



Annual Report on Zoonoses in Denmark 2003



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The cover page illustration:
*Incidence of human campylobacteriosis,
1980-2003*

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Introduction

This report on zoonoses presents a summary of the occurrence of zoonotic agents in feeding stuffs, animals, food stuffs and humans in Denmark. The report is based on data compiled according to the zoonoses directive 92/117/EEC, supplemented by data obtained from the Danish surveillance and control programmes as well as data from relevant research projects from the institutions, which have contributed to the preparation of this report. This report is also available at www.dvfv.dk. In addition to the information available in the printed report, the web edition includes figures depicting the age and gender distribution of the major human zoonotic infections.

On 1 January 2004, the Danish Veterinary Institute and the Institute of Food Safety and Nutrition of the Danish Veterinary and Food Administration were merged to establish a new governmental research institute: The Danish Institute for Food and Veterinary Research. This new institute unites food and veterinary research under the Danish Ministry of Food, Agriculture and Fisheries. For a list of the contributing institutions and their appropriate abbreviations, please refer to the back of this report.

Profile of the year

The most remarkable feature of this year was an almost 20% decrease in the number of human *Campylobacter* cases. This coincided with the implementation of a *Campylobacter* strategy aiming at reducing the number of *Campylobacter* in Danish broiler meat (p. 20). The strategy included a number of different interventions at the farm-, slaughterhouse- and consumer-level. It is believed that the most important factor contributing to the decrease was the attempt to allocate all *Campylobacter* negative flocks to the production chilled products and positive flocks to the production of frozen products. In 2003, the number of human infections caused by *Salmonella* continued to decrease. However, in contrast to infections caused by most other *Salmonella* serotypes, the number of human *S. Typhimurium* infections increased by 18%.

Outbreaks

While there were fewer outbreaks caused by *S. Enteritidis* in 2003 than in the preceding year, the number of *S. Typhimurium* outbreaks increased. Several *S. Typhimurium* outbreaks occurred, but a particularly large one caused by *S. Typhimurium* phage type U302 was traced to a restaurant in an amusement park. The outbreak investigation strongly suggested that a member of the kitchen staff was the source of the outbreak (p. 13). A large outbreak caused by *S. Enteritidis* phage type 8 was also identified and involved at least 50 persons at a music festival. This outbreak was caused by cross contamination in a kitchen at the festival (p. 13). A small outbreak caused by *Yersinia* was also identified in 2003. *Yersinia* outbreaks are extremely rare in Denmark and this outbreak was found to be associated with meat from a butcher shop (p. 21).

Surveillance

The *Salmonella* surveillance programmes in the poultry and pig production continued as described in previous Annual Reports on Zoonoses in Denmark. However, in July 2003 the Danish Veterinary and Food Administration agreed on changing the surveillance programme for multi-drug resistant *S. Typhimurium* DT104 (MRDT104), thereby changing the way the MRDT104 infected pig herds are handled (pp. 9-10). The surveillance programmes for *S. Dublin* in cattle continued as described in 2003 (pp.16-17) as did the programme for transmissible spongiform encephalopathy (pp. 24-25). The latter was, however, affected by the implementation of several new regulations and now also includes testing of all fallen sheep and goats older than 18 months.

1. Salmonella

Feeding stuffs

The Danish Plant Directorate (PD) monitors all Danish feed compounders for presence of *Salmonella*. The monitoring includes sampling of compound feeds and feed materials as well as sampling during feed processing (environmental samples). Further details were described in the Annual Report on Zoonoses in Denmark, 2000. The monitoring of compound feeds was discontinued in October 2003.

As in previous years, the prevalence of *Salmonella* in feed in 2003 was low. The results are shown in Table 1.

In 2003, the rare serotype S. Idikan was detected in environ-

mental samples and in samples of compound feed for pigs and cattle at a specific feed mill over a period of five months. This serotype was also detected in pen-faecal samples, collected as part of the *Salmonella* control programme in pigs (see pp. 8-9), from five different pig farms. Feed from four of these farms could be linked to the feed mill in question. The bacterial isolates were DNA typed by Pulsed Field Gel Electrophoresis (PFGE). The isolates from the feed mill were found to be indistinguishable from the isolates at the five farms, but different from the DNA-profiles of unrelated isolates obtained from feeding stuffs and pork.

Rendering Plants

The EU Regulation laying down rules concerning animal by-products not intended for human consumption (Regulation no. 1774/2002 of 3rd Oct.2002, The Animal By-product Regulation) deals with three different categories of meat and bone meal. Category 1 and 2 material is processed at category 1 and 2 processing plants and the end product is not intended for feeding purposes. Category 3 processing plants produce processed animal protein that may be used for feeding purposes.

The hygiene at the processing plants is mainly monitored by the plants' own-check programmes, which are inspected by the Regional Veterinary and Food Control Authorities (RVFCA). *Salmonella* positive findings are reported to the RVFCA.

In 2003, 339 samples of fishmeal and 27 samples of meat and bone meal were examined for *Salmonella* by the RVFCA. None of these were found positive.

At two major category 3 processing plants, a total of 5,388 samples were collected as part of the plants' own check programmes and 15 (0.3%) samples were found positive for *Salmonella*. Isolates from the samples were serotyped and the following serotypes were found: S. Montevideo (7), S. Kentucky (3), S. Livingstone (2), S. Llandoff (2) and S. Poona (1).

Poultry and poultry products

From the 1st of January 2003, the Danish Poultry Council (DPC) took over the administration of the National *Salmonella* Control Programme for poultry. The

Table 1. Control of *Salmonella* in compound feeds, feed processing and feed materials, 2003.

	Number of samples/ % <i>Salmonella</i> positive			Serotypes 2003
	2001	2002	2003	
Compound feeds in total	2,616 / 0.2	2,708 / 0.1	1,363 / 0.1	
Feed for pigs	1,552 / 0.1	1,498 / 0.0	796 / 0.1	S. Idikan
Feed for cattle, horses, sheep and rabbits	741 / 0.4	754 / 0.3	378 / 0.3	S. Idikan
Feed for poultry	262 / 0	350 / 0	164 / 0	
Pet food	61 / 0	106 / 0	25 / 0	
Feed materials in total	332 / 1.8	349 / 3.7	151 / 1.3	
Farm animals	244 / 0.8	269 / 1.9	144 / 1.4	S. Kentucky, S. Putten
Pets	88 / 4.5	80 / 10	-	
Feed processing plants (process control)				
Ordinary inspections	2,697 / 1.0	2,740 / 1.2	2,409 / 1.4	S. 4.12:b:-, S. Cubana, S. Falkensee, S. Give, S. Havana, S. Idikan, S. Infantis, S. Kentucky, S. Lexington, S. Meleagridis, S. Mbandaka, S. Oranienburg, S. Orion var 15, S. Putten, S. Senftenberg, S. Tennessee, S. Typhimurium DT 66
Additional inspections	267 / 20.5	262 / 18.3	241 / 19.1	

Source: PD

programme is, however, still monitored by the Danish Veterinary and Food Administration (DVFA). Slaughter or destruction of parent flocks in compliance with the Zoonosis Directive is covered by governmental funds. Furthermore, the government reimburses the value of hens sampled from suspected flocks and 75% of the test costs in small flocks. All other expenses in connection with routine and suspicion samples are paid by the poultry industry.

In 2003, the Danish *Salmonella* surveillance and control programme generally continued as described in the Annual Report on Zoonoses in Denmark 2000, 2001 and 2002. The numbers of establishments in the broiler and table-egg production are shown in Table 2 and the sampling scheme is summarised in Table 3.

Table egg production

No central-rearing flocks or breeding flocks were found infected with *Salmonella* in 2003. Four out of 367 (1.1%) examined pullet-rearing flocks were found infected with *Salmonella*. Two flocks were infected with *S. Enteritidis* phage type (PT) 8, one flock with *S. Derby* and one flock was declared infected based on serology (mix ELISA, Table 4).

In flocks producing eggs for certified egg packing stations, 14 (2.3%) out of 611 tested table egg flocks were found infected with *Salmonella*. In comparison, 2.6% (16 out of 619) of the investigated flocks were found infected with *Salmonella* in 2002.

Out of 167 examined battery flocks, nine (5.4%) were found infected: *S. Enteritidis* PT8 (3), *S. Enteritidis* PT6 (3), *S. Enteritidis* PT4 (2) and *S. Derby* (1). One of the *S. Enteritidis* PT6 infected flocks was also found infected with *S. Infantis*. As in previous years, the overall prevalence was higher among battery flocks than in any other type of production.

A total of 191 free-range flocks were examined and two (1.0%) flocks were infected with *S. Enteritidis* PT4 and PT8, respectively. Of 71 examined deep litter flocks, two (2.8%) flocks were infected with *S. Enteritidis* PT4. One organic flock (out of 173 examined, 0.6%) was found infected with *S. Enteritidis* PT1b.

Out of 516 examined flocks producing eggs for barnyard sale, five (1.0%) flocks were declared infected with *Salmonella* based on the serological results alone. Owners with hobby flocks, producing eggs for personal use only, are not required to participate in the *Salmonella* control programme, and consequently collection of suspi-

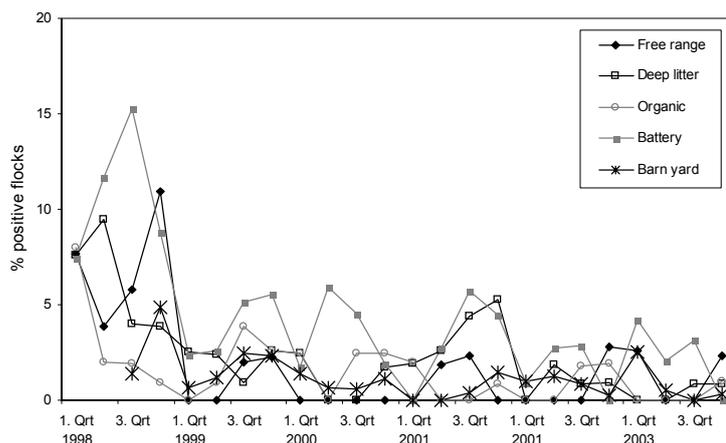


Figure 1. Quarterly percentage of layer flocks infected with *Salmonella* according to type of production, 2003.

Source: DVFA

Table 2. Number of farms in the broiler and the table-egg production, 2003.

	No. of farms	No. of houses	No. of animals
Broiler production			
Central rearing	21	107	1,244,000
Broiler breeders	52	165	503,250
Hatcheries	6		
Broilers	303	730	129,860,683 ^{a)}
Table-egg production			
Central rearing	7	8	60,000
Layer breeders	7	13	57,353
Hatcheries	6		
Rearing	105	168	1,493,935 ^{a)}
Layers, excl. barnyard sale	282	421	3,576,221 ^{b)}

a) Hatched for use in Denmark.

b) Including hens imported as day-old chickens.

Source: DVFA and DPC

Table 3. Salmonella surveillance of the broiler and table-egg production, 2003.

CENTRAL REARING FLOCKS		Central rearing flocks	Breeding segment, central rearing flocks
Time	Sample taking	Material	Material
Day-old	Per delivery	10 samples of crate material and 20 dead chicks ^{a)}	10 samples of crate material and 20 dead chicks ^{a)}
1 st week	Per unit	40 chicks	-
2 nd week	Per unit	2 pairs of sock samples	-
4 th week	Per unit	60 faecal samples ^{a)}	60 faecal samples ^{a)}
8 th week	Per unit	2 pairs of sock samples	2 pairs of sock samples
2 weeks before being moved	Per unit	2 pairs of sock samples ^{a)} and 60 blood samples	60 faecal samples ^{a)}
HATCHING EGG PRODUCTION		Hatching egg production	Breeding segment, hatching egg production
Every two weeks	Per flock	250 meconium samples or 50 dead chickens collected at the hatchery ^{a)+b)}	250 meconium samples or 50 dead chickens collected at the hatchery ^{a)+b)}
Every week	Per unit	2 pairs of sock samples ^{c)}	-
HATCHERY			
After each hatching	1-4 hatchers may be pooled	At least 25 grams of wet dust per hatcher	At least 25 grams of wet dust per hatcher
a) Requirements of the Zoonosis Directive (92/117/EEC).			
b) Samples collected by the RVFCA every 8 weeks			
c) Samples collected by the RVFCA every three months			
REARING STOCK			
Time	Sample taking	Material	
Day-old	Per delivery	10 samples of crate material and 20 dead chicks	
Week 3	Per flock	5 pairs of sock samples or 300 faecal samples, if sock samples cannot be collected. Flocks of less than 200 birds: 2 pairs of sock samples or 60 faecal samples	
Week 12	Per flock	Flocks of 500 or more: 60 blood samples and 5 pairs of sock samples or 300 faecal samples of sock samples cannot be collected ^{a)} Flocks of 200-499: 55 blood samples and 5 pairs of sock samples ^{a)} Flocks of less than 200 birds: Blood samples ^{b)} and 2 pairs of sock samples or 60 faecal samples ^{a)}	
TABLE EGG PRODUCTION FOR CERTIFIED PACKING STATIONS			
Every 9 weeks	Per flock	Egg samples ^{b)} and 2 pairs of sock samples or faecal samples, equal to the number of eggs, if sock samples cannot be collected	
BARNYARD AND HOBBY FLOCKS			
3 times a year	Per flock	Egg samples ^{b)}	
a) Samples collected by the RVFCA			
b) According to Table 1 in Governmental Order No. 44, Jan. 23rd 2003			
Source: DVFA			

Table 4. Occurrence of Salmonella in the table-egg production, 2003.

Zoonotic pathogen	Central Rearing		Layer Breeders		Rearing		Table-egg production	
	Examined flocks	Positive flocks	Examined flocks	Positive flocks	Examined flocks	Positive flocks	Examined flocks	Positive flocks
<i>Salmonella</i> spp.	24	0	15	0	367	4 (1.1)	611	14(2.3)
<i>S. Enteritidis</i>	-	-	-	-	-	2 (0.6)	-	13 (2.1)
<i>S. Typhimurium</i>	-	-	-	-	-	-	-	-
Other serotypes	-	-	-	-	-	2 (0.6)	-	1 (0.2)
Source: DVFA								

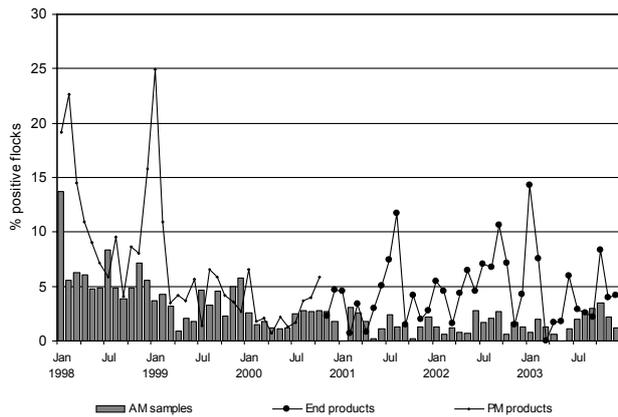


Figure 2. Percent *Salmonella* positive broiler flocks detected at the mandatory ante-mortem (AM) and end-product examination, 1998-2003. Post-mortem (PM) examinations were replaced by end-product examinations in November, 2000. Source: DVFA and DPC

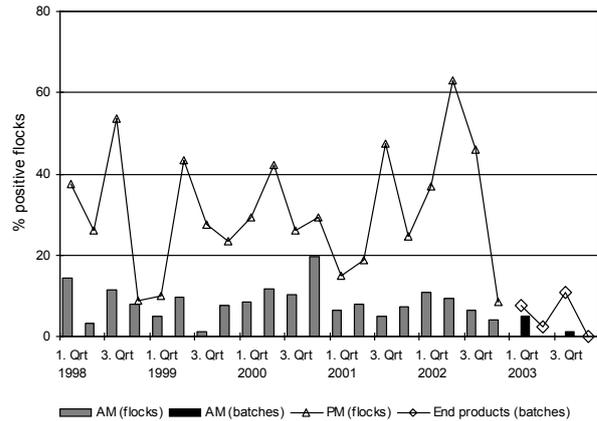


Figure 3. Quarterly percentage of *Salmonella* positive turkey flocks detected at the mandatory AM and PM examinations, 2003. PM examinations were replaced by end-product examinations in January, 2003. Source: DVFA

on samples is only sporadically done. Out of 139 examined hobby flocks, three were found infected. The distribution of infected flocks in the different types of production is shown in Figure 1. The sero- and phage type distribution is shown in the Appendix, Table A7 and A8.

Broiler production

In 2003, three out of 226 (1.3%) examined central rearing flocks were found infected with *S. Typhimurium* DT170 (Table A1). Out of 182 examined breeder flocks, four (2.2%) were found infected with *Salmonella*. Three flocks, reared on the same premises, were infected with *S. Typhimurium* DT41, while one flock was infected with *S. Enteritidis* (phage type could not be determined).

All broiler flocks were monitored for *Salmonella* by the mandatory ante-mortem (AM) examination. Two-three weeks prior to slaughter, five pairs of sock samples were collected from each flock. In 2003, the percentage of positive flocks ranged from 0.0% to 3.4% per month with a mean of 1.7% (Table A1, Figure 2). This is a slight increase compared to 2002, where the prevalence was 1.5%. The most frequently occurring

serotype was *S. Typhimurium*, which was found in 26.0% of the infected flocks. The sero- and phage type distribution is presented in Table A7, A8 and A9.

The mandatory examination, after slaughter, was done by sampling batches of chicken cuts close to packaging. A batch is defined as the amount of meat from animals slaughtered between two cleanings and disinfections. *Salmonella* was detected in 77 of 1,552 (5.0%) investigated batches of chicken meat.

Turkey production

All turkey flocks were monitored for *Salmonella* by the mandatory AM examination. *Salmonella* was detected in 12 (5.7%) of 211 investigated flocks (Table A2). Of these, five flocks were infected with *S. Kottbus*, two flocks with *S. Saintpaul*, two flocks with *S. 4.12:d:-* and three flocks with other serotypes. *S. Enteritidis* was not isolated from any turkey flocks in 2003. The serotype distribution is shown in Table A7.

Prior to 2003, testing for *Salmonella* in turkeys post slaughter was carried out by testing a pool of 10 neck-skin samples from each flock. However, since January 1st, 2003, the mandatory examination of turkeys after slaughter has been

carried out by sampling turkey cuts close to packaging (as described for broilers). In 2003, *Salmonella* was detected in 17 of 271 (6.3%) batches, (Table A2, Figure 3), which is a decrease compared to 2002, where *Salmonella* was detected in 27 of 323 (8.4%) turkey flocks. It should be noted, however, that results from previous years (neck-skin samples) and 2003 (meat samples) may not be completely comparable. The decrease in prevalence may be explained by the fact that in 2003 all AM *Salmonella* positive flocks were exported prior to slaughter.

Duck production

Duck flocks were monitored by the mandatory AM examination prior to slaughter. In 2003, 220 flocks were examined. *Salmonella* was isolated from 161 (73.2%) of the flocks, representing an increase in prevalence compared to 2002 where 54.7% of the flocks were found positive. In several cases, more than one serotype was isolated from each flock (Table A7). *S. Anatum* was the most frequently isolated serotype in the infected flocks (Table A7).

The public plan for control of *Salmonella* in poultry, 1996-2002

The public *Salmonella* control programme was implemented in December 1996 and has continuously been evaluated and revised accordingly as described in previous issues of the Annual Report on Zoonoses in Denmark. Initially, the programme was planned to run for three years with a total budget of 188.1 mill. DKK (25.2 mill. EUR), of which the poultry industry contributed 30 mill. DKK (4 mill. EUR). In 1999, 62 mill. DKK had still not been used and the programme was extended for another three years. The public funding ran out by the end of 2002 and all administrative and economic responsibilities were taken over by the industry. The Government continues to set the targets for the programme in order to ensure a continuous reduction of *Salmonella*, and the DVFA supervise that the programme runs satisfactorily. The Government also continues to compensate certain losses incurred by the producers including compensation of parent flocks infected with *S. Enteritidis* or *S. Typhimurium*.

Results of the programme

In both the table-egg and broiler sector, the results are good. The prevalence of infected table-egg layer flocks was reduced from 13.4% in 1998 to 2.3% in 2003. In the broiler production, positive results were already seen before 1996 as part of the existing voluntary control programme, but the prevalence continued to decrease after implementation of the new programme and was reduced from 12.9% in 1997 to 1.7% in 2003. The occurrence of *Salmonella* in breeding and parent flocks has throughout the period been around 1.2%, but the top-down eradication strategy, where infected flocks are culled or slaughtered, ensures that the spread of *Salmonella* through the breeding pyramids is minimal.

Public-health effects

The good results in the primary production are also reflected in the number of reported human *Salmonella* infections, which has been reduced with 66% from 5,015 in 1997 to 1,713 in 2003. The reduction is, in particular, a result of fewer infections caused by the consumption of table eggs. The Danish Zoonosis Centre (DZC) estimates that the number of egg-associated cases was reduced from 3,009 cases in 1997 to 271 in 2003, corresponding to a 90% reduction. Assuming that the incidence of egg- and broiler-associated infections had remained at the 1997 level in case of no control, the DZC further estimates that the society has saved 330-865 mill. DKK (45-116 mill. EUR) in public-health costs including medical consultation, laboratory analyses, hospitalisation, and lost labour.

In conclusion, the *Salmonella* control programme implemented in the poultry sector has been successful and the money invested has been more than returned. The results show that salmonellosis, to a wide extent, can be prevented because we have the tools to investigate the epidemiology as well as control measures and strategies that are cost-effective.

Pigs and pork

A serological surveillance programme for detection of *Salmonella* infection in slaughter pig herds was implemented in 1995. The programme has been changed through the years, adjusting to a continuously decreasing level of infection in the pig herds. The programme has

previously been described in Annual Report on Zoonoses in Denmark 2000-2002. Originally, the DVFA handled the administration of the programme. However, since May 2002, the daily administration has been taken over by the Danish Bacon and Meat Council (DBMC). The programme is, however, still monitored by the DVFA.

Herds producing more than 200 slaughter pigs per year are monitored by serological testing of meat juice (approx. 600,000 meat juice samples per year). The serological results are used to assign each herd to one of three levels based on the proportion of sero-positive meat juice samples over the last three months. The

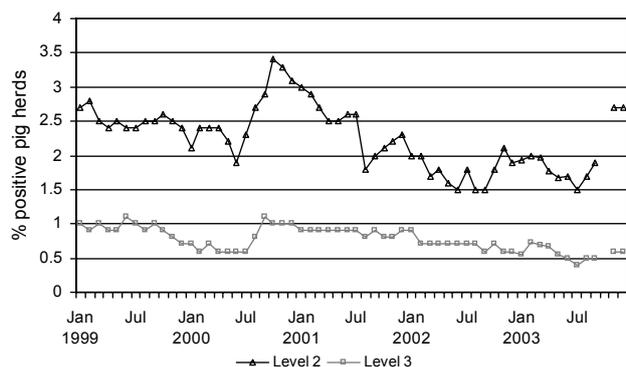


Figure 4. Serological surveillance of *Salmonella* in pig herds. Percent positive pig herds within level 2 and 3, 1996-2003. The level at which a serological test is considered positive was reduced in August 2001. No herds were assigned to higher *Salmonella* levels in November 2003. Source: DVFA

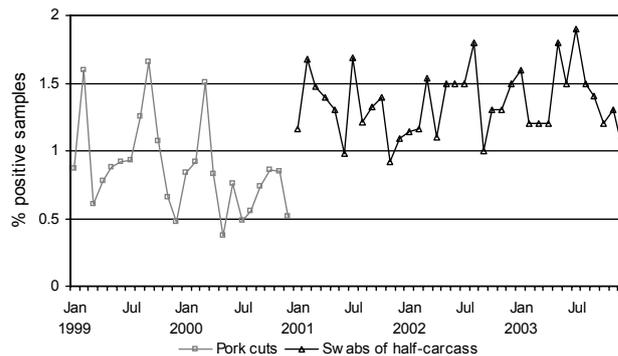


Figure 5. *Salmonella* in pork monitored at slaughterhouses, 1999-2003. From January 2001 monitored by swab samples of half-carasses. Source: DVFA

samples are weighted, so that results from the most recent month are given more weight. In Level 1 are herds with no or few reactors; Level 2 contains herds with a higher proportion of reactors, and Level 3 are herds with an unacceptably high proportion of reactors from which pigs must be slaughtered under special hygienic precautions. Herds in Level 2 and 3 are obligated to collect pen-faecal samples in order to clarify the distribution and type of the *Salmonella* infection in the herd.

In September 2003, a rise in the numbers of positive meat juice samples, from 5.6% to approx. 10.1% in the following month was observed. The increase was so abrupt that a technical problem was suspected. Consequently, no herds were assigned to a higher *Salmonella* level in November. Investigations into the matter identified a technical laboratory problem beginning in August 2002. The problem caused some of the samples to be analysed incorrectly resulting in an artificially low level of positive meat juice samples. The high number of slaughter-pig herds in Level 2 and 3 observed after September 2003 is considered to be valid. The increase may in part be explained

by an accumulation of positive herds that were not correctly assigned to level 2 or 3 during the period where the technical problem persisted. However, a general increase in the occurrence of *Salmonella* in slaughter-pig herds cannot be excluded. The procedure of assigning herds to *Salmonella* levels was resumed in December. By the end of 2003, 96.7% of the herds fell within level 1; 2.7% within level 2 and 0.6% within level 3 (Figure 4, Table A3).

Breeding and multiplying herds are monitored monthly by serological testing of blood samples. If a specific cut-off level is reached, the herd owner is obligated to collect pen-faecal samples. This serological test was unaffected by the disorder described above, because a different procedure for analysing these samples is used. However, a rise in the numbers of herds reaching the cut-off level has also been observed for these herds, supporting a general increase in the *Salmonella* prevalence in pig herds. This will be further investigated.

With a few exceptions, all sow herds supplying piglets to slaughter pig herds in level 2 or 3 are obligated to collect pen-faecal samples in order to determine the distribution of *Salmonella* within

the herd, and to clarify possible transmission of *Salmonella* from the sow herd to the slaughter pig herd.

Clinical salmonellosis was recorded in 66 herds (Table 5). This represents the number of herds submitting material from clinically affected animals to the laboratory. Of these, 14 herds were placed under official veterinary supervision including four herds that were placed under Zoonosis Supervision, due to isolation of multi-drug resistant *Salmonella* Typhimurium DT104 (MRDT104) from the herds.

All data from the surveillance of *Salmonella* in pigs are registered in a central database, the Zoonosis Register. This register is a part

Table 5. Isolation of *Salmonella* from outbreaks of clinical disease in pig and cattle herds, 2003

Serotype	Pigs herds	Cattle herds
Derby	7	-
Dublin	-	40
Infantis	5	1
London	1	-
Typhimurium MRDT104	4	1
Other		
Typhimurium	48	32
Worthington	1	-
Total	66	74

Source: DVFA

of the Central Husbandry Register and is administered by the DVFA.

Monitoring of *Salmonella* in pork is based on swab samples taken from three designated areas of chilled half-carcasses. The samples are pooled, each pool consisting of samples from five carcasses. However, in the smallest slaughterhouses the samples are analysed individually. In 2003, 34,250 samples were pooled into 6,850 pools, which were then analysed. *Salmonella* was found in 292 of these. Furthermore, 210 samples were collected and analysed individually, and *Salmonella* was found in two of these samples (Table A3). When determining the prevalence of pooled samples, the loss of sensitivity and the probability of more than one sample being positive in each pool has to be taken into consideration. A conversion factor of three has been determined on the

basis of comparative examinations, as described in Annual Report on Zoonoses in Denmark, 2001. In 2003, the overall prevalence of *Salmonella* in pork was estimated to 1.4% (Figure 5), which is at the same level as in 2001 and 2002. The most common serotypes encountered in pork were *S. Typhimurium*, *S. Derby* and *S. Infantis* (Table A7). The phage type distribution of *S. Typhimurium* is shown in Table A9.

Cattle and beef

The *S. Dublin* programme is described on pp. 10-11. In 2003, salmonellosis was recorded in 74 herds (Table 5). This figure represents the number of herds submitting material from clinically affected animals to the laboratory. Of these, 47 herds were placed under official veterinary supervi-

sion, and four more herds were subject to hygienic slaughter due to solitary findings of *S. Dublin*.

Monitoring of *Salmonella* in beef and veal at slaughterhouses is based on swab samples taken from three designated areas of chilled half-carcasses. The samples are pooled, each pool consisting of samples from five carcasses, except in the smallest slaughterhouses, where samples are analysed individually. In 2003, 11,660 samples were pooled into 2,332 pools. *Salmonella* was found in 25 of these. Furthermore, 937 samples were collected and analysed individually, and *Salmonella* was found in four of these. Using the same correction factor reported for pork, the overall sample prevalence for 2003 was estimated to be 0.4% (Figure 6, Table A4). This represents an increase compared to 2002, where the estimated prevalence was

National surveillance of *Salmonella* Dublin in cattle

Since October 2002, there has been a national surveillance programme for *S. Dublin*. The programme is based on Order no. 974 and was developed by the DVFA, the DFVF and the cattle industry.

The surveillance is based on serological testing of blood or milk samples from the currently running BVD and IBR programmes and on reports from veterinary surgeons on clinical salmonellosis in cattle herds. The programme aims at identifying herds free of *S. Dublin* infection. This approach was chosen, because epidemiological analyses showed that the serological test was better at identifying non-infected herds than infected herds. The overall

objective was to decrease the spread of *S. Dublin* between herds by setting up a system, which encourages trade between negative herds.

Based on serological results, cattle herds are divided into three levels. Level 1: Most likely free of *S. Dublin*. Level 2: *S. Dublin* is most likely present (or unknown status). Level 3: *S. Dublin* has been isolated from the herd, or the herd has purchased animals from known level 3 herd. The *S. Dublin* status is revised at least every three months. Test results are valid for four months (milk samples and blood samples in herds with more than 25 animals; blood samples in herds with less than 25 animals are valid for 12

months). The percentage of herds in levels 1-3 in December 2003 is shown in Table I.

No detectable change in the proportion of level 2 and 3 herds was observed in 2003. However, the trade among herds assigned to level 1 has been changed significantly. Presently, only approx. 2.5% of the level 1 herds receive cattle from level 2 or level 3 herds compared to approx. 40% of the herds at the time of introduction of the programme. During the first year it was shown that a level 1 herd had a higher risk (15.5%) of being infected with *S. Dublin* when level 2 cattle were introduced compared to when level 1 cattle was introduced (5.6%) or when no cattle was introduced (5.2%).

Table I. Percentage of cattle farms in each level

S. Dublin Level	Milk producing herds	Non-milk producing herds
Level 1	74.0	76.5
Level 2	25.9	23.5
Level 3	0.2	<0.0

a) Not all non-milk producing herds had to be tested from the starting point of the programme.

Source: DVFA

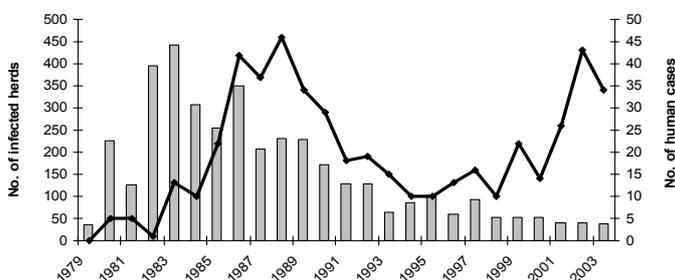


Figure I. Number of human cases and cattle herds with clinical salmonellosis caused by *S. Dublin*, 1979-2003

Line: No. of human cases Cols: *S. Dublin* infected cattle herds

Source: DVFA and SSI

Herds with salmonellosis are placed under official restrictions and supervision and the animals are slaughtered under special hygienic precautions. Each carcass is swabbed and tested bacteriologically and positive carcasses must be heat-treated.

In 2003, *Salmonella* was isolated in 74 cattle herds in Denmark. The predominant serotypes were *S. Dublin* (40 cases, 54.1%) and *S. Typhimurium* (33 cases, 42.9%), including one case of MDRT104 (Table 5).

A slaughterhouse investigation of 1,712 calves from the 35 largest feed lots in Denmark showed that 54% of the herds delivered animals carrying *S. Dublin* in the intestinal content. A total of 2.5% of all animals were positive for *S. Dublin* with an in-herd prevalence of 1-15%.

Occurrence of *S. Dublin* in beef

In 2003, a retail investigation for *S. Dublin* was carried out in cuts of Danish and imported beef. One of 2,035 Danish beef samples (0.1%) was found positive for *S. Dublin*. No other *Salmonella* serotypes were isolated from Danish beef. While none of 1,112 imported beef samples were found positive for *S. Dublin*, one sample (0.1%) was found positive for *S. Heidelberg*. These numbers are lower than what was found in a survey, carried out in Denmark in 2002, of 1,380 minced beef samples (1.5% positive).

Human infections

In contrast to infections caused by other *Salmonella* serotypes such as *S. Enteritidis* and *S. Typhimurium*, the number of human cases

caused by *S. Dublin* increased from 2001 (26 cases) to 2002 (43 cases). However, in 2003 the number of cases fell to 34. The number of human cases and the number of clinical infected cattle herds caused are depicted in Figure I.

PFGE typing of *Salmonella Dublin*

PFGE typing has been used for many years at both Statens Serum Institut (SSI) and DFVF for molecular epidemiological analysis of *Salmonella* spp. These institutes have now established an internet-based data sharing tool in which PFGE profiles derived from bacterial strains of human and animal origin can be stored in a common database and compared, to investigate, identify and quantify potential sources of infection.

In 2003, 77 *S. Dublin* strains of human (n=32), porcine (n=3), cattle (n=41) and unknown (n=1) origin were assigned to PFGE types by use of the restriction enzyme *Xba*I. The PFGE profiles of *S. Dublin* is relatively conserved and profile variation depends upon minor differences in band mobility.

PFGE types 1 and 3 predominate among human infections and these are also found in porcine and bovine sources. However, type 1 is also common in cattle, whereas type 3 is relatively infrequent in bovine material. Two porcine strains and one strain of unknown origin were assigned to PFGE type 1 and one porcine strain to PFGE type 3. In the material only one strain originating from foreign production was included and this strain was assigned to PFGE type 1. The three porcine strains may represent cross contamination or may be regarded as passants in pigs kept close to cattle.

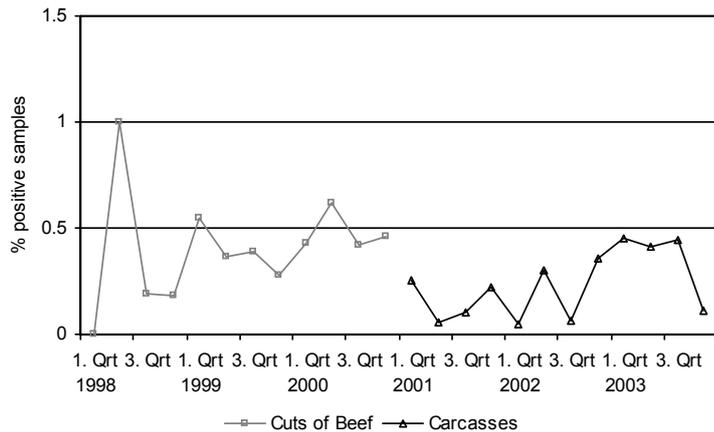


Figure 6. *Salmonella* in beef, monitored at slaughterhouses, 1998-2003. Swab samples taken from 3 designated areas of chilled half carcass. Source: DVFA

0.2%. The most common serotype in beef was *S. Dublin* (Table A7). The phage type distribution of *S. Typhimurium* is shown in Table A9.

Occurrence of multi-drug resistant *S. Typhimurium* DT104

The Danish surveillance programme for MRDT104 was described in Annual Report 2001. In July 2003, the DVFA and the pig-industry agreed on a new way of handling pig herds infected with MRDT104. The change was based on a risk assessment made by the DFVF and the DVFA, and an assessment of the human health consequences made by SSI. The basic idea of the changes was that the manner of handling MRDT104 infected herds and herds infected with other types of *Salmonella* should approach each other. These requirements were met by tightening the general programme, based on the assumption that measures affecting the occurrence of *Salmonella* in general would also have an effect on MRDT104. Changes were made mainly in the primary production. The plan for handling MRDT104 herds at slaughter remained the same and the zero-tolerance of the bacteria in food was maintained.

In the primary production, the main elements were lifting the Zoonosis Supervision in pig herds, including the elements of restriction on sales of animals, handling of manure, and the examinations needed in order to lift the sanctions. The demand for an action plan for reducing *Salmonella* in infected herds was repealed and the bacteriological testing of trade contacts was replaced by serological testing. Similar changes were not made in the cattle production. Since the occurrence of MRDT104 in Danish cattle is very low and because there is no specific procedure for eliminating MRDT104 in cattle at slaughter, the cattle industry wished to maintain the restrictions on cattle herds with MRDT104 in order to reduce the risk of spreading the infection between herds. Selling pigs from MRDT104 infected herds, or from herds that have had trade contacts with herds infected with MRDT104, to cattle herds was also prohibited.

The tightening of the general programme included further restrictions on Level 3-herds, including regulation on how to handle manure from these herds. At slaughter the changes include hot water treatment of all carcasses from MRDT104 herds with a *Salmonella* index above 20, reduc-

tion of the amount of analyses after hot water treatment and setting new targets for the plan.

The number of herds infected with MRDT104 was the same in 2003 as in 2002 (22 herds). From January to July, 15 pig herds were found infected with MRDT104 and from August and until the end of 2003, seven herds were found infected. In addition to this, one cattle herd was found infected.

Monitoring of MRDT104 in fresh meat imported from the EU and third countries continued in 2003. The overall prevalence of MRDT104 in imported meat was 0.4% (18 of 4,250 samples, Table 6).

Wildlife and pet animals

The DFVF monitors the occurrence of *Salmonella* in pet animals and wild mammals and birds. The group of wild mammals and birds consists mainly of dead animals submitted by hunters, veterinarians and others. As in 2002, pet animals were investigated on clinical indication only. The findings of *Salmonella* in pets, wildlife and zoo animals, in 2003, are shown in Table A5.

Products from retail outlets

At the retail level, the RVFCA collect samples for routine surveillance of *Salmonella* in meat and products hereof. As part of a new strategy, the number of samples collected for control purposes at this level was reduced in 2002 compared to previous years and 20% of the resources, previously used for microbiological examinations of foods at retail, are now allocated to so-called „centrally coordinated

Table 6. Number of Salmonella positive samples obtained from imported poultry, pork and beef, 2003.

Imported product	Number of samples	Positive (%)	Positive for DT104 (%)
Poultry	2,110	225 (10.7)	2 (0.1)
Pork	810	59 (7.3)	11 (1.4)
Beef	1,280	11 (0.9)	5 (0.4)
Lamb	20	0	0
Others	30	3 (10.0)	0
Total	4,250	298 (7.0)	18 (0.4)

Source: DVFA

projects". These projects focus on collecting data on prevalence and concentration of specific pathogens in foods during processing and at retail. The purpose of the strategy is to provide supplementary data for ongoing and future risk assessment analyses.

A total of 31 broiler and broiler products, 10 samples of turkey cuts and turkey products, 183 samples of pork and pork products, and 4,788 samples of beef and beef products were examined in 2003. In samples from non heat-treated meat and meat products the prevalence was 0.0%, 0.0%, 0.6%, and 0.1% respectively. *Salmonella* was not isolated from any heat-treated products (Tables A1, A2, A3 and A4).

At retail, an investigation of 2,035 of Danish beef cut samples and 1,273 samples of imported beef showed a prevalence of 0.05% and 0.06% respectively.

Salmonellosis in humans

The number of human *Salmonella* infections reached an all time high in 1997, but has since then decreased steadily. In 2003 this positive trend continued when a total of 1,713 laboratory confirmed episodes of salmonellosis were reported (32 cases per 100,000 inhabitants, Table A6). The incidence hereby reached the lowest level since 1985 (Figure 7 and Figure 8) representing a

decrease of 17% compared to 2002 and a five-fold decrease relative to the peak year 1997.

Compared to the previous year the overall figure constitutes a 33% decline for *S. Enteritidis*, an 18% increase for *S. Typhimurium* and an 8% decline for the group of other serotypes.

In 2003, the number of *S. Enteritidis* episodes was 737 (13.7 cases per 100,000, Table A6). The phage type distribution among 473 randomly selected *S. Enteritidis* isolates from human infections is presented in Table A8. The most common phage types were PT4 (32%), PT8 (24%), PT21 (12%), PT1 (9%) and PT14b (6%). The proportion of the two previously dominating types, PT8 and PT6, continues to decrease while PT4 continued its increase and is now

the dominating phage type. Figure 10 shows the geographical distribution of the *S. Enteritidis* cases.

The number of *S. Typhimurium* episodes was 450 (8.4 per 100,000) in 2003. Figure 11 shows the geographical distribution of *S. Typhimurium* cases and the phage type (DT) distribution of 445 cases is presented in Table A9. The most common phage types were DT104 (12.7%), DTU302 (12.4%), DT120 (10.7%) and DT170 (9.6%). The many DTU302 cases are explained by a single large outbreak. A total of 65 DT104 and DT104b cases were registered in 2003 and 49 (75.4%) of these were caused by multi-drug resistant strains. Fifteen of the 65 cases (23.0%) were acquired abroad (Figure 9) and all of these were multi-drug resistant.

The remaining 526 (9.8 cases per 100,000) *Salmonella* cases were distributed among 88 different serotypes. The most commonly encountered of these serotypes were *S. Virchow* (40 cases), *S. Agona* (38 cases), *S. Dublin* (34 cases), *S. Newport* (27 cases), *S. Derby* (26 cases), *S. Infantis* (26 cases), *S. Stanley* (26 cases) and *S. Uganda* (19 cases)(Table A7).

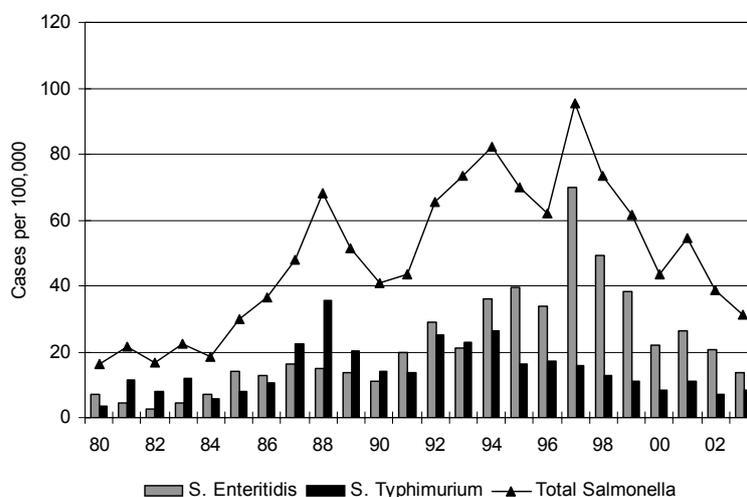


Figure 7. Registered cases of human salmonellosis in Denmark, 1980-2003.

Source: SSI

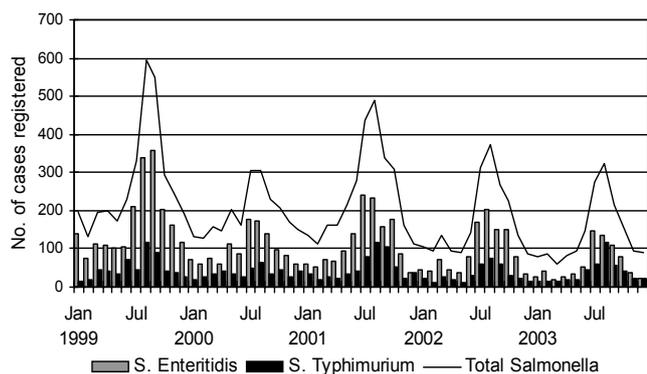


Figure 8. Seasonal variation in registered cases of human salmonellosis, 1997-2003.
Source: SSI

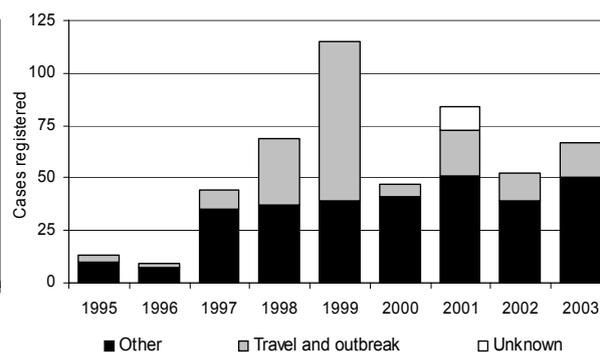


Figure 9. Registered cases of human *S. Typhimurium* DT104 (including DT104b) in Denmark, 1995-2003.
Source: SSI

Outbreaks of zoonotic gastrointestinal infections

In Denmark, outbreaks of food- and water-borne infections caused by zoonotic agents are reported in three different systems. First, general practitioners and hospitals are obligated to notify all infections suspected to be food-borne, without awaiting microbiological analysis. These early notifications of suspected outbreaks are submitted to the Regional Medical Officer of Health with a copy to the Department of Epidemiology at SSI (Table 7). Secondly, gastrointestinal

pathogens identified at clinical microbiology laboratories are reported to the Unit of Gastrointestinal Infections at SSI, which houses the reference laboratory for enteric pathogens and is in charge of the laboratory surveillance system (Table 8). Thirdly, individuals who experience food poisoning may report these incidents to the RVFCA. Such reports and results of the outbreak investigations are collated at DVFA (Table 9). No systematic evaluation of the overlap between these three parallel systems exists.

In general, there were fewer outbreaks reported with *Campylobacter* and *S. Enteritidis*, but more with *S. Typhimurium* in 2003 than in the preceding years. This is in accordance with the general trend in the number of infections with these types of bacteria. Outbreaks reported by physicians decreased from 81 in 2002 to 48 in 2003 (Table 7). Twelve outbreaks with a total of 103 laboratory confirmed cases were found through the laboratory based surveillance system (Table 8). This compares to 61 cases in 2002. An investigation carried out

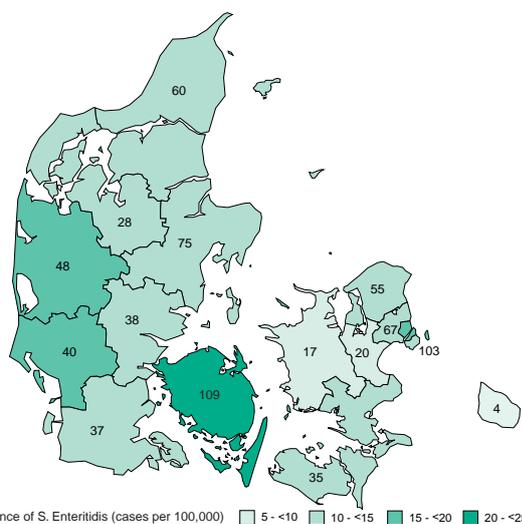


Figure 10. Geographical distribution of the number of cases per county and incidence of human infections with *S. Enteritidis*, 2003.
Source: SSI

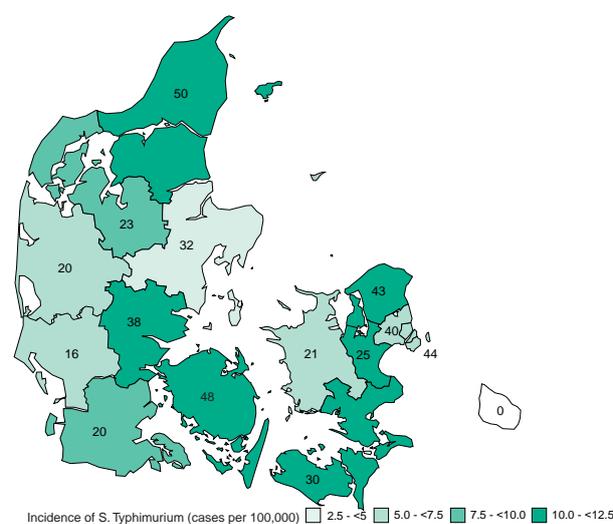


Figure 11. Geographical distribution of the number of cases per county and incidence of human infections with *S. Typhimurium*, 2003.
Source: SSI

in 2003 into family outbreaks occurring among cases registered in the laboratory based surveillance system showed that a substantial and otherwise not registered number of these occur. Such outbreaks are not included in Table 8.

In 2003, 27 outbreaks reported by the RVFCA were investigated. Of these, nine outbreaks were caused by zoonotic bacteria, eight outbreaks were of unknown cause and in 10 outbreaks other agents were identified as the cause (Table 9).

During 2003, several outbreaks with *S. Typhimurium* occurred. Many of these were first detected on the basis of specific resistance patterns (Table 8). The largest outbreak took place during a period of three weeks in the summer and was traced to the

buffet served in a restaurant popular among tourists. It involved 40 laboratory verified cases from Denmark and 22 and four from Sweden and Norway respectively. Based on the attack rate among the patrons it was estimated that the true number of cases was around 400. The strain was of phage type U302 with the resistance profile: Amp, Strep, Sulfa, Tet. The outbreak strain was also isolated from a member of staff from the kitchen in the restaurant. This fact in combination with other evidence strongly indicated that this person was, at least in part, responsible for propagating the outbreak. Another large nationwide outbreak was caused by *S. Enteritidis* PT8 and also had food at a single restaurant as its source. This outbreak involved a

minimum of 50 persons of which 10 were laboratory confirmed. The outbreak took place during the course of a large music festival and was due to cross contamination in the kitchen at a time with an unusual high number of patrons.

Sources of human salmonellosis

To obtain a better understanding of the dynamics of the occurrence of human *Salmonella* infections, the DZC has applied a mathematical model to estimate the contribution of the major animal and food sources to human cases of salmonellosis. The model is based on a comparison of the number of human cases caused by different *Salmonella* sero- and phage types with the prevalence of the *Salmonella* types isolated from the

Table 7. Clinical based surveillance of suspected outbreaks of food-borne zoonotic diseases notified to SSI, 2003.

Zoonotic pathogen	No. of outbreaks	General outbreaks		Outbreaks within the household	
		Suspected source	No. of outbreaks	Suspected source	No. of outbreaks
<i>S. Enteritidis</i>	4	beef, tuna burger	4	eggs, fried plaice with lobster sauce	4
<i>S. Typhimurium</i>	1	buffet meal	4	beef, eggs	4
<i>S. Typhimurium</i> DT 104	1	various food items	0		0
Other zoonotic <i>Salmonella</i> spp.	2	chicken	0		0
<i>Campylobacter</i>	5	beef	7	eggs, chicken, kebab, beef	7
<i>Yersinia</i>	2	buffet	0		0
Virus or food toxin	1	fish	0		0
Unknown	12	beef, pizza, scallops, veal, chicken	5	eggs, beef, sausages	5
Total					

Source: SSI

Table 8. Outbreaks identified in the laboratory-based surveillance of zoonotic diseases, SSI, 2003.

Occasion and pathogen	Estimated no. of cases	Confirmed cases	Suspected source
General outbreak (music festival), <i>S. Enteritidis</i>	>50	10	Chinese restaurant
Restaurant, <i>S. Enteritidis</i>	8	2	Tiramisu
Regional outbreak, <i>S. Ibadan</i>	?	3	Imported tomatoes
Regional outbreak, <i>S. Typhimurium</i>	?	4	Butcher shop
General outbreak, <i>S. Typhimurium</i> , DT40	?	7	?
Private party, <i>S. Typhimurium</i> , DT104	>20	2	Sirloin served at party
Private party, <i>S. Typhimurium</i> , DT104	>30	2	?
General outbreak, <i>S. Typhimurium</i> , DT170	?	15	Slaughterhouse
General outbreak, <i>S. Typhimurium</i> , DT130	?	5	?
General outbreak, <i>S. Typhimurium</i> , U302	400	40	Restaurant, buffet
General outbreak, <i>S. Uganda</i>	?	17	?
Regional outbreak, <i>Yersinia enterocolitica</i>	?	8	Butcher shop

Source: SSI

Table 9. Outbreaks of food-borne zoonotic diseases caused by zoonotic bacteria registered by the RVFCA, 2003.

Zoonotic pathogen	Total no. of outbreaks	Total number of sick persons	Suspected source (no. of outbreaks)	Confirmed by culture in foodstuffs/patients
Salmonella Enteritidis	4	26	Foie gras, cheese and salad (1) Sandwiches with salad, eggs, shrimps and dressing (1) Dish with buttermilk (1) Chicken salad (1)	no / yes no / yes yes / yes no / yes
Salmonella Typhimurium	2	68	Buffet (1) Different meat products (1)	yes / yes no / yes
Yersinia enterocolitica	1	8	Fresh pork meat or meat products	no / yes
Virus	2	37	Different meat products and salads (1) Veal steak, potatoes, sauce and cream dessert (1)	no / yes no / yes
Other agents	10	74	Many different foodstuffs involved	
Unknown	8	191	Many different foodstuffs involved	
Total	27	404		

Source: DVFA

various animal-food sources, weighted by the amount of food source consumed. For 2003, we also included data on antimicrobial susceptibility testing of *S. Typhimurium* isolates from animals, food and humans.

In 2003, the estimated mean number of human cases (per 100,000 inhabitants) that could be attributed to various sources, was as follows: table eggs: 5.0; broilers: 0.7; pork: 3.8; turkeys: 0.1; ducks: 0.4; beef: 0.3; imported poultry products: 4.3; imported beef: 0.9; imported pork: 0.3; cases related to outbreaks: 1.4; travel: 9.8 (see comment below); unknown source: 5.0 (Figure 12). Figure 13 shows the estimated number of cases caused by three major sources of infection (broilers, eggs and pork) from 1988 to 2003. Compared to 2002, the number of egg-associated cases continued to decline, which is believed to be a result of the surveillance and control programme implemented in the table-egg production. In contrast, cases related to domestically produced pork increased. The reason for this was at the

time of finishing this report still being investigated, but the previously described rise in the number of sero-positive pig herds may be part of the explanation.

The number of cases reported, as having acquired their infection abroad is known to be underreported. In previous years, the number of travel-related cases among those with unknown travel history was estimated using data from cases with a known travel history (i.e. yes or no to travel). In 2003, however, this approach proved extremely difficult as a very high proportion of cases (71%) had no information regarding travelling. Furthermore, this proportion varied between *Salmonella* types. For instance, almost all cases with an antibiotic-resistant *S. Typhimurium* infection had travel information, whereas this information was missing for 77% of the *S. Enteritidis* infections. We, consequently, based our estimation of the total number of travel-associated cases in 2003 on data from the previous years being aware though that this may not be very precise. For

2003, we estimated that approximately 526 (9.8 per 100,000) cases were travel associated. Of these, 281 cases had positively reported travelling before disease onset.

Specifically, for the 450 reported *S. Typhimurium* cases, 89 were estimated to be associated with travelling and 53 were estimated to be outbreak related. Of the domestically and sporadically occurring cases, 198 were associated with Danish produced food and 61 with imported food, whereas the remaining 49 cases had an unknown source of origin. Based on the antimicrobial susceptibility testing, it was furthermore estimated that 10% of infections from Danish produced food were multi-drug resistant (resistant to four or more drugs), 43% resistant (resistant to less than four drugs) and 47% susceptible. The same proportions for the imported food were 49%, 40% and 11%, respectively. Overall, this indicates that the majority of multi-drug resistant *S. Typhimurium* infections were acquired from food produced outside Denmark.

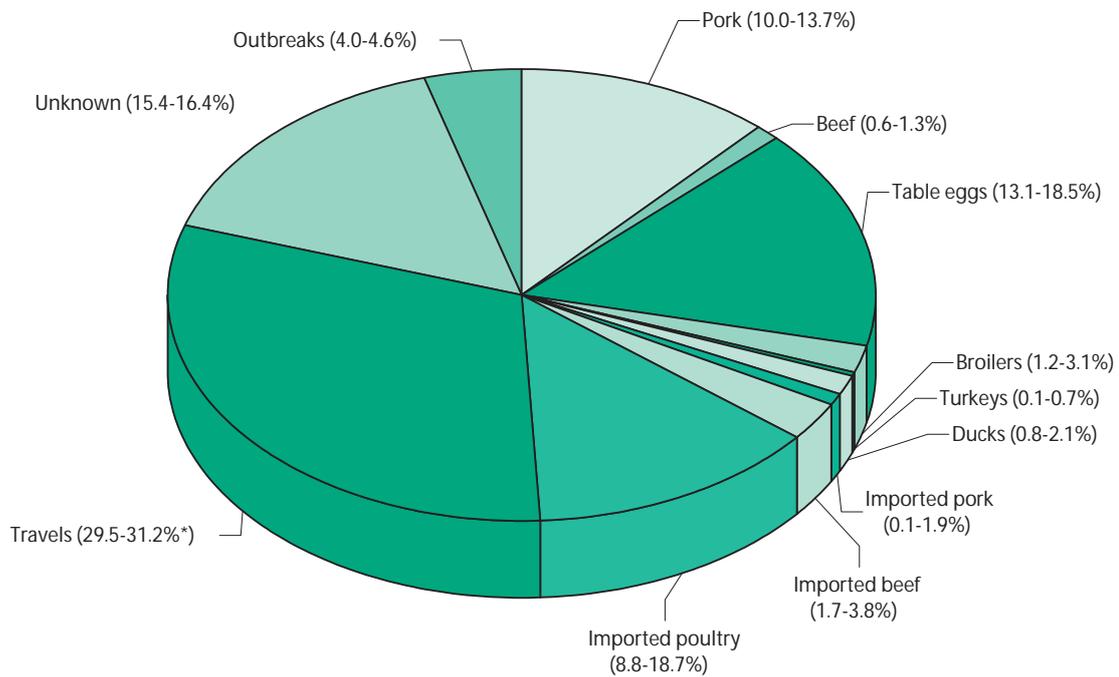


Figure 12. Estimated sources of 1,713 cases of human salmonellosis in Denmark, 2003. The estimated mean number of cases per source: 271 from table eggs, 526 travel associated*, 230 from imported poultry, 202 from pork, 13 from imported pork, 4 from turkeys, 36 from broilers, 17 from beef, 48 from imported beef, 74 from outbreaks and 271 of unknown source. * Estimate should be interpreted carefully, since data concerning travel history were very poor in 2003
Source: DZC

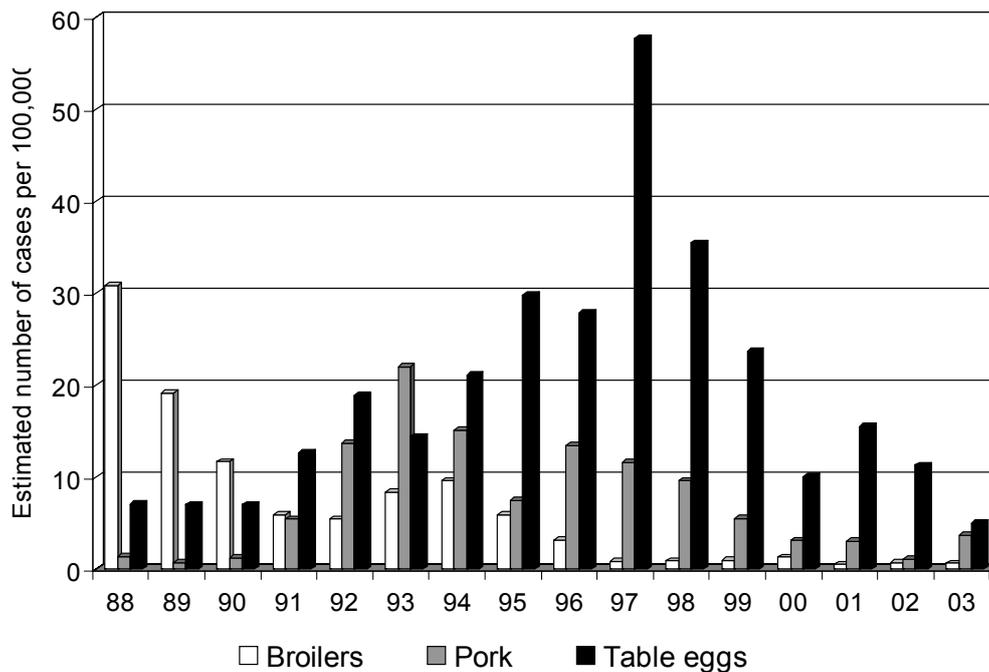


Figure 13. Estimated major sources of human salmonellosis in Denmark, 1988-2003.
Source: DZC

2. Campylobacter spp.

Poultry

In 2003, the national surveillance of *Campylobacter* spp. in broilers was continued. Ten cloacal swabs were collected from each broiler flock/batch at the time of slaughter (a flock may be slaughtered in several batches) and cloacal swabs from individual flocks/batches were pooled to one sample. A total of 5,150 pooled samples were analysed by use of a PCR detection method. Overall, the prevalence was 35.0% (Table A1) with percentages of positive batches per month ranging from 11.4% to 78.0% (Figure 14). No hens, ducks or turkeys were tested for *Campylobacter* in 2003.

As part of the monitoring programme for the occurrence of antimicrobial resistance in zoonotic bacteria from broilers (DANMAP), one flock from each broiler house in Denmark was examined for *Campylobacter*. Each sample, consisting of a pool of 10 cloacal swabs, was analysed by conventional microbiological methods. Out of 349 investigated samples, 113 (32.4%) were found positive for *Campylobacter*. Of

these, 93.0% were identified as *C. jejuni*, 6.2% were *C. coli* and one isolate (0.9%) was *C. Lari*.

Pigs and cattle

As part of the DANMAP programme caecal contents were sampled at slaughterhouses and examined for thermophilic *Campylobacter*. In pigs the prevalence was 93.4% (Table A3) and in cattle 63.6% (Table A4).

Wildlife and pet animals

Monitoring of *Campylobacter* in wild mammals and birds, previously carried out by the DFVF, was discontinued in 2002.

As in 2002, pet samples were not routinely monitored for *Campylobacter* in 2003 and only samples submitted specifically for *Campylobacter* analysis (i.e. clinical cases) were examined. *Campylobacter* were found in seven of 14 samples from dogs and in one of six examined cats (Table A5).

Products from retail outlets

In 1996, the DVFA established a national surveillance programme for thermophilic *Campylobacter* spp. in retail foods. This programme was continued in 2003 using a new semi-quantitative method based on pre-enrichment in Bolton broth followed by plating on modified charcoal cephaloridine desoxycholate agar or Abeyta-Hunt-Barker agar.

In 2003, the surveillance comprised mainly samples of raw chicken meat and gas-packed vegetables, but also a few samples of turkey meat, pork and beef. In total, 1,360 retail samples were analysed, of which 563 samples were raw chicken meat. The prevalence of thermophilic *Campylobacter* spp. in raw chicken and in turkey meat were 37.3% and 42.0%, respectively (Tables A1-A2). The numbers of thermophilic *Campylobacter* spp. in chilled and frozen chicken meat are shown in Figure 15. Thermophilic *Campylobacter* spp. were not found in pork and beef (Tables A3-A4), nor in vegetables comprising 501 gas-packed products and 47 non-gas-packed products.

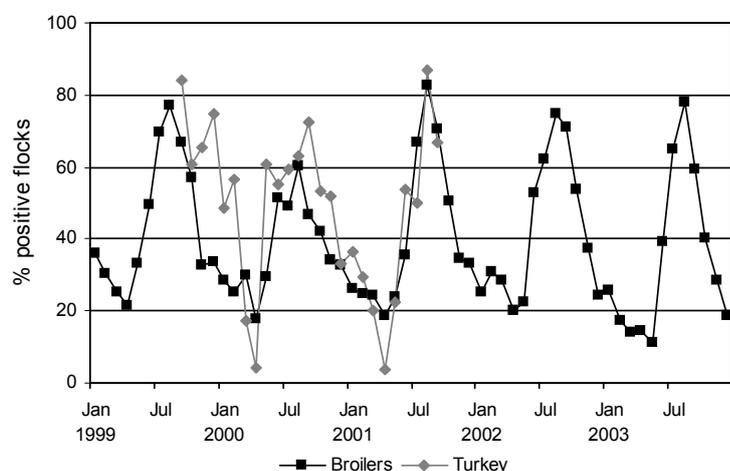


Figure 14. Percent *Campylobacter* positive poultry flocks, 1999-2003
Source: DFVF

Campylobacteriosis in humans

Even though the number of human campylobacteriosis cases decreased markedly in 2003, it remains the single most important cause of bacterial gastrointestinal disease in Denmark. There were 3,542 laboratory confirmed

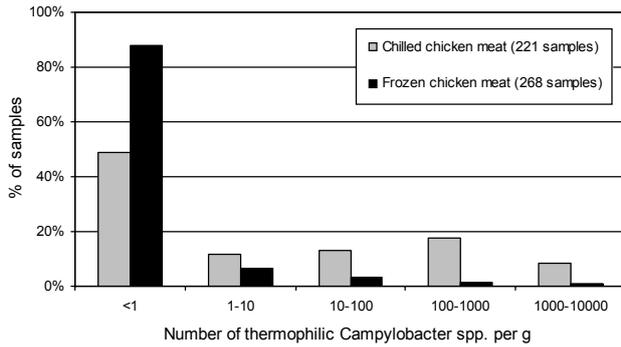


Figure 15. The number of thermophilic *Campylobacter* in Danish produced and imported chicken products from retail outlets, 2003.

Source: DVFA

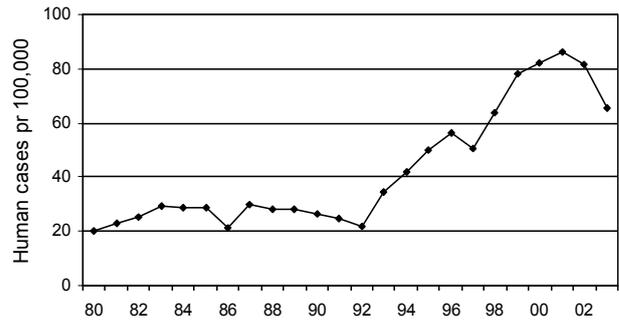


Figure 16. Incidence per 100,000 of human campylobacteriosis in Denmark, 1980-2003.

Source: SSI

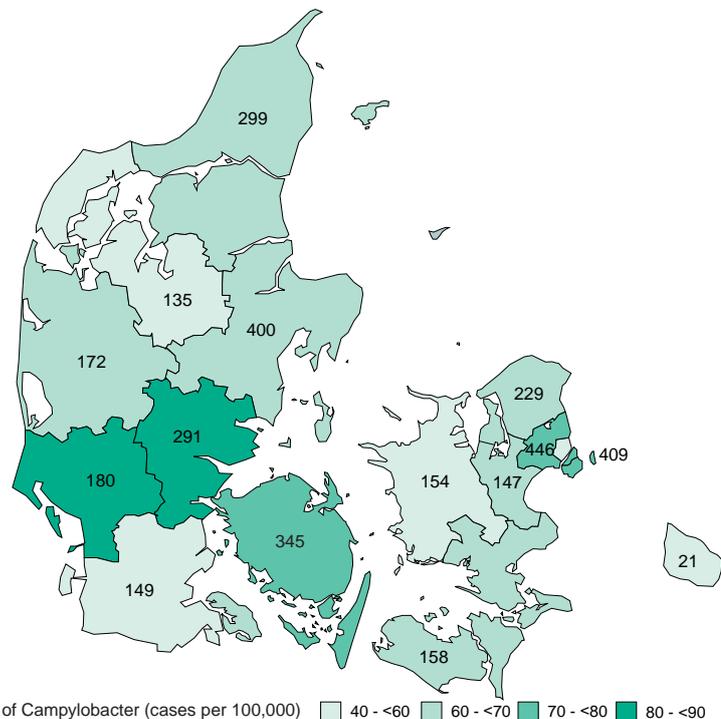
episodes of campylobacteriosis in 2003 (65.8 cases per 100,000 inhabitants). The number of human *Campylobacter* infections rose continuously from 1992 to 2001 (Figure 16), but dropped 5% in 2002 and a further 19% in 2003. Consumption of poultry and poultry products is believed to be the primary source of human campylobacteriosis in Denmark, though other sources also exist. Over the last one to two years an effort has been made by the poultry industry, in collaboration with the DVFA and the DFVF, to reduce the number of fresh retail chickens contaminated with *Campylobacter* (see p. 20). It is thought that the observed decline in the number of human infections is attributable to this effort. It is assumed that approximately three quarters of the infections are domestically acquired. The geographical distribution of human infections caused by *Campylobacter* spp. is shown in Figure 17.

Outbreaks of human campylobacteriosis are relatively rare. They are identified and recorded in the same manner as *Salmonella* outbreaks and summarised in Tables 7, 8 and 9.

Serotyping of *Campylobacter*

Isolates of *C. jejuni* and *C. coli* were serotyped using the 'Penner serotyping scheme' for heat-stable antigens. *C. jejuni* was the predominant species among humans, poultry and cattle. Serotype 2, serotype 1,44 and the 4-complex were the most common *C. jejuni* serotypes among human isolates (Table 10). These serotypes are also common in

other sources of human infections. *C. coli* is the prevailing species in pigs, whereas *C. coli* accounts for less than 10% of the thermophilic *Campylobacter* species in humans, broilers and cattle. In 2003, 6.5% of the human isolates that were speciated were *C. coli* and the remaining *C. jejuni*. The most common *C. coli* serotypes in pigs were serotype 30 and 5 (Table 11). Only few *C. coli* isolates were obtained from other sources.



Incidence of Campylobacter (cases per 100,000) 40 - <60 60 - <70 70 - <80 80 - <90

Figure 17. Geographical distribution of the number of cases per county and incidence of human campylobacteriosis, 2003.

Source: SSI

Table 10. Serotype distribution (%) of *Campylobacter jejuni* from humans, food and animals, 2003.

Serotype	Human n = 115	Cattle n = 56	Broilers n = 96	Food n = 80
2	27.8	17.9	21.9	18.8
4-complex ^{a)}	24.3	33.9	14.6	22.5
1.44	10.4	5.4	9.4	7.5
5	7.0	0.0	2.1	2.5
19	5.2	5.4	3.1	1.3
11	3.5	3.6	2.1	6.3
23.36	3.5	14.3	1.0	1.3
6.7	2.6	1.8	8.3	3.8
21	2.6	0.0	2.1	1.3
53	2.6	0.0	2.1	1.3
Others	7.0	14.3	21.9	18.8
Not typable	3.5	3.6	11.5	15.0
Total	100	100	100	100

a) 4-complex: Reaction with one or more of the following antisera: 4, 13, 16, 43, 50, 64, 65.
Source: DFVF

Table 11. Serotype distribution (%) of *Campylobacter coli* from humans, food and animals, 2003.

Serotype	Human n = 8	Pigs n = 119	Food n = 21
30	12.5	24.4	4.8
5	0	16.0	0
46	0	14.3	9.5
24	25	10.9	9.5
59	12.5	6.7	0
34	0	5.9	14.3
54	12.5	5.0	4.8
56	12.5	5.0	0
25	0	2.5	4.8
49	12.5	2.5	0
Others	12.5	4.2	38.1
Not typable	0	2.5	14.3
Total	100	100	100

Source: DFVF

Strategy for preventing campylobacteriosis in humans

In 2003, the Danish Ministry of Food, Agriculture and Fisheries adopted a strategy against *Campylobacter* that focuses on *Campylobacter* in broilers, as the first step. The strategy was developed in collaboration between DVFA, DFVF, The Consumers Board and the DPC. It was prepared on the basis of experiences from the other Nordic countries, results from research projects in the primary production, and based on the results from a risk assessment prepared by the DFVF (previously DVFA, www.fdir.dk).

The strategy aims at reducing the number of human cases by:

- Reducing the prevalence of *Campylobacter* infected flocks entering the slaughterhouses by improving biosecurity in and around the poultry houses. To encourage implementation of hygiene and biosecurity measures at the farms, an economic incentive was introduced by the industry in 1998, rewarding farmers supplying *Campylobacter* negative flocks. This bonus was increased in 2003, and compliance with the industry code of practice was reinforced.
- Reducing the number of *Campylobacter* in broiler meat after slaughter. In 2003, this was attempted by allocating *Campylobacter* negative flocks to production of fresh chilled products and positive flocks to production of frozen products.
- Preventing cross-contamination in domestic kitchens by educating consumers. Besides providing consumers with general advices on food hygiene, the DVFA initiated a specific consumer campaign on *Campylobacter* in February 2003. This campaign was directed at younger people, and focused specifically on how to prevent cross-contamination in domestic kitchens.

In 2003, one of the largest Danish poultry producers marketed a *Campylobacter* free fresh chilled chicken. The production is based on testing in the flocks a week before slaughter and intensive testing of the flock, when it enters the slaughter line. Furthermore, the industry is currently testing methods for reducing the concentration of *Campylobacter* in fresh chicken meat by a mild heat treatment with steam and ultrasound.

The *Campylobacter* strategy is a voluntary strategy and no regulations concerning *Campylobacter* have been prepared. However, the DVFA has asked the slaughterhouses to provide data as to which extent *Campylobacter* negative flocks have been allocated to be used for chilled products. The implementation of the strategy has coincided with a significant reduction in the number of human cases of campylobacteriosis. The prevalence of positive flocks decreased from 43% in 2002 to 35% in 2003 and is most likely due to reinforcement of strict hygiene and biosecurity measures. It is also likely that the practice of allocating negative flocks to chilled products and positive flocks to frozen products, although not totally consistent, has contributed to the reduction in human cases, and that the effect of this allocation was strengthened by the lower prevalence of positive flocks.

3. Yersinia enterocolitica

Pigs

As part of the DANMAP programme, caecal contents were sampled from randomly selected pig herds at slaughterhouses and examined for *Yersinia enterocolitica*. In 2003, a total of 497 animals were tested and 63 (13%) were found positive for *Y. enterocolitica* (Table A3).

Products from retail outlets

Analysis of the presence of *Y. enterocolitica* in meat and meat products at the retail level is not part of the routine surveillance carried out by the DVFA and information on the prevalence of this organism in various types of food is, therefore, scarce. In 2003, no samples were analysed.

Yersiniosis in humans

A total of 245 cases of infection with *Y. enterocolitica* were registered in year 2003 (4.5 cases per 100,000 inhabitants), which is 2% more than in 2002. Overall, the number of infections with *Y. enterocolitica* has decreased

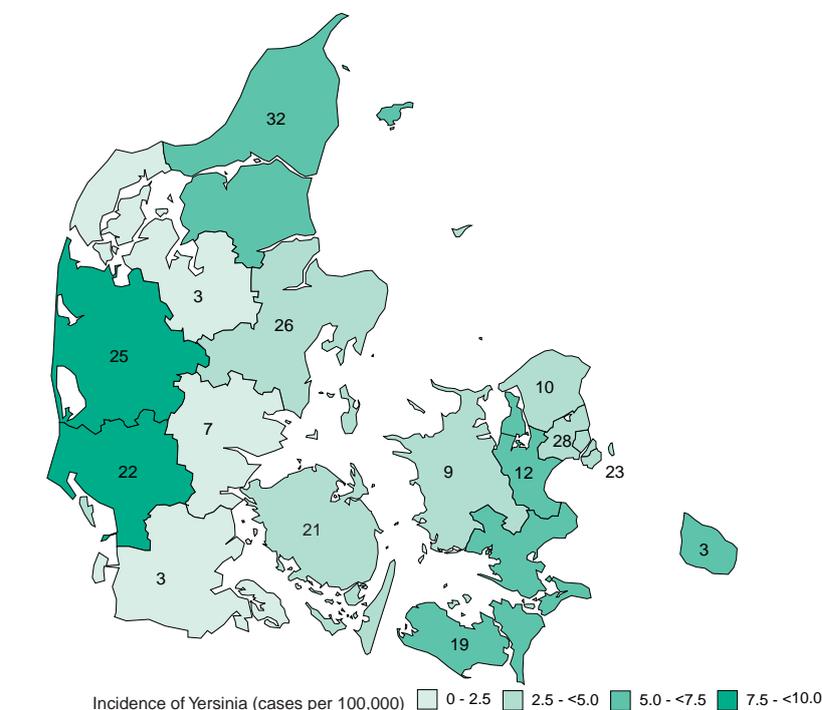


Figure 19. Geographical distribution of the number of cases per county and incidence of human yersiniosis, 2003. Source: SSI

steadily since 1985 where more than 1,500 cases were reported (Figure 18, and Table A6). As in previous years, the majority of isolates (95%) were serotype O:3. The vast majority of the infections were domestically acquired and most of the patients were children (the median age was 10). The

primary source of yersiniosis in Denmark is thought to be pork. The geographical distribution of the human *Y. enterocolitica* cases in 2003 is presented in Figure 19.

Outbreaks with *Y. enterocolitica* are rare, but during August 2003 a cluster of patients appeared in North Jutland County. A case-control study involving eight patients and 16 age, gender and municipality matched controls found a specific butcher shop to be associated with disease and furthermore implicated consumption of wiener/cocktail sausages and ground pork. Samples taken at the premises were negative and the outbreak resolved within a few weeks.

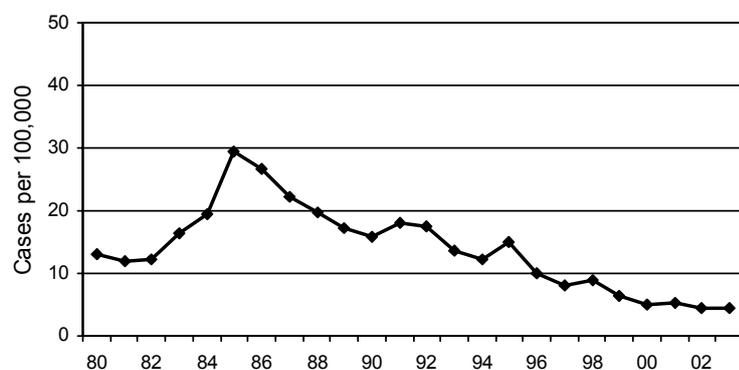


Figure 18. Incidence per 100,000 of human yersiniosis in Denmark, 1980-2003. Source: SSI

4. *Listeria monocytogenes*

Products from retail outlets

Data describing the occurrence of *Listeria monocytogenes* in food at the retail level in Denmark in 2003 were reported by the RVFCA to the DVFA (Table 12). From 2001 to 2002, the number of samples containing more than 100 *L. monocytogenes* per gram decreased from 2% to 0% in gravad, smoked, salted, not heat-treated or slightly heat-treated fish products. Otherwise no major changes were observed.

It should be noted that the number of samples tested for *L. monocytogenes* has declined, especially in the group of heat-treated products and products containing mayonnaise (Table 12). This is a reflection of the change in strategy in the use of microbiological analyses at the retail level. The number of samples exceeding 100 cfu/g is still low and not significantly different from 2002.

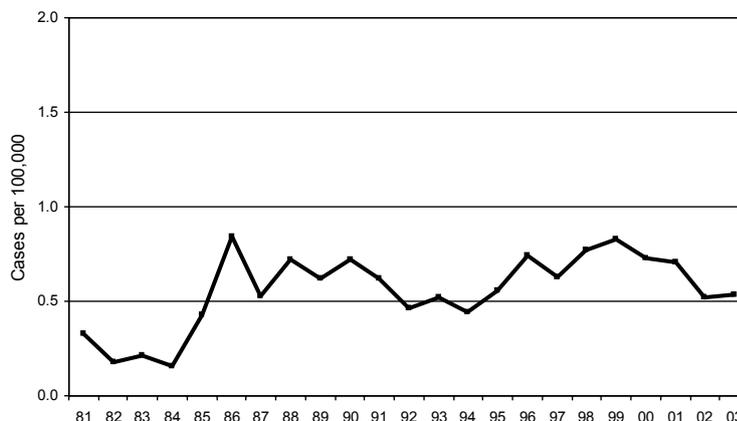


Figure 20. Incidence per 100,000 of human listeriosis in Denmark, 1981-2003.
Source: SSI

Listeriosis in humans

In 2003, 29 cases of listeriosis were registered (Table A6 , Figure 20). Nineteen cases presented with septicaemia, four with meningitis, and four were classical maternofoetal cases. Two cases presented with pleuritis/peritonitis. Geographically, the

patients were spread all over the country. Based on serogrouping, riboprinting and PFGE typing no clustering of cases could be identified. Eleven cases were caused by serogroup 1 and 17 cases by serogroup 4, while the serogroup was undetermined for one case.

Table 12. Percentage distribution of the number of *Listeria monocytogenes* in selected foods, sampled at retail level in Denmark by the RVFCA in 1999-2003.

	2003		2002		2001		2000	
	Number of samples	Percent of samples with cfu>100 per g	Number of samples	Percent of samples with cfu>100 per g	Number of samples	Percent of samples with cfu>100 per g	Number of samples	Percent of samples with cfu>100 per g
Heat-treated products of pork, beef, chicken and turkey handled after heat treatment	799	0.1	1,331	0.2	2,952	0.2	3,861	0.4
Preserved, not heat-treated or slightly heat-treated products of pork, beef, chicken and turkey	77	2.6	244	0.8	115	0.9	162	2.5
Gravad, smoked, salted, not heat-treated or slightly heat-treated fish products	222	0.0	157	0.0	152	2.0	120	0.8
Sprouts or sliced vegetables	84	0.0	71	0.0	87	0.0	160	0.0
Products containing mayonnaise	225	0.0	573	0.3	1,664	0.1	2,163	0.2
Cheese and cheese products	8	0.0	34	0.0	31	0.0	44	0.0
Ready to-eat dishes	284	0.0	482	0.0	1,239	0.2	1,410	0.2

Source: DVFA

5. Verocytotoxigenic Escherichia coli

Cattle

The occurrence of verocytotoxin producing *Escherichia coli* O157 (VTEC O157) has been monitored in cattle by DFVF since 1997 by investigating faecal samples from slaughter calves. The samples are collected at the slaughterhouses and as part of the DANMAP programme. In 2003, VTEC O157 was detected in 7.1% (15/212) of faecal samples from slaughter calves.

Another study of the occurrence of five serogroups of *E. coli* (O26, O103, O111, O145, and O157) was conducted by the DFVF in collaboration with the DVFA. A total of 750 faecal samples from cattle at slaughter (primarily slaughter calves and cows originating from dairy farms). The samples were analysed by methods, which included an immunomagnetic separation step. In this study, a total of 58 isolates were verified as belonging to serogroups O26 (14), O103 (4), O111 (0), O145 (5), and O157 (35). Three O26 isolates (0.4%) and 23 O157 (3.1%) isolates were identified as verocytotoxin producers.

Products from retail outlets

Analysis of the presence of VTEC in meat and meat products at the retail level is not part of the routine surveillance carried out by the DVFA. In 2003, only few samples were analysed and none were found positive.

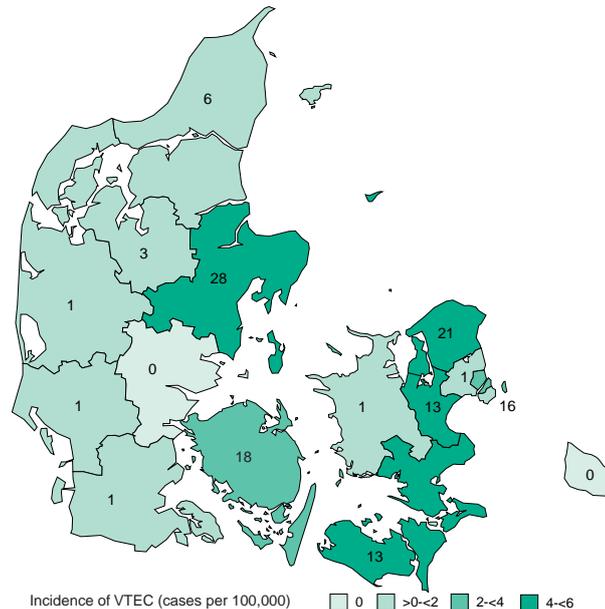


Figure 22. Geographical distribution of the number of cases per county and incidence of human infections with verocytotoxigenic *E. coli* (VTEC), 2003. Source: SSI

Human infections

In 2003, 128 episodes of VTEC infections were identified (incidence 2.4 per 100,000). Of these, 27 cases (22%) were O157 (Figure 21, Table 13 and A6). This represents an overall decrease in the number of diagnosed episodes of 11% compared to 2002 and it is the first time since the registration of cases began in 1997 that the number has not increased.

Haemolytic uraemic syndrome (HUS) has been notifiable in Denmark since spring 2000. In 2003, three cases were reported, none of them fatal. From these cases VTEC strains of serotype O157:H7 and O157:H+ were isolated. All strains were *vtx1* negative and *vtx2* positive.

The method used for microbiological diagnosis was slide agglutination of suspect colonies with OK-antisera against the most common VTEC and EPEC serotypes. In counties where diagnostics are performed by SSI (covering approximately half the Danish population) colony hybridisation using probes for verocytotoxin- and *eae*-genes was used for screening of all diarrhoeagenic *E. coli* prior to slide agglutination. The geographical distribution of human infections with VTEC is presented in Figure 21. The very low incidence seen in several counties is by and large a reflection of the differences in diagnostic practices mentioned above.

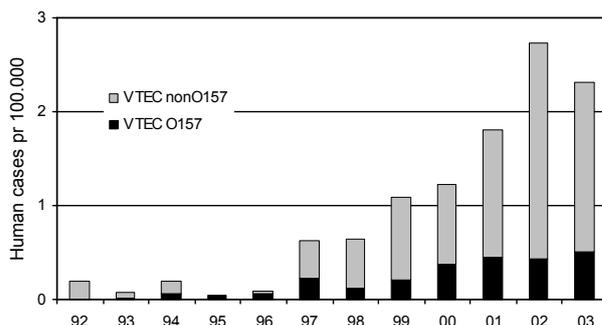


Figure 21. Incidence of human infections with verocytotoxigenic *E. coli*, 1992-2003. Source: SSI

Table 13. Human VTEC serotypes resulting in more than 5 episodes, 2003.

Serotype	Number of episodes
O157	27
O103	23
O146	13
Rough	13
Other	52
Total	128

Source: SSI

6. Transmissible Spongiform Encephalopathy

The Danish Surveillance programme continued throughout 2003. The major changes included the implementation of TSE Regulations/Decisions (nr. 260/2003, 652/2003, 1053/2003, 1128/2003, 1139/2003, 1234/2003, 1809/2003, 1874/2003, 100/2003, 1915/2003). The most important consequence of this was, that from 1st of October 2003, all fallen sheep and goats older than 18 months must be tested.

Cattle

BSE testing of samples from normal slaughter animals is performed at three approved private laboratories in Denmark. Two of these laboratories use the Enfer Test (ELISA) with spinal cord as test material, while the third uses the Prionics Check Test (western blotting) with brain stem as test material. All

risk animals in Denmark are tested with western blotting (bovine categories presented in Table 14). Fallen stock is generally tested at the approved private laboratory using western

blotting. The remaining samples from the Danish cattle population at risk are examined at the DFVF. Furthermore, a small part of the fallen stock tests are examined at the DFVF to maintain a testing routine, since it is the national reference laboratory. The DFVF also receives clinically suspected animals for diagnosis and performs confirmatory testing on initially positive samples from the private laboratories.

In 2003, Denmark tested a total of 289,702 bovine animals. Among these, two were positive for BSE, including one clinical case and one healthy slaughter animal (Table 14). Furthermore, BSE was diagnosed in an animal exported to Portugal.

The geographical distribution of BSE positive herds in Denmark, 2000-2003 is shown in Figure 23. The case exported to Portugal is not included in Table 14 and Figure 23, because cases of BSE are reported to EU from the country of diagnosis.

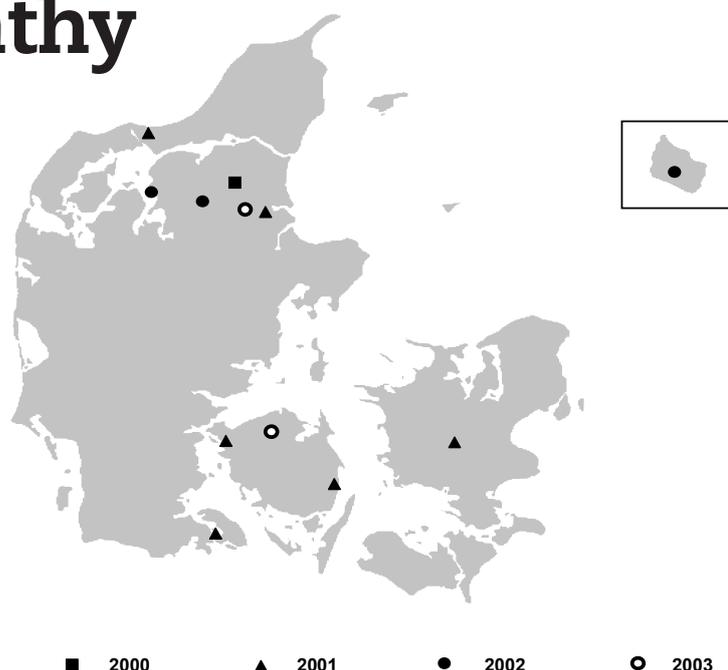


Figure 23. Geographical distribution of BSE positive herds in Denmark, 2000-2003
Source: DVFA

Table 14. The BSE surveillance programme in Denmark, 2003.

Active surveillance	No of tests	No. of positive animals
Healthy slaughtered animals (<30 mo.)	250,558	1
Risk categories		
Emergency slaughters (>24 mo.)	1,739	0
Animals>24 mo., where ante-mortem inspection at the slaughterhouse reveals signs of disease or zoonotic infection	17	0
Fallen stock (>24 mo.)	35,570	0
Feed cohort investigation(animals born between Aug. 1995 and July 1997 in herds receiving feed of same origin as the first Danish BSE-case from Jan. 2000)	1,444	0
Imported UK animals	6	0
Animals from herds under restriction	330	0
Passive surveillance	No of tests	No. of positive animals
Animals clinically suspected of having BSE	38	1
Total	289,702	2

Source: DVFA

Table 16. The TSE surveillance programme of sheep and goats in Denmark, 2003.

	No of tests sheep and goats	No. of positive tests
Active surveillance		
Fallen stock (>18 mo.)	1,672	0
Healthy slaughtered animals (>18 mo.)	962	0
Passive surveillance		
Animals clinically suspected of having TSE	8	0
Total	2,634	0

Source: DVFA

Of the three Danish-borne cases identified in 2003, one was born in August 1997, after implementation of the 1997 feed ban in Denmark. This feed ban was reinforced in 2001.

For a period of time the EU Commission has tried to influence the OIE in order to harmonise legislation for classification of the countries with regards to BSE status. At the end of 2002, the Veterinary Laboratory Agencies (VLA) in Weybridge, UK, were asked to bring forward a new way of calculating this. A mathematical model to calculate BSE prevalence in a country was presented in 2003 and discussions on the topic have since then taken place within the framework of EU. Together with the DVFA, the DFVF played an important role in the

evaluation of the model and in the discussions and negotiations on the issue.

Sheep and goats

It has been demonstrated that sheep can contract BSE under experimental conditions and there is, therefore, concern that this may also occur under field conditions though it has never been recorded. Some genotypes of sheep are resistant to scrapie and - although based on less evidence - also to BSE. The concentration of pathogenic prion in the resistant sheep is much lower than in non-resistant sheep and as a result of this, the resistant sheep will pose a minor risk to public health than the non-resistant. A survey of the genotypes present in Danish breeding sheep was performed at DFVF in 2003 (Table 15).

On the 1st October 2003 the EU Commission officially approved an extended Danish national surveillance programme for scrapie in sheep and goats. All fallen sheep and goats in Denmark are now to be tested for scrapie when delivered to a rendering plant. On account of this programme the breeding programme for resistance against scrapie in sheep has become optional for sheep breeders in Denmark.

The general procedures for BSE/scrapie testing of fallen stock are carried out as described for cattle.

Brain stem is used as material in all cases. When a rapid test show inconclusive or positive results, material is subjected to confirmatory testing at DFVF by histopathology and immunohistochemistry (IHC). Scrapie has never been detected in sheep and goats in Denmark. In future, it is the plan that if the confirmatory test is found positive for scrapie, material will be sent to the VLA for molecular typing to determine whether it is scrapie or BSE. In 2003, a total of 2,634 sheep and goats were tested for scrapie (Table 16).

Humans

Since 1997, the human form of the disease, variant Creutzfeldt-Jakob Disease (vCJD) has been notifiable in Denmark. No cases of vCJD have been reported to date.

7. Cryptosporidium Mammals

At present, 13 species of *Cryptosporidium* are regarded as valid. Of these, the most extensively studied species, *C. parvum*, has a very wide host spectrum, whereas some of the other species appear to be more host-specific. Currently, genotyping of *Cryptosporidium* species is not offered as a routine diagnostic tool in Denmark, but has been completed as a part of ongoing research projects.

In 2003, 3,584 faecal samples from mammals were screened for *Cryptosporidium*. Of the bovine samples 16% were positive for *Cryptosporidium*. Among other animal species, the occurrence of *Cryptosporidium* did not exceed 2.1%. All mammalian samples (from all age groups), submitted to DFVF for routine parasitological analysis, were screened for *Cryptosporidium*. Thus, many samples from animals outside the group of risk (i.e. young animals) were analysed, thereby decreasing the relative prevalence of *Cryptosporidium* in the samples. The relative number of

Table 15. Survey of prion protein genotypes in Danish breeding sheep. Type 1 sheep (homozygous for the ARR-allele) are resistant to scrapie, while type 3 and 4 sheep are the most susceptible. For certain breeds the ARR-allele is uncommon (10% or less).

Breed	Type 1 ARR/ARR	Type 2 ARR/AXX	Type 3 AXX/AXX	Type 4 VRQ/XXX
Dorset	17	12	4	2
Finnsheep	0	3	41	2
Gotland Pels	0	0	99	33
Danish Landrace	20	15	4	0
Leicester	25	11	3	7
Danish Marsh Sheep	33	0	0	4
Oxford down	26	12	2	0
Shropshire	4	6	44	0
Suffolk	11	20	16	10
Texel	4	16	21	1

Source: DFVF

Cryptosporidium positive samples has increased in cattle from 10.9% in 2002 to 16.0% in 2003, causing an increase in the total percentage of positive samples from 5.7% in 2002 to 7.6% in 2003.

Ongoing prevalence studies in Danish livestock have revealed *Cryptosporidium* herd prevalences of 100% in both cattle and pig herds, with an individual prevalence of approximately 33% in young animals.

The 'bovine' genotype of *C. parvum* causes the majority of *Cryptosporidium* infections in Danish cattle herds, but novel genotypes have also been detected. In Danish pig herds two distinct and apparently host-adapted genotypes of *Cryptosporidium* have been identified so far. One of these (*C. parvum* pig genotype I) has also been reported in humans, whereas the other (*C. parvum* pig genotype II) has been found in human sewage. Considering the high density of livestock in certain regions of Denmark, the widespread prevalence of *Cryptosporidium*, and the finding of zoonotic species/genotypes in both cattle and pig herds, animals constitute a major reservoir of these organisms. This gives rise to further questions concerning transmission routes under Danish conditions.

Humans

Two *Cryptosporidium* spp., the zoonotic species *C. parvum* and the anthroponotic species *C. hominis*, are responsible for most human *Cryptosporidium* infections. However, several other species have been shown to infect humans, apparently without immunosuppression being a prerequisite. So far, *C. parvum* and *C. hominis* have been detected in Denmark in addition to a few cases caused by *C. meleagridis*.

Cryptosporidiosis is not a notifiable disease in Denmark and the incidence in humans is therefore unknown. At most diagnostic laboratories in Denmark only patients with persistent diarrhoea or

a history of recent travel are routinely examined for cryptosporidiosis. At SSI 58 cases of cryptosporidiosis were diagnosed in 2003 (Table A6). Previous surveys have shown that approx. 80% of the diagnosed cases are acquired abroad. The distribution of different *Cryptosporidium* species/genotypes in the Danish human population, routes of transmission, risk factors, and potential correlation between genotype and pathogenicity are being investigated in an ongoing study.

8. *Mycobacterium bovis*/tuberculosis

In accordance with Commission Decision 99/467/EEC as amended by Decisions 2000/69/EEC, 2000/442/EEC, 2000/694/EEC Danish cattle herds have been declared officially free from bovine tuberculosis (TB) since 1980. TB in cattle is a notifiable disease in Denmark. Monitoring is performed by meat inspection, which means that all slaughter animals are examined for lesions indicative of TB. Bulls at semen collection centres are subject to pre-entry and annual intradermal tuberculin testing. The last case of TB in cattle was diagnosed in 1988.

Since December 1994, bovine tuberculosis has not been diagnosed in deer in Denmark.

Humans

Bovine tuberculosis in humans has been a notifiable disease since May 1, 2000. In 2003, one case of human tuberculosis caused by *M. bovis* was registered (Table A6). The patient was of foreign origin and had previously suffered from tuberculosis outside Denmark. The disease is believed to result from a reactivation of an earlier infection acquired abroad.

9. *Brucella* spp.

Cattle

In accordance with Commission Decision 99/466/EEC as amended by

Decision 2000/69/EEC Denmark has been regarded officially free from brucellosis in cattle since 1979.

Brucellosis is a notifiable disease, and clusters of abortions are notifiable. Monitoring is performed by examination of abortion material. Bulls are subject to serological testing before entering bovine semen collection centres. After entry they are examined annually for brucellosis.

Pigs

Boars at porcine semen collection centres are subject to pre-entry testing, followed by testing at least every 18 months and before they leave the centre. Due to serological cross-reactions with *Yersinia enterocolitica* serotype O:9 a surveillance programme for this infection has been implemented in breeding herds with monthly serologic analyses. With this programme potential false positive *Brucella* reactive herds are identified prior to testing of pigs in quarantine for export or entry into semen collection centres. Brucellosis in pigs was not recorded in 2003.

Sheep and goats

In accordance with Commission Decision 94/877/EEC Denmark has been declared officially free from brucellosis. Ovine and caprine brucellosis (*B. melitensis*) has never been recorded in Denmark. Monitoring is performed by testing for *Brucella* antibodies in blood samples from sheep and goats submitted as a part of a voluntary control programme for lentivirus. In 2003, 4,731 blood samples from 657 herds were examined.

Humans

Human *Brucella* infections are not notifiable in Denmark. At SSI 14 persons were found positive by serology in 2003 (three positive for *B. abortus*, one for *B. melitensis* and 10 for both). One was culture confirmed. No information on travel association was available.

10. *Leptospira* spp.

Leptospirosis in animals is not a notifiable disease in Denmark.

Pigs

Examination for leptospire in pigs is performed by antigen detection, culture and serology. Suspicion of leptospirosis is often based on increased incidence of abortions or other reproductive problems in a herd. In 2003, a total of 340 samples were investigated by immunofluorescence testing and leptospire were detected in one herd. This herd was infected with *L. interrogans* serovar bratislava.

Humans

Leptospirosis in humans is notifiable in Denmark. In year 2003, 28 patients were diagnosed by serology; all recovered. Infections caused by *L. interrogans* serovar ichterohaemorrhagiae were most common, but infections caused by serovar bratislava, ballum, hardjo and poi were also detected.

11. *Trichinella* spp.

Infection has not been recorded in domestic animals since 1930.

Pigs and wild boars

All pigs slaughtered at Danish export approved slaughterhouses are examined for *Trichinella* spp. in accordance with Council Directive 64/433/EEC. During 2003 samples from 22,375,420 pigs were examined, and none of the samples were found to contain *Trichinella* spp.

It is also compulsory to examine slaughtered wild boars. The DVFA was informed of 1,280 examinations, all of which were negative.

Horses

All horses which are slaughtered at Danish export approved slaughterhouses are examined for *Trichinella*

spp. During 2003, samples from 1,441 horses were examined, and none of the samples were found to contain *Trichinella* spp.

Humans

Human trichinosis is not a notifiable disease. At SSI no domestically acquired cases of human trichinosis were recorded in 2003.

12. *Echinococcus granulosus/multilocularis*

Echinococcus granulosus/multilocularis infections in all animals are notifiable. Surveillance for *E. granulosus* is performed through meat inspection. In 2003, there were no findings.

In 2003, 34 foxes were examined for *E. multilocularis* at the DFVF. None were positive.

Human infections are not notifiable. At SSI, less than 10 cases were diagnosed in 2003.

13. *Toxoplasma gondii*

Toxoplasmosis in humans is not a notifiable disease in Denmark, and the incidence of toxoplasmosis in humans is unknown. However, Denmark has a nationwide neonatal screening system for toxoplasmosis. In 2003, 64,682 newborns were tested through this system and 13 were found positive.

14. Psittacosis (Ornithosis)

At the DFVF all caged birds submitted for diagnosis are tested for *Chlamydia psittaci*. In 2003, a total of 11 birds, all of which were psittacines, were found positive for *C. psittaci*.

Humans

Human infections are notifiable in Denmark. In 2003, 14 patients were reported. The diagnosis was verified by serology for seven patients and by PCR for *C. psittaci* -DNA for four patients. For three patients the diagnosis could not be verified.

15. Rabies

Rabies is a notifiable infection in both humans and animals. The classic sylvatic rabiesvirus (lyssa virus type 1) is not found in Denmark, nor has it been reported from areas close to Danish borders for a number of years. It is, however, endemic in Greenland, where arctic foxes spread the disease to sleigh dogs and other mammals.

The European bat lyssa virus (EBL) is found in the Danish bat population and in 2003, 32 wild bats were submitted for diagnosis and three were found infected with EBL. A total of 12 other animals, including one dog, three cats, two racoons and four sheep were examined and were all found negative.

No human cases of rabies were reported in 2003, but 13 people were treated by post-exposure prophylaxis after suspected exposure in Denmark. Of these, 10 suffered from bat bites (none of the bats were examined), two from bites by racoons and one was bitten by a sheep. The two racoons and the sheep tested negative for rabies and the treatments were subsequently interrupted. In addition, 53 people were treated by prophylactic vaccination after exposure abroad to bites from animals suspected of being infected.

Antimicrobial resistance

For information on antimicrobial resistance in zoonotic bacteria we refer to the yearly report: DANMAP - Consumption of antimicrobial agents and occurrence of antimicrobial resistance in bacteria from food animals, food and humans in Denmark. The 2003 report will be available at: www.dfvf.dk or can be ordered from the Danish Zoonosis Centre (dzc@dzc.dk) by the end of June 2004.

Appendix

Demographic data

Area of Denmark: 44,000 sq km

Human population, 2003

Age group (years)	Male	Female	Total
< 1	32,990	31,179	64,169
1-4	137,025	130,862	267,887
5-14	349,582	331,408	680,990
15-24	302,343	292,812	595,155
25-44	793,861	770,805	1,564,666
45-64	709,405	702,884	1,412,289
> 65	337,217	461,134	798,351
Total	2,662,423	2,721,084	5,383,507

Source: The Statistical Yearbook 2003, Danmarks Statistik

Average number of livestock and herds in Denmark, 2003

	Livestock	Herds
Cattle	1,759,192	32,380
Pigs	12,554,538	18,567
Laying hens excl. Barn yard	3,651,883	336
Broilers	23,531,000	392
Turkeys	452,739	47
Sheep	199,060	10,101
Goats	18,817	2,283

Source: The Central Husbandry Register

Approximate total number of animals slaughtered, 2003

Pigs	22.5 mill.
Broilers	130.4 mill.
Turkeys	546,120
Cattle	574,426
Sheep, lambs and goats	75,870
Horses	2,673

Source: DVFA

Table A1. Occurrence of Salmonella and Campylobacter in the broiler production in Denmark, 2003.

	Flock level		Slaughterhouse		Retail - cuts and products of broiler meat						Note:
	Broiler breeders		Broiler flocks a)		Cuts		Not heat treated		Heat treated		
	Flocks examined	% positive flocks	Flocks examined	% positive flocks	N	% positive flocks	N	% positive samples	N	% positive samples	
<i>Salmonella</i> spp.	182	2.2	4,385	1.7	1,552	5.0	4	0.0	27	0.0	a
Danish	182	2.2	4,385	1.7	1,552	5.0	1	0.0	8	0.0	-
S. Enteritidis	-	1.6	-	0.1	-	-	-	-	-	-	-
S. Typhimurium	-	0.0	-	0.5	-	-	-	-	-	-	-
Other serotypes	-	0.6	-	1.1	-	-	-	-	-	-	-
Imported	-	-	-	-	-	-	1	0.0	19	0.0	-
Unknown origin	-	-	-	-	-	-	2	0.0	0	-	-
<i>Campylobacter</i> spp.	-	-	5,150	35.0	-	-	563	37.3	1	0.0	b
Danish	-	-	-	-	-	-	407	32.9	0	-	-
Imported	-	-	-	-	-	-	151	50.3	0	-	-
Unknown origin	-	-	-	-	-	-	5	0.0	1	0.0	-

a) Parent flocks were examined according to Table 3. Broiler flocks were monitored by sock-samples 2-3 weeks prior to slaughter and by end-product samples after slaughter.

b) Flocks investigated by cloacal swabs collected at slaughter, ten birds per flock were examined. Summed up in batches, where one flock may be slaughtered in up to six batches.

Source: DPC and DVFA

Table A2. Occurrence of Salmonella and Campylobacter in the turkey production in Denmark, 2003.

	Flock level		Slaughterhouse		Retail - cuts and products of turkey meat						Note:
	Turkey flocks		Meat cuts		Not heat treated		Heat treated				
	Flocks examined	% positive flocks	Batches examined	% positive flocks	N	% positive samples	N	% positive samples			
<i>Salmonella</i> spp.	211	5.7	271	6.3	5	0.0	5	0.0	-	a	
Danish	211	5.7	271	6.3	-	-	0	-	-	-	
S. Enteritidis	-	-	-	0.0	-	-	-	-	-	-	
S. Typhimurium	-	-	-	0.0	-	-	-	-	-	-	
Other serotypes	-	5.7	-	6.3	-	-	-	-	-	-	
Imported	-	-	-	-	-	-	0	-	-	-	
Unknown origin	-	-	-	-	5	0.0	5	0.0	-	-	
<i>Campylobacter</i> spp.	-	-	-	-	50	42.0	3	0.0	-	-	
Danish	-	-	-	-	10	70.0	0	-	-	-	
Imported	-	-	-	-	38	36.8	0	-	-	-	
Unknown origin	-	-	-	-	2	0.0	3	0.0	-	-	

a) Flocks monitored by sock samples 2-3 weeks prior to slaughter and by end-product samples after slaughter. One flock may be slaughtered in several batches.

Source: DPC and DVFA

Table A3. Occurrence of zoonotic pathogens in pigs and pork in Denmark, 2003.

Zoonotic pathogen	Herd level			Slaughterhouse		Retail				Note:
	Examined			Carcass samples		Not heat treated		Heat treated		
	Herds	Animals	%positive herds	N	%positive samples	N	%positive samples	N	%positive samples	
<i>Mycobacterium bovis</i>	18,567	22.5 mill	0.0	-	-	-	-	-	-	a
<i>Brucella abortus</i>	-	-	0.0	-	-	-	-	-	-	b
<i>Trichinella</i> spp.	18,567	22.5 mill	0.0	-	-	-	-	-	-	c
<i>Salmonella</i> spp.	14,290	578,228	3.3	34,460	1.4	183	0.6	228	0.0	d,e
<i>S. Typhimurium</i>	-	-	-	-	0.5	-	-	-	-	-
<i>S. Derby</i>	-	-	-	-	0.4	-	-	-	-	-
Other serotypes	-	-	-	-	0.5	-	-	-	-	-
<i>Campylobacter</i> spp.	259	259	93.4	-	-	50	0.0	88	0.0	f
<i>C. jejuni</i>	-	-	-	-	-	-	-	-	-	-
<i>C. coli</i>	-	-	-	-	-	-	-	-	-	-
<i>C. lari</i>	-	-	-	-	-	-	-	-	-	-
<i>Y. enterocolitica</i>	497	497	12.7	-	-	0	0.0	0	0.0	f,g

a) All slaughter pigs were examined in connection with meat inspection.

b) Serological examination of boars on admission to semen collection centres and before leaving the station.

c) All pigs slaughtered at export slaughterhouses were examined in connection with meat inspection.

d) Herds were monitored by serological testing. Herds belonging to Level 2 and 3 were defined as *Salmonella* positive.

e) At the slaughterhouse swabs are taken from three areas of the half-carcass. Five samples are pooled except at slaughter houses where less than five pigs are slaughtered per day, in which case the samples are analysed individually.

f) Herds examined by caecal samples from one animal per herd collected at slaughter (from the DANMAP-programme).

g) Isolates obtained at the retail level were not sero- or biotyped.

Source: DVFA and DFVF

Table A4. Occurrence of zoonotic pathogens in cattle and beef in Denmark, 2003.

Zoonotic pathogen	Herd level			Slaughterhouse		Retail				Note:
	Examined			Carcass samples		Not heat treated		Heat treated		
	Herds	Animals	%positive herds	N	%positive samples	N	%positive samples	N	%positive samples	
<i>Mycobacterium bovis</i>	-	574,426	0.0	-	-	-	-	-	-	a
<i>Brucella abortus</i>	-	-	0.0	-	-	-	-	-	-	b
<i>Salmonella</i> spp.	308	308	2.6	12,570	0.4	2,035	0.1	-	-	c,d
<i>S. Dublin</i>	-	-	2.3	-	0.0	-	0.0	-	-	-
<i>S. Typhimurium</i>	-	-	0.0	-	0.0	-	0.0	-	-	-
Other serotypes	-	-	0.3	-	0.1	-	0.1	-	-	-
<i>Campylobacter</i> spp.	88	88	63.6	-	-	45	0.0	12	0.0	c
<i>C. jejuni</i>	-	-	-	-	-	-	-	-	-	-
<i>C. coli</i>	-	-	-	-	-	-	-	-	-	-
Not speciated	-	-	-	-	-	-	-	-	-	-
<i>E. coli</i> (O157)	-	750	7.7	-	-	-	-	-	-	c

a) Slaughter animals examined in connection with meat inspection. Notifiable disease.

b) Bulls examined on admission to semen collection centres and annually after entry. Clusters of abortions are notifiable. Notifiable disease in cattle.

c) Herds were investigated by caecal samples from one animal per herd collected at slaughter (from the DANMAP-programme).

d) At the slaughterhouse swabs are taken from three areas of the half-carcass. Five samples are pooled except at the smallest slaughterhouses where the samples are analysed individually.

Source: DVFA and DFVF

Appendix

Table A5. Occurrence of Salmonella and Campylobacter in pet animals, wild mammals and birds in Denmark, 2003.

Zoonotic pathogen	Pet animals						Wild mammals						Wildlife birds Zoo animals					
	Dog		Cat		Others		Hare		Ruminants		Fox		Others		Water fowl		Zoo animals	
	N	%posi- tive	N	%posi- tive	N	%posi- tive	N	%posi- tive	N	%posi- tive	N	%posi- tive	N	%posi- tive	N	%posi- tive	N	%posi- tive
Salmonella spp.	20	0.0	9	0.0	19	33.3	18	0.0	9	11.1	32.0	0.0	256	5.5	148.0	4.1	259.0	6.6
S. Enteritidis	-	-	-	-	-	0.0	-	-	-	0.0	-	-	-	4.3	-	0.0	-	0.0
S. Typhimurium	-	-	-	-	-	0.0	-	-	-	0.0	-	-	-	1.2	-	3.4	-	0.4
Others	-	-	-	-	-	33.3	-	-	-	11.1	-	-	-	0.0	-	0.7	-	6.2
Campylobacter spp.	14	50.0	6	16.7	3	0.0	-	-	-	-	-	-	-	-	1.0	0.0	15.0	0.0
C. jejuni	-	14.3	-	0.0	-	-	-	-	-	-	-	-	-	-	-	-	-	-
C. coli	-	0.0	-	0.0	-	-	-	-	-	-	-	-	-	-	-	-	-	-
C. upsaliensis	-	14.3	-	0.0	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Others/not speciated	-	21.4	-	16.7	-	-	-	-	-	-	-	-	-	-	-	-	-	-

Source: DFVF

Table A6. Zoonoses in humans, 2003 - number of cases over a ten-year period.

Zoonotic pathogen	Incidence		Registered no. of cases										Note:
	2003		2003	2002	2001	2000	1999	1998	1993				
Mycobacterium bovis	0.0		1	2	4	12	2	9	-	-	-	a	
Brucella abortus/melitensis	0.3		14	16	18	-	-	-	-	-	-	b	
Trichinella spp.	-		-	-	-	-	-	-	-	-	-	c	
Salmonella spp.	31.8		1,713	2,071	2,918	2,308	3,268	3,880	3,811	-	-	d	
S. Enteritidis	13.7		737	1,104	1,416	1,182	2,025	2,607	1,186	-	-	d	
S. Typhimurium	8.4		450	378	589	436	584	678	1,193	-	-	d	
Other Serotypes	9.8		526	589	913	690	659	595	1,432	-	-	d	
Campylobacter coli/jejuni	65.8		3,542	4,378	4,620	4,386	4,164	3,372	-	-	-	e	
E. multilocularis/granulosus	-		-	-	-	-	-	-	-	-	-	c	
Leptospira spp.	0.2		13	13	6	21	23	12	-	-	-	f	
Listeria monocytogenes	0.4		19	28	38	39	44	41	-	-	-	f	
Rabies	-		-	-	-	-	-	-	-	-	-	g	
Toxoplasma gondii	-		-	-	-	-	-	-	-	-	-	b	
Cryptosporidium spp.	1.1		58	38	84	-	-	-	-	-	-	b	
Yersinia enterocolitica	4.6		245	240	286	265	339	464	710	-	-	-	
VTEC	2.4		128	141	92	60	51	34	-	-	-	-	
VTEC-O157	0.5		27	23	24	18	10	12	0	-	-	-	

a) Notification mandatory. Cases of tuberculosis are due to reactivation of latent infections in elderly or imported disease.

b) Notification not mandatory.

c) Notification not mandatory. A few travel associated cases occur.

d) Only first isolations registered.

e) A sample (n=123) of the isolates were identified to the species level: Of these 115 were *C. jejuni* and 8 were *C. coli*.

f) Notification mandatory (since 1986).

g) Notification mandatory. No domestic or imported cases.

Source: SSI

Table A7. Serotype distribution (%) of Salmonella from animals, carcasses at slaughterhouses, imported meat and humans in Denmark, 2003. In some cases more than one serotype was found per positive herd/batch and therefore the number of typed units may be greater than the number of positive herds/batches.

Serotype										Imported meat ^{f)}			
	Human n=1,713	Pig herds ^{a)} n=619	Pork ^{b)} n=296	Cattle herds ^{c)} n=70	Beef ^{b)} n=23	Layer flocks ^{d)} n=14	Broiler flocks ^{e)} n=77	Turkey flocks ^{e)} n=17	Duck flocks ^{e)} n=161	Pork n=66	Chicken n=130	Turkey n=78	Duck n=49
S. Enteritidis	43.0	0.2	0.3	0.0	0.0	92.9	3.9	0.0	14.3	0.0	38.5	1.3	4.1
S. Typhimurium	26.3	69.6	39.5	41.4	4.3	0.0	26.0	0.0	0.0	66.7	2.3	6.4	42.9
S. Virchow	2.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.8	0.0	0.0
S. Agona	2.2	0.8	0.7	0.0	0.0	0.0	0.0	29.4	0.0	0.0	3.8	5.1	0.0
S. Dublin	2.0	0.0	0.3	54.3	69.6	0.0	0.0	0.0	0.0	0.0	0.0	1.3	0.0
S. Newport	1.6	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	2.6	0.0
S. Stanley	1.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	5.1	0.0
S. Infantis	1.5	4.5	5.7	1.4	8.7	0.0	22.1	0.0	0.0	1.5	0.8	1.3	0.0
S. Derby	1.5	15.3	28.4	1.4	4.3	7.1	5.2	0.0	0.0	10.6	2.3	10.3	0.0
S. Hadar	1.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	5.0	0.0	5.4	19.2	6.1
S. Saintpaul	0.7	0.0	0.0	0.0	0.0	0.0	0.0	23.5	0.6	0.0	2.3	9.0	12.2
S. Blockley	0.6	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	3.1	3.8	0.0
S. Kottbus	0.5	0.0	0.0	0.0	0.0	0.0	2.6	29.4	7.5	0.0	2.3	14.1	2.0
S. Kentucky	0.5	0.0	0.0	0.0	0.0	0.0	5.2	0.0	0.0	0.0	0.0	0.0	0.0
S. Anatum	0.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	53.4	0.0	0.8	0.0	0.0
S. Heidelberg	0.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	14.6	1.3	0.0
S. Poona	0.4	0.0	0.0	0.0	0.0	0.0	1.3	0.0	0.0	0.0	0.0	0.0	0.0
S. Montevideo	0.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
S. Schwarzengrund	0.3	0.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.8	0.0	0.0
S. Indiana	0.2	0.0	0.0	0.0	0.0	0.0	10.4	0.0	8.1	0.0	2.3	5.1	28.6
S. Rissen	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	3.0	0.8	0.0	0.0
S. Brandenburg	0.2	0.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	12.1	0.0	0.0	0.0
NT	0.4	2.1	13.9	0.0	4.3	0.0	1.3	0.0	1.9	0.0	6.2	1.3	2.0
Others	11.8	6.8	11.1	1.4	8.7	0.0	22.1	17.6	9.3	6.1	13.1	12.8	2.0
Total	100	100	100	100	100	100	100	100	100	100	100	100	100

- a) Isolates obtained from sampling in slaughter-pig herds placed in Level 2 or 3.
b) Representative swab samples of pork and beef carcass from the surveillance programme at slaughterhouses.
c) Cattle herds examined on clinical indications. The data are not representative for the Danish cattle population.
d) Representative samples from the surveillance programme in production flocks.
e) Representative faecal or sock samples from the mandatory AM inspection prior to slaughter.
f) Monitoring of imported meat and meat products.
Source: DVFA, DFVF and SSI

Table A8. Phage-type distribution (%) of S. Enteritidis from humans, animals, carcasses and slaughterhouses and imported meat, 2003

Phage type	Human n=737	Layers ^{d)} n=13	Ducks n=23	Imported meat ^{f)}	
				Chicken n=50	Duck n=21
FT8	14.9	30.8	0	2.0	0.0
FT4	20.2	38.5	0	50.0	0.0
FT6	3.3	23.1	0	10.0	0.0
FT1	6.0	0.0	0	18.0	0.0
FT21	7.7	0.0	0	10.0	0.0
FT21B	0.5	0.0	0	0.0	0.0
FT35	0.1	0.0	0	4.0	0.0
FT1B	0.3	7.7	0	0.0	0.0
FT9B	0.0	0.0	100	0.0	95.2
NT	39.3	0.0	0	6.0	0.0
Others	7.6	0.0	0	0.0	4.8
Total	100	100	100	100	100

Footnotes: See table A7.
Other phage typed S. Enteritidis isolates: 3 broiler flocks (PT1B), 1 turkey isolate (PT4) and 2 imported duck isolates (PT4, PT9B).
Source: DVFA, DFVF and SSI

Table A9. Phage-type distribution (%) of S. Typhimurium from humans, animals, carcasses at slaughterhouses and imported meat, 2003.

Phage type	Human n=450	Pig herds ^{a)} n=440	Pork ^{b)} n=117	Cattle herds ^{c)} n=30	Broilers ^{e)} n=20	Imported meat ^{f)}	
						Pork n=44	Duck n=21
DT120	10.7	9.5	18.8	20.0	0.0	11.4	0.0
DT104	12.7	4.5	5.1	20.0	10.0	6.8	0.0
DT12	7.3	31.1	17.9	23.3	30.0	0.0	0.0
DTU302	12.4	0.7	0.9	0.0	0.0	25.0	0.0
DT193	4.4	5.2	2.6	0.0	0.0	9.1	0.0
DT170	9.6	14.3	19.7	10.0	15.0	0.0	0.0
DT135	0.4	1.1	0.9	0.0	10.0	0.0	0.0
DT104b	1.6	0.0	2.6	0.0	0.0	0.0	0.0
DT66	0.9	5.2	8.5	0.0	5.0	2.3	0.0
DT17	1.8	7.5	5.1	10.0	10.0	0.0	0.0
DT8	0.9	0.0	0.0	0.0	0.0	0.0	95.2
DT10	0.2	0.9	0.9	3.3	0.0	0.0	0.0
DTU288	2.7	2.5	1.7	3.3	0.0	0.0	0.0
DT110	0.2	0.9	1.7	0.0	15.0	0.0	0.0
DTU312	0.2	1.1	0.0	0.0	0.0	0.0	0.0
NT	18.4	3.2	0.0	0.0	0.0	15.9	0.0
Others	15.6	12.0	13.7	10.0	5.0	29.5	4.8
Total	100	100	100	100	100	100	100

Footnotes: See Table A7.
Other phage typed S. Typhimurium isolates: 1 beef isolate (DTU312), 6 imported beef isolates (5 DT104, 1 other), 3 imported chicken isolates (1 DTU302, 2 others), 5 imported turkey isolates (2 DT 104, 3 others).
Source: DVFA, DFVF and SSI

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