.* Dublin in cattle and beef.

All plans have been outlined in cooperation between industry, research institutes and authorities, and are followed by a technical working group and a steering committee. This ensures progress, that new knowledge is incorporated in the plans, and an assessment of achievement of targets.

At EU level, harmonized surveillance programs and common targets have been set for the broiler and laying egg production. An overview on the status on the targets can be seen in Table A30.

Salmonella surveillance and control programmes for poultry, pigs and cattle are presented in Appendix Tables A33-A38. Sample analysis is performed at authorised private laboratories, the Danish Food and Veterinary Administrations laboratory, the National Food Institute and the National Veterinary Institute at the Technical University of Denmark. 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 A39.

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

- Results from the table egg production are presented in Appendix Tables A9-A10.
- Results from the broiler production are presented in Appendix Tables A6-A7, A11 and A18.
- Results from the duck and turkey productions are presented in Appendix Tables A6 and A12.
- Results from the pig production are presented in Appendix Tables A6-A7, A15, A18 and Figures A1-A3.
- Results from the cattle production are presented in Appendix Tables A6, A16-A17 and Figure A4.
- Results from the feeding stuff production are presented in Appendix Tables A19-A20.
- Results from the rendering plants are presented in Appendix Table A21.
- Results based on suspicion of diseases in pets, zoo animals and wild life are presented in Appendix Tables A23-A24.

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

- Results from the broiler production are presented in Appendix Tables A13-A14 and A18.
- Results based on suspicion of diseases in pets, zoo animals and wild life are presented in Appendix Table A24.

a) The Danish Veterinary and Food Administration (DVFA) is one authority but operates from more locations throughout the country. To be able to distinguish the locations the terms DVFA is used synonymous with the location in Glostrup and Food Control Office followed by the location synonymous with the location in guestion.



Pig and cattle carcasses are screened for *Mycobacterium* and *Echinococcus* during meat inspection at the slaughterhouse. Although swine kept under controlled housing conditions in Denmark are exempted from examination for *Trichinella* at slaughter, all slaughter pigs, sows and boars are still examined at slaughter. Free range pigs, horses, wild game (e.g. wild boar) and other species susceptible to *Trichinella* must still be tested. 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, and 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.

### 7.4 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 2016.

For further information consult the website of the Danish Veterinary and Food Administration, www.fvst.dk.

### 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, 2014-2016

	2016		2015		2014	
Source	Estimated no. of reported cases (95% credibility interval <sup>a</sup> )	Percentage of reported cases	Estimated no. of reported cases (95% credibility interval <sup>a</sup> )	Percentage of reported cases	Estimated no. of reported cases (95% credibility interval <sup>a</sup> )	Percentage of reported cases
Domestic pork	64 (46-91)	6,0	35 (16-60)	3.7	172 (126-214)	15.4
Domestic beef	17 (9-27)	1,6	13 (3-23)	1.4	25 (20-31)	2.2
Domestic table eggs	22 (1-55)	2,0	Oc	0	33 (22-47)	3.0
Domestic broilers	9 (1-21)	0,9	Oc	0	22 (1-69)	2.0
Domestic ducks	5 (0.2-17)	0,5	No data	-	No data	-
Imported pork	40 (12-70)	3,7	61 (35-86)	6.6	13 (0-44)	1.1
Imported beef	$O_P$	0	Oc	0	3 (0-7)	0.2
Imported broilers	43 (24-64)	4,0	27 (9-49)	2.9	33 (14-53)	2.9
Imported turkey	3 (0-9)	0,3	11 (1-28)	1.2	$O_{q}$	0
Imported duck	4 (0-11)	0,4	15 (5-27)	1.6	22 (11-34)	2.0
Travels	573 (566-579)	53,3	522 (516-528)	56.5	538 (528-549)	48.0
Unknown source	233 (196-267)	21,7	220 (185-254)	23.8	216 (178-252)	19.2
Outbreaks, unknown source	61	5,7	21	2.3	45	4.0
Total	1,074		925		1,122	

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

b) No samples from imported beef were found positive for Salmonella in 2016

c) No samples from domestic table egg layers, domestic broiler meat and imported beef were found positive for Salmonella in 2015

d) No samples from imported turkey meat were found positive for Salmonella in 2014.

Source: National Food Institute, Technical University of Denmark

### Human disease and outbreak data

Table A2. Zoonoses in humans, number of laboratory-confirmed cases, 2011-2016

	Incidence per 100,000 inhabitants	Reported r	no. of cases	;			
Zoonotic pathogen	2016	2016	2015	2014	2013	2012	2011
Bacteria							
Brucella abortus/melitensis <sup>a,d</sup>	-	3	6	4	4	2	7
Campylobacter coli/jejuni <sup>b</sup>	81.7	4,677	4,348	3,782	3,766	3,728	4,068
Chlamydia psittaci <sup>b</sup>	0.4	24	25	16	12	12	7
<i>Leptospira</i> spp.⁵	0.2	10	5	10	3	7	11
Listeria monocytogenes <sup>b</sup>	0.7	39	43	92	50	50	49
Mycobacterium bovis⁵	0.03	2	1	1	0	0	1
Salmonella total <sup>b</sup>	18.8	1,074	925	1,122	1,136	1,198	1,166
S. Enteritidis <sup>b</sup>	4.3	246	258	268	346	242	293
S. Typhimurium <sup>b,c</sup>	5.6	300	233	427	337	415	386
Other serotypes <sup>b</sup>	8.9	508	434	427	453	541	487
VTEC total <sup>b</sup>	4.7	269	228	248e	186	190	224
0157	0.6	37	33	37	23	36	27
Other O-groups or non-typeable	3.6	204	195	192	163	154	197
Yersinia enterocolitica <sup>b</sup>	10.0	573	539	432	345	291	224
Viruses							
Lyssavirus <sup>b</sup>		0	0	0	0	0	0

a) Not notifiable, hence the incidence cannot be calculated.

Source: Statens Serum Institut.

Table A3. VTEC O-group distribution in humans<sup>a</sup>, 2016

0-group	Number of episodes	O-group	Number of episodes
0157	37	063	6
0103	22	0113	6
0146	20	0154	5
026	15	0121	5
027	12	Notification <sup>b</sup>	65
0128	8	Other O-groups or not-typed	55
0145	6	Isolate not available but presence of vtx genes confirmed by PCR	1
091	6		
Continued in the nex	xt column	Total	269

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

Source: Statens Serum Institut.

b) Notifiable.

c) S. Typhimurium and the 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. 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) Includes 19 cases verified by PCR only (see Table A3).

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

Table A4. Food- and waterborne disease outbreaks<sup>a</sup> reported in the Food- and waterborne Outbreak Database (FUD) (n=49), 2016

Pathogen	No. of patients	Patients labora- tory confirmed	Setting	Source	FUD no.
Bacillus cereus	50		Restaurant	Composite meal	1509
Campylobacter	4	4	Private party	Chicken	1552
Campylobacter jejuni, ST19	8	8	Regional	Unknown	1519
Campylobacter jejuni, ST48	103	19	Catering	Duck meat (Imp)	1513
Clostridium perfringens	21		Private party	Composite meal	1486
Clostridium perfringens	68		Catering	Composite meal	1506
Clostridium perfringens	16		Catering	Composite meal	1512
Clostridium perfringens	186		Catering	Composite meal	1522
Clostridium perfringens	9		Private party	Composite meal	1541
Clostridium perfringens	15		Catering	Composite meal	1547
Clostridium perfringens	38		Sport event	Composite meal	1551
Histamin	2	1	Shop	Blue cheese	1485
L. monocytogenes ST6	3	3	Regional	Unknown	1574
L. monocytogenes ST4	7	7	National	Cold cuts of meat	1525
_ectines / cyanogenic glycosides	15		Canteen	Elderberries (raw)	1537
Norovirus	3		National	Oysters	1484
Norovirus	58	3	National	Oysters (Imp)	1497
Norovirus	412	28	National	Lettuce (Imp)	1500
Norovirus	23	6	Canteen	Buffet meal	1502
Norovirus	25	5	Restaurant	Buffet meal	1503
Norovirus	8	1	Private party	Buffet meal	1507
Norovirus	28	6	Restaurant	Buffet meal	1508
Norovirus	34	3	Canteen	Sandwiches	1510
Norovirus	13		Restaurant	Lettuce (Imp)	1534
Norovirus	284	14	Canteen	Buffet meal	1536
Norovirus	103	7	Restaurant	Buffet meal	1538
Norovirus	31	9	Restaurant	Composite meal	1539
Norovirus	6		Shop	Open sandwiches	1542
Norovirus	31	4	Restaurant	Oysters (Imp)	1549
Norovirus	20	3	Private party	Cake	1560
Norovirus	22		Restaurant	Buffet meal	1561
Norovirus	32	4	Catering	Open sandwiches	1565
Norovirus	45		Canteen	Buffet meal	1578
Salmonella Enteritidis, MLVA0051 <sup>d</sup>	6	6	Restaurant	Unknown	1531
Salmonella 0:4,12;H:i:-, MLVA1776	5	5	National	Unknown	1548
Salmonella 0:4,5,12;-:-, MLVA0126	16	16	Regional	Pork meat	1521
Salmonella 0:4,5,12;H:i:-, MLVA0338b	12	12	International	Dried spicy snack sausage	1504
Salmonella 0:4,5,12;H:i:-, MLVA0478b	9	9	National	Unknown	1515
Salmonella 0:4,5,12;H:i:-, MLVA0617 <sup>b,e</sup>	5	5	National	Composite meal	1558
Salmonella Reading	4	4	National	Unknown	1527
Salmonella Szentes	3	3	National	Unknown	1514
	6	6	National	Unknown	1526
Salmonella Typhimurium, MLVA0642	U	O	racional	OTIKITOWIT	1310

Table A4. Food- and waterborne disease outbreaks<sup>a</sup> reported in the Food- and waterborne Outbreak Database (FUD) (n=49), 2016 (Continued from previous page)

Pathogen	No. of patients	Patients laboratory confirmed	Setting	Source	FUD no.
VTEC 0103:H2, ST17	6	6	National	Unknown	1511
VTEC 0121:H19, vtx2a, eae, ehxA, ST655	5	2	Farm	Unknown	1520
VTEC 0157:H7, vtx1a, vtx2a, eae, ehxA, ST11	2	2	Regional	Unknown	1528
VTEC 0157:H7, vtx1a, vtx2c, ST11	6	6	Regional	Beef/Kebab <sup>c</sup>	1530
Outbreaks related to travel					
Salmonella enteritidis, MLVA0004	9	9	Travel (Greece)	Unknown	1529
Salmonella enteritidis, MLVA0121	5	5	Travel (Hotel, Turkey)	Unknown	1517
Total	1.825	234			

Note: (imp)= imported product.

Source: Food- and waterborne Outbreak Database (FUD).

Table A5. Outbreaks reported in 2015 but where additional patients were reported in 2016

Pathogen	No. of patients	Patients laboratory confirmed	Setting	Source	FUD no.
Salmonella 0:4,5,12;i:-, MLVA0479	4	4	National	Unknown	1487
Total	4	4			

a) ST=MLST Sequence Type.

b) MLVA profiles for the most common human MLVA-types can be found in tables A6, A7 and A8.

c) The beef/kebab was produced in Denmark with raw material of multiple origins.

d) This outbreak strain clusters with the strains found in an international outbreak related to eggs of Polish origin. However the source of the outbreak in Denmark could not be correlated with Polish eggs, egg-products or poultry meat.

e) Only patients from 2016 is reported for this outbreak. The outbreak was not concluded at the time of reporting and additional cases will be reported in 2017.

### Monitoring and surveillance data

Table A6. Top 15 (humans) serotype distribution (%) of Salmonella from humans, animals, carcasses, Danish and imported meat, 2016. N=number of culture positive units<sup>a</sup>

	Human	Pig⁵	Pork	Beefd	Broilere	Layere	Ducksf	Imported	meat (ba	atches)	
	cases	animals	batches	batches	flocks	flocks	flocks	Porkg	Broilerg	Ducksf	Turkeyg
Serotype	N=1074	N=129	N=107	N=11	N=21	N=3	N=24	N=20	N=29	N=5	N=9
Enteritidis	22.9	0	0	0	0	0	0	0	27.6	40.0	0
1,4,[5],12:i:-	17.9	20.9	36.4	0	71.4	66.7	0	65.0	0	0	0
Typhimurium	10.1	22.5	11.2	0	4.8	0	25.0	10.0	0	20.0	0
Stanley	3.9	0	0	0	0	0	0	0	0	0	0
Newport	2.7	8.0	0	0	0	0	75.0	0	0	40.0	0
Java	2.5	0	0	0	0	0	0	0	0	0	0
Infantis	2.1	8.0	2.8	0	4.8	0	0	0	27.6	0	0
Dublin	1.9	0	0	72.7	0	0	0	0	0	0	0
Kentucky	1.8	0	0	0	0	0	0	0	0	0	0
Saintpaul	1.8	0	0.9	0	0	0	0	0	0	0	0
Agona	1.4	0	0	0	0	0	0	0	0	0	0
Virchow	1.2	0	0	0	0	0	0	0	3.4	0	0
Bovismorbifi- cans	1.1	0	0	0	0	0	0	0	0	0	0
Derby	1.1	48.8	32.7	9.1	9.5	33.3	0	20.0	0	0	0
Poona	1.1	0	0	0	0	0	0	0	0	0	0
Other	23.6	6.2	5.6	9.1	9.5	0	0	5.0	41.4	0	100
Unknown	2.9	0	10.3	9.1	0	0	0	0	0	0	0
Total	100	100	100	100	100	100	100	100	100	100	100

a) One isolate per serotype per unit is included, thus the number of isolates may exceed the number of units. Thus, in 2015 more isolates were included from broiler flocks.

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

b) Isolates collected from coecum samples taken randomly at slaughter. Where more than one *Salmonella* positive pig with different serotypes was randomly selected from a herd, one pig per serotype was included.

c) Sampling of pork carcasses at slaughterhouses according to the surveillance programme (Table A38).

d) Sampling of beef carcasses at slaughterhouses according to the surveillance programme (Table A37).

e) Sampling of production flocks prior to slaughter according to surveillance programmes (Tables A34).

f) Centrally coordinated study (see section 7.4 and Table A28 for description).

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

Table A7. Top 10 (humans) MLVA<sup>i</sup> distribution (%) of Salmonella Typhimurium including the monophasic S. 1,4,[5],12:i:-from humans, animals, carcasses and danish and imported meat, 2016. N= number of isolates

MLVA type <sup>i</sup>	Human	Pork <sup>c</sup>	Layere	Meat (batch	Meat (batches)		
	cases	batches	flocks	Pork (imp)g	Pork (DK)g	Ducks (imp)f	Ducks (DK)f
STTR 9 5 6 10 3	N=294	N=50	N=2	N=15	N=9	N=6	N=24
3 14 10 NA 211	6.1	0	0	0	0	0	0
3 12 9 NA 211	5.4	0	0	0	0	0	0
3 12 8 NA 211	5.1	0	0	0	0	0	0
3 11 10 NA 211	4.1	0	0	0	0	0	0
3 14 8 NA 211	3.1	0	0	0	0	0	0
3 13 9 NA 211	3.1	0	0	0	0	0	0
3 15 9 NA 211	2.7	0	0	0	0	0	0
3 11 9 NA 211	2.7	0	0	0	0	0	0
3 12 10 NA 211	2.7	0	0	0	0	0	0
Other	61.6	100	100	100	100	100	100
Total	100	100	100	100	100	100	100

For footnotes a-g see Table A6.

Table A8. Top 10 (humans) MLVA<sup>i</sup> distribution (%) of Salmonella Enteritidis from humans and imported meat, 2016. N= number of isolates

MLVA type <sup>i</sup>	Human cases	Broiler meat batch (imp) <sup>g</sup>	Duck meat batch (Imp) <sup>f</sup>
SE 1 5 2 9 3	N= <sup>j</sup> 240	N=8	N=2
3 10 7 2 2	16.3	12.5	0
4 10 5 3 1	10.8	50.0	0
4 9 5 3 1	8.8	0	0
4 11 5 3 1	8.3	0	0
3 11 7 2 2	4.6	0	50.0
3 10 8 2 2	3.8	0	0
4 11 4 3 1	3.3	0	0
3 9 7 2 2	2.5	0	0
5 10 8 2 2	2.5	0	0
4 10 4 3 1	2.5	0	0
Other	36.7	37.5	50.0
Total	100	100	100

For footnotes a-g see Table A6.

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

i) The isolates are analysed for the following loci: STTR9|STTR5|STTR6|STTR10|STTR3 and the results are reported in the same order in the table. "NA" = locus missing.

j) In total, 320 human cases of *S*. Typhimurium was reported in 2016 (Table A2), only 294 isolates were analysed for MLVA type. Source: Danish Veterinary and Food Administration, Statens Serum Institut, and National Food Institute.

i) The isolates are analysed for the following loci: SE1|SE5|SE2|SE9|SE3 and the results are reported in the same order in the table .

j) In total, 246 human cases of *S*. Enteritidis was reported in 2016 (Table A2), only 240 isolates were analysed for MLVA type.

Table A9. Occurrence of Salmonella in the table egg production, 2006-2016

		ng period <sup>b</sup> nt flocks)	Adult period <sup>c</sup> (parent flocks)		Pullet-r		Table egg la	Table egg layer flocks	
	N	Positive	N	Positive	N	Positive	N	Positive	
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	359	3	
2013	10	0	7	0	173	0	373	4	
2014	22	0	8	0	150	0	347	2	
2015	15	0	8	0	123	0	344	0	
2016	15	0	10	0	132	0	426	3 <sup>d</sup>	

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

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

Table A10. Occurrence of Salmonella in the table egg layer flocks sorted by type of production, 2006-2016

	Dee	Deep litter		Free range		rganic	Battery	
	N	Positive	N	Positive	N	Positive	N	Positive
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	136	1	131	1
2013	108	0	37	1	137	3	94	0
2014	97	0	30	0	125	1	95	1
2015	108	0	29	0	172	0	86	0
2016	125	<b>1</b> ª	31	0	196	<b>1</b> <sup>b</sup>	74	<b>1</b> <sup>c</sup>

a) S. Derby (1).

 $Source: Danish\ Agriculture\ and\ Food\ Council,\ and\ Danish\ Veterinary\ and\ Food\ Administration.$ 

b) Salmonella was not detected in grandparent flocks during rearing period (3 flocks).

c) Salmonella was not detected in grandparent flocks during adult period (4 flocks).

d) S. Derby (1), S. 4.5.12:i:- (2).

b) S. 4.5.12:i:- (1).

c) S. 4.5.12:i:- (1).

Table A11. Occurrence of Salmonella in the broiler production<sup>a</sup>, 2006-2016

		Rearing period <sup>b</sup> (parent flocks)		Adult period <sup>c</sup> (parent flocks)		Broiler flocks		hterhouse s/batches)
	N	Positive	N	Positive	N	Positive	N	Positive
2006	190	0	282	5	3,621	71	875	17
2007	152	0	258	3	3,703	60	884	10
2008	146	0	293	2	3,845	43	518 <sup>d</sup>	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	3,448	27	368	0
2013	128	0	152	1	3,498	34	288	0
2014	121	2	131	3	3,470	26	277	4
2015	91	0	289	1	3,631	23	148	0
2016	184	0	182	3e	3,606	21 <sup>f</sup>	203	<b>1</b> g

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

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

Table A12. Occurrence of Salmonella in turkey and duck flocks, 2006-2016

, ,						
	Turkey flocks <sup>a</sup>					
	N	% pos				
2006	11	0				
2007	13	0				
2008	10	10.0				
2009	15	0				
2010	24	4.2				
2011	38	2.6				
2012	23	0				
2013	56	3.6				
2014	10	0				
2015	80	1.3				
2016	76	0				

a) See Table A36 for description of the surveillance programme for turkey flocks. The major turkey slaughterhouse in Denmark closed down in 2004. Therefore, most commercially reared turkey flocks are transported abroad for slaughter.

Source: Danish Veterinary and Food Adnimistration

b) Salmonella was not detected in grandparent flocks during rearing period (12 flocks).

c) Salmonella was not detected in grandparent flocks during adult period (6 flocks).

d) 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.

e) S. Give (1), S.Derby (1), S.Stourbridge (1).

f) S. Typhimurium (1), S. 4.5.12:i: - (11), S. 1,4,5,12:i:-DT193 (4), S.Infantis (1), S. Bispeblerg (1), S.Derby (2), S. Onderstepoort (1).

g) S. Newport (1).

Table A13. Occurrence of Campylobacter in broiler flocks, 2006-2016<sup>a</sup>

	Cloacal swabs at s	laughter	Sock samples at fai	rm
	N (Flocks)	% pos	N (Flocks)	% pos
2006	4,522	30.8	-	-
2007	4,527	26.8	-	-
2008	4,950	26.3	-	-
2009	4,591	29.4	-	-
2010	-	-	3,132	16.5
2011	-	-	3,379	14.4
2012	-	-	3,376	11.6
2013	-	-	3,508	13.1
2014	3,474	27.7	-	-
2015	3,274	19.6	-	-
2016	3,184	21.8	-	-

a) See Tables A33 for description of the surveillance programmes. In 2014 the sampling method changed back from boot swabs collected in the stable 7-10 days before slaughter to cloacal swabs at slaughter according to Regulation no. 1512 of 13/12/2013.

Source: Danish Agriculture and Food Council and National Veterinary Institute (until 2009).

Table A14. Occurrence of Campylobacter in non-heat treated broiler meat at slaughter and retail<sup>a</sup>, 2012-2016

		Chilled bro	iler meat (sa	amples)			
		At slaught	er	At retail			
		Denmark		Denmark		Im	port
		N	% pos	N	% pos <sup>b</sup>	N	% pos⁵
2012	Conventional	1,044 <sup>c</sup>	21.5	-	-	-	-
	Organic/free-range	-	-	-	-	-	-
	In total	-	-	521	9.7	154	28.2
2013	Conventional	870 <sup>d</sup>	28.2	849	12.1	170	12.8
	Organic-free-range	93⁴	90.3	35	42.9	38	71.1
	In total	-	-	884	17.8	208	31.9
2014	Conventional	927	25.7	-	-	-	-
	Organic/free-range	108	75.0	-	-	-	-
2015	Conventional	960	20.1	-	-	-	-
	Organic/free-range	115	78.2	-	-	-	-
2016	Conventional	999	21.3	1,339	12.8	232	37.9
	Organic/free-range	117	87.2	93	71.0	245	78.8
	In total	-	-	1,432	17.4	477	57.5

a) Centrally coordinated studies (see Table A28 and section 7.4 for description). Limit of quantification: 10 cfu/g.

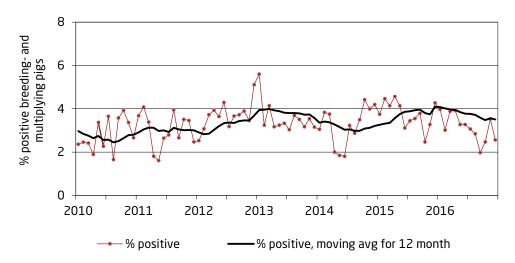
b) The prevalence is calculated as a mean of quarterly prevalences., except organic/free-range results.

c) Included are 238 leg-skin samples, prevalence = 24,4%.

d) Leg-skin samples only.

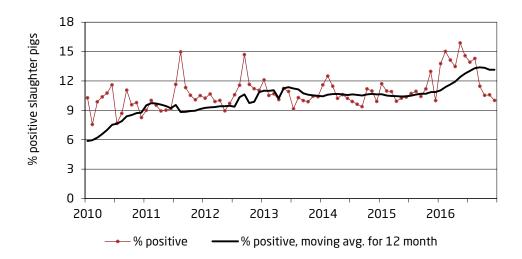
Source: National Food Institute.

Figure A1. Serological surveillance of Salmonella in breeding and multiplying pigs<sup>a</sup> based on monthly testing of blood samples, 2010-2016



a) For more information about the surveillance programme, see Table A38. Source: Danish Agriculture and Food Council.

Figure A2. Serological surveillance of Salmonella in slaughter pigs°, 2010-2016. Percentage of seropositive meat juice samples (first sample per herd per month)<sup>b</sup>



a) For more information about the surveillance programme, see Table A38.

b) The peaks in January 2010 and August 2011 were due to data transfer problems. The reason for the increase in late summer 2012 is unknown. Source: Danish Agriculture and Food Council.

Table A15. Occurrence of zoonotic pathogens in pigs and pork in Denmark, 2016

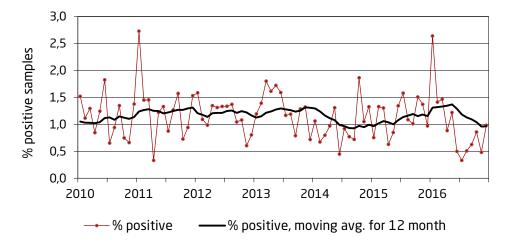
	Herds		Animals/Sample	<u> </u>	
Zoonotic pathogen	N	Pos		Pos	% pos
At farm					
Brucella abortusº	-	-	32,702	0	-
Leptospira sppsuspicion <sup>b</sup>	57	2	65	2	-
Leptospira bratislava <sup>c</sup>	100	90	-	-	
At slaughterhouse (slaughter pigs)					
Salmonella spp. <sup>d,e</sup>	6,313	337 <sup>f</sup>		-	-
Salmonella spp.dg (slaughtering >30.000 pigs/year)	-	-	17,950	-	$0.98^{h}$
Salmonella spp. $^{\rm dg}$ (slaughtering 1.000 or more and less than 30.000 pigs/year)	-	-	378	-	0.26
Salmonella spp. <sup>d,i</sup>	-	-	707	129	18.2
Trichinella spp. <sup>j</sup>	-	-	17,751,135	0	-
Mycobacterium bovish	-	-	17,843,548	0	-
Echinococcus granulosis/multilocularis	-	-	17,843,548	0	-

a) 5-8 ml blood samples were analysed using either the SAT, RBT or ELISA methods.

- d) See Table A38 for description of the Salmonella surveillance programme.
- e) Data are from December 2016. Slaughter pig herds monitored using serological testing of meatjuice samples collected at slaughter.
- f) Includes herds belonging to Salmonella level 2 and 3 only (See Table A38).
- g) Swab samples from four designated areas of the half-carcass were collected at the slaughterhouse after min. 12 h chilling. Sample size is 4x100 cm<sup>2</sup>. Samples from five animals were pooled, at slaughterhouses where 30.000 pigs or more were slaughtered per month. In slaughterhouses slaughtering 30.000 pigs or less per year, samples were analysed individually.
- h) 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.
- i) Coecum samples are randomly collected from slaughter pigs at slaughter.
- j) Samples collected from slaughter pigs at slaughter were examined using the method described in Directive 2075/2005/EEC. In 2014, an amendment to EU regulation (EC) No 2075/2005 came into force stating that slaughter pigs, sows and boars kept under "controlled housing conditions" in Denmark are extempted testing for *Trichinella*. Free range pigs must be tested for *Trichinella*.
- h) Slaughter pigs were examined by meat inspectors at slaughter.

Source: Danish Veterinary and Food Administration, National Veterinary Institute and National Food Institute, Technical University of Denmark.

Figure A3. Salmonella in pork, monitored at slaughterhouses, 2010-2016



a) For more information about the surveillance programme, see Table A38. Source: Danish Veterinary and Food Administration.

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) Serological analyses were performed for L. bratislava, L. grippotyphosa, L. hardjo, L. tarassovi, L. icterohaemorrhagiae, L. pomona and L. sejroe. Antibodies were only detected against L. bratislava (MAT titer >= 40). In addition, 223 herds (10.260 samples) was analysed for L. pomona in connection with export. All were negative.

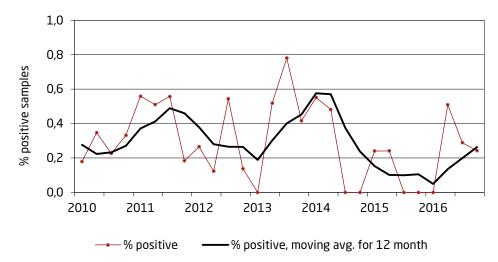
Table A16. Occurrence of zoonotic pathogens in cattle and beef in Denmark, 2016

	Animals/Sample	S	
Zoonotic pathogen	N	Pos	% pos
At farm			
Brucella abortus <sup>a</sup>	2,324	0	-
Mycobacterium bovis <sup>b, c</sup>	2,324	0	-
Coxiella burnetii	130 <sup>d</sup>	10	-
At slaughterhouse			
Salmonella spp.e(slaughtering >=7.500 cattle/year)	7,175	-	0.23 <sup>f</sup>
Salmonella spp.e (slaughtering 250 or more and 7.500 or less cattle/year)	269	-	0.37
Mycobacterium bovis <sup>b, g</sup>	539,600	0	-
Echinococcusus granulosis/multilocularis <sup>a</sup>	539,600	0	-

a) Denmark has been declared officially brucellosis free since 1979. The last outbreak was recorded in 1962. 5-8 ml blood samples were analysed using either the SAT or CFT methods.

Source: Danish Veterinary and Food Administration, National Veterinary Institute, and National Food Institute, Technical University of Denmark.

Figure A4. Salmonella in beef, monitored at slaughterhouses<sup>a</sup>, 20010-2016



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

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 interdermal 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) Samples analysed using an ELISA method.

e) See Table A37 for description 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 at slaughterhouses slaughtering 7,500 or more cattle per year. At slaughterhouses slaughtering 250 or more and less than 7,500 cattle per year, samples were analysed individually.

f) 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.

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

Table A17 Cattle herds in the Salmonella Dublin surveillance programme<sup>a</sup>, December 2016

			Non-milk producing h	erds	Milk produci herds	ng
Salmonell	<i>a</i> Dublin le	evel	N	%	N	%
Level 1		On the basis of milk samples	-	-	2,850	91.9
		On the basis of blood samples	13,761	97.3	-	-
	Total	Probably Salmonella Dublin free	13,761	97.3	2,850	91.9
Level 2		Titer high in blood- or milk samples	119	8.0	195	6.3
		Contact with herds in level 2 or 3	155	1.1	26	8.0
		Other causes	93	0.7	16	0.5
Level 3		Salmonellosis, official supervision	9	0.1	15	0.5
	Total	Non <i>Salmonella</i> Dublin free	376	2.7	252	8.1
Total num	ber of her	ds	14,137	100	3,102	100

a) See Table A37 for description of the surveillance programme.

Source: Seges.

Table A18 Results from the intensified control of Salmonella and Campylobacter in fresh meat based on case-by-case risk assessments, 2016

		Batches tested	No. of batches positive	No. of batches deemed unsafe based on a risk assessment	Batches de- emed unsafe based on other criteria <sup>a</sup>	Mean prevalence in positiv batches	Mean relative human risk in batches <sup>bc</sup>
Campylobacte	r						
Danish	Broiler	122	32	1	-	35.2 <sup>d</sup>	3.5 (n=17)
Imported	Broiler	154	87	7	-	36.9 <sup>d</sup>	5.4 (n=43)
Salmonella							
Danish	Pork	151	14	1	-	44.8 <sup>e,c</sup>	9.5 (n=6)
	Broiler	98	0	0		-	-
Imported	Pork	144	10	2	-	25 <sup>e,c</sup>	27.5 (n=3)
	Broiler	130	12	-	3	45 <sup>f</sup>	8.5 (n=2)
	Turkey	25	3	-	0	60 <sup>f</sup>	-

a) Microbiological criteria specified in regulation (EC) No 2073/2005 as amended. For Danish broiler meat there is a zero-tolerance for Salmonella and all positive batches must be heat treated before being put on the marked (Order no. 1512 of 13/12/2013).

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

b) 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.

c) Only batches subjected to risk assessment (n) have been included.

d) The Campylobacter prevalence in each batch of broiler meat and turkey meat is based on the proportion of positive samples (12 samples per batch). Include all positive batches.

e) The Salmonella prevalence in each batch of pork is based on the proportion of positive pooled samples (1-5 subsamples per pool, 10 pools per batch). Includes all positive batches send to risk assessment.

f) The Salmonella prevalence in each batch of broiler meat and turkey meat is based on the proportion of positive samples (5 samples per batch). Include all positive batches.

Table A19. Feed business operators own sampling of Salmonella in compound feeds, feed processing and feed material (batch-based data), 2014-2016

	2016		2015		2014	
	N	Positive	N	Positive	N	Positive
Feed processing plants (process control) <sup>a</sup> :						-
Ordinary inspections - clean zone	7,062	9 <sup>d</sup>	7,307	6	7,557	17
Ordinary inspections - unclean zone	10,009	30e	602	29	456	63
Compound feed, farm animals	700	0	1,148	1	858	0
Feed materials, farm animals <sup>b</sup>	1,386	13 <sup>f</sup>	1,416	17	1,656	28
Transport vehicles, clean zone/hygiene samples <sup>c</sup>	1,166	<b>1</b> g	1,190	5	1,143	1
Transport vehicles, unclean zone/hygiene samples <sup>c</sup>	144	4 <sup>h</sup>	63	10	235	7

Note: Data are from one feed and grain trade organisation only, representing a proportion of feed at the Danish market.

- a) Presence of Salmonella in compound feed is indirectly monitored by environmental samples collected during feed processing.
- b) Predominantly poducts of soy (e.g. soybean meal) but also products of rape (e.g. rapeseed cake) and sunflower (e.g. sunflower meal).
- c) Samples from transport vehicles (hygiene samples) prior to loading of feed compounds.
- d) S. Havana, S. Idikan, S. Senftenberg, S. Putten, S. Mbandaka, S. Falkensee
- e) S. Putten, S. Havana, S. Senftenberg, S. Agona, S. Mbandaka, S. Rissen, S. Idikan, S. Kralingen, S. 13.23, S. Falkensee, S. Infantis, S. 4.5.12:i
- f) S. 6.7:r:-, S. Mbandaka, S. Agona, S. Amsterdam, S. Derby, S. Idikan, S. Senftenberg, S. Oranienburg
- g) S. Putten.
- h) S. Putten.

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

Table A20. Control of Salmonella in compound feeds, feed processing and feed material (batch-based data), 2013-2016

	2016		2015		2014		2013	
	N	Positive	N	Positive	N	Positive	N	Positive
Feed processing plants (process control) <sup>a</sup> :								
Ordinary inspections <sup>b</sup>	278	<b>7</b> <sup>d</sup>	319	17 <sup>d</sup>	402	10	333	7
Feed materials, farm animals <sup>c</sup>	64	1e	71	3e	90	4	99	2

a) Presence of Salmonella in compound feed is indirectly monitored by environmental samples collected during feed processing. Companies are sampled one to four times per year.

- b) Primarily findings of *Salmonella* in the unclean zone.
- c) Predominantly soybean meal and rapeseed cake.
- d) S. Infantis (2), S. Putten (1), S. Havana (2), S. 4.12:b:- (1), S. Rissen (1).
- e) S. Infantis (1).

Source: Danish Veterinary and Food Administration.

Table A21. Salmonella in three categories of meat and bone meal by-products not intended for human consumption, 2016

Category	of	Own-che	ck samples	Product samples		
processing	g plant	N	Positive	N	Positive	
1+2	By-products of this material cannot be used for feeding purposes	520	9	98	2	
2	By-product of this material may be used for feed for fur animals	50	0	14	0	
3	By-products from healthy animals slaughtered in a slaughterhouse. Products of these may be used for petfood <sup>b</sup> and for feed for fur animals	520	7	1,151	0	
	Total					

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

Source: Daka Denmark A/S.

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

Table A22. Pathogens in batches<sup>a</sup> of ready-to-eat vegetables <sup>b,c</sup>2016

	Campylo	bacter
Type of sample	N	Pos
Vegetables <sup>c</sup>	961	3 <sup>d</sup>

a) Five samples per batch.

Source: Danish Veterinary and Food Administration.

Table A23. Occurrence of zoonotic pathogens in pets and zoo animals in Denmark<sup>a</sup>, 2016

	Pet	animal	S				Zoo	animals		
	Dog	gs	Cats		Othe	ers	Mamı reptil	nals & es	Birds	
Zoonotic pathogen	N	Pos	N	Pos	N	Pos	N	Pos	N	Pos
Salmonella spp.	1	0	2	0	<b>1</b> <sup>b</sup>	0	<b>4</b> <sup>c</sup>	2 <sup>de</sup>	8 <sup>f</sup>	0
Chlamydia psittaci	2	0	0	-	3g	0	$2^h$	0	0	-
Cryptosporidium spp.	2	0	0	-	3 <sup>i</sup>	0	2 <sup>J</sup>	0	0	-
Lyssavirus (classical)	1	0	3	0	$1^{k}$	0	0	-	0	-
European Bat <i>Lyssavirus</i>	1	0	3	0	<b>1</b> <sup>k</sup>	0	0	-	0	-

a) All samples are analysed based on suspicion of disease, and does not reflect the country prevalence.

Source: National Veterinary Institute, Technical University of Denmark, and Danish Veterinary and Food Administration.

b) Centrally coordinated study (See section 7.4 for description) to control and investigate Campylobacter in Danish and imported ready-to-eat vegetables.

c) In 2016 only Danish and imported salads were analysed.

d) 1 Lactuca sativa and 2 Lactuca sativa var. crispa 'Lollo rossa' were positive with Campylobacter jejuni.

b) Pool from 3 pet rats.

c) Lesser mouse-deer 1265 (1), leopard gecko(1), red-footed tortoise (1), corn snake(1).

d) Leopard gecko (1), corn snake (1).

e) S. Enteritidis (1), S. Poano (1).

f) Doves (2), penguins (3), ibis (1), cockatiel (1), budgerigar (1).

g) Rabbits (3).

h) Ring-tailed lemur (1), southern pig-tailed macaque (1).

i) Rabbits (3).

j) Ring-tailed lemur (1), southern pig-tailed macaque (1).

k) Cattle (1).

Table A24. Occurrence of zoonotic pathogens in wild and farmed wildlife in Denmarka, 2016

	Farmed wi	ldlife			Wildlife			
	Wild boar		Minks & o	chincillas	Mammals		Birds	
Zoonotic pathogen	N	Pos	N	Pos	N	Pos	N	Pos
Salmonella spp.	26	0	0	0	7 <sup>b</sup>	0	<b>1</b> <sup>c</sup>	0
Campylobacter spp.	0	-	0	-	0	-	0	-
Chlamydia psittaci	0	-	0	-	0	-	10 <sup>d</sup>	0
Cryptosporidium spp.	0	-	0	-	68e	6 <sup>f</sup>	0	-
Echinococcus multilocularis	0	-	0	-	26 <sup>g</sup>	$2^h$	0	-
Trichinella spp <sup>i</sup>	589	0	34	0	48 <sup>j</sup>	0	0	-
Lyssavirus (classical)	0	-	0	-	5 <sup>k</sup>	0	0	-
European Bat Lyssavirus	0	-	0	-	5 <sup>k</sup>	0	0	-

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

Source: National Veterinary Institute, Technical University of Denmark, and Danish Veterinary and Food Administration.

b) Badgers (7).

c) Great cormorant (1).

d) Doves (9), great cormorant (1).

e) Bisons (4), fallow deer (1), red deers (7), raccoon dogs (9), roe deers (47).

f) Fallow deer (1), raccoon dog (1), roe deers (4).

g) Raccoon dogs (17), foxes (9).

h) Foxes (2).

i) In 2014, an amendment of EU regulation (EC) No 2075/2005 came into force stating that slaughter pigs, sows and boars kept under "controlled housing conditions" in Denmark are extempted testing for *Trichinella*. Free range pigs, horses and wild game and toher species susceptible to *Trichinella* must be tested.

j) Dolphin (1), raccoon dogs (15), foxes (28), harbour seal (1), porpoises (3).

k) Bats (5).

Table A25. The Bovine Spongiform Encephalopathy (BSE) surveillance programme<sup>a</sup> for cattle, 201

Table N.23. The bovine Spongijonn encephalopatry (b3c) surveinance	programme joi cattic, Le	71
Type of surveillance	N <sup>b</sup>	Positive
Active surveillance		
Healthy slaughtered animals	21	0
Risk categories:		
Emergency slaugthers	1,307	0
Slaughterhouse antemortem inspection revealed suspicion or signs of disease	0	-
Fallen stock	19,367	0
Animals from herds under restriction	0	-
Passive surveillance		
Animals suspected of having clinical BSE	<b>1</b> <sup>c</sup>	0
Total	20,696	0

a) According to the EU Regulation (EC) 999/2001 as amended, Commission Decision 2009/719/EC as amended and Danish Order no. 878 of 01/07/2013 as amended.

Table A26. The Transmissible Spongiform Encephalopathy (TSE) surveillance programme<sup>a</sup> for sheep and goats, 2016

Type of surveillance	N <sup>b</sup>	Positive
Active surveillance		
Fallen stock (>18 months)	812	<b>1</b> <sup>c</sup>
Animals from herds under restriction	0	-
Passive surveillance		
Animals suspected of having clinical TSE	0	-
Total	812	1°

a) According to the EU Regulation (EC) 999/2001 as amended, Commission Decision 2009/719/EC as amended and Danish Order no. 1288 of 20/12/2011 as amended.

Source: National Veterinary Institute, Technical University of Denmark, and Danish Veterinary and Food Administration.

Table A27. Distribution (%) of prion protein genotype of sheep randomly selected, 2016

	Genotype	Sheep n=100
NSP 1	ARR/ARR	36.0
NSP 2	ARR/AHQ, ARR/ARH, ARR/ARQ	15.0
NSP 3 (ARQ/ARQ)	ARQ/ARQ	28.0
NSP 3 (Other)	AHQ/AHQ, AHQ/ARH, AHQ/ARQ, ARH/ARQ, ARH/ARH, ARQ/ARH, ARH/AHQ, ARQ/AHQ	13.0
NSP 4	ARR/VRQ	0
NSP 5	ARH/VRQ, ARQ/VRQ, VRQ/VRQ, AHQ/VRQ	8.0
Total		100.0

a) The genotypes were grouped in the NSP classification system according to their different susceptibility:

b) Samples (brain stem material) are tested using a IDEXX technique. Confirmatory testing is carried out using histopathology or immunohistochemistry. Further confirmation on autolysed material is performed at the European Union TSE reference laboratory.

c) In addition, one suspicious case was rejected without testing according to TSE Regulation 999/2001 as later amended, Article 12(1).

Source: National Veterinary Instistute, Technical University of Denmark, and Danish Veterinary and Food Administration.

b) Samples (brain stem material) are tested using a IDEXX technique. Confirmatory testing is carried out using histopathology or immunohistochemistry. Further confirmation on autolysed material is performed at the European Union TSE reference laboratory.

c) The Western immunoblotting profile of the samples from the sheep shows molecular migra-tion and banding ratios consistent with those seen in atypical scrapie. This result provides sufficient evidence to classify this case as POSITIVE ATYPICAL SCRAPIE. Genotype pending. All material analysed at the EU Community Reference laboratory for TSE, APHA, Weybridge, UK.

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, Technical University of Denmark, and Danish Veterinary and Food Administration.

Table A28.	Centrally	coordinated	studies	conducted	in	2016

<u>onducted in</u>	2016	
No. of planned samples	Pathogen surveyed	Further information
1000	Campylobacter spp.	Appendix Table A14
100	Campylobacter spp.	Appendix Table A14
2000	Campylobacter spp.	Appendix Table A14
640 batches	Campylobacter spp., Salmonella spp.	Appendix Table A18
1000	Campylobacter spp.	Appendix Table A22
880	Salmonella spp.	Appendix Tables A6 and A15
2500	Listeria monocytogenes, Salmo- nella spp., staphylococci, Esche- richia coli, aerobic plate count, Enterobacteriaceae.	Data are being processed <sup>a</sup>
300	ESBL, AmpC, carbapenemaseproducing <i>E. coli</i>	Data are being processed <sup>a</sup>
300	Enterobacteriaceae, E. coli, Enterococcus spp., ESBL, AmpC, carbapenemaseproducing E. coli	Data are being processed <sup>a</sup>
366	Campylobacter spp., Escherichia coli	Results are presented in the 2016 DANMAP report
300	ESBL, AmpC, carbapenemaseproducing <i>E. coli</i>	Results are presented in the 2016 DANMAP report
700	Escherichia coli, Campylobacter spp., Enterococcus faecalis, ESBL, AmpC, carbapenemase-producing E. coli	Results are presented in the 2016 DANMAP report
300	Salmonella spp., ESBL	Results are presented in the 2016 DANMAP report
100	Salmonella spp.	Data are being processed <sup>a</sup>
140	Listeria monocytogenes, Sal- monella spp., Escherichia coli, staphylococci	Data are being processed <sup>a</sup>
50	Listeria monocytogenes, Sal- monella spp., Escherichia coli, staphylococci	Data are being processed <sup>a</sup>
100	Norovirus (froszen strawberries)	Data are being processed <sup>a</sup>
100	Listeria monocytogenes, Sal- monella spp., Escherichia coli, staphylococci	Data are being processed <sup>a</sup>
50	Salmonella spp., Escherichia coli	Data are being processed <sup>a</sup>
100	Salmonella spp., Escherichia coli	Data are being processed <sup>a</sup>
480	Salmonella spp.	Data are being processed <sup>a</sup>
400	Listeria monocytogenes	Data are being processed <sup>a</sup>
Cor	ntinued on the next page	
	No. of planned samples 1000 1000 20000 8800 25000 3000 3000 3000 3000 1000 1400 1000 10	planned samples  1000

Table A28. Centrally coordinated studies conducted in 2016 (Continued from previous page)

Title of project	No. of planned samples	Pathogen surveyed	Further information
Listeria in the production environment	400	Listeria monocytogenes	Data are being processed <sup>a</sup>
MRSA in pork meat	300	MRSA	Results are published on the DVFA website www. fvst.dk (in Danish)
Sourrce of infection baseline for campylo- bacter infections	1350	Campylobacter spp.	Data are being processed <sup>a</sup>
Virus in frozen berries	200	Norovirus, hepattis A virus	Data are being processed <sup>a</sup>
Baseline Norovirus in oisters (continued in 2017).	4	Norovirus	Data will be proccesed together with 2017 data.

a) Results will be published on the DVFA website WWW.fvst.dk ( in Danish)

Table A29. Listeria monocytogenes in Danish produced ready-to-eat (RTE) foodso, 2016

				lysed by nethod <sup>b</sup>	y a		es analy tative m		a
		Batch	iesc	Single sampl		Batch	nesc	Single	
Food category	Sampling place	N	Pos	N	Pos	N	Pos	N	Pos
Bakery products	At processing	-	-	-	-	-	-	-	-
Egg and egg products	At processing	6	0	30	0	10	0	50	0
Cheese, RTE	At processing	23	0	115	0	20	0	100	0
Milk and dairy products excluding cheeses), RTE	At processing	25	0	125	0	15	0	75	0
Products made from broiler meat, RTE	At processing	-	-	-	-	1	0	5	0
Products made from turkey meat, RTE	At processing	-	-	-	-	-	-	-	-
Products made from pork, RTE	At processing	29	2	145	10	18	0	90	0
Products made from beef, RTE	At processing	2	0	10	0	8	0	40	0
Meatproducts, unspecified, RTE	At processing	7	0	35	0	8	0	40	0
Fruit, RTE	At processing	4	0	20	0	2	0	10	0
Vegetables, RTE	At processing	15	0	75	0	15	0	75	0
Fish and Fishery products, RTE	At processing	25	1	130	3	8	0	40	0
Shellfish and products thereoff, RTE	At processing	-	-	-	-	13	0	65	0
Other RTE products	At processing	53	2	265	2	78	1	390	5
Babyfood and food for special medical purpose, RTE	At processing	2	0	10	0	0	0	0	0
Environmental samples	At processing	-	-	841	95	-	-	-	-
Suspect productsamples, RTEd	At processing	58	5	306	8	48	1	252	1

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

Source: Danish Veterinary and Food Administration.

b) Listeria monocytogenes present in a 25 g sample of the product.

c) Five samples from each batch, analysed individually.

d) Several samples/batches have been analyzed by both a qualitative method and a quantitative method.

Source: Danish Veterinary and Food Administration.

Table A30. Status on targets for Campylobacter and Salmonella, 2016

National Action Plans	Target	Status
Campylobacter in broilers 2013-20	17ª	
Flocks at farm	20% reduction in prevalence of positive flocks in 2016 compared ot 2012	A reduction of 9.4% was obtained for the period 2011-2013 <sup>b</sup> A reduction of 25% was obtained for the period 2014-2016 <sup>b</sup>
Fresh meat at slaughterhouse	Reduction of the relative human risk (RR) compared to the level in 2013 <sup>b</sup> 2014: RR reduced by 25% 2016: RR reduced by 50%	A reduction of 28% was obtained in 2014 compared with 2013 <sup>c</sup> A reduction of 37% was obtained in 2016 compared to 2013 <sup>c</sup>
Salmonella in poultry <sup>d</sup>		
Laying hen flocks of <i>Gallus</i> gallus	Initially eradication, later a reduction strategy in the table egg production	3 positive flocks (Table A9-A10) Eggs from positive flocks are de- stroyed or heat treated
Carcasses at slaughterhouse	Initially eradication, later a reduction strategy in the broiler production Zero-tolerance in Danish broiler meat.	No positive batches (Table A11) Positive batches are heat treated
Salmonella in pigs 2014-2017		
Carcasses at slaughterhouse	Max. 1% <i>Salmonella</i> at carcass level in 2014-2017	1.0% (Table A15)
Estimated human cases from pigs	No considerable increase in the number of estimated human cases from pigs in the Danish <i>Salmonella</i> source account	In total 64 cases were attributed to Danish pork in 2016 (CI95%: 46-91) vs. 35 cases in 2015 (CI95%: 16-60). (Chapter 1 and Table A1)
Salmonella Dublin in cattle 2012-2	016	
Herds at farm	Eradication of S. Dublin in all herds by 2016., i.e. all herds in level $1^{\rm e}$ by the end of 2016	8,1% of milk-producing herds and 2,7% of non-milk producing herds are in level 2 or 3 (December 2016) (Table A17)
EU Regulations		
Regulation (EC) No. 1190/2012		
Breeding and fattening turkey flocks	Max. 1% positive for <i>S.</i> Enteritidis and <i>S.</i> Typhimurium <sup>e</sup>	No fattening flocks positive with target serovars (N=76). (Table A12)
Regulation (EC) No. 200/2010		
Breeding flocks of Gallus gallus	Max. 1% adult flocks positive for <i>S</i> . Typhimurium <sup>f</sup> , <i>S</i> . Enteritidis, <i>S</i> . Hadar, <i>S</i> . Infantis and <i>S</i> . Virchow	0% (no flocks) <sup>h</sup> (Table A9 and A11)
Regulation (EC) No. 1168/2006		
Laying hen flocks of <i>Gallus</i> gallus	MS specific targets, for Denmark: Max. 2% adult flocks positive for <i>S.</i> Typhimurium <sup>f</sup> and <i>S.</i> Enteritidis	0.5% (Table A9)
Regulation (EC) No. 646/2007		
Broiler flocks of <i>Gallus gallus</i>	Max. 1% positive <i>S.</i> Typhimurium <sup>e</sup> and <i>S.</i> Enteritidis	0.4% (16 flocks) positive with target serovars (Table A11)

a) The duration of the action plan has been prolonged to the end of 2017, to ensure that new knowledge from ongoing projects is included.

b) As a consequence of a change in sampling from AM testing 7-10 days before slaughter to cloacal swabs at the point of slaughter, 2013 and 2014 data cannot be compared. The target has therefore been changed to cover firstly the period 2011-2013 and secondly the period 2014-2016. For the two periods as a whole, the target is 20%.

c) 2013 is agreed as the baseline since 2012 data are not compareable with data from 2013 and onwards due to a nessessary improvent in the data collection.

d) Supplementary to EU-regulations.

e) See Table A37for explanation of the herd levels.

f) Including the monophasic strains S. 1,4,[5],12:i:-.

h) No positive flocks for S . Enteritidis, S . Hadar, S . Infantis ,S . Virchow and S . Typhimurium Including the monophasic strains S. 1,4,[5],12:i:-Source: Danish Veterinary and Food Administration.

## Monitoring and surveillance programmes

Table A31. Overview of notifiable and non-notifiable human diseases presented in this report, 2016

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

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.

Table A32. Overview of notifiable and non-notifiable animal diseases presented in this report, 2016

Patogen	Notifiable	EU legislation	Danish legislation
Bacteria			
<i>Brucella</i> spp. Cattle	1920 <sup>a</sup> OBF in 1979 <sup>b</sup>	Decision 2003/467/EC	Order no 305 of 3/5 2000
Sheep and goats	ObmF in 1995°	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	-	-
Leptospira spp. (only in production animals)	2003	-	Order no. 132 of 18/11 2016
Mycobacterium bovis/tuber- culosis Cattle	1920ª OTF in 1980 <sup>d</sup>	Decision 2003/467/EC	Order no. 1417 of 11/12 2007
Coxiella burnetii	2005	-	Order no 1332 of 18/11 2016
<i>Salmonella</i> spp. Cattle Swine Poultry	1993°	-	Order no.537 of 01/06/2016 Order no. 1280 of 04/12/2014 Order no. 1512 of 13/12/2013
VTEC	no	-	-
Yersinia enterocolitica	no	-	-
Parasites			
Cryptosporidium spp.	no	-	-
Echinococcus multilocularis	2004	Council Directive 64/433/EC	Order no. 1332 of 18/11/2016
Echinococcus granulosus	1993	Council Directive 64/433/EC	Order no. 1332 of 18/11/2016
Trichinella spp.	1920ª	Regulation 2075/2005/EC (as amended)	Order no. 544 of 28/05/2014
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 <sup>f</sup>	Regulation 999/2001/EC (as amended)	Order no. 1326 of 26/11/2015

a) Clinical cases, observations during the meat inspection at the slaughterhouse, positive blood samples or finding of agents 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 since 1962.

c) Officially Brucella melitensis Free (ObmF) according to Council Directive 91/68/EC and Commision Decision 2003/467/EC. the disease has never been 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 since 1988 or in deer since 1994.

e) Only clinical cases notifiable.

f) Denmark was recognized as a country with neglible risk for BSE at World Organisation for Animal Health (OIE) general session in May 2011. Source: Danish Veterinary and Food Administration.

Table A33. Salmonella surveillance programme for the rearing flocks and adult flocks of the grandparent and parent generation of the broiler and table egg production, 2016

Samples taken	Material	Material
	Grandparent generation	Parent generation
Per delivery	5 transport crates from one delivery: crate liners (>1 m² in total) or swab samples (>1 m² in total). Analysed as one pool	5 transport crates from one delivery: crate liners (>1 m² in total) or swab samples (>1 m² in total). Analysed as one pool
Per unit	-	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample of 60 g
Per unit	5 pairs of boot swabs (analysed as two pooled samples), or 1 faeces sample consisting of 2x150 g	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample of 60 g
Per unit	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample of 60 g	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample of 60 g
Per unit	5 pairs of boot swabs (analysed as two pooled samples), or 1 faeces sample consisting of 2x150 g	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample of 60 g
	Grandparent generation	Parent generation
Per flock	Hatcher basket liners from 5 baskets (>1 m² in total) or 10 g of broken eggshells from each of 25 hatcher baskets (reduced to 25 g sub-sample). Analysed as one pool	Hatcher basket liners from 5 baskets (>1 m² in total) or 10 g of broken eggshells from each of 25 hatcher baskets (reduced to 25 g sub-sample). Analysed as one pool
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
Per unit	-	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample of 60 g
Per unit	5 pairs of boot swabs (analysed as two pooled samples), or 1 faeces sample consisting of 2x150 g	5 pairs of boot swabs (analysed as two pooled samples), or 1 faeces sample consisting of 2x150 g
Per unit	5 pairs of boot swabs (analysed as two pooled samples), 2 dust samples (250 ml) and 5 birds (analysed for antimicro- bial substances)	5 pairs of boot swabs (analysed as two pooled samples), 2 dust samples (250 ml) and 5 birds (analysed for antimicro- bial substances)
	Per unit Per unit Per unit Per lunit Per unit Per unit Per unit Per unit Per flock Per hatch Per unit	Per unit Per lock Per lock Per unit S pairs of boot swabs (analysed as two pooled samples), or 1 faeces sample consisting of 2x150 g Per unit S pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample of 60 g Per unit S pairs of boot swabs (analysed as one pooled sample), or 1 faeces sample of 60 g  Per unit S pairs of boot swabs (analysed as two pooled samples), or 1 faeces sample consisting of 2x150 g  Grandparent generation  Per flock Hatcher basket liners from 5 baskets (>1 m² in total) or 10 g of broken egg-shells from each of 25 hatcher baskets (reduced to 25 g sub-sample). Analysed as one pool  Per hatch Wet dust samples. Up to four hatchers of the same flock can be pooled  Per unit S pairs of boot swabs (analysed as two pooled samples), or 1 faeces sample consisting of 2x150 g  Per unit S pairs of boot swabs (analysed as two pooled samples), 2 dust samples (250 ml) and 5 birds (analysed for antimicro-

a) Sampling requirements set out by Regulation (EC) No 200/2010.

Source: Danish Veterinary and Food Administration.

b) Samples collected by the food business operator.

c) Sampling requirements set out by Order no 952 of 10/07/2013.

d) Samples collected by the Danish Veterinary and Food Administration.

e) When eggs from a flock exceed the capacity of one incubator, each incubator should be sampled as described.

f) If samples are negative, sampling is repeated 14 days later.

Table A34. Salmonella and Campylobacter surveillance programme for the broiler flocks, 2016

Time	Samples taken	Material
Salmonella		
15 - 21 days before slaughter <sup>a,c,d</sup>	Per flock	5 pairs of boot swabs. Analysed in subsamples of 2 and 3 pairs
7 - 10 days before slaughter <sup>b,e</sup>	Per flock	5 pairs of boot swabs. Analysed in supsamples of 2 and 3 pairs
After slaughter <sup>b,c,f</sup>	Per batch	From slaughterhouses slaughtering 1000 chickens or hen pr day or more: 300 neck skin samples of 1 gram, pooled into subsamples of 60 gram. From slaughterhouses slaughtering less than 1000 chickens or hen pr day: 15 neck skin samples of approx. 10 gram pooled into 5 subsamples of 25 gram
Campylobacter <sup>e</sup>		
After slaughter	Per flock	12 cloacal swabs from 24 animals, analysed in one pool <sup>h</sup>

a) Sampling requirements set out by Regulation (EC) 200/2012.

Table A35. Salmonella surveillance programme for the pullet-rearing, table egg layer and barnyard/hobby flocks in the table egg production, 2016<sup>a</sup>

Time	Samples taken	Material
Pullet-rearing		
Day-old <sup>a,c</sup>	Per delivery	5 transport crates from one delivery: Crate liner (> 1 m² in total) or swab samples (> 1 m² in total) (Analysed as one pooled sample)
4 weeks old <sup>a,c</sup>	Per flock	5 pairs of boot swabs (analysed as two pooled samples) or 5 faeces samples of 60 gram
2 weeks before moving <sup>a,b</sup>	Per flock	5 pairs of boot swabs (analysed as two pooled samples) or 5 faeces samples of 60 gram. 60 blood samples (serology)
Table egg layers (Production for certif	fied packing statio	ns)
24 weeks old <sup>a,b</sup>	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 a dust sample by a cloth of min. $900$ cm <sup>2</sup>
Every 2 weeks from age 20 weeks <sup>a,c,d,</sup>	Per flock	2 pairs of boot swabs (analysed as one pooled sample) or 1 faeces sample consisting of 2x150 g.
After positive serological findings <sup>b</sup>	Per flock	5 pairs of boot swabs (analysed as two pooled samples) or 5 faecal samples consisting of 60 gram each
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 monophasic strains <i>S</i> . 1,4,[5],12:i:-b	Per flock	5 pairs of boot swabs (analysed as two pooled samples) or 5 faeces samples consisting of 60 gram each, 2 dust samples (250 ml) and 5 birds (analysed for antimicrobial substances)
Barnyard and hobby flocks <sup>e</sup>		
Every 18 weeks <sup>a,c,f</sup>	Per flock	Egg samples (serology)

a) Sampling requirements set out by Order no 517/2011, replaced by Order no. 227 of 02/03/2015.

b) Sampling requirements set out by Order no. 1512 of 13/12/2013 replacing 1105 of 18/09/2013 replacing 1462 of 16/12/2009.

c) Samples collected by the food business operator.

d) Once a year, one pair of socks is collected by the Danish Veterinary and Food Administration.

e) Samples are collected by a representative of the slaughterhouse, laboratorium or the Danish Veterinary and Food Administration.

f) sampling requirements set out by Regulation (EC) 2073/2005.

g) For flocks to be slaughtered outside Denmark, 1 pair of boot swabs is collected by the owner 10 days before slaughter at the latest.

h) If the flock is slaughtered over several days, the last batch is sampled.

Source: Danish Veterinary and Food Administration.

b) Samples collected by the Danish Veterinary and Food Administration.

c) Samples collected by the food business operator.

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

e) Voluntary for hobby flocks.

f) For flocks with 30 birds or less: No testing if only delivered to a well-known circle of users.

Source: Danish Veterinary and Food Administration.

Table A36. Salmonella surveillance programmes for turkey flocks, 2016

Time	Samples taken	Material
Turkey production		
Max. 21 days before slaughter <sup>a,b</sup>	Per flock	2 pairs of boot swabs. Analysed individually

a) Sampling requirements set out by Regulation (EC) 584/2008 and Order no. 1512 of 13/12/2013.

Table A37. Salmonella surveillance programme<sup>a</sup> for the cattle production, 2016

No. of samples	Samples taken	Purpose/Comment
Milk producing herds		
4 samples distributed over 18 months	Bulk tank samples	Calculation of herd level <sup>b</sup>
Non-milk producing herds		
1 sample every 180 days at slaughter <sup>c</sup>	Blood samples	Calculation of herd level <sup>b</sup>
4-8 samples depending on herd size	Blood samples	Consecutive negative samples required for level 1 <sup>d</sup>
Beef carcasses at the slaughterhol	use	
5 samples daily, pooled into one analysis	Swab samples from 4 designated areas after 12 hours chilling (4x100cm²)	Slaughterhouses slaughtering 7.500 or more cattle per year
5 samples every second month, analyzed individually	Swab samples from 4 designated areas after 12 hours chilling (4x100cm²)	Slaughterhouses slaughtering 2.500 or more and less than 7.500 cattle per year
5 samples every 6th month, analyzed individually	Swab samples from 4 designated areas after 12 hours chilling (4x100cm²)	Slaughterhouses slaughtering 250 or more and less than 2.500 cattle per year
No sampling		Slaughterhouses slaughtering less than 250 cattle per year

a) Order no. 537of 01/06/2016 as ammended. In 2013 and 2014, the programme for eradication of *Salmonella* Dublin from the Danish cattle production was intensified. This implies regionalisation of the country according to prevalence and compulsory eradication plans in Level 2 herds.

Level 1: Herd assumed free of infection based on bulk milk samples (milk producing herd) or blood samples (non-milk producing herd),

Level 2: Herd not assumed free of infection,

Level 3: Herd infected based on culture and clinical signs or bacteriological findings in the intensified sampling.

- 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.

b) Samples collected by the food business operator or the Danish Veterinary and Food Administration.

Source: Danish Veterinary and Food Administration.

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

Table A38. Salmonella surveillance programme<sup>a</sup> for the pig production, 2016

Time	Samples taken	Purpose/Comment
Breeding and multiplier herds		
Every month	10 blood samples per epidemiological unit	Calculation of <i>Salmonella</i> -index based on the mean seroreaction from the last three months with more weight to the results from the more recent months (1:3:6) <sup>b</sup>
Max. twice per year	Herds with <i>Salmonella</i> -index 5 or above: Pen-faecal samples	Clarify distribution and type of infection in the herd <sup>c</sup>
Sow herds		
When purchaser of piglets is assigned to level 2 or 3, max. twice per year	Pen-faecal samples	Clarify distribution and type of infection in the herd, and possible transmission from sow herds to slaughter pig herds
Herds positive with <i>S.</i> Typhimurium, <i>S.</i> Infantis, <i>S.</i> Derby and <i>S.</i> Choleraesuis are considered positive for the following 5 years <sup>d</sup>	No samples are collected from the herd during the 5 year period when the herd is considered positive, 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 e: one meat juice sample per month	Calculation of slaughter pig index based on the mean proportion of positive samples from the last three months with most weight to the result from the most recent month (1:1:3) <sup>f</sup> . Assigning herds to level 1-3 and assigning herds to risk-based surveillance (RBOV) <sup>e, g</sup>
Slaughter pigs, animals		
At slaughter <sup>h</sup>	Coecum samples, avg. 54 samples per month, 12 months per year	Random collection of samples for monitoring of the distribution of serotypes and antimicrobial resistance.
Pork carcasses at the slaughterhous	e	
5 samples daily, pooled into one analysis	Swab samples from 4 designated areas after 12 hours chilling (4x100cm²)	Slaughterhouses slaughtering more than 200 pigs per day
5 samples every second month	Swab samples from 4 designated areas after 12 hours chilling (4x100cm²)	Slaughterhouses slaughtering 10.000 or more pigs and less than 30.000 pigs per year
10 samples per year, 5 each 6 month	Swab samples from 4 designated areas after 12 hours chilling (4x100cm²)	Slaughterhouses slaughtering 1.000 or more pigs and less than 10.000 pigs per year
No sampling		Slaughterhouses slaughtering less than 50 pigs per month

a) Sampling requirements set out by Order no. 539 of 03/06/2016.

b) Herds with index above 10 have to pay a penalty for each pig sold.

c) The herd owner must inform buyers of breeding animals about the infection level and type of Salmonella.

d) These serotypes are primarily spread by live trade, and are known to persist in herds. S. Typhimurium includes the monophasic S. 1,4,[5],12:i:-.

e) RBOV: risk-based surveillance in herds with a slaughter pig index of zero (no positive samples in the previous three months) the sample size is reduced to one sample per month. Increasing seroprevalence from level 1 to level 3.

f) Since November 2014: based on the proportion of seropositive samples from the last three months. Both the number of seropositive and total number of samples are weighted with most weight to samples from the most recent month (1:1:5).

g) Pigs from herds with highest level of infection (Level 3) must be slaughtered under special hygienic precautions.

h) Centrally coordinated study (Table A28).

Source: Danish Veterinary and Food Administration.

Table A39. Typing methods used in the surveillance of foodborne pathogens in Denmark, 2016

Methods	Human	Food	Animal
Salmonella enterico	1		
Serotype	All	AII	All
Phage type	None	Few <i>S.</i> Typhimurium and <i>S.</i> Enteritidis	Few S. Typhimurium and S. Enteritidis, all isolates danish from poultry
Antimicrobial resistance	All <i>Salmonella</i> except <i>S.</i> Enteritidis	Almost all isolates	Almost all isolates
MLVA	S. Typhimurium <sup>a</sup> and S. Enteritidis	S. Typhimurium <sup>a</sup> , S. Enteritidis and S.Dublin for the Salmo- nella source account, outbreak investigations and research	S. Typhimurium <sup>a</sup> , S. Enteritidis and S. Dublin for the Salmonella source account, outbreak investigations and research
PFGE	Outbreak investigations	Outbreak investigations	Outbreak investigations
WGS	Outbreak investigations	Some for outbreak investigation and research	Some for outbreak investigation and research
Campylobacter coli	/jejuni		
Antimicrobial resistance	Isolates from 4 districts for DANMAP surveillance	For DANMAP surveillance purposes and the case-by-case program	Only for DANMAP surveillance purposes
FlaA-SVR	None	Outbreak investigations	None
MLST, WGS	Outbreaks investigations and research project	None	None
VTEC			
Serotype	Based on WGS	None	All (0157)
Virulence profile	Based on WGS	None	All (0157)
PFGE	Few	None	Outbreak investigations
WGS	All	None	None
Listeria			
Serogroup	All	None	None
PFGE	All	All	All
WGS	All	All	All
Yersinia enterocolit	ica		
0-group	All isolates send to SSI	None	None

a) Including the monophasic strains  $\it S.\,1,4,[5],12:i:-$ .

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

### Population and slaughter data

Table A40. Human population, 2016

Age groups (years)MalesFemalesTotal0-4153,180144,883298,0635-14340,137323,368663,50515-24379,123361,549740,67225-44726,302709,6551,435,95745-64759,701755,6991,515,40065+501,735593,4371,095,172Total2,860,1782,888,5915,748,769				
5-14       340,137       323,368       663,505         15-24       379,123       361,549       740,672         25-44       726,302       709,655       1,435,957         45-64       759,701       755,699       1,515,400         65+       501,735       593,437       1,095,172	Age groups (years)	Males	Females	Total
15-24       379,123       361,549       740,672         25-44       726,302       709,655       1,435,957         45-64       759,701       755,699       1,515,400         65+       501,735       593,437       1,095,172	0-4	153,180	144,883	298,063
25-44       726,302       709,655       1,435,957         45-64       759,701       755,699       1,515,400         65+       501,735       593,437       1,095,172	5-14	340,137	323,368	663,505
45-64       759,701       755,699       1,515,400         65+       501,735       593,437       1,095,172	15-24	379,123	361,549	740,672
65+       501,735       593,437       1,095,172	25-44	726,302	709,655	1,435,957
	45-64	759,701	755,699	1,515,400
Total 2,860,178 2,888,591 5,748,769	65+	501,735	593,437	1,095,172
	Total	2,860,178	2,888,591	5,748,769

Source: Statistics Denmark, 1 January 2016.

Table A41. Number of herds/flocks, livestock and animals slaughtered, 2016

	Herds/flocks (capacity)	Livestock (capacity)	Number slaughtered
Slaughter pigs (>30 kg)	6,459	5,907,986	17,843,548
Cattle	18,370	1,567,213	539,600
Broilers	251	22,600,679	101,594,600
Layers (excl. barnyard)	183	3,367,000	-
Turkeys	33	347,401	3,300
Sheep & lambs	6,575	145,010	78,659
Goats	2,985	19,625	1,340
Horses	-	-	1,597

Source: The Central Husbandry Register and Danish Veterinary and Food Administration.

Table A42. Number of farms in the broiler production, 2016

	No. of holdings	No. of houses/flocks	Livestock (capacity)
Rearing period (grandparent)	2	10	90,000
Adult period (grandparent)	3	8	90,000
Rearing period (parent)	20	97	130,000
Adult period (parent)	42	147	720,000
Hatcheries	5	-	-
Broilers	240	612	n.a

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

Table A43. Number of farms in the table egg production, 2016

	331		
	No. of holdings	No. of houses/flocks	Livestock (capacity)
Rearing period (grandparent)	1	3	48,000
Adult period (grandparent)	7	7	75,000
Rearing period (parent)	8	10	38,000
Adult period (parent)	8	10	56,000
Hatcheries	6	-	-
Pullet-rearing	42	65	1,072,000
Layers (excl. barnyard)	182	183	3,367,000

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

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