trends and sources

report on zoonotic agents in belgium in 2006

2006

working group on foodborne infections and intoxications

.be

- Federal Agency for the Safety of the Food Chain (FAVV-AFSCA)
- Scientific Institute of Public Health (WIV-ISP)
- Veterinary and Agrochemical Research Centre (CODA-CERVA)

Executive summary

Controlling zoonoses remains an enormous task and opportunity for all competent authorities. Measures and systems of disease surveillance, diagnosis and control must be implemented on a national level and have to be based on a suitable regulatory framework and an appropriate level of funding. Active collaboration between all actors of the food chain, stakeholders, industry, scientists, experts of the national reference laboratories and other laboratories, specialists of the competent authorities, technical committees have to bring together their experiences, methods and findings. Only collaborative approach and effective partnership at all levels will achieve success to control zoonoses and improvement of food safety.

The most commonly reported zoonotic infections in humans are those caused by bacterial zoonotic agents that can be shed by asymptomatic farm animals. Campylobacteriosis remained for the second time the most frequently reported zoonotic disease in humans. Broiler and other poultry meat is an important source of foodborne Campylobacter infections. Salmonellosis is the second most frequently reported zoonosis. Salmonella control remains an important task.

Salmonella was the most important cause of foodborne outbreaks. The major sources of Salmonella in foodborne outbreaks are eggs, poultry meat and pig meat.

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Preface

The Belgian authority, like all European member states, has the obligation to yearly submit an official Trends & Sources report to the European Food Safety Authority (EFSA) based on article 9 of Directive 2003/99/EC of the European Parliament and the Council on the monitoring of zoonoses and zoonotic agents. In that report all the relevant official monitoring programmes in primary production as well as on feed and food are presented. The report specifies all available data from monitoring and research activities, as well as laboratory findings from the previous year and includes results from antimicrobial susceptibility testing and foodborne outbreaks. Similarly, data on zoonotic infections in humans are officially reported each year to the European Centre for Disease prevention and Control (ECDC).

Based on these two official reports, the Federal Agency for the Safety of the Food Chain, together with the scientific institutions CODA-CERVA and WIV-ISP agreed to regularly publish a booklet which contains this same information, but presented to professional readers as well as to those who have a general interest in animal and human infections and in the safety of our food.

We hope that the reader will enjoy this fifth edition of the Belgian report on zoonotic agents.

Hein Imberechts	Luc Vanholme	Katelijne Dierick
		Geneviève Ducoffre
CODA – CERVA	FAVV - AFSCA	WIV - ISP

Introduction

This report compiles the available data for 2006 on zoonoses and zoonotic agents, and is derived from the official documents reported to EFSA and ECDC. For this reason, it is a unique document in which laboratory results from the primary production, from food and from clinical, public health sources are combined. In addition to the compulsory reporting on zoonoses and zoonotic agents as listed in the European Directive 2003/99/EC, this document contains data on other foodborne agents that may be of interest to the reader, e.g. on avian influenza, transmissible spongiform encephalopathies (TSE, e.g. mad cow disease) or norovirus infections.

Together with the general descriptive information on the infections themselves, their evolution over time, and some recommendations on prevention of the infection, this booklet should meet the expectations of everybody who is concerned with the possible contamination of our daily food with bacteria, viruses, parasites and prions.

The Federal Agency for the Safety of the Food Chain organises diverse monitoring programmes in, among others, the primary production and in the transformation and distribution sectors. From their description follows that much effort is being paid to control the contamination of foodstuffs with pathogens. Some infectious diseases have successfully been reduced or even eliminated (for instance salmonellosis, brucellosis, mad cow disease) and for others (for instance campylobacteriosis) further programmes should be set up. In addition to the continuous effort from the authorities, the consumer should be aware that she or he has also an important role to play. Indeed, respect for the cold chain and simple hygiene measures in the kitchen may be very efficient in preventing foodborne contaminations and unpleasant clinical sequels.

Most of the data in this report are from the following sources:

- The Federal Agency for the Safety of the Food Chain (FAVV-AFSCA);
- The Scientific Institute of Public Health (WIV-ISP);
- The Veterinary and Agrochemical Research Centre (CODA-CERVA).

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- A. Linden, Faculty of Veterinary Medicine, Bacteriology and Pathology of Bacterial diseases Department, University of Liège;
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- E. Vanopdenbosch and S. Roels, Department of Biocontrol, Veterinary and Agrochemical Research Centre;

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National Reference Laboratories (NRL) for zoonotic agents

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Avian Influenza	T. van den Berg	CODA-CERVA Groeselenberg 99, 1180 Brussels	thvan@var.fgov.be http://www.var.fgov.be/
Brucella, public and animal health	K. Walravens	CODA-CERVA Groeselenberg 99, 1180 Brussels	karl.walravens@var.fgov.be http://www.var.fgov.be/
Biotoxins	J.-Y. Michelet	WIV-ISP, Food Section J. Wytsmanstraat 14, 1050 Brussels	Jean-Yves.Michelet@iph.fgov.be http://www.iph.fgov.be/
BSE / TSE	S. Roels	CODA-CERVA Groeselenberg 99, 1180 Brussels	stroe@var.fgov.be http://www.var.fgov.be/
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Escherichia coli VTEC and EHEC, public health	D. Pierard	UZ Brussel , Microbiology Laarbeeklaan, 101, 1090 Brussels	labomicro@uzbrussel.be http://www.uzbrussel.be
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Listeria monocytogenes	M. Yde	WIV-ISP, Bacteriology Section J. Wytsmanstraat, 14, 1050 Brussels	Marc.Yde@iph.fgov.be http://www.iph.fgov.be/
Mycobacterium	M. Fauville-Dufaux (public health) F. Portaels (public health) K. Walravens, M. Govaerts (animal health)	WIV-ISP, Pasteur Institute Dpt Rue Engeland, 642, 1180 Brussels ITG, Mycobacteriology Nationalestraat, 155, 2000 Antwerpen CODA-CERVA Groeselenberg, 99, 1180 Brussels	Mfauville@pasteur.be http://www.pasteur.be/pasteur_en/index.html Fportaels@itg.be http://www.itg.be/itg/GeneralSite/Generalpage.asp Karl.Walravens@var.fgov.be Marc.Govaerts@var.fgov.be http://www.var.fgov.be/
Phage typing centre (Salmonella, Staphylococcus)	C. Wildemauwe	WIV-ISP, Pasteur Institute Dpt Rue Engeland, 642, 1180 Brussels	Cwildemauwe@pasteur.be http://www.pasteur.be/pasteur_en/index.html
Q-Fever (Coxiella burnetii)	M. Van Esbroeck	ITG, Klinische Biologie Nationalestraat, 155, 2000 Antwerpen	mvesbroeck@itg.be http://www.itg.be/itg/GeneralSite/Generalpage.asp
Rabies	I. Le Roux S. Van Gucht	WIV-ISP, Pasteur Institute Dpt Rue Engeland, 642, 1180 Brussels	Ileroux@pasteur.be Svangucht@pasteur.be http://www.pasteur.be/pasteur_en/index.html
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Salmonella, animal health	H. Imberechts	CODA-CERVA Groeselenberg, 99, 1180 Brussels	Hein.Imberechts@var.fgov.be http://www.var.fgov.be/
Toxoplasmosis	S. Decraeye	WIV-ISP, Pasteur Institute Dpt Rue Engeland, 642, 1180 Brussels	sdecraeye@pasteur.be http://www.pasteur.be/pasteur_en/index.html
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general information

Susceptible human population

The evolution of the total human population in Belgium categorised per age, sex and region from 2002 to 2006 is shown in table 1.

Table 1. *Evolution of the total human population in Belgium categorised per age, sex and region from 2002 to 2006*
Source: National Institute for Statistics http://statbel.fgov.be/

	2002	2003	2004	2005	2006
Total	**10 309 725**	**10 355 844**	**10 396 421**	**10 445 852**	**10 511 382**
0-19	2 408 943	2 407 368	2 408 456	2 414 041	2 428 706
20-64	6 154 390	6 186 086	6 207 845	6 232 311	6 273 659
65+	1 746 392	1 762 390	1 780 120	1 799 500	1 809 017
Male	**5 042 288**	**5 066 885**	**5 087 176**	**5 111 325**	**5 143 821**
0-19	1 231 221	1 230 382	1 230 570	1 233 688	1 241 251
20-64	3 094 653	3 110 779	3 120 599	3 131 390	3 150 333
65+	716 414	725 724	736 007	746 247	752 237
Female	**5 267 437**	**5 288 959**	**5 309 245**	**5 334 527**	**5 367 561**
0-19	1 177 722	1 176 986	1 177 886	1 180 353	1 187 455
20-64	3 059 737	3 075 307	3 087 246	3 100 921	3 123 326
65+	1 029 978	1 036 666	1 044 113	1 053 253	1 056 780
Brussels	***978 384***	***992 041***	***999 899***	***1 006 749***	***1 018 804***
Flanders	***5 972 781***	***5 995 553***	***6 016 024***	***6 043 161***	***6 078 600***
Wallonia	***3 358 560***	***3 368 250***	***3 380 498***	***3 413 978***	***3 413 978***
Foreigners	***846 734***	***850 077***	***860 287***	***870 862***	***900 473***

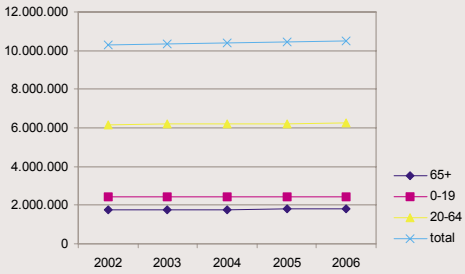

Figure 1. *Evolution of human population 2002 - 2006*

- Susceptible human population
- Susceptible animal populations

Susceptible animal populations

Ruminants and pigs

The origin of the following table is SANITEL, the computerised registration and identification database of farm animals, as managed and centralised by the Federal Agency for the Safety of the Food Chain.

Table 2. *Total number of herds and animals in 2004, 2005 and 2006*

	2004		2005		2006	
	Herds	Animals	Herds	Animals	Herds	Animals
Cattle	44 555	2 781 676	42 204	2 492 757	40 640	2 697 824
Pigs	10 614		10 792		10 631	
Breeding sows[1]		664 316		657 998		653 358
Fattening pigs[2]		4 998 124		4 989 016		4 850 501
Sheep	31 405	214 612	32 323	219 274	30.924	220.600
Goats	13 736	37 666	14 247	43 727	13.025	46.950
Deer	2 965	13 427	3 093	14 655	2 021	12 805

1 total number of available places for sows and gilts in all herds
2 total number of available places for fattening pigs in all herds

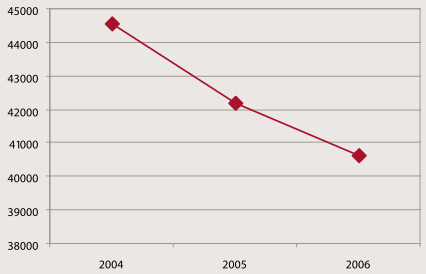

Figure 2. *Evolution total number of cattle herds 2004 - 2006*

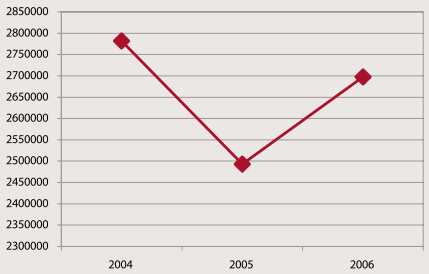

Figure 3*. Evolution total number of bovine animals 2004 - 2006*

Poultry

***Table 3**. Total number of holdings and total number of available places for fowl in 2004, 2005 and 2006*

	2004		2005		2006	
	Herds	Animals	Herds	Animals	Herds	Animals
Gallus Gallus						
Layers	529	14 364 922	386	10 562 160	472	13 377 548
Broilers	1 097	27 873 988	1 024	26 754 817	978	25 894 597
Elite, Parent, Breeding	193	2 255 085	156	2 144 874	232	3 170 815
Total	2 284	50 947 719	1 566	39 461 851	1 682	42 442 960
Ducks	31	33 949	17	45 140	27	77 140
Geese	8	4 843	5	3 800	6	4 900
Guinea fowl	27	87 440	16	71 400	12	39 200
Partridges	2	123 300	4	129 000	4	136 000
Pheasants	14	206 649	16	226 049	23	268 000
Pigeons	4	1 520	2	1 300	2	2 500
Quails	7	56 020	1	1 700	/	/
Turkeys	63	498 146	37	246 076	41	248 006

Animals slaughtered in 2003, 2004, 2005 and 2006

Table 4. *Number of animals slaughtered in 2003, 2004, 2005 and 2006. Source: Data from the Federal Agency for the Safety of the Food Chain*

	2003	2004	2005	2006
Cattle	570 000	564 266	523 795	496 181
Calves	317 000	317 269	313 115	327 467
Pigs	11 609 933	11 229 149	10 861 234	10 794 757
Solipeds	12 304	11 655	11 542	10 728
Sheep	83 112	87 119	112 771	148 767
Goats	2 514	3 814	2 585	3 036
Broiler	222 327 256	244 064 267	237 670 666	247 721 072
Layer	19 711 279[1]	28 577 233	29 907 674	32 265 603

1 The small number of layers slaughtered in 2003 is associated with the outbreak of avian influenza in March 2003 and the consequent d epopulation of poultry houses.

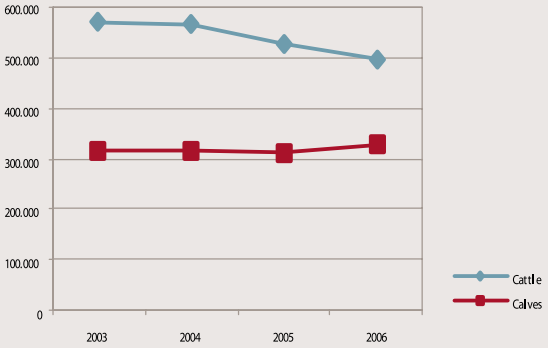

Figure 4. *Evolution in slaughtered bovines 2003 - 2006*

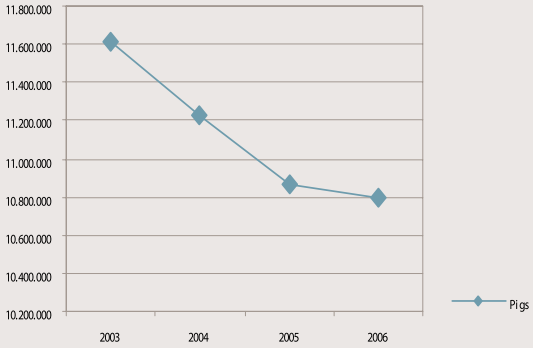

Figure 5*. Evolution in slaughtered pigs 2003 - 2006*

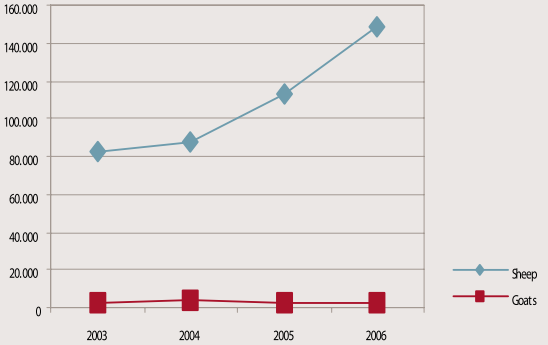

Figure 6. *Evolution in slaugtered sheep and goats 2003 - 2006*

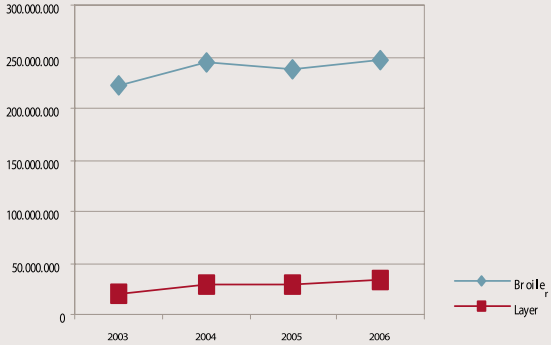

Figure 7*. Evolution in slaughtered poultry 2003 - 2006*

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campylobacteriosis

Campylobacteriosis

Campylobacter is worldwide the most common cause of bacterial gastroenteritis in man. Campylobacteriosis overtakes salmonellosis as the most reported animal infection transmitted to humans. The incidence of Campylobacter peaks during infancy and early adulthood. The infection may cause Guillain-Barré syndrome.

The consumption of undercooked poultry meat represents the main mode of contamination, but other food sources such as pork and beef, unpasteurised milk, or contaminated drinking water are also reported. Contacts with faeces of infected pets may also be a source of contamination. This chapter focuses on Campylobacter jejuni and Campylobacter coli which are the most frequently reported pathogens in humans.

The contamination of poultry carcasses and meat with Campylobacter are monitored by the Federal Agency for the Safety of the Food Chain since 2000. The rate of positive poultry samples is high, but stable. Broiler and layer meat have to be well cooked and cross-contamination should be avoided during preparation.

- Campylobacter in food
- Antimicrobial resistance in strains isolated from meat and meat products
- Campylobacter in humans

Campylobacter in food

Monitoring programme

In 2006, the Federal Agency for the Safety of the Food Chain selected for its monitoring programme more than 200 Belgian slaughterhouses, more than 100 meat cutting plants and more than 100 retail trades representative of the Belgian production of carcasses and meat.

Samples for Campylobacter were taken from carcasses, meat preparations and fillets of broilers, carcasses of layers, carcasses and minced meat from pork, dairy products and live bivalve molluscs. Specially trained staff of the Federal Agency for the Safety of the Food Chain performed the sampling. Five contamination levels, 25g, 10g, 1g, 0.01g and 600cm2 were analysed. For broiler carcasses and fillets, approximately 300 independent samples were taken per matrix in order to detect a minimal contamination rate of 1% with 95% confidence.

Results of the 2006 monitoring

The results of the monitoring of the Federal Agency for the Safety of the Food Chain are shown in the next table.

Table 5. *Zoonosis monitoring programme – Campylobacter in food*

Sample	Quantity of sample analysed	Percentage of positive samples
Broiler		
Carcasses at slaughter (n=6443)	25g (caeca)	55.5%
Carcasses at slaughter (n=315)	0.01g	1.9.%
Carcasses at retail (n=40)	0.01g	42.5%
Meat cuts (skinned or with skin) at retail (n=40)	0.01g	5%
Meat preparation at processing plant (n=162)	0.01g	2.5%
Meat preparation at retail (n=102)	0.01g	2.0%
Fillets at processing plant (n=326)	1g	12.4%
Layer		
Carcasses at slaughter (n=1017)	25g (caeca)	86.4%
Carcasses at slaughter (n=246)	0.01g	6.5%
Carcasses at retail (n=32)	0.01g	25.0%
Pork		
Carcasses (n=418)	600 cm2	13.4%
Minced meat at processing plant (n=50)	25g	2.0%
Minced meat at retail (all species) (n=26)	10g	0.0%
Raw milk cheese at retail (n=55)	25g	0.0%
Raw milk cheese (fresh) at farm (n=75)	25g	0.0%
Live bivalve molluscs (n=55)	25g	1.8%

Table 6. *Evolution of the pork Campylobacter prevalence 2004-2006*

		Sampling level	2004	2005	2006
Pork	Carcasses	600 cm2	4.9%	7.2%	13.4%
	Minced meat	25g	2.8%	0.7%	2.0%

The contamination rate of pork carcasses raised again in 2006. Pork carcasses are sampled at the end of the slaughter line; cooling decreases greatly the number of Campylobacter. The contamination rate of minced pork meat was also higher in 2006 compared to the previous year but the contamination rate remains low.

Antimicrobial resistance in Campylobacterstrains isolated from meat and meat products

Antimicrobial resistance in Campylobacter strains isolated from meat and meat products

Surveillance programme and method used

In 2006, 187 Campylobacter strains isolated in the zoonoses monitoring programme and originating from poultry and pork were send for their antimicrobial susceptibility.

Fifty-six strains were isolated from pork meat or carcasses, 104 strains were isolated from broiler meat or carcasses, 20 strains were isolated from spent hens and in 7 strains the animal species was unknown. C. coli was the most prevalent strain isolated from pork carcasses (87.5%), while for poultry meat C. jejuni was the most isolated Campylobacter strain (63.5%) and C. coli represented 23% of the isolates. Minimum Inhibitory Concentrations (MIC) were determined by the use of E-test on blood agar plates. The antimicrobials tested and the breakpoints (following the CLSI standards) used are listed in the following table.

Table 7. *Campylobacter in meat and meat products: list of antimicrobials tested and breakpoints used.*

Antimicrobial	Breakpoints (mg / ml)
Ampicillin	8 – 32
Tetracycline	4 – 16
Nalidixic acid	16 – 32
Ciprofloxacin	1 – 4
Erytromycin	1 – 8
Gentamycin	4 – 16

The percentage of resistant strains of Campylobacter in food is reported in the next table.

Table 8*. Antimicrobial susceptibility testing of Campylobacter in food: Percentage of resistant strains*

	Poultry meat		Pork
	C. jejuni (n=66)	C. coli (n=25)	C. coli (n=49)
Tetracycline	41	100	86
Ciprofloxacin	36	56	33
Nalidixic acid	32	52	33
Gentamicin	0	0	4
Erythromycin	2	8	16
Ampicillin	30	24	10

Antimicrobial resistance in Campylobacter from poultry meat

91 Campylobacter strains were isolated in poultry meat and carcasses and tested for antimicrobial susceptibility (66 Campylobacter jejuni and 25 Campylobacter coli strains). In total 32% of the C. jejuni strains were sensitive for all tested antibiotics. Tetracycline resistance was present in 41% of the strains followed by ciprofloxacin (36%) and nalidixic acid (32%) resistance. Ampicillin resistance was noticed in 30% of the C. jejuni strains and 2% of the strains were resistant against erythromycin. Overall the antibiotic resistance within C. coli was higher than in C. jejuni, with a much higher percentage of resistance against ciprofloxacin, nalidixic acid and tetracycline. Resistance against erythromycin was found in 8% of the C. coli strains. Campylobacter isolates from broiler meat did not show resistance to gentamycine.

The ampicillin resistance is much higher in strains isolated from broiler meat and carcasses than in strains isolated from pork meat.

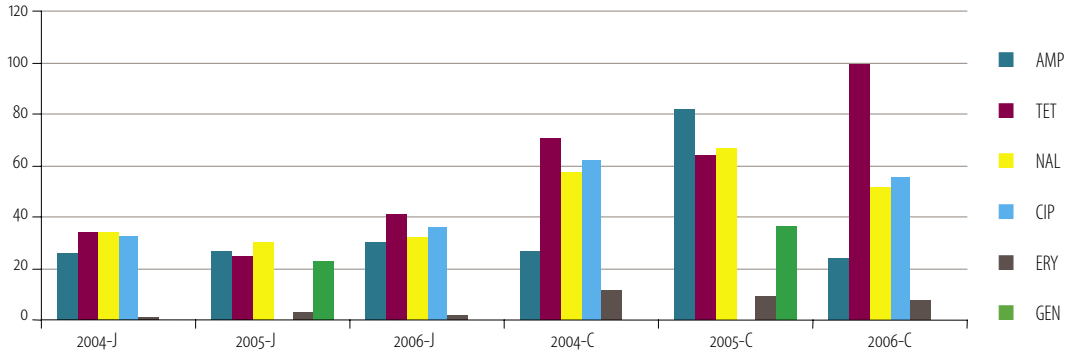

Figure 8. *Percentage of antimicrobial resistance in Campylobacter jejuni (J) and Campylobacter coli (C) strains in poultry meat*

Antimicrobial resistance in Campylobacter from pork

In the C. coli isolates (49) from pork, resistance was observed for all tested antibiotics . Only 2 strains were sensitive to all tested antibiotics. The resistance against tetracycline (86%) was high followed by ciprofloxacin and nalidixic acid (33%). Multi-resistance, which means resistance against 4 antibiotics or more was observed in 2 strains with following resistance profile : 'Ciprofloxacin-Erythromycine-Nalidixic acid-Tetracycline'.

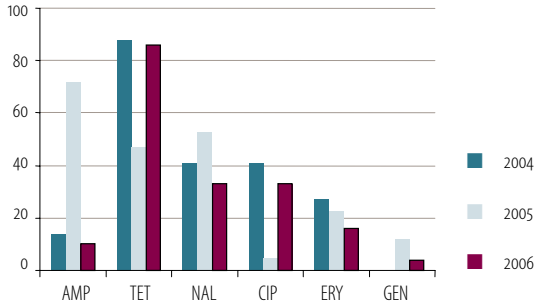

Figure 9. *Percentage of antimicrobial resistance in Campylobacter coli strains isolated from pork*

Campylobacter in humans

In 2006, the Belgian Sentinel Laboratory Network consisted of 110 laboratories reporting Campylobacter. 5,771 strains of Campylobacter were isolated which represent at country level an isolation rate of 55 per 100 000 inhabitants. The number of Campylobacter infections shows a significant decreasing trend since 2000 at national and regional level ($p<0.05$; figure 10). Since 2005 Campylobacteriosis remains the most frequently reported zoonosis in humans.

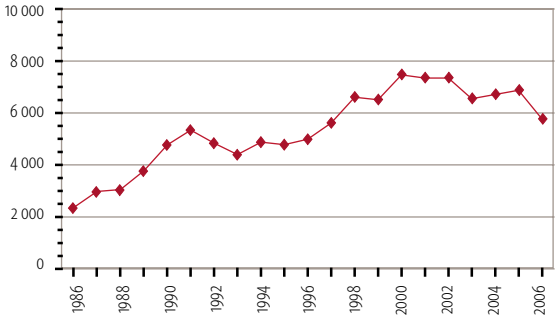

Figure 10. *Total number of Campylobacter infections in humans by year (1986-2006)*
Source: Sentinel Laboratory Network

Cases are reported during the entire year, with a peak in the summertime (figure 11).

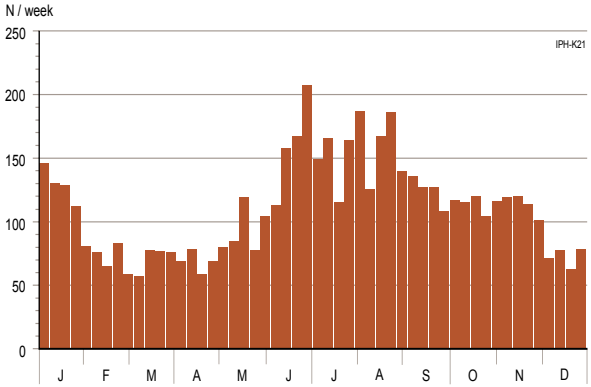

Figure 11. *Weekly number of cases of Campylobacter in 2006, Belgium.*
Source: Sentinel Laboratory Network

Campylobacter isolation rates are higher in children under 5 years of age. Under 15 years of age, boys appear to be significantly more affected than girls. There is no explanation for this observation, but it is also reported in other countries (Table 9).

Table 9. *Number of cases of Campylobacter by age groups, 2006*
Source: Sentinel Laboratory Network

Age groups	Males		females		Total	
(year)	N	%	N	%	N	%
<1	168	6,1	128	5,1	313	5,6
1-4	689	22,8	496	19,6	1185	21,3
5-14	475	15,7	325	12,8	800	14,4
15-24	317	10,5	368	14,5	685	12,3
25-44	558	18,5	594	23,4	1152	20,7
45-64	489	16,2	321	12,7	810	14,6
≥65	309	10,2	302	11,9	611	11,0
Total	3022	100,0	2534	100,0	5556	100,0

Since the beginning of the registration, the incidence in Flanders, especially in the province Antwerp, is twice as high as compared to Wallonia. This was confirmed in 2006 with an estimated incidence of 68/100,000 inhabitants in Flanders, 34/100,000 inhabitants in Wallonia and 38/100,000 inhabitants in Brussels-Capital Region.

trends and sources

report on zoonotic agents in belgium in 2006



salmonellosis

Salmonellosis

In Belgium, as in many countries, Salmonella is a major cause of registered bacterial foodborne infections, both in individuals and in communities. Salmonella infections provoke a gastro-intestinal illness with nausea, vomiting, abdominal cramps, diarrhoea and fever. In susceptible persons bacteraemia and septicaemia may occur. Often, food prepared with contaminated raw eggs, egg products or insufficiently heated poultry meat or pork are the source of the human Salmonella infection. Therefore, surveillance programmes that in time detect Salmonella contaminations in the whole food chain (feed, living animals, slaughterhouses, cutting plants, retail sector, restaurants) together with sanitary measures to reduce contamination are essential. In addition, good hygiene practices during food preparation in the kitchen, adequate refrigeration and adequate heating also help to prevent Salmonella infections.

In 2006, the total number of reported Salmonella cases in humans was significantly lower compared to the three previous years :. 3 693 records in 2006, 4 916 records in 2005, 9 543 records in 2004, and 12 792 in 2003. This evolution was mainly due to a significant decrease of Salmonella Enteritidis isolated in humans.

- Salmonella in animal feed
- Salmonella in poultry
- Salmonella in pigs
- Salmonella in cattle
- Salmonella in food (meat and meat products)
- Salmonella in humans
- Antimicrobial resistance

Salmonella in animal feed

Each year, an official monitoring for the detection of Salmonella in compound feeding stuffs and in raw materials is organised by the Federal Agency for the Security of the Food Chain. Microbiological testing on 25g samples is done in the FASFC laboratories. In case of isolation of Salmonella in official samples no certification is provided.

Out of forty-one feed materials of animal origin (2 dairy products, 23 meat and bone meal, 1 poultry offal meal, 1 feather meal, 3 blood meal, 9 animal fat and 2 egg powder), only one meat and bone meal sample was found positive (S. Livingstone). No fish meal and fish oil samples were analysed in 2006.

A total of 137 vegetal samples were analysed in 2006. Two cereal samples (one maize and one other cereal grain derived) were tested and were found free of Salmonella. Two out of 135 samples from feed materials of oil seed origin were found contaminated, i.e. one out of 24 samples from rape seed origin (contamination with Salmonella Tennessee) and one out of 58 soya derived samples (Salmonella Anatum). Two palm kernel derived samples, 22 sunflower derived samples, 10 linseed derived samples and 19 other oil seeds derived samples were all found negative for Salmonella.

In addition, 297 compound feedingstuffs were tested. Only one compound feed for poultry breeders was found contaminated with Salmonella (S. Mbandaka was identified). The following samples were all found negative for Salmonella: 3 compound feedingstuffs for cattle, 20 for pigs, 44 for poultry (not specified), 27 for laying hens, 35 for poultry broilers, one for sheep, 28 not specified feedingstuffs, 1 pet food and another 130 complementary feedingstuffs.

Salmonella in poultry

Salmonella in breeders and hatcheries

Surveillance programme in breeders

The regional animal health associations (i.e. "Association Régionale de Santé et d'Identification Animales" [ARSIA (http://www.arsia.be/)] and "Dierengezondheidszorg Vlaanderen" [DGZ Vlaanderen (http://www.dgz.be/)]) organise the official sampling in the framework of the Belgian Salmonella control programme in breeders.

All breeder flocks are routinely examined for Salmonella at delivery as day-old birds (imported and domestic flocks). At the farm, pieces (5 by 5 cm) of the inner linings of the delivery boxes of the day-old chickens are taken by the owner, i.e. one sample for the hen-chicks and one for the cock-chicks. Each sample consists of 20 pieces of inner linings. The two samples are analysed separately. In addition, 20 living hen-chicks and 20 living cock-chicks are tested serologically. The samples have to be taken the day of the delivery and have to reach the lab within 24h of sampling. Breeders during the rearing period are sampled at the age of 16 weeks by technicians of

DGZ and ARSIA. For this purpose, a pooled faecal sample of 60 x 1g or, alternatively, 2 pairs of overshoes is taken. Technicians of DGZ and ARSIA also officially sample all breeders in production; i.e. a pooled faeces sample of 60 x 1g, or 2 pairs of overshoes every six weeks. In addition, every two weeks each flock is sampled on mandatory basis with 2 pairs of overshoes by the owner. All samples are immediately analysed in the laboratories of DGZ or ARSIA according to ISO 6579:2002 FDAM 1.

The official programme also controls the hygiene level of hatcheries 4 times a year. These are done during visits of the technician at non-hatching days and comprise various sites of the hatchery, including hatching drawers. Rodac samples are taken and both total bacteria and moulds are counted. After appropriate incubation, an index or code is given to the number of colonies per surface of approximately 22 cm2 in order to facilitate comparisons. In addition, a specific Salmonella control is done 4 times a year, on pooled samples from dead-in-shell chicks and on fluff and meconium. These samples are sent to the laboratory by the owner.

In 1999 the royal and ministerial decrees concerning the sanitary qualification (Gezondheidskwalificatie - Qualification sanitaire, Royal Decree of 10 August 1998, Ministerial Decree of 19 August 1998) came into force. They prescribe minimal requirements for infrastructure and general hygienic measures including specific sampling for Salmonella detection on farms with more than 5 000 birds. Thus, all poultry flocks before arrival at the slaughterhouse (i.e. breeders, layers and broilers) undergo a bacteriological examination.

Case definition, notification, sanitary measures and vaccination

A poultry breeding flock is considered Salmonella positive when Salmonella Enteritidis or Salmonella Typhimurium is isolated from one-day-old chickens, at 16 weeks (rearing) or at the occasion of one of the official samplings during production. If at least one sample in a flock is positive, the whole flock is considered as positive.

Confirmatory samples during rearing or production may be requested by the farmer, and are taken by the competent authority. The results of these analysis are binding.

The isolation of zoonotic Salmonella is notifiable since January 2004 and should be reported to the Federal Agency for the Safety of the Food Chain.

Several measures are taken on the positive breeder flock: the hatching eggs are no longer incubated, but are removed and destroyed, and not yet incubated hatching eggs may be pasteurised. In addition, positive flocks are logistically slaughtered and after removal the houses are thoroughly cleaned and disinfected.

Vaccination against Salmonella Enteritidis and / or Salmonella Typhimurium is strongly recommended for parent flocks. Both attenuated and inactivated vaccines are available.

Epidemiological investigations and results of 2006 surveillance

In 2006, 13 parent flocks (both layer and broiler breeders) were tested as day-old chicks and none was found positive for Salmonella. Also at 16 weeks of age, and during production, the flocks were negative. Layer breeders were found free of Salmonella Enteritidis and Salmonella Typhimurium from 2003 on.

In 2006, only 3 grandparent flocks were tested, and all were negative for Salmonella. In addition, 35 layer breeder flocks, 724 broiler breeder flocks and 109 breeder flocks (unspecified) were sampled. 13 broiler breeders were found contaminated during production with Salmonella (2 Salmonella each from serotypes Agona, Anatum Braenderup, Mbandaka and Senftenberg, and 1 Salmonella Panama, Rissen and Schwartzengrund).

Salmonella in layers and broilers

Surveillance programme in commercial poultry flocks

The national control programme for Salmonella in layers and broilers is performed according to the sanitary qualification act, which is applicable to farms with more than 5 000 birds. Sampling is done by the farmer and consists of an exit sample for Salmonella, within 3 weeks of slaughter. The owner can sample in 3 ways: (1) pooled faeces (60 x 1g) taken with swabs, (2) a pooled faeces (60 x 1g) taken by hand, or (3) two pairs of overshoes, pooled. All samples have to be examined by an accredited laboratory within 48h.

In addition, layer and broiler flocks may be sampled as day-old chicks at the farm (entry control). In this purpose, the owner samples pieces of inner linings of the delivery boxes in the same way as is done for breeder flocks. After transport of layers to the production unit, a 60 x 1g faecal sample may be taken from the delivery boxes. Every flock is sampled taking into account the different origins of rearing.

From October 2005 to September 2006, the European coordinated monitoring of broilers flocks was undertaken according to article 5 of Directive 2003/99/EC. Details of a report of this baseline study on the prevalence of Salmonella in laying hen flocks can be found at the website of the European Food Safety Authority). (http://www.efsa.europa.eu/en/science/monitoring_zoonoses/reports/zoon_report_finbroilers.html).

Case definition, notification, sanitary measures and vaccination

A poultry layer flock is declared positive if Salmonella Enteritidis is isolated at one day of age or during rearing. In addition, the flock is positive if Salmonella belonging to any serotype is isolated within 3 weeks before slaughter. As for broilers, a flock is declared positive if in one of the samples Salmonella is isolated. Salmonella is notifiable to the Federal Agency for the Safety of the Food Chain since January 2004.

In case of positive findings in layers, the poultry house must be cleaned and disinfected after removal of the positive flock. If Salmonella was detected in a broiler flock at 3 weeks before slaughter, the birds were slaughtered at the end of the day (logistic slaughter).

Vaccination is strongly recommended for layers. Both attenuated and inactivated vaccines are available.

Epidemiological investigations and results of 2006 surveillance

In laying hen flocks within 3 weeks before slaughter, 34 out of 844 samples were positive for Salmonella, corresponding to 33 out of 676 flocks (4.9%) and 32 out of 349 farms. Serotype data were not available; see results from NRL. Testing of 181 flocks at one day of age and 40 flocks during rearing resulted negative for Salmonella.

The figures of layer flock contamination for 2006 are comparable to those of 2005 when approximately 6% of the flocks were found positive. The contrast with the figures from 2004 (27% of laying hen flocks) is significant, and probably in part due to the recommended vaccination of the layers.

As for broilers, 5 003 flocks were tested as one-day old chickens, of which 16 (0.3%) were found infected. Three weeks before slaughter 312 (3.6%) of 8 593 flocks were found to be contaminated with Salmonella, corresponding to 162 out of 1 065 farms. Serotype data were not available; see results from NRL.

Laboratory findings of the NRL show that more than 90% of the strains from poultry were isolated in the context of the European monitoring among broilers. The proportion of serotype Enteritidis isolates (27.7%) among poultry Salmonella remained almost the same as in 2005 (28.3%); that of Salmonella Typhimurium decreased slightly from 8.5% in 2005 to 5.2% in 2006. However, the proportion of Salmonella Paratyphi B (both tartrate positive and negative strains; all from broilers) raised from 7.8% in 2005 to 23.2% in 2006. Also Salmonella Bredeney (all from broilers) raised from 1.3% in 2005 to 11.1% in 2006. Nine isolates originated from layers; all were serotype Enteritidis.

During the last ten years, the number of poultry isolates sent to the laboratory was situated between approximately 700 and 1 100, except for 2005 when the European co-ordinated monitoring among layers caused a significant rise of isolates (almost 1 500 in total). In 2006, when a similar monitoring among broilers ran, almost thousand strains were tested. The figures show that Salmonella Enteritidis is still of major concern, also among broiler flocks. Salmonella Paratyphi B becomes more and more prevalent (among broilers). These isolates are frequently (70% of the isolates) multiple resistant to antimicrobials. Salmonella Typhimurium fluctuated between 5.5% and 13.0%

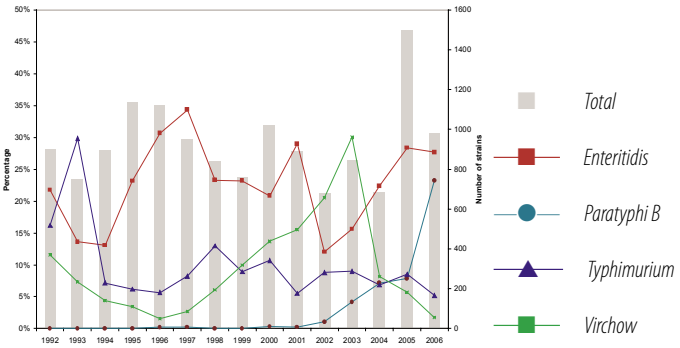

Figure 12. *Evolution of the percentages of the principal Salmonella serotypes isolated from poultry between 1992 and 2006. The bars represent the total number of poultry isolates per year, and refer to the right axis; the lines represent the percentage of each serotype per year and refer to the left axis.*

Salmonella in turkeys

Surveillance programme and sampling

The national control programme for Salmonella in turkeys is performed according to the sanitary qualification act (see before). Sanitary Qualification A is mandatory for all commercial breeding flocks. Flocks are at least sampled as day-old chickens, at the age of 26 weeks when entering the production unit if this is on a different farm than the rearing unit, and within the last 3 weeks before slaughter. Meat production flocks are sampled within three weeks of slaughter if the holding has a capacity of more than 5 000 birds (Sanitary Qualification B). On a voluntary basis, one-day-old birds may be sampled also.

Samples for day-old-birds are taken at the farm, and consist of pieces (5 by 5 cm) of the inner linings of delivery boxes. Two samples, each composed of 20 pieces of inner linings, are taken for each flock, one for the hen chicks and one for the cock chicks. The two samples are analyzed separately according to ISO 6579:2002.

At 26 weeks, 60 blood samples were taken of each breeder flock. If one or more blood sample are positive, faecal samples are taken to confirm the results. The owner takes faeces samples from the delivery boxes at time of delivery. A sample consists of 60 x 5 to 10g sub-samples taken from every flock with different origin of rearing. The samples have to be examined by an accredited laboratory within 48 hours.

Within 3 weeks before slaughter, the owner takes a pooled faecal sample consisting of 60 x 1g sub-samples of each flock. Alternatively, the sampling may consist of a pooled faecal sample of 60 x 1g taken by hand, or recovered from two pair of overshoes that were pooled for analysis.

Case definition, sanitary measures and vaccination policy

A turkey flock is considered positive if zoonotic Salmonella serotypes were isolated. Measures are taken only at time of slaughter: if the flock is Salmonella positive, it is slaughtered at the end of the day (logistic slaughter). There is no vaccination policy for breeding flocks, nor for meat production flocks.

Notification of zoonotic Salmonella to the Federal Agency for the Safety of the Food Chain is compulsory since January 2004.

Results of the investigation in 2006

Two breeding flocks were tested and were found negative for Salmonella. As for the 13 meat producing flocks which were analysed, 2 were positive for Salmonella: one Salmonella Kottbus and one Salmonella Stanleyville.

Salmonella in ducks and partridges

The surveillance programme for breeder animals of ducks, and for meat producing ducks is similar to that of turkeys (sanitary qualification A for breeders and B for meat production).

Two duck breeding flocks were tested and found negative for Salmonella. In addition, 24 meat production flocks of ducks were tested, and 4 were positive for Salmonella (serotypes Kottbus and Typhimurium).

Finally, the 2 parent flocks of partridges that were tested were free of Salmonella.

Salmonella in pigs

Serology

Surveillance programme in fattening pigs

Similar to 2005, in 2006 the blood samples from fattening and growing pigs that were taken in the framework of the monitoring of Aujeszky's disease in 2006 were also analysed for Salmonella. Blood samples from pigs were taken every 4 months. Depending on the number of pigs in the farm, 1 to 12 blood samples were taken. The analysis for Salmonella-specific antibodies was done in the veterinary laboratories ARSIA and DGZ by means of a commercially available ELISA kit, following the manufacturer's instructions.

The aim of the current voluntary surveillance programme is to identify maximum 10% of pig farms with the highest Salmonella prevalence and the identification Salmonella-specific risk factors in these herds. Indeed, it is likely that those herds, when participating in the supportive control programme, will benefit the best results in terms of decreasing the risk for Salmonella infections. Statistical modelling on the available data from 2005 and 2006 will serve to choose the best possible algorithm for identifying the problem herds.

Pigs were not vaccinated in 2006, since no vaccine was authorised in Belgium.

Results in 2006

A total of 207 843 serological analyses were performed. Of these, 21 026 samples (10.2%) had a S/P ratio above 1, which is lower than the 12.7% samples in 2005. On the basis of these preliminary results, the Federal Agency for the Safety of the Food Chain will consider in 2007 a way to identify pig holdings at risk taking into account the sampling plan and trends observed within the serological data.

Bacteriology

There was no surveillance system for Salmonella in pigs based on bacteriology. However, several samples were taken for research activities.

Laboratory findings from the National Reference Laboratory showed that almost a similar number of pig Salmonella strains were typed in 2006 as compared to former years, i.e. n=481. Among these, Salmonella Typhimurium (69.0%) [74.4% belong to Classic variant O5+] was the most prominent serotype, followed by Salmonella Derby (16.4%). Salmonella Typhimurium continues to be the most prevalent serotype among pig isolates, with a tendency to increase in importance.

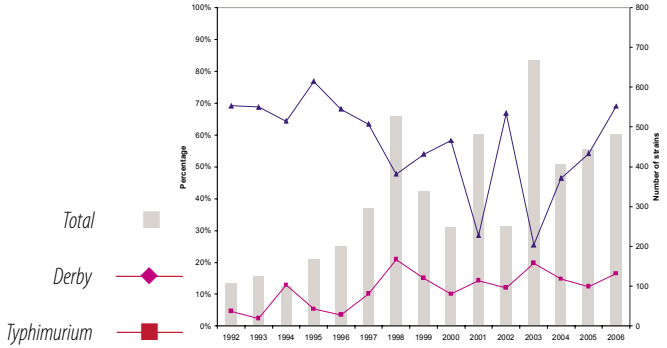

Figure 13. *Evolution of the percentages of the principal Salmonella serotypes isolated from pigs between 1992 and 2006. The bars represent the total number of pig isolates per year, and refer to the right axis; the lines represent the percentage of each serotype per year and refer to the left axis*

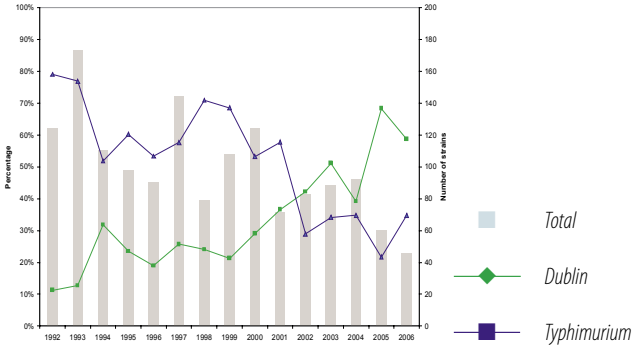

Figure 14*. Evolution of the percentages of the principal Salmonella serotypes isolated from cattle between 1992 and 2006. The bars represent the total number of cattle isolates per year, and refer to the right axis; the lines represent the percentage of each serotype per year and refer to the left axis.*

Salmonella in cattle

There was no official monitoring programme for Salmonella in cattle in 2006. Salmonella iolates were sent on a voluntary basis to the National Reference Laboratory for serotyping.

In Belgium no Salmonella vaccine was authorised in cattle.

According to the National Reference Laboratory. the number of cattle Salmonella isolates analysed decreases each year: n=92 in 2004, n=60 in 2005 and 46 in 2006. Most frequently found serotypes were Salmonella Dublin (59.7%) and Salmonella Typhimurium (30.4%), which is in line with the remarkable increasing trend of Salmonella Dublin since 2000. Salmonella Dublin is the principal serotype in cattle since 2002. Salmonella Typhimurium is on a second place.

Salmonella in food (meat and meat products)

Surveillance programme

In 2006, the Federal Agency for the Safety of the Food Chain selected for its monitoring programme more than 200 Belgian slaughterhouses, more than 100 meat cutting plants and more than 100 retail points representative of the Belgian production of carcasses and meat.

Sampling for Salmonella was done on the following matrices: carcasses, trimmings and minced meat of pork, minced meat and meat preparations of beef, carcasses and fillets of broilers and layer carcasses. Sampling of pork carcasses was done by

means of swabs. The carcass samples of broilers and layers consisted of 10g of neck skin. The following samples were analysed: 25g (trimmings, minced meat of pork, chicken and beef), 600 cm2 (pork carcasses), 1g (broiler carcasses) and 0.1g (layer carcasses). Sampling was done by specially trained staff. For most matrices, approximately 100 - 300 independent samples were taken per matrix in order to detect a minimal contamination rate of 1% with 95% confidence.

Notification is mandatory since March 2004 (Ministerial Decree on mandatory notification in the food chain). For Salmonella, absence in 25g in ready-to eat food is requested.

Epidemiological investigations and results of 2006 surveillance

Table 10. *The results of the monitoring – Salmonella in meat and meat products*

Species	Quantity of sample analysed	Preva-lence	Pre-dominant serotype	Other serotypes(in decreasing order)
Beef				
Minced meat at processing plant (n=125)	25g	1.3%	Typhimurium	
Minced meat at retail (n=35)	25g	0.0%		
Meat preparation (steak tartare) at retail (n=124)	25g	0.0%		
Pork				
Carcasses at slaughter (n=154)	600cm^2	7.1%	Typhimurium	Derby, Mbandaka
Trimmings (n=328)	25g	2.4%	Derby	Typhimurium
Minced meat (n=142)	25g	3.5%	Typhimurium	Derby
Raw meat product (n=21)	25g	0.0%		
Broilers				
Carcasses at slaughter (n=69)	1g	1.4%	Bredeney	
Carcasses at slaughter (n=6432)	25g (caeca)	9.3%	Paratyphi B	Bredeney, Blockley, Typhimurium, Enteritidis, Hadar, Infantis, Indiana, Agona, Seftenberg, Virchow, Livingstone, Bivismorbificans, Minnesota, Cleveland, Kottbus, Saintpaul, Anatum, Brandenburg, Heidelberg
Carcasses at retail (n=40)	1g	2.5%	Typhimurium	
Fillets (n=293)	1g	13.3%	Bredeney	Paratyphi B, Enteritidis, Typhimurium, Blockey, Virchow
Layers				
Carcasses at slaughter (n=101)	0.1g	26.1%	Enteritidis	Livingstone
Carcasses at slaughter (n=1017)	25g (caeca)	26.2%	Enteritidis	Livingstone, Infantis, Typhimurium, Agona, Derby, Havana, Worthington, Rissen, Hadar, Bredeney, Seftenberg, Mbandaka

The contamination of pig carcasses, trimmings and minced meat decreased in 2006.

The contamination of layer carcasses increased in 2006.

The contamination of broiler carcasses and broiler fillets decreased in 2006

The contamination of minced meat of beef with Salmonella is limited.

***Table 11**. Evolution of the food Salmonella prevalence 2000-2006*

	Samples	Sampling level	2000	2001	2002	2003	2004	2005	2006
Pork	Carcasses	600cm2	24.1%	20.8%	15.4%	14.6%	12.3%	9.3%	7.1%
	Trimmings	25g	32.3%	17.7%	11.2%	6.1%	10.4%	7.3%	2.4%
	Minced meat	25g	16.6%	10.3%	11.0%	6.4%	9.4%	6.5%	3.5%
Broilers	Carcasses	1g	6.6%	11.4%	7.0%	12.1%	7.9%	5.7%	1.4%
	Minced meat	25g			21.0%	29.3%	18.5%	15.9%	
	Fillets	25g	12.7%	15.1%	12.6%	11.7%	20.6%	14.2%	13.3%
Layers	Carcasses	0.1g	26.7%	21.9%	20.3%	18.6%	19.6%	14.0%	26.1%
Beef	Carcasses	1600 cm2		2.7%	0.0%				0.0%
	Minced meat	25g	6.1%	2.7%	3.3%	0.3%	2.1%	0.6%	1.3%

Salmonella in other food

In the national monitoring of milk and dairy products, no Salmonella was found in 25g samples of raw cows' milk cheese at farm (n=194), at processing plant (n=18) and at retail (n=98), raw goats' milk cheese at farm (n=12) and at retail (n=10), raw sheep's milk cheese at farm (n=7) and at retail (n=10), ice cream at farm (n=10) and at processing plant (n= 10), butter made from raw milk at farm (n=30) and at retail (n=16).

In the national monitoring of other food, 25g samples of egg products (n=135), desserts containing raw eggs (n=126), bakery products with egg fillings (n=162), species and herbs at retail (n=59) and at processing (n=69), ready-to-eat pre cut fruits and vegetables at retail (n=87) and at processing (n=34), ready-to-eat prepared dishes at retail (n=113) and at processing (n=48), chocolate or confectionary containing chocolate at retail (n=97) or at processing (n=23), live bivalve molluscs (n=92) were analysed. Salmonella was only found in species and herbs at processing (7.2%).

Salmonella in humans

Surveillance programme and methods used

Data about human salmonellosis cases and human isolates were obtained from 161 clinical laboratories. All isolates were serotyped by slide agglutination with commercial antisera following the Kauffmann-White scheme. When necessary, additional biochemical tests were performed to confirm the identification or to differentiate between the subspecies. Phage typing and antimicrobial susceptibility testing were performed on isolates randomly sampled from the four serotypes Enteritidis, Typhimurium, Hadar and Virchow. Two additional serotypes (Brandenburg and Derby) were also randomly sampled, all isolates of Salmonella Infantis, Newport, Typhi and Paratyphi selected and tested for their antimicrobial susceptibility.

The objective of the national surveillance programme is to document the occurrence and trends of serotypes, to detect local, regional, national or even international outbreaks, to find and eliminate the source and to suggest preventive actions to the Federal Agency for the Safety of the Food Chain. This national Salmonella surveillance also intended to rapidly interact at the international level via electronic communication (with the Enter-net international surveillance network) and helped detecting outbreaks and targeting preventive strategies.

Epidemiological investigations and results of 2006 surveillance

From 1987 on, a remarkable increase in the number of human salmonellosis cases was registered, consecutively to the rise of the serotype Enteritidis, leading to a peak of 15 774 cases in 1999 (Figure 1, Table 1). In that year, exceptionally high numbers of Salmonella Enteritidis. Between 2000 and 2004, the total number of laboratory-confirmed cases varied between 14 088 and 9 543 (Table 1). In 2003, the high number of salmonellosis cases mainly resulted from the increase of the serotype Enteritidis. These isolates exceeded for the first time 70% of the total number of Salmonella strains analysed. From 2005 a substantial decrease of Salmonella Enteritidis infections compared with the annual number of cases in the period 2000-2004 was recorded. This decrease persisted in 2006 where the total number of cases caused by Salmonella spp. and by Salmonella Enteritidis decreased to 3693 and 1052 cases, respectively.

In recent years, the number of Salmonella Typhimurium isolates remained at a level of about 2 500 strains per year, but started to decrease from 2005 (Table 1). After decreasing over the last years, Salmonella Infantis increased in 2004 up to more than 100 cases to become the third serotype in human cases in 2004, but decreased to 58 and 37 cases in 2005 and 2006, respectively. Regarding Salmonella Virchow, about 140 to 150 isolates were annually registered from 2000 to 2003, whereas from 2004 less than 100 strains were yearly reported. A remarkable drop of Salmonella Brandenburg (322 in 2000 vs about 60 from 2003 to 2006) cases was noted over the last years. Similarly, the number of Salmonella Derby cases is shrinking since the beginning of 2000 but remained stable over the period 2004-2006.

***Table 12**. Trends for the most prevalent Salmonella serotypes from 1986 to 2006*

	1986	1987	1988	1989	1990	1991	1992	1993	1994	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006
Total	6092	6360	8247	9752	11695	10891	10391	10840	11294	10754	12008	14239	14514	15774	14088	11065	10075	12792	9543	4916	3693
Enteritidis	298	320	1163	2236	3382	4721	4084	5260	5700	5138	6145	8284	9003	10492	9503	7112	6398	9118	6075	2226	1052
Typhimurium	3512	3233	3699	4018	4756	3652	3835	3528	3418	3623	3522	3347	3221	3348	2799	2370	2438	2486	2459	1659	1826
Others	2282	2807	3385	3498	3557	2518	2472	2052	2176	1993	2341	2608	2290	1934	1786	1583	1239	1188	549	691	815
Derby	131	169	168	177	161	134	139	103	113	107	118	157	162	138	169	158	92	100	64	67	52
Brandenburg	167	151	159	255	302	176	161	147	204	241	214	296	274	279	322	200	148	66	63	76	47
Virchow	152	170	235	293	302	224	295	273	308	245	178	114	115	86	147	143	132	152	91	65	46
Infantis	168	173	168	275	249	224	225	160	150	174	267	263	180	169	120	126	74	52	107	58	37

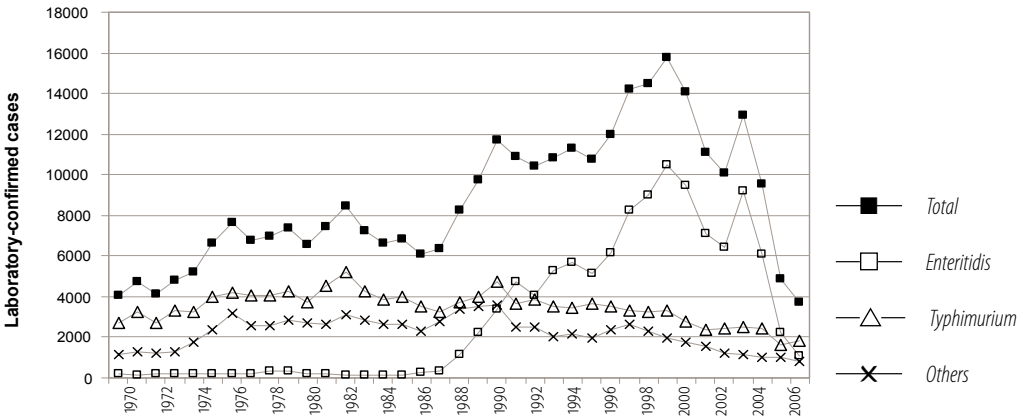

***Figure 15.** Trend of the human Salmonella isolates and of the two major serotypes Enteritidis and Typhimurium over the last thirty six years in Belgium: number of laboratory confirmed cases*

Age and seasonal distribution

Most cases of salmonellosis were reported in children less than 5 years old (44.4% of cases), with no significant gender difference.

Table 13. *Human cases of Salmonella: Age and gender distribution, 2006. Note that the gender of all salmonellosis cases is not known. M: male; F: female; SR: sex ratio*

	Salmonella				Salmonella Enteritidis				Salmonella Typhimurium			
Age	Total	M	F	SR	Total	M	F	SR	Total	M	F	SR
< 1 year	321	170	147	1.2	63	32	31	1.0	130	70	56	1.3
1 to 4 y	1320	689	615	1.1	314	161	150	1.1	838	429	399	1.1
5 to 14 y	613	317	287	1.1	167	88	77	1.1	380	188	185	1.0
15 to 24 y	194	93	98	0.9	70	37	33	1.1	64	27	36	0.8
25 to 44 y	301	130	168	0.8	109	44	64	0.7	76	31	44	0.7
45 to 64 y	261	125	135	0.9	105	38	67	0.6	71	37	34	1.1
≥ 65 y	340	134	203	0.7	119	43	75	0.6	113	39	72	0.5
unknown	343	124	116	1.1	105	40	32	1.3	154	53	53	1.0
Total	3693	1782	1769	1.0	1052	483	529	0.9	1826	874	879	1.0

Regarding the seasonal distribution (Figure 2), about 200 to 400 cases were monthly reported between January and July 2006. From August until September, the monthly number of isolates increased, to reach about 500 isolates. From October to December, the monthly number of isolates gradually decreased.

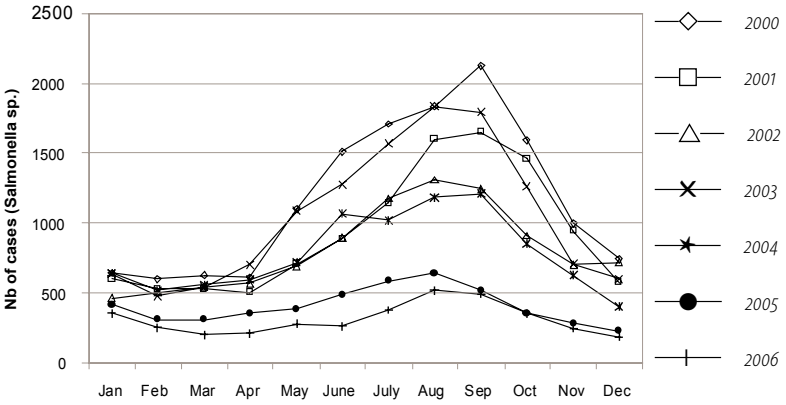

Figure 16. *Seasonal distribution of Salmonella isolates among humans from 2000 to 2006.*

Antimicrobial resistance

Antimicrobial resistance in isolates from living animals

Methods used

Data on antibiotic resistance of Salmonella strains from livestock came from the National Reference Laboratory. Susceptibility tests were performed by the disk diffusion test, using Neo-Sensitabs (Rosco). Only one isolate per file (LIMS: electronic laboratory information management system) was selected for susceptibility testing (see further). Tests and interpretation were done according to the manufacturers guidelines using an inoculum and breakpoints as described by CLSI (formerly NCCLS) (Kirby-Bauer). Internal control was performed with quality control strain E. coli ATCC25922. Results were accepted when results with the QC strain were within the limits as proposed by Rosco.

Table 14. *Animal Salmonella: list of antimicrobials tested. For all susceptibility tests Neo-Sensitabs from Rosco were used according to the providers instructions*

Antimicrobial	Amount of antimicrobial	Breakpoints (mm)
Ampicillin	33µg	17 - 19
Ceftiofur	30µg	20 – 22
Streptomycin	100µg	23 – 25
Neomycin	120µg	20 – 22
Gentamicin	40µg	20 – 22
Tetracycline	80µg	20 – 22
Sulfonamides	240µg	20 – 22
Trimethoprim - sulfonamides	5.2µg + 240µg	27 – 31
Nalidixic acidid	130µg	21 – 24
Enrofloxacin	10µg	20 – 22
Chloramphenicol	60µg	21 – 24
Florfenicol	30µg	15 - 18

Epidemiological investigations and results of 2006 surveillance

The susceptibility of 1 278 Salmonella strains was tested. Isolates were to a reasonable extent independent from each other: within the same file (LIMS) only one isolate from a group of isolates with the same serotype was selected for susceptibility testing.

A total of 839 Salmonella isolates (65.6%) was fully susceptible to all antimicrobial drugs tested. Most resistance was found against ampicillin (24.5%), sulfonamides (22.2%), streptomycin (15.3%), tetracyclin (14.6%), nalidixic acid (12.8%) and trimethoprim - sulfonamides (11.3%).

Sixty-four strains were found resistant against chloramphenicol (5.0%); about 62% of these isolates were also resistant against florfenicol. Moreover, 55 isolates were found ceftiofur resistant (4.3%). These cephalosporin resistant strains only originated from poultry (n=52) and from food (n=3). In addition, nine enrofloxacin resistant strains (0.7%) (seven Salmonella Typhimurium, one Group E1-E2-E3 and one non typable) were detected. Finally, 5 neomycin resistant strains were found. All the isolates were found sensitive to gentamicin.

Most (95,5%) Salmonella Agona isolates (n=66) were fully susceptible for all antimicrobials tested.

About 46.7% of Salmonella Blockley isolates (n=30; all from poultry) were completely sensitive, but 12 isolates had profile ampicillin, nalidixic acid, sulfonamides, tetracycline, trimethoprim - sulfonamides.

Most of Salmonella Derby strains (n=28) were sensitive (82.1%), although some resistance against sulfonamides (17.9%), streptomycin (10.7%) and trimethoprim - sulfonamides (10.7%) was noticed.

As for Salmonella Dublin isolates (n=15; all from cattle), 53.3% were found completely susceptible. Resistance against sulfonamides (33.3%), chloramphenicol (33.3%) and nalidixic acid (26.7%) was noticed.

Most Salmonella Enteritidis isolates (n=165) were susceptible (93.9%). Some resistance was found against nalidixic acid (3.6%; 6 isolates) and against ampicillin (1.8%).

All Salmonella Hadar (n=27) strains were found resistant against nalidixic acid (100%). In addition, tetracyclin (85.2%) and ampicillin (51,9) were frequently found; most strains (51.9%; 14 isolates) were resistant to all three antimicrobials.

Half of the tested Salmonella Indiana strains (n=6; all from broilers) were fully susceptible. Three had the profile ampicillin, chloramphenicol, tetracycline, sulfonamides, trimethoprim - sulfonamides.

About half of the Salmonella Infantis strains (n=24) were fully susceptible (58.3%). Strains were mainly resistant against ampicillin (41.7%), ceftiofur (25.0%) and streptomycin (16.7%). Some isolates (8.3%) were nalidixic acid resistant.

As for Salmonella Paratyphi B (all originated from broilers and from food with chicken), tartrate positive (i.e. var. Java) and tartrate negative strains seem to have slightly different antibiotic resistance profiles. Salmonella Paratyphi B var. Java (n=90) were in 90% of cases resistant to one or more antibiotic, with most resistance against ampicillin (72.2%), streptomycin, sulfonamides and trimethoprim - sulfonamides (all three about 66%) and nalidixic acid (55.6%). Fourty (44.4%) of the isolates showed profile ampicillin, streptomycin, sulfonamides, trimethoprim - sulfonamides. As for Salmonella Paratyphi B, tartrate negative isolates (n=14), 21.4% were fully sensitive, and especially ampicillin, sulfonamides, trimethoprim - sulfonamides and nalidixic acid resistance (all 57.1%) was registered. Profile ampicillin, sulfonamides, trimethoprim - sulfonamides was most abundant (35.7%).

About 54.6% of Salmonella Typhimurium isolates (n=174) were found susceptible; classic variant (O5+) strains were found slightly more often susceptible (35.9%) than Copenhagen variant (O5-) isolates (31.6%). The multiresistance profile ampicillin,

streptomycin, tetracycline, sulfonamides was encountered in only 12.6% of O5+, whereas this profile could be detected in 44.7% of O5- isolates. Pentaresistance ampicillin, streptomycin, tetracycline, sulfonamides, chloramphenicol in Classic and Copenhagen variants reached 7.1% and 26.3%, respectively.

All of the Salmonella Virchow isolates (n=25) were resistant to nalidixic acid (100%). Also ampicillin (36.0%) and ceftiofur (20.0%) resistances were noteworthy.

Some strains belonging to other serotypes were also tested, but to a lesser extent. Most of these isolates were fully sensitive for all the antimicrobials tested.

Antimicrobial resistance in strains isolated from meat

During 2006, all 203 strains of Salmonella enterica isolated from poultry meat and from pork during the zoonosis monitoring program were sent to the Scientific Institute of Public Health for serotyping and determination of antimicrobial resistance. Not one Salmonella was isolated from beef. Meat samples included carcasses, meat cuts and minced meat. Minimum Inhibitory Concentrations (MIC) were determined by the use of E-test. The antimicrobials tested were ampicillin, ceftriaxon, chloramphenicol, ciprofloxacin, kanamycin, nalidixic acid, streptomycin, sulfamethoxazole, tetracycline, trimethoprim and trimethoprim – sulfonamides. Interpretation of the results was according to CLSI. Quality control was performed by using an Escherichia coli ATCC 25922 strain. Breakpoints used are listed in the following table.

Table 15. *Salmonella from meat and meat products: list of antimicrobials tested with their breakpoints*

Antimicrobial	Breakpoints(mg / ml)
Ampicillin	8 – 32
Ceftriaxone	8 – 64
Streptomycin	8 – 32
Kanamycin	16 – 64
Tetracycline	4 – 16
Sulfamethoxazole	256 – 512
Trimethoprim	8 – 16
Trimethoprim - sulfonamides	2 – 4
Nalidixic acid	16 – 32
Ciprofloxacin	1 – 4
Chloramphenicol	8 – 32

The level of resistance of Salmonella isolates from broilers and pork is influenced by the serotype distribution in the corresponding meat. The presence of highly resistant serotypes as Hadar, Virchow, Paratyphi B and Typhimurium contributed mainly to the high resistance levels in some matrices. The results for poultry meat and pork are summarized in the next table.

Table 16. *Antimicrobial susceptibility testing of Salmonella spp. isolated from meat: percentage of resistant strains*

Antimicrobial tested	Poultry meat (n=132)	Pig meat (n=21)
Ampicillin	15	19
Ceftriaxon	3	0
Streptomycin	13	19
Kanamycin	0	0
Tetracycline	14	38
Sulfamethoxazole	14	24
Trimethoprim	15	14
Trimethoprim+sulfonamides	14	14
Nalidixic Acid	11	0
Ciprofloxacine	0	0
Chloramphenicol	7	5

Antimicrobial resistance in strains isolated from poultry meat

In 2006, 132 Salmonella enterica isolates from poultry meat were tested for their antimicrobial susceptibility. Of all tested strains 70% were sensitive for all tested antibiotics. Most resistance was found to sulfamethoxazole (14%), tetracycline (14%), streptomycin (13%) trimethoprim and trimethopri m+sulfonamides (15%), ampicillin (14%) and nalidixic acid (11%). Chloramphenicol resistance was observed in 7% of the Salmonella strains isolated from poultry meat. Four strains (3%) were resistant against the cephalosporin ceftriaxon. No resistance was found for ciprofloxacin and kanamycin. From the Salmonella isolates from broiler the percentage of resistance decreased considerably for almost all the antibiotics tested except for ceftriaxon and chloraphenicol where a slight increase in the resistance was noticed in comparison with 2005.

For 2006, 51 Salmonella Enteritidis isolates from poultry meat were tested for their susceptibility to all antimicrobials. The resistance in this serotype is very low as was found in previous years. Only two strains showed resistance, one against ampicillin and the other strain against streptomycin and nalidixic acid.

All Salmonella Paratyphi B (n=9) isolates were resistant against at least one or more antimicrobials. The serotypes Agona (1) Derby (1) and Infantis (7) were fully sensitive against all tested antimicrobials.

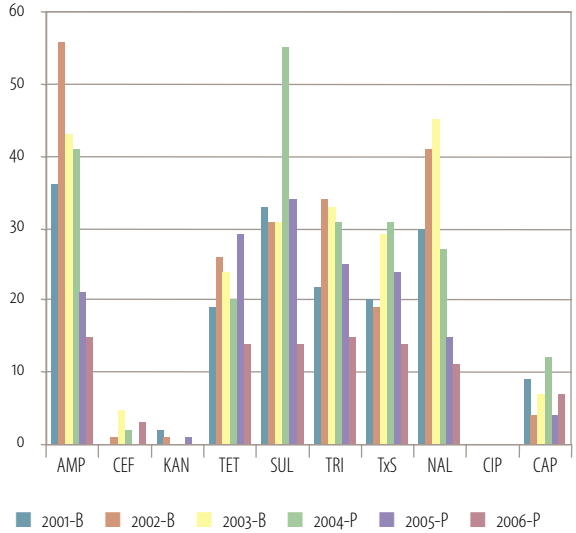

Figure 17. *Percentage resistant Salmonella strains in broiler meat (2001-2003) and poultry meat (2004-2006)*

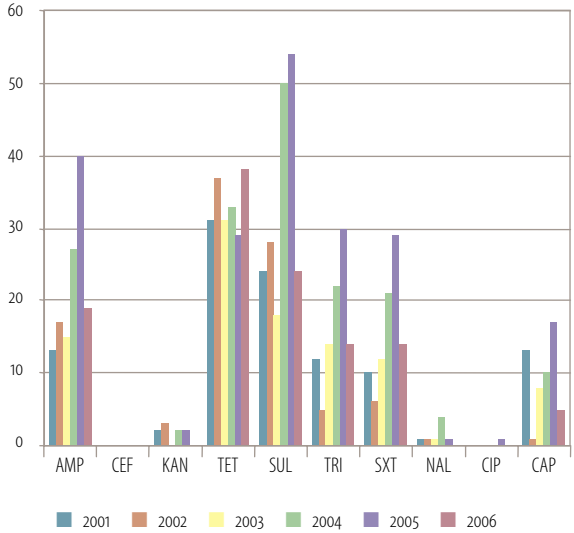

Figure 18. *Percentage resistant Salmonella strains in pork (2001-2006)*

Antimicrobial resistance in strains isolated from pork

In total 21 Salmonella strains from pork were tested for their susceptibility. Salmonella Typhimurium (10) and Salmonella Derby (7) are the two most frequently isolated serotypes from pork. In total 57% of the strains were sensitive to all tested antibiotics. A high degree of resistance was determined for tetracycline 38%, sulfamethoxazole 24% and streptomycin 19%. No resistance was noticed to ceftriaxon, ciprofloxacin, kanamycin and nalidixic acid. Only 1 strain was resistant against chloramphenicol. Multi-resistance was observed in 19% of the strains (> 4 antimicrobials). Compared to 2005 a general decrease in antimicrobial resistance was observed except for tetracycline.

Antimicrobial resistance and phage typing of human isolates

Methods used

A total of 1075 human Salmonella isolates randomly selected from the six most important serotypes in 2006 (Enteritidis, Typhimurium, Hadar, Virchow, Brandenburg and Derby), comprising as well all isolates of the serotypes Infantis, Newport, Typhi and Paratyphi, were examined for their resistance. Thirteen antibiotics of therapeutic or epidemiological interest were tested in disk diffusion according to Kirby-Bauer, following CLSI procedures.

Table 17. *List of antimicrobials used for susceptibility testing of Salmonella*

Antimicrobial	Amount of antimicrobial	Breakpoints (mm)
Ampicillin	10 µg	14 - 16
Amoxicillin + clavulanic acid	20/10 µg	14 - 17
Cefotaxime	30 µg	15 - 22
Streptomycin	10 UI	12 - 14
Kanamycin	30 UI	14 - 17
Neomycin	30 UI	15 - 17
Gentamicin	10 µg	13 - 14
Tetracycline	30 µg	12 - 14
Sulfonamides	300 µg	16 - 13
Trimethoprim	5 µg	15 - 11
Trimethoprim + sulfamethoxazole	1,25/ 23,75 µg	11 - 15
Nalidixic acid	30 µg	14 - 18
Ciprofloxacin	5 µg	16 - 20
Chloramphenicol	30 µg	13 - 17

Epidemiological history and results of 2006 surveillance

Resistance was mostly found to tetracycline (24.2%), sulfonamides (23.4%), ampicillin (23.2%), streptomycin (21.8%), and to a lesser extent to trimethoprim (10.3%).

The vast majority (89.5%) of human Salmonella Enteritidis isolates (n=493) was fully sensitive to all antimicrobials tested.

Salmonella Typhimurium (n=316) showed a high level of resistance; especially resistances to ampicillin (56.6%), sulfonamides (53.8%), tetracycline (59.8%) and streptomycin (49.1%) are striking. About half of the isolates (45.8%) were found resistant to four or more antimicrobial agents. In addition, almost 21% of the isolates showed multi-resistance to at least ampicillin, chloramphenicol, streptomycin, sulfonamides and tetracycline. About 66% of these multi-resistant isolates (ACS-SuT) were of phage type DT104.

Except one strain, all Salmonella Hadar isolates (n=15) were resistant to at least one antibiotic. Resistance to tetracycline, nalidixic acid, ampicillin and streptomycin reached values from 73% up to 93%. Simultaneous resistance to these four antibiotics was observed in 53.4% of these isolates. Resistance to sulfonamides significantly increased (up to 56%). However, isolates from this serotype remained fully sensitive to cefotaxime, ciprofloxacin, chloramphenicol and gentamicin.

In Salmonella Virchow (n=45), multi-resistance was less common as compared to 2003 (22.1% of the strains in 2006 instead of 60% of the 2003 isolates). The highest incidence of resistance was observed for nalidixic acid (57.8%). Resistances to ampicillin, tetracycline, sulfonamides, trimethoprim and trimethoprim+sulfonamides were common (approximately 30%). Two strains of Salmonella Virchow showed resistance to cefotaxime due to the presence of TEM-52 β-lactamase.

In contrast, the vast majority of Salmonella Brandenburg (n=46) and Salmonella Derby (n=67) isolates remained sensitive to the vast majority of tested antibiotics: 78.2% and 74.9% sensitive or resistant to one antibiotic, respectively.

Salmonella Infantis (N= 36) displayed in general a low level of multi-resistance.

The vast majority of Salmonella Paratyphi B var Java (N=25) were multi-resistant (72%). Resistance to nalidixic acid, trimethoprim, ampicillin and streptomycin reached values from 52% up to 80 %.

In contrast, the vast majority of Salmonella Newport (n=16) isolates remained sensitive to the vast majority of tested antibiotics: 87.5% were fully sensitive to all antimicrobials tested. However, two isolates displayed resistance to at least 8 antibiotics but remained sensitive to amoxicillin + clavulanic acid, cefotaxime and ciprofloxacin.

No tendency could be highlighted from the results on Salmonella Typhi. That could be due to the fact that most of isolates are travel-associated and that the origins (country/region) of the isolates were different.

In general, resistance patterns and levels of Salmonella isolated in 2006 were comparable to those from 2002-2005.

A total of 489 human Salmonella Enteritidis isolates were phage typed. Of these, 29.2% were PT 21 and 23.1% were PT 4. In addition, 316 Salmonella Typhimurium isolates were phage typed and most prevalent types were DT120 (28.7%), DT104 (15.8%), DT193 (10.1%), DT12 (5.1%), and U302 (2.8%).

Table 18. *Antimicrobial resistance in human Salmonella of serotypes Enteritidis, Typhimurium, Brandenburg, Derby, Hadar, Virchow, Infantis, Typhi, Newport, Paratyphi B and A isolated in 2006*

Serotype	Total	N												
			Amp	Amc	Ctx	Tet	Cip	Tmp	Neo	Nal	Chl	Gen	Kan	Str	Sul
Enteritidis	1052	493	5.1	0	0.2	0.8	0	1.0	0	5.5	0.2	0	0	0.8	1.0
Typhimurium	1826	316	56.6	2.2	0	59.8	0.3	16.5	0.9	3.8	25.0	0.9	1.3	49.1	53.8
Derby	52	52	3.8	0	0	23.1	0	3.8	0	1.9	1.9	0	0	26.9	34.6
Brandenburg	47	46	2.2	0	0	39.1	0	17.4	2.2	4.3	2.2	0	2.2	4.3	19.6
Virchow	46	45	28.9	2.2	2.2	33.3	0	28.9	2.2	57.8	2.2	2.2	2.2	11.1	33.3
Infantis	37	36	19.4	0	0	13.9	0	19.4	2.8	11.1	0	0	2.8	16.7	33.3
Typhi	19	20	20	0	0	15	0	20	0	55	20	0	0	75	20
Newport	16	16	12.5	0	0	12.5	0	12.5	6.25	12.5	12.5	12.5	6.25	12.5	12.5
Hadar	22	15	46.7	6.7	0	93.3	0	6.7	0	73.3	0	0	0	93.3	0
Paratyphi B var Java	14	14	64.3	0	7.1	14.3	0	57.1	0	57.1	14.3	0	0	64.3	78.6
Paratyphi B	4	4	0	0	0	0	0	25	0	25	0	0	0	25	0
Paratyphi A	12	13	0	0	0	0	0	0	0	84.6	0	0	0	0	0

Abbreviations antimicrobial; AMP, ampicillin; AMC, amoxicillin + clavulanic acid; CTX, cefotaxime; TET, tetracycline; CIP, ciprofloxacin; TMP, trimethoprim; NEO, Neomycin; NAL, nalidixic acid; CHL, chloramphenicol; GEN, gentamicin; KAN, kanamycin; STR, streptomycin; SUL, sulfonamides

trends and sources

report on zoonotic agents in belgium in 2006



listeriosis

Listeriosis

National evaluation of the recent situation, the trends and sources of infection

Listeria monocytogenes is of major concern to the food industry and public health authorities. Ingestion of food contaminated with L. monocytogenes may cause either a serious invasive illness affecting people with altered or deficient immune responses, or a non-invasive febrile gastro-enteritis. In Belgium, listeriosis most commonly takes the form of an infection of the uterus or the newborn (10%), the bloodstream (70%) or the central nervous system (10-20%). In pregnant women, this can result in spontaneous abortion, stillbirth or the birth of a severely ill baby. Although the incidence of listeriosis is low, the high case fatality rate, which often reaches as high as 20-30%, requires early diagnosis and appropriate antimicrobial therapy.

L. monocytogenes is also pathogenic for cattle and sheep where it may cause abortion and encephalitis.

Listeria is ubiquitous and widely distributed in the environment (soil, vegetables, meat, milk, fish) and is mostly transmitted to humans via consumption of contaminated food. Vulnerable people are advised not to eat food with a proven elevated risk of L. monocytogenes contamination. Unfortunately, the specific source of contamination is rarely demonstrated with cases of listeriosis in Belgium. The annual number of cases varies slightly but remains comparable with data from neighbouring countries (3-8 cases per million inhabitants).

Relevance of the findings in animals, feedingstuffs and foodstuffs to human cases (as a source of infection)

A monitoring programme was organised by the Federal Agency for the Safety of the Food chain. More than 100 meat cutting plants and more than 200 retail trades representative of the Belgian production of food, were selected for this study. The matrixes were minced meat of pork, beef, cooked ham, pâté, salami, smoked salmon and milk and dairy products.

- Listeria monocytogenes in food
- Listeria monocytogenes in humans

Recent actions taken to control the zoonoses

General food hygiene rules are essential for the prevention of human listeriosis. As some persons are at high risk (pregnant women, the eldery, immuno-compromised people), they are advised not to eat certain categories of food with proven elevated risk of L. monocytogenes contamination, such as unpasteurized milk and butter, soft cheeses and ice cream made from unpasteurized milk, any soft cheese crust, smoked fish, pâté, cooked ham, salami, cooked meat in jelly, raw minced meat from beef, pork and poultry, steak tartar, raw fish and shellfish (oysters, mussels, shrimps), fish, meat and surimi salads, insufficiently rinsed raw vegetables, unpeeled fruit. People should be made aware of the risk to all ready-to-eat food products.

Listeria monocytogenes in food

Surveillance programme and methods used

Monitoring programme

In 2006, the Federal Agency for the Safety of the Food Chain selected for its monitoring programme more than 100 meat cutting plants and more than 100 retail trades representative of the Belgian production of carcasses and meat.

The matrices for Listeria isolation were minced meat, meat preparations and meat products from pork, beef and chicken, cheeses and other diary products, smoked salmon and other food products and prepared dishes.

Results of the 2006 monitoring

See table 19 on the following page.

Table 19. *Zoonosis monitoring programme - Listeria monocytogenes in food (2006)*

Sample		Quantity analysed	Percentage of positive samples
Beef	Minced meat at processing plant (n=67)	25g	14,9%
	Minced meat at retail (steak tartare) (n=36)	Enumeration (M=100 cfu/g)	0.0%
	Meat preparation at retail intended to be eaten raw (prepared steak tartare) (n=117)	Enumeration (M=100 cfu/g)	0.0%
Pork	Minced meat at processing plant (n=123)	1g	11.4%
	Cooked ham at processing plant (n=69)	25g	1.4%
	Cooked ham at retail (n=69)	Enumeration (M=100 cfu/g)	0.0%
	Pâté at processing plant (n= 79)	25g	1.3%
	Pâté at retail (n=72)	Enumeration (M=100 cfu/g)	0.0%
	Sausages at processing plant (n= 25)	25g	44%
	Sausages at processing plant (n= 48)	Enumeration (M=100 cfu/g)	0.0%
	Sausages at retail (n=41)	Enumeration (M=100 cfu/g)	0.0%
Poultry	Meat preparation at retail intended to be eaten cooked (n=377)	1g	40.6%
	Meat preparation at processing plant intended to be eaten cooked (n=386)	1g	19.9%
	Minced meat at retail (all species) (n=44)	1g	13.6%
Other food products and prepared dishes	Unspecified RTE foods (n=1303)	Enumeration (M=100 cfu/g)	0.0%
Cheeses	Cheeses made from raw or low heated cow milk at retail (n=126)	Enumeration (M=100 cfu/g)	0.0%
	Cheeses made from pasteurised cow milk at retail (n=144)	Enumeration (M=100 cfu/g)	0.0%
	Cheeses made from raw or low heated cow milk at farm (n=235)	1g	4.3%
	Cheeses made from raw or low heated cow milk at processing plant (n=29)	25g	0.0%
	Cheeses made from pasteurised cow milk at farm (n=32)	1g	0.0%
	Cheeses made from pasteurised cow milk at processing plant (n=79)	25g	1.3%
Dairy products	Butter made from raw or low heat-treated milk at farm (n=118)	1g	0.0%
	Butter made from raw or low heat-treated milk at retail (n=37)	Enumeration (M=100 cfu/g)	0.0%
	Butter made from pasteurised cow milk at processing plant (n=112)	25g	0.0%
	Ice cream at farm (n=66)	1g	0.0%
	Ice cream at processing plant (n=68)	1g	0.0%
Fish	Smoked salmon at processing plant (n=150)	25g	21.3%
	Smoked salmon at retail (n=142)	Enumeration (M=100 cfu/g)	0.7%

The results of the monitoring and the trends of Listeria monocytogenes prevalence since 2000 are shown in Table 20.

Table 20. *Evolution of the food Listeria monocytogenes prevalence 2000-2006*

		Sampling level	2000	2001	2002	2003	2004	2005	2006
Pork	Minced meat	1g	25.0%	18.3%	20.7%	21.5%	17.6%	10,2%	12.0%
	Cooked ham	25g	6.0%	4.6%	3.0%	2.5%	3.8%	4.5%	1.4%
	Pâté	25g	4.3%	4.9%	5.4%	4.0%	1.2%	1.4%	1.3%
Beef	Minced meat	1g	16.0%	14.8%	13.7%	10.7%	13.6%	6.7%	13.3%
Chicken	Meat preparation	1g			33.8%	60.0%			40.6%
		0.01g					7.9%	7.5%	
Fish	Smoked salmon	25g			23.1%	22.1%	8.0%	15.7%	21.3%

Listeria monocytogenes in humans

In 2006, the Sentinel Laboratory Network and the National Reference Laboratory reported 67 cases of listeriosis. This number is less than in 2003 and 2004, when particularly high numbers of listeriosis cases were recorded. For the period 1994-2006, the annual number of cases reported to the Network is depicted in Figure xx, corresponding to an annual mean number of 55 cases. Geographic distribution of the cases in 2006 is as follows: six cases were reported in Brussels, 46 in Flanders and 13 in Wallonia (2 from unknown geographic origin). People older than 65 year represent more than 50% of the cases.

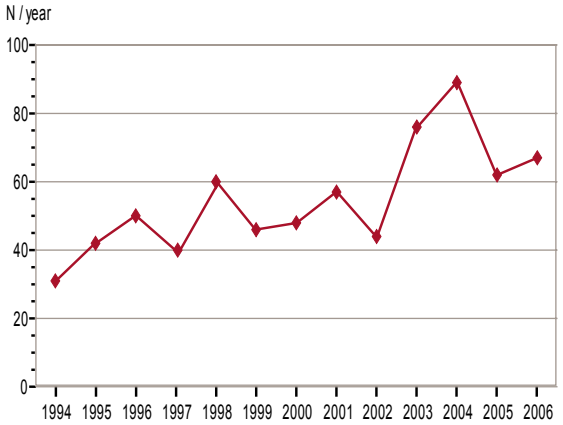

Figure 19. *Total number of Listeria monocytogenes infections in humans by year (1994-2006). Sources: Sentinel Laboratory Network and National Reference Laboratory*

In 2006 the National Reference Laboratory serotyped 56 clinical strains of L. monocytogenes; in addition 2 strains of L. ivanovii of human origin were received. The serovar 1/2a and 4b were the most prevalent (51.8% and 33.9% respectively). Four strains were related to perinatal cases (isolated in the child), 6 strains were isolated from cerebro-spinal fluid (conclusive for a meningo-encephalitis form), 44 strains were isolated from blood and one from urine. Two outbreaks of listeriosis were recognized with 4 and 9 cases respectively: no source of contamination could be determined.

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yersiniosis

Yersinia enterocolitica

Y. enterocolitica is a cause of diarrhea and abdominal pain. Infection with Y. enterocolitica occurs most often in young children. Common symptoms in children are fever, abdominal pain and diarrhea, which is often bloody. Symptoms typically develop 4 to 7 days after exposure and may last 1 to 3 weeks or longer. In older children and adults, right-sided abdominal pain and fever may be the predominant symptoms and may be confused with appendicitis. In a small proportion of cases, complications such as skin rash, joint pains, or spread of bacteria to the bloodstream may occur.

Only strains of Y. enterocolitica belonging to certain biotypes cause illness in humans. Pigs are considered as the major reservoir for pathogenic Y. enterocolitica. In infected pigs, the bacteria is most likely to be found in the tonsils. Infection is most often acquired by eating contaminated food, especially raw or undercooked pork. Drinking contaminated unpasteurised milk or untreated water can also transmit the infection.

- Yersinia enterocolitica in food
- Yersiniosis in humans

Yersinia enterocolitica in food

Monitoring programme

The Federal Agency for the Safety of the Food Chain organised a monitoring of meat since 1997, which showed a very low prevalence of Yersinia enterocolitica in pork, beef and poultry. In 2006, like in 2005, the monitoring programme concentrated on one matrix, i.e. pork minced meat intended to be eaten cooked and one contamination level (1g).

***Table 21**. Monitoring programme for Yersinia enterocolitica in food*

Sample		Quantity analysed	Percentage of positive samples
Pig meat	Minced meat at processing plant (n=103)	1g	0.0%
	Minced meat at retail (n=85)	1g	0.0%

Yersiniosis in humans

In 2006, the Belgian Sentinel Laboratory Network registered 264 cases, corresponding to a national incidence estimated at 2.5 per 100 000 inhabitants. Cases were observed all over the year. Forty percent of cases were 0 to 4 year old children.

As already reported in former years, the incidence in Flanders is higher than in Wallonia. In 2006, the incidence was 3.0 per 100 000 inhabitants in Flanders, 1.8 per 100 000 inhabitants in Wallonia and 1.4 per 100.000 inhabitants in Brussels-Capital Region.

Since 1986, when 1.514 cases were reported by this network, the number of human infections in Belgium significantly decreased (Figure XX).

Bio-serotyping was performed by the National Reference Laboratories. In 2006, 70% of the 430 isolates tested belonged to pathogenic bio-serotypes (including 9 Y. pseudotuberculosis) with serotype O: 3 / biotype 4, accounting for 63 % of the total. The remaining 128 strains (30 %) belonged to non-pathogenic bio-serotypes and their number did not vary markedly during the last years, in contrast to the obvious decrease of the pathogenic strains.

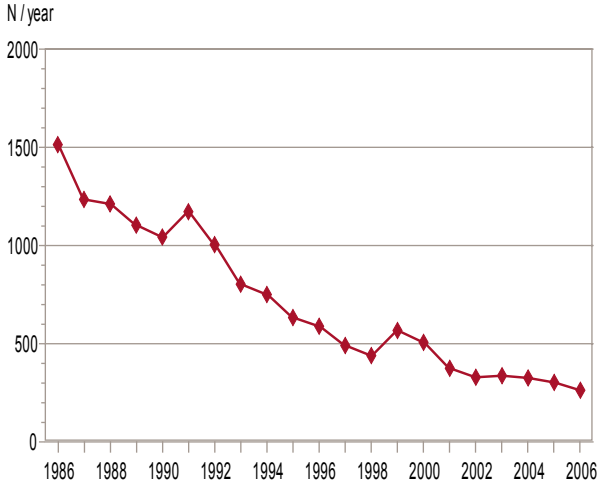

Figure 20: *Total number of Yersinia enterocolitica infections in humans by year (1986-2006). Source: Sentinel Laboratory Network*

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verotoxin producing escherichia coli (vtec)

Verotoxin producing Escherichia coli

Infection with zoonotic verotoxin producing E. coli is a life-threatening disease in young children, in immunocompromised or in elderly people. Especially in the United Kingdom, but also in other European countries, the disease is well known and is caused by virulent E. coli O157. Other serotypes, e.g. O26, O91, O103 and O145 may be involved also.

Cattle is the principal reservoir of VTEC, but are not clinically affected by zoonotic E. coli infection. The organism is excreted in the faeces, which represents a potential risk to people working closely with farm animals and their environment.

Human infections occur after consumption of contaminated food, after contact with contaminated water, or by direct transmission of VTEC from infected humans or animals. The clinical symptoms range from mild diarrhoea through haemorraghic colitis and renal insufficiency or haemolytic uremic syndrome (HUS). In some cases death may follow. Prevention mainly relies on bio-security measures at farm-level and hygienic measures at the level of the slaughterhouses.

- Verotoxin producing Escherichia coli in cattle
- Escherichia coli O157 in food
- Verotoxinogenic Escherichia coli in humans

In Belgium, approximately 40 mostly sporadic cases are registered per year. In 2006, two relatives were infected with E. coli O157 during their stay on a cattle farm. This was the first case in Belgium where a VTEC infection could unequivocally be traced back to excreting animals.

Since August 2005, the sampling of cattle at farms that had sent E. coli O157 positive animals to the abattoir is not compulsory any more. Only a few food samples (carcasses, cheese from raw milk) have been found positive for E. coli O157 in 2006.

Verotoxin producing Escherichia coli in cattle

Surveillance programme, measures and methods used

The surveillance starts when a typical E. coli O157 (stx1, stx2, eaeA, enterohemolytic) is isolated from a carcass at the slaughterhouse. In such case, the farm of origin was traced back via Sanitel, the computerised registration and identification database for farm animals, managed by the Federal Agency for the Safety of the Food Chain. FASFC officials inform the owner that typical E. coli O157 circulate on his farm and encourage the implementation of hygienic measures, i.e. cleaning and disinfection of milk reservoirs and milking equipment, and cleaning of animals before transport to the slaughterhouse.

Carcasses contaminated with typical E. coli O157 should be destroyed or may be heat treated. In all other cases, no specific measures are taken.

The method used for isolation of E. coli O157 is described in ISO 16654:2001. Briefly, the samples were enriched in mTSB with novobiocin and treated by immunomagnetic separation. Subsequently, the suspected colonies on CT-SMAC were latex agglutinated for the detection of E. coli O157. Confirmation of serotype (O group) was done by means of slow tube agglutination after heating of the bacterial cultures. Virulence factors were determined by PCR for toxin genes stx1 and stx2 and for eae (intimin). Enterohemolysis was done on appropriate culture media.

Epidemiological investigations and results of 2006 surveillance

Since August 2005, herds are not longer monitored after E. coli O157 is isolated from the surface of a carcass.

Escherichia coli O157 in food

Monitoring programme and method used.

In 2006, the Federal Agency for the Safety of the Food Chain selected for its monitoring programme more than 200 Belgian slaughterhouses, more than 100 meat cutting plants and more than 100 retail trades representative of the Belgian production of carcasses and meat.

Carcasses (1600cm2), trimmings (25g) and minced meat and meat preparations of beef (25g) were sampled for E. coli O157.

The Belgian official detection method (SP-VG-M001), according to ISO16654 was used for analysis. After a pre-enrichment in mTSB with novobiocin at 42°C for 7 hours, enrichment was done in CT-Mac Conkey at 37°C for 16-18 hours and subsequent testing in the immunoassay O157 (VIDAS ECO, bioMérieux). Subsequent selective immunomagnetic enrichment was performed (Dynabeads, Dynal or VIDAS ICE, bioMérieux) followed by isolation on sorbitol-Mac Conkey,

incubated at 42°C for 18 h. Serological confirmation was done by means of agglutination of latex particles (Oxoid). Suspected isolates were sent to the National Reference Laboratory for detection of genes encoding virulence factors.

A sample was considered to be positive when E. coli O157 was isolated and when specific virulence genes sequences were confirmed by PCR.

Notification is mandatory since March 2004 (Ministerial Decree on mandatory notification in the food chain). For enterohemorrhagic E. coli, absence in 25g in ready-to-eat food products put on the market is compulsory.

Results of the 2006 monitoring

The results by the monitoring of the Federal Agency for the Safety of the Food Chain are shown in the following table.

Table 22. *Zoonosis monitoring programme - E. coli O157 (2006)*

	Sample	Prevalence
Beef	Carcasses (n=1 214)	0.9%
	Fresh meat at cutting plant (n=243)	0.0%
	Minced meat at processing plant (n=55)	0,0%
	Minced meat at retail (n=31)	0.0%
	Meat preparations (steak tartare) at retail, intended to be eaten raw (n=97)	0.0%
Milk	Raw or low heat-treated cows' milk at farm (n=123)	0.0%
Cheese	From raw or low heat-treated cows' milk, at farm (n=234)	2.1%
	From raw or low heat-treated cows' milk, at processing (n=27)	0.0%
	From raw or low heat-treated cows' milk, at retail (n=126)	0.0%
	From raw or low heat-treated sheep' milk, at farm (n=7)	0.0%
	From raw or low heat-treated sheep' milk, at retail (n=10)	0.0%
	From raw or low heat-treated goats' milk, at farm (n=12)	0.0%
	From raw or low heat-treated goats' milk, at retail (n=10)	0.0%
Butter	From raw or low heat-treated milk, at farm (n=79)	0.0%
	From raw or low heat-treated milk, at retail (n=13)	0.0%
	From raw or low heat-treated milk, at processing plant (n=70)	0.0%
Icecream	At farm (n=66)	0.0%
	At processing plant (n=69)	0.0%

Verotoxin producing Escherichia coli in humans

Only few clinical laboratories examine human stools for the presence of E. coli O157. Therefore, a correct incidence of VTEC in human populations cannot be given.

In 2006, the National Reference Laboratory confirmed 46 verotoxigenic E coli. Among these:

- 36 typical VTEC isolates, positive for two factors of additional virulence: (i) the presence of the gene eae (intimin) gene and enterohemolysin (EHEC virulence plasmid) gene
- 10 atypical VTEC isolates, negative for intimin and enterohemolysin.

The number of isolates analysed annually by the NRL has been rather constant, corresponding to a large rate of underdiagnosis.

***Table 23**. E. coli : evolution in number of isolates in humans since 1998*

	1998	1999	2000	2001	2002	2003	2004	2005	2006
Number of O serogroups	48	53	47	46	46	47	45	47	46
Number of typical isolates	38	46	33	36	37	40	36	36	36

In 2006, 9 strains (3 from serotype O157:H7, 3 O145, one O26, one O175 and one O1:K1:H7) were associated with haemolytic uremic syndrome (HUS). Eight patients were less then 5 years old and one was a 13 year old girl.

According the information available at the NRL, all but two of these cases were not related. The infection source in the 13 year old girl and her 11 year old sister, who presented only bloody diarrhea without complications, could be traced. Laboratory analysis (enterohemolysis, presence of stx2 and eae, PFGE and PHIA typing of the stx2 gene cluster) showed that the isolates from the two girls were indistinguishable from strains isolated from cattle and from the environment (dust) of the farm the girls visited shortly before becoming sick. This was the first time a VTEC infection in humans was traced back to a contact with animals.

Beside the cases confirmed by culture, VTEC was also serologically confirmed in nine children aged 1 to 11 years old and presenting HUS (8 serogroup O157 and 1 serogroup O121).

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zoonotic tuberculosis

Zoonotic tuberculosis (Mycobacterium bovis)

Tuberculosis in humans caused by M. bovis is rare.

- In regions where M. bovis infections in cattle are largely eliminated, only few residual cases occur among elderly persons as a result of the reactivation of dormant M. bovis within old lesions and among migrants from high-prevalence countries. Agricultural workers may acquire infection of M. bovis by inhaling aerosols from coughing infected cattle and may subsequently develop typical pulmonary or genito-urinary tuberculosis. Such patients may infect cattle through cough or urine. Evidence for human-to-human transmission is only rarely reported.
- In developing countries, where M. bovis is largely prevalent among cattle, some studies reported that 3-6% of all tuberculosis cases are due to M. bovis and that mostly young people get infected through the ingestion of contaminated raw milk. Also occupational contacts should be regarded as a risk factor for transmission to humans, although companion animals can provide a less common indirect route of infection.

- Mycobacterium bovis in cattle
- Mycobacterium in other animals
- Mycobacterium bovis in humans

In human, the disease caused by M. bovis is clinically indistinguishable from that caused by M. tuberculosis. Pulmonary tuberculosis is frequently observed and cervical lymphadenopathy, intestinal lesions, chronic skin tuberculosis and other non pulmonary forms are particularly common.

In 2006, the National Reference Laboratory identified only 1 human case of bovine tuberculosis. However, the molecular identification of Mycobacterium performed in 25 laboratories of the country only identified the complex M. tuberculosis, without distinction between bovis and tuberculosis. The number of M. bovis reported by laboratories is thus underestimated.

Human tuberculosis (Mycobacterium tuberculosis)

The incidence of human tuberculosis shows little variation over the last years. In 2001, 2002, 2003, 2004, 2005 and 2006 respectively 1321, 1309, 1128, 1226, 1144 and 1127 new notified cases of active human tuberculosis were detected. Over the 60% were male patients. In 2006, 51 % of the tuberculosis cases were foreigners.

Groups at risk are persons with a marginal existence, asylum seekers and refugees. Alcoholism and a co-infection with HIV are known as specific risk factors. Human tuberculosis cases are mainly concentrated in urban populations.

Belgium is officially free from bovine tuberculosis (Mycobacterium bovis) since 25 June 2003 (Commission Decision 2003/467/EC establishing the official tuberculosis, brucellosis and enzootic bovine leucosis free status of certain Member States and regions of Member States as regards bovine herds).

Mycobacterium bovis in cattle

Surveillance programme

The control of tuberculosis is based on Council Directive 64/432/EEC, which is implemented and adapted in the national legislation since 1963 and was last adapted by Royal Decree of 17 October 2002.

The control implies:

- Skin testing of animals at purchase (mandatory),
- In case of a positive reactor, skin testing of all the animals of the holding and skin testing of all contact animals (tracing on and tracing back),
- Systematic post mortem examinations at the slaughterhouse; in case a suspected lesion is identified, a sample is sent to the National Reference Laboratory for analysis.

The Federal Agency for the Safety of the Food Chain is informed about any doubtful or positive result of the skin test and may decide to re-examine (additional tests) the animals or to kill them (test slaughter, additional tests). If M. bovis is isolated as a consequence of post mortem examinations or of mandatory test-slaughter, all animals in the herd of origin are skin tested and a complete epidemiological investigation is performed.

An animal is defined as infected with bovine tuberculosis if the skin testing is positive or if M. bovis is isolated by culture or confirmed by laboratory testing (PCR). A holding is defined as infected if M. bovis was isolated or detected by PCR from an animal of the holding.

Isolation of M. bovis and biochemical testing is exclusively performed in the National Reference Laboratory where also IFN-gamma and molecular typing by means of IS6110 RFLP, spoligotyping and MIRU-VNTR are done.

In Belgium, vaccination against tuberculosis is prohibited.

Epidemiological investigations and results of 2006 surveillance

At the slaughterhouse, 883 tissue samples from individual animals were taken. The samples originated from animals suspected of being infected with M. bovis, i.e. skin test reactors, animals that had been in contact with M. bovis infected animals or animals that showed suspicious lesions at meat inspection. The samples were submitted to the National Reference Laboratory where culture, PCR and confirmatory tests were done. M. bovis was detected in 88 animals all belonging to the 8 outbreak herds.

Table 24. *Evolution of bovine tuberculosis outbreaks in cattle herds in Belgium*

1999	2000	2001	2002	2003	2004	2005	2006
13	24	23	13	7	8	5	8

The National Reference Laboratory performs routine IS6110 RFLP typing and spoligotyping of M. bovis field isolates. Since 1995, the dates of 96% of the outbreak herds are typed by both methods. More recently, all strains typed by RFLP and spoligotyping were additionally analysed by MIRU-VNTR, which is done in collaboration with Pasteur Institute Brussels. As a consequence, a comprehensive database of the vast majority of M. bovis types isolated in Belgium since 1995 is available.

For 2006, M. bovis isolates originating from 8 outbreak herds were typed by the three molecular typing methods available at CODA-CERVA (Spoligotyping, VNTR and RFLP IS6110). In one herd a total new type of strain was observed (never observed in the Belgian's typed collection since 1995), characterized by the spoligotype SB1398 (M. bovis org). Two groups of two herds shared similar M. bovis strain types. In the last 3 herds, individual types were observed. All types observed in these 7 herds have already been observed in Belgium: spoligotypes SB0120, SB0162, SB0134 and SB0824. Interestingly, spoligotype SB0134 (3 herds) was clearly subdivided into two group of unrelated strains by the VNTR typing.

Mycobacterium in other animals

In the South of Belgium, the control of wild animal diseases is carried out by « the Network of Wildlife Disease Surveillance" of the Faculty of Veterinary Medicine (University of Liège).

In 2006, targeted organs of 432 wild cervids (Cervus elaphus and Capreolus capreolus) and 215 wild boars (Sus scrofa) were checked for suggestive lesions of tuberculosis. In the same way, 7 badgers (Meles meles) found dead were analysed by the network. In all cases of suspected lesions, samples were sent to the National Reference Laboratory for analysis (CODA-CERVA, Uccle). In 2006, no suspected case was detected positive for Mycobacterium bovis in wildlife.

Mycobacterium bovis in humans

In 2006, 1 human case of bovine tuberculosis was identified by the National Reference Laboratory.

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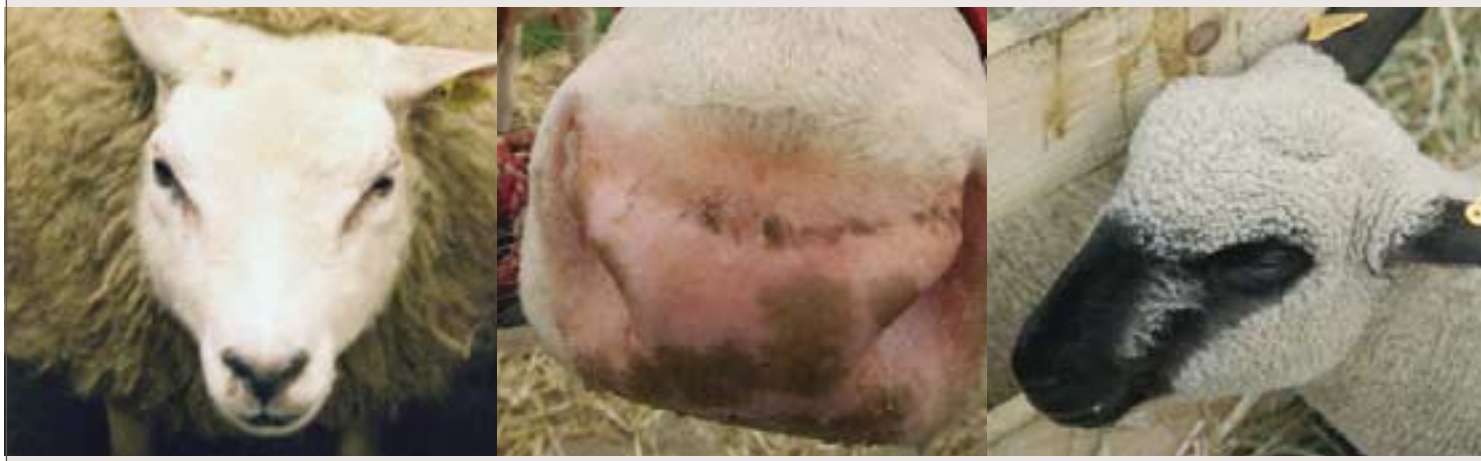

zoonotic brucellosis

Zoonotic brucellosis

(Brucella melitensis, Brucella abortus, Brucella suis)Bacteria of the genus Brucella may infect sheep, goats, cattle, deer, elk, pigs, dogs and several other animals, where they cause disease. Humans become infected by contact with infected animals or with contaminated animal products. Brucella infections in humans may cause a range of symptoms that are similar to that of flu and may include fever, sweats, headaches, back pains and physical weakness. Several infections of the central nervous systems or lining of the heart may occur.

- In the non-"officially brucellosis free" Mediterranean countries, the consumption of raw milk or raw cheese from sheep and goats is thought to be the major source of contamination (B. melitensis).
- In Northern European countries, besides some occupational human cases of B. abortus infections, the majority of brucellosis cases are imported and are mainly caused by B. melitensis.
- In Belgium, less than 10 cases/year of imported B. melitensis infections have been reported over the past few years. In 2006, 2 cases were reported..

- Brucellosis in cattle
- Brucellosis in sheep and goats
- Brucellosis in pigs
- Brucellosis in wildlife
- Brucellosis in humans

Brucellosis in cattle

Belgium is officially free from bovine brucellosis since the 25th of June 2003 (Commission Decision 2003/467/EC establishing the official tuberculosis, brucellosis and enzootic-bovine-leucosis-free status of certain Member states and regions of Member states as regards bovine herds).

Surveillance programme and methods used

Since the official brucellosis free status, the eradication programme has been changed in a surveillance programme. Beef cattle older than 2 years are serologically monitored once every three years. The herds are selected on the basis of geographical localisation. Dairy cattle are checked at least 4 times a year via tank milk. Furthermore, all female animals older than 1 year and breeding bulls are serologically tested at purchase. Each abortion or premature birth in animals at risk is subject to compulsory notification to the Federal Agency for the Safety of the Food Chain and testing for brucellosis is obligatory. Aborting females should be kept in isolation until the results of the investigation exclude Brucella infections.

Tank milk is examined by means of the milk ring test. For animals older than 2 years, serology (i.e. micro-agglutination as screening test; in case of a positive result, an indirect ELISA test is performed as confirmatory test) is used if no sufficient milk ring tests are done (at least 4 ring tests a year). Bacteriological examination is done in case of serological and/or epidemiological suspicion.

Allergic (brucellin) tests may be carried out if serological cross-reactions are suspected. These tests are performed by the Federal Agency for the Safety of the Food Chain in collaboration with the National Reference Laboratory.

An animal is legally suspected of brucellosis in case of a positive ELISA. If, according to the epidemiology and the results of the skin test, an animal or herd is found to be at risk, a bacteriological investigation always takes place. Hence, a brucellosis animal is defined as an animal in which Brucella has been isolated and a cattle herd is considered as infected if one of its animals is positive for brucellosis by culture.

	Individual serological tests	Bulk milk tests
2004	488 548	102 267 pools
2005	579 390	80 025 pools
2006	500 766	73 482 pools

For individual serological testing, the SAT micro-agglutination test has been used for routine testing whereas the indirect ELISA is accepted for confirmation.

Vaccination has been prohibited in Belgium since 1992.

Epidemiological investigations
and results of 2006 surveillance

The intensified bovine brucellosis eradication programme started in Belgium in 1988. In case of active brucellosis, i.e. excretion of Brucella, the plan consisted in the culling of all animals of the infected herd (total depopulation), the slaughtered animals were compensated for based on the replacement value.

The annual herd prevalence notified at the end of the year was 1.13% in 1988 and has fallen below 0.01% since 1998. In March 2000, the last case of bovine brucellosis was identified. No infected herd was detected in Belgium since then.

In 2006, the Federal Agency for the Safety of the Food Chain didn't have to instruct, the test slaughter of animals, positive by repeated serological testing, for additional analysis.

Brucellosis in sheep and goats

Belgium is official free for sheep and goat brucellosis (B. melitensis) since 29 March 2001 (Commission Decision 2001/292/EC amending Decision 93/52/EEC recording the compliance by certain Member States or regions with the requirements relating to brucellosis (Brucella melitensis) and according them the status of a Member State or region officially free of the disease).

Surveillance programme

Serum samples taken in the framework of national monitoring for Visna-Maedi and at export were examined for Brucella melitensis specific antibodies by means of ELISA (5% of the total population). Positive samples were subsequently tested with Rose Bengal test and Complement Fixation test. A sample is classified as positive for brucellosis only if it is positive in all three tests. If this is the case, a skin test should be performed on the seropositive animals and the congeners. A positive skin test leads to the bacteriological investigation of the animal.

Since 2001, yearly serum samples from about 5% of the sheep and goats populations were tested at the National Reference Laboratory. In addition, serum samples from sheep for export were analysed. In 2006, 7.986 samples were tested. Serological positive reacting animals after serial and repeated testing were finally negative. The National Reference laboratory has confirmed infections of Yersinia enterocolitica 0:9 in sheep. Those infections are associated with false positive serology in the tests ELISA, Rose Bengal and possibly CFT of brucellosis. The phenomenon of FPSR (false positive serological reactors) as documented for bovines is also observed in sheep. In absence of clinical, bacteriological and epidemiological evidence, an infection with Y. enterocolitica 0:9 can be retained to explain FPSR met in small ruminants in our country.

Brucellosis in pigs

Surveillance programme in pigs
and epidemiological investigations

Serological screening for Brucella is done in breeding pigs that are brought together (e.g. at a fair), at artificial insemination centres or in animals intended for trade. The methods used are Rose Bengal test (RBT), Slow Agglutination test (SAT) according to Wright, complement fixation test (CFT) and ELISA. Bacteriological examination for Brucella and Yersinia is done in case of positive serology.

Regularly, false positive serological reactions are reported. These are due to a Yersinia enterocolitica O9 infection and are confirmed by Yersinia spp. isolation in the absence of Brucella spp. isolation.

The domestic pig population is free of brucellosis (last Brucella isolation in pigs in Belgium was in 1969). In 2006, all 239 samples were negative.

Brucellosis in wildlife

Regional control programme

Since 2002, an annual surveillance programme is organised by the Network of Wildlife Disease Surveillance (Faculty of Veterinary Medicine, Liege) in collaboration with the National Reference Laboratory (CODA - CERVA, Uccle) with the aim to analyse brucellosis in wild boars (Sus scrofa) and lagomorphs in the South of Belgium. Blood samples and organs of hunted or found dead animals are analysed in order to follow seroprevalence and identify isolates of Brucella in these species. In 2006, 271 hunted wild boars were sampled and the apparent seroprevalence was 46 % (IC95 = 39.1 – 52.9). Brucella suis biovar 2 was isolated from spleen and tonsil of wild boars sampled in 2003. In hares, no Brucella was isolated from 154 spleen analysed between 2003 and 2006.

Recommendation.

Further attention should be given to brucellosis in wild species, as the potential for contact with B. suis can be high, particularly for people handling and/or slaughtering game animals. The species to be considered should include at least wild boar, deer and other wild ruminants as well as hares.

Brucellosis in humans

The last indigenous case of Brucella was reported in 1997. It is helpful to note that B. suis biovar 2, the only biovar circulating in Belgium among wild boars, shows only limited pathogenicity for humans, if pathogenic at all.

In 2006, the National Reference Laboratory confirmed two cases of Brucella melitensis 3. The country of origin of these two imported cases was not known.

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Q-fever

Coxiella burnetii

Q-fever (Q for query) is a systemic disease caused by an obligate intracellular bacterium Coxiella burnetii that is highly resistant to chemical and physical agents. Coxiella burnetii occurs worldwide with the exception of New Zealand.

Natural reservoirs are more than 40 species of ticks and free-living vertebrates, primarily rodents. Ticks or their excreta spread the disease to domestic animals, e.g. sheep, goats, cattle and dogs. These animals may display a cycle that does not involve ticks since coxiellae can multiply in the trophoblast of the placenta. The placentas and amniotic fluids of these animals contain large numbers of bacteria which contaminate pastures and soil. Once animal secreta or excreta have dried, infectious dust is created.

In animals, the infection is most often latent. In cattle and sheep, abortion may occur.

- Coxiella in animals
- Coxiella in humans

Coxiella in animals

Q-fever is a zoonotic disease caused by Coxiella burnetii, a bacteria that resists to heat, drying and many common disinfectants. This resistance enables the bacteria to survive for a long period in the environment. Cattle, sheep, and goats are the main reservoirs but a wide variety of other animals can be contaminated, including domesticated pets. Coxiella burnetii does not usually cause clinical disease in these animals, although an increased abortion rate and fertility problems in cattle, sheep and goats are observed. The emergence of these common symptoms over a longer period of time leads finally to the diagnosis of Q-fever.

Organisms are excreted in milk, urine, and faeces by infected animals. Animals shed the organisms especially during parturition within the amniotic fluids and the placenta. Airborne transmission can occur in premises contaminated by placental material, birth fluids or excreta from infected animals. Airborne inhalation is the most important transmission route of infection.

In 2006, 166 bovine animals, 4 sheep and 2 goats were analysed. Four latent infected bovines were detected at import.

Recommendations for prevention and control of Q-fever.

- Public education and information on sources of infection
- Advice to persons 'at risk', especially persons with pre-existing cardiac valvular disease or individuals with vascular grafts and pregnant women
- Restrict access to barns and laboratories used in housing potentially infected animals
- Quarantine aborted animals
- Analyse of placenta and aborted foetuses in case of any abortion
- Appropriate disposal of placenta, birth products, foetal membranes and aborted foetuses
- Use only pasteurised milk and milk products
- Infected holding facilities should be located away from populated areas. Measures should be implemented to prevent airflow to other occupied areas.

Coxiella in humans

Transmission in people is either airborne or results from direct or indirect contact with infected animals or their dried excreta. Consumption of infected food such as unpasteurised milk or dairy products leads to infection and seroconversion but rarely to clinical symptoms.

Infection with Coxiella burnetii is either inapparent, acute, or chronic. The incubation period of acute Q-fever ranges from 2 to 4 weeks. The infection has an abrupt onset and patients present usually with high fever, hepatitis or pneumonia. The spontaneous evolution is usually a complete recovery but in immunocompromised hosts a chronic infection can develop with endocarditis as the major clinical form.

Consumption of pasteurized milk or raw milk only from Q-fever free herds as well as proper hygiene when in contact with infected animals are the best preventive measures.

In the Institute of Tropical Medicine (National Reference Laboratory), a total of 1448 human sera have been examined for the presence of phase I and II IgM and IgG antibodies to Coxiella burnetii by IFAT (Focus Technologies). The samples originated from 666 men and 779 women. The median age of the patients was 41 years (range 1 month – 90 years).

No confirmed or probable cases have been detected during the year 2006. Eight possible cases (5 men and 3 women) have been found merely on the basis of one serological result (due to the lack of follow-up samples) and without clinical information. The age of the patients ranged from 22 to 68 years with a median age of 40 years. At least two of the patients stayed abroad before the start of their illness.

trends and sources

report on zoonotic agents in belgium in 2006



foodborne outbreaks

Foodborne outbreaks in humans

A 'foodborne outbreak' means an incidence, observed under given circumstances, of two or more human cases of the same disease and/or infection, or a situation in which the observed number of human cases exceeds the expected number and where the cases are linked, or are probably linked, to the same food source (Directive 2003/99/EC, Article 2(d)). This includes outbreaks caused by any virus, bacteria, algae, fungus, parasite, other biological entity or their toxins which is likely to cause foodborne illness. Outbreaks caused by ingestion of drinking water are also considered foodborne (Regulation 178/2002/EC, Art. 2).

In Belgium different authorities are dealing with foodborne outbreaks:

The Federal Agency for the Safety of the Food Chain (FASFC) deals with safety of foodstuffs, epidemiological investigation on foodstuffs and animal health issues in case of a foodborne outbreak.

- Major etiological agents
- Foodborne outbreaks 2006
- Working Group on Foodborne Infections

The Communities (Flemisch, French and German speaking Community) that deal with person related matters as human health, can start an epidemiological investigation by its Public health medical inspectors in case of a foodborne outbreak. They can also take human stool samples.

The Scientific Institute of Public Health - IPH in Brussels (National reference laboratory on Foodborne Outbreaks) analyses all suspected food samples, collects all data on foodborne outbreaks and gives scientific support to the FASFC officers and the Public Health Inspectors.

A national "Platform Foodborne outbreaks", approved by the National Conference of Ministers of Public Health, was created in 2004 to advance data exchange between different competent authorities on food safety, animal health and public health.This report contains data collected from FASFC, the Flemish Community, the French community, the Brussels Common Community Committee, the Sentinel Laboratory Network for human clinical microbiology, and the Federal Reference Centres for Foodborne outbreaks, Salmonella and Shigella, Listeria and C. botulinum.

In case of an outbreak the source of contamination, the cause and the etiological agent need to be determined to take adequate measures to prevent more human cases.

The etiological agent can be a bacterium, a toxin, a parasite or a virus. The symptoms and the time of onset after the meal can give an indication of the responsible etiological agent.

Major etiological agents

Foodborne bacteria

Salmonella enterica

Although the number of human salmonellosis drastically decreased since 2005 in Belgium, it remains the most frequently reported pathogen in foodborne outbreaks. The onset time varies between 6 and 48 hours after ingestion of the contaminated food. Nausea, vomiting, abdominal cramps, diarrhoea, fever and headache are the symptoms in an acute outbreak and last for 1-2 days or longer. In case of an outbreak human samples (stool) and suspected food samples are tested for Salmonella. If Salmonella is detected, PFGE typing can confirm the clonal relationship between the human isolates and those isolated from food products. Raw or undercooked meat, poultry meat, eggs, shrimps, cream-filled desserts and chocolate are frequently associated with foodborne Salmonella outbreaks. The food can be the origin of contamination or transmit the infection from a contaminated food handler.

Shigella

Shigella is principally a disease of humans. The organism is frequently found in water polluted with human faeces. The symptoms are abdominal pain, cramps, diarrhoea, fever, vomiting, blood in stools. Some strains produce enterotoxin and Shiga toxin (very much like the verotoxin of E. coli O157:H7). Water, salads and raw vegetables are frequently associated with outbreaks. Water contaminated by faeces and unsanitary

handling by food handlers are the most common causes of contamination.

Campylobacter jejuni and coli

Since 2005 Campylobacter is the most frequently reported foodborne pathogen in humans in Belgium. Campylobacter jejuni and coli infections cause diarrhoea, which may be watery or sticky and can contain blood. Other symptoms often observed are fever, abdominal pain, constipation, nausea, headache and muscle pain. The illness usually occurs 2-5 days after ingestion of the contaminated food or water and generally lasts 7-10 days, but relapses are not uncommon (about 25% of cases). Campylobacter frequently contaminates raw poultry meat and raw pork. Raw milk and cheeses made from raw milk are also sources of infections.

E. coli O157

E. coli serotype O157:H7 is a variety of E. coli that produces large quantities of one or more potent toxins (verotoxin, shiga-like toxin) that cause severe damage to the mucosal lining of the intestine. The illness is characterized by severe abdominal pain and diarrhoea which is initially watery but becomes bloody. The illness is usually self-limited and lasts for an average of 8 days. Some victims, particularly young children, develop the haemolytic uraemic syndrome (HUS), characterized by renal failure and haemolytic anaemia. The disease can lead to permanent loss of kidney function.

Undercooked or raw hamburger (ground beef), unpasteurized fruit juices, raw vegetables, and raw milk are known food vehicles in outbreaks.

Yersinia enterocolitica

Yersiniosis is frequently characterised by symptoms as gastroenteritis with diarrhoea and/or vomiting; however, fever and abdominal pain are typical symptoms. Yersinia infections can also cause pseudo-appendicitis and arthritis. Illness onset is usually between 24 and 48 hours after ingestion of food or water, which are the usual vehicle of infection. Contaminated and undercooked pork is a common source of infection, but also ice-cream has been reported as the source of infection.

Clostridium perfringens

The common form of Clostridium perfringens poisoning is characterized by intense abdominal cramps and diarrhea which begin 8-22 hours after consumption of foods containing large numbers vegetative cells of strains capable of producing the food poisoning toxin. Toxin production in the human digestive tract is associated with sporulation. The illness is usually over within 24 hours but less severe symptoms may persist in some individuals for 1 or 2 weeks. In most instances, the actual cause of poisoning by C. perfringens is temperature abuse of prepared foods. Small numbers of the organisms are often present after cooking and multiply to food poisoning levels during cooling and storage of prepared foods under anaerobic conditions (e.g. fat layer on stock). Meat, meat products, and gravy are the foods most frequently implicated.

Staphylococcus aureus

Some Staphylococcus strains are capable of producing a highly heat-stable enterotoxin that causes illness in humans.

The toxin is preformed in the food. The onset of symptoms in staphylococcal food poisoning is usually rapid and in many cases acute, depending on individual susceptibility to the toxin, the amount of contaminated food eaten, the amount of toxin in the ingested food, and the general health of the victim. The most common symptoms are nausea, vomiting and abdominal cramping. Recovery generally takes two days. Food at risk for staphylococcal food poisoning are those that require considerable handling during preparation and that are kept at slightly elevated temperatures after preparation. Contamination occurs by infected food handler or by the food itself (e.g. milk)

Bacillus cereus

Although Bacillus cereus is a well-known cause of foodborne illness it is not commonly reported because of its usually mild symptoms. It can cause two types of food poisoning known as the emetic and the diarrhoeal types. For the emetic type, a heat-stable emetic toxin named cereulide, preformed in the food, is responsible for the symptoms similar to those of Staphylococcus aureus intoxication, and is characterised by a short incubation period. This type is probably the most dangerous since it has been associated with life-threatening acute conditions like acute liver failure. Heat-unstable enterotoxins, produced in the gut by vegetative cells cause the diarrhoeal type, with symptoms parallel to those of the Clostridium perfringens food poisoning, with a 6 to 24h incubation period. The emetic type is frequently associated with the consumption of food rich in carbohydrates such as rice and pasta whereas the diarrhoeal type is often associated food rich with cooked meat and meat products.

Foodborne viruses

Foodborne and water-borne viral infections are increasingly recognized as causes of illness in humans. This increase is partly explained by changes in food processing, consumption patterns, and globalisation of the food trade. Bivalve molluscs, especially oysters because they are consumed raw, are notorious as a source of foodborne viral infections (filter-feeding shellfish can concentrate viruses up to 100-fold from large volumes faecally contaminated water). Several other foods, however, have also been implicated as vehicles of transmission (fruits, berries, vegetables, salads, sandwiches). Raw and minimally processed fruits and vegetables are high risk food products.

Viruses cannot grow in or on food but may be present on fresh products by contact with polluted water in the growing area or during processing. Unhygienic handling during distribution or final preparation is also reported as a cause of contamination. People can be infected without showing symptoms. Person to person transmission is common and the high frequency of secondary cases following a foodborne outbreak results in amplification of the problem. It is often difficult to identify whether the food is contaminated at the source, as is common with oysters, or whether the food is contaminated by a sick food handler, or whether person to person transmission occurred.

Although there are numerous faecal-orally transmitted viruses, the risk of foodborne transmission is highest for hepatitis A virus and norovirus. European data show that oysters are frequently reported as a main source of contamination, but water, fruits and food handler contamination are also reported. Increased awareness towards viral infections and improved

detection methods due to advances in molecular techniques, especially real-time RT-PCR which allow quantification, has made diagnosis and outbreak management easier.

Focus on Noroviruses

Noroviruses are among the most important causes of gastroenteritis in adults and often occur as outbreaks which may be foodborne. They are the most common cause of non-bacterial foodborne outbreaks recognised in Europe and United States and have been diagnosed worldwide. Noroviruses can be transmitted from person to person, or indirectly via food or water contaminated with faeces or vomit. They are responsible of mild, self-limited gastroenteritis but attack rates are high.

Marine biotoxins

Marine biotoxin poisoning in humans is caused by ingestion of shellfish containing algae toxins. Bivalve molluscs like mussels, oysters and scallops eat phytoplankton. Some kinds of phytoplankton produce, under certain climatic and hydrographic circumstances, natural toxins which are thus absorbed by the bivalve molluscs. According to the effects that they cause, they are classified in different groups: the 3 main groups are the paralytic shellfish poisoning toxins (PSP), the diarrhoeic shellfish poisoning toxins (DSP) and the amnesic shellfish poisoning toxins (ASP).

The effects of these toxins are generally observed as acute intoxications: paralytic shellfish toxins are causing paralysis in man, in extreme case resulting in death. These PSP toxins are accumulated by shellfish grazing on algae producing these toxins. Symptoms of human PSP intoxication vary from a slight tingling or numbness to complete respiratory paralysis. In fatal cases, respiratory paralysis occurs within 2 to 12 hours of consumption of the PSP contaminated food. The responsible toxins are produced by worldwide present dinoflagellates.

Diarrhoeic shellfish toxins are characterised by the diarrhoea they produce in man, unpleasant but not lethal. Symptoms include diarrhoea, nausea, vomiting and abdominal pain starting 30 minutes to a few hours after ingestion and complete recovery occurs within three days. Here too, worldwide present dinoflagellates are responsible for the production of the toxins. Europe and Japan seem to be the most affected areas.

Amnesic shellfish toxins have till now been detected at the American and Canadian east-coast. The symptoms of the intoxication include abdominal cramps, vomiting, disorientation and memory loss (amnesia). A permanent loss of memory is possible. In extreme cases, for older people, a lethal result has been reported. These toxins are produced by a diatom and the main ASP toxin is domoic acid .

Parasites

Giardia lamblia

Giardia lamblia is a protozoan that may cause diarrhea within 1 week of ingestion of the cyst, which is the environmental survival form and infective stage of the organism. Normally illness lasts for 1 to 2 weeks, but there are cases of chronic infections lasting months to years. Illness is most frequently associated with the consumption of contaminated water, contaminated vegetables that are eaten raw or food contamination by infected or infested food handlers. Cool moist conditions favor the survival of the organism.

Foodborne outbreaks 2006

Prevention of foodborne outbreaks

Since the most frequent causes of foodborne outbreaks are disruption of cold chain, insufficient heating of the food, lack of personal hygiene, bad hygiene in the kitchen, long delay between preparation and consumption and raw materials of poor microbiological quality, outbreaks can be prevented by the application of simple hygienic rules like adequate refrigeration of the food, hand washing before and during preparation, clean surfaces and materials in the kitchen, separation of raw and cooked food and sufficient heating during preparation.

Reported outbreaks in 2006

During 2006, a total of 116 outbreaks of foodborne infections and intoxications were recorded in Belgium. More than 1038 people were ill, at least 110 persons were hospitalised.

The geographic distribution is shown in Figure 21.

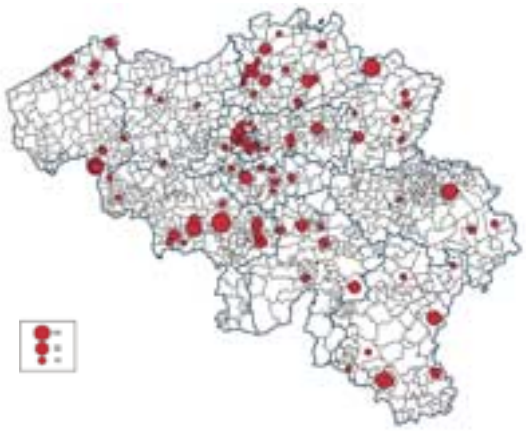

***Figure 21**. Geographical distribution with number of human cases in foodborne outbreaks in Belgium - 2006*

Causative agents

In 12% of the outbreaks Salmonella was the causative agent (n=14) and 134 persons were affected. This figures confirm the decrease in importance of Salmonella as causative agent noticed in 2004 (53%) and 2005 (20%). Salmonella Enteritidis was still the most dominant serotype and was detected in 64.5 % of the Salmonella outbreaks. The only other serovar isolated in foodborne outbreaks was Typhimurium. In one outbreak the serovar was unknown.

The consumption of contaminated eggs or egg products was the most important source of salmonellosis especially in outbreaks where the serotype Enteritidis was involved.

The second most isolated agent was coagulase positive Staphylococcus spp.. Toxine A and C were produced by most of the strains.

Thermotolerant Campylobacters were responsible for 4 % of the outbreaks which remains the same as in 2005.

B. cereus was the causative agent in six outbreaks (5% of the outbreaks) and 175 persons became ill. A general outbreak was observed in a hospital where 60/280 persons became ill after eating spaghetti bolognaise which was contaminated with a B. cereus strain, that produced the emetic toxin cereulide.

Four foodborne Norovirus outbreaks were identified. In two cases Norovirus could be detected in the stool samples from human patients and in three cases Norovirus could be detected in the food. All the Norovirus outbreaks reported were linked with institutional catering and to an infected person that prepared or distributed the food. The small number of cases is very likely an artefact that may be explained by under-reporting of gastroenteritis and the few analysis demands to detect noroviruses.

Other causative agents were Giardia (n=4), Shigella (n=4), E. coli O157 (n=1) histamine (n=2). Listeria monocytogenes (n=3) was responsible for the stillbirth of 3 babies.

In 56% of the outbreaks no causative agent could be identified. An important reason for this is the absence of left-overs of the meal in most of those outbreaks.

Table 25. *Foodborne outbreaks in humans in Belgium in 2006*

Causative agent	Outbreaks	Ill	Died	Hospitalised	Sources
Salmonella	14	134	0	49	Preparations with raw eggs, mixed meals, pastry
Shigella	4	9	0	-	unknown
Campylobacter	5	48	0	8	unknown
E. coli O157	1	2	0	2	raw milk
Yersinia enterocolitica	0	0	0	0	
B. cereus	6	175	0	0	mixed meals, bakery, pasta, milk
S. aureus	7	48	0	0	mixed meals
C. perfringens	0	0	0	0	
C. botulinum	0	0	0	0	
L. monocytogenes	3	7	3	7	unknown
Giardia	4	9	0	-	unknown
Norovirus	4	154	0	0	Mixed meals
Histamine	2	8	0	3	Fish
Marine biotoxins	0	0	0	0	
Unknown	66	466	0	6	
Total	116	1060	3	75	

Source of the foodborne outbreaks

In only 4% of the outbreaks, preparations with raw eggs were identified as the source of the illness. In 2005 and 2004 this was respectively 8% and 36%. Meat and meat based products were responsible for 17% of the cases. Remarkable was the appearance of pasta (5%), pizza (4%), pita donar kebab(7%) and Chinese food (7%) as food vehicle.

Setting of the foodborne outbreaks

Restaurants were the most important location of exposure. It was the case of 32% of foodborne outbreaks in Belgium in 2006, with almost one fourth of it being Chinese restaurants. Take-away restaurants were responsible for 13% of outbreaks. Private households were as important locations as institutional catering with each 10% of foodborne outbreaks. Shops (butchers', bakeries, …) were at the origin of 9%. Other locations of exposure were camping (4%), a recreation place (1%) and a farm (1%) with a small outbreak of E. coli O157. In 20% of the outbreaks the place of exposure was unknown.

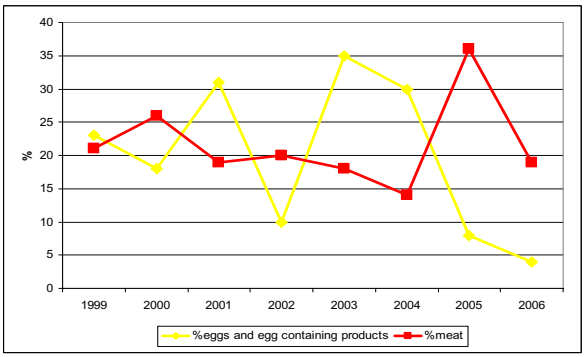

Figure 22 *: Relative importance of eggs and meat in foodborne outbreaks from 1999 until 2006*

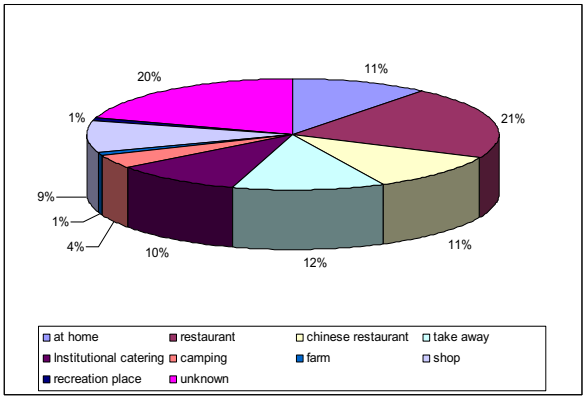

Figure 23*. Settings of foodborne outbreaks 2006*

Working group on foodborne infections

Presentation of the working group

The working group was created in 1995 by the Scientific Institute of Public Health (WIV–ISP) and brings together, on a voluntary basis, the main actors in the field of foodborne infections and intoxications in Belgium.

Since its final reform in 1993, Belgium consists of Communities and Regions, each with their specific responsibilities and competences. Since food and food hygiene is a federal matter and matters related to persons such as illness are the competence of the Flemish, French or German community, data on foodborne outbreaks are dispersed. As a consequence, there was a need for a working group that assures the coordination, the streamlining of policy and the harmonization of the approach between the different partners implicated in outbreaks.

The group is composed of delegates representing

- the Federal Public Service Public Health, Food Chain Safety and Environment,
- the Federal Agency for the Safety of the Food Chain,
- the Scientific Institute of Public Health,
- the Health Inspection Services of the Communities,
- the Brussels Community Coordination Commission,
- the Anti-poison centre,
- the Department of Veterinary Public Health and Food Safety, University of Ghent,
- the National Reference Laboratory for food microbiology at the University of Liège and
- the Veterinary and Agricultural Research centre.

The Scientific Institute of Public Health houses the working group and is represented by the Epidemiology section, the Reference centres for Salmonella and Shigella, for Listeria and for Foodborne Infections and Intoxications.

The main goals of the working group are to exchange field information on detection, epidemiological investigation, controlling and reporting of outbreaks and eventually of sporadic cases of foodborne infections in the country. Significant effort has been put on the improvement of outbreak data collections and case-control studies. The working group also provides scientific support to the mandatory annual Belgian Trends and Sources Report to the European Food Safety Agency (EFSA).

In 2004, the Belgian authorities recognized the working group as 'Platform for foodborne infections and intoxications and food related zoonoses' reporting to the National Conference of Ministers of Public Health.

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trichinellosis

Trichinella

Trichinella is an intestinal parasite whose larvae can be present in the muscles of different animal species. It is transferred to humans by the consumption of contaminated raw or undercooked meat or meat products from an infested animal. Particularly, the following animals represent a risk for humans:

- game, in particular wild boar and carnivorous hosts such as the bear and fox;
- backyard pigs and pigs with extensive outdoor access including pigs from organic farms;
- horses.

Therefore, pork, wild boar and horse meat should always be examined before marketing. Carcasses found positive for the presence of Trichinella are declared unfit for consumption. Commission Regulation (EC) N° 2075/2005 imposes systematic Trichinella examination of all pig carcasses intended for export and all horses, wild boar and other susceptible wildlife animals.

After 1 to 4 weeks incubation, trichinellosis in humans causes myalgia, fever, eosinophilia, facial oedema and possibly fatal myocarditis.

Trichinella has not been detected in carcasses of pigs and horses destined for human consumption in Belgium for many years. Improvements in the monitoring and the reporting of Trichinella in wildlife should be considered.

It is recommended to travellers not to import raw meat of susceptible animals, e.g. sausages or bear meat. Also the consumption abroad of meat of unknown quality should be avoided.

- Trichinella in food animals
- Trichinella in other wildlife

Trichinella in food animals

Surveillance programme and methods used

Pig carcasses intended for intra community trade or export, except when frozen, all locally slaughtered horses and wild boars placed on the market were checked for Trichinella.

The analysis is done by artificial digestion: the magnetic stirrer method of pooled 100 gram sample as described in Commission Regulation (EC) N° 2075/2005, 1 gram per fattening pig, 2 grams per breeding sow or boar and 5 grams per horse or wild boar. Serology may be done in live pigs and for epidemiological studies on wildlife.

Notification to the Federal Agency for the Safety of the Food Chain is compulsory.

Results of the 2006 surveillance

A total of 10 158 164 pigs, 8 205 solipeds (mainly horses) and 9 284 wild boars were examined. All samples were negative.

Trichinella in other wildlife

In 2006, 42 foxes and 15 badgers were analysed for Trichinella, and all tested negative.

An important measure to avoid spreading of trichinellosis among wildlife is not to leave offal of animal carcasses in the field after skinning of hunted animals.

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echinococcosis

Echinococcosis

Echinococcosis is caused either by Echinococcus granulosus or Echinococcus multilocularis.

- Echinococcus granulosus, the agent of cystic echinococcosis, produces unilocular human hydatidosis. It is a small tapeworm (6 mm) that lives in the small intestine of domestic and wild canids. Sheep, goats, pigs, cattle and wild boar serve as intermediate hosts for the infection. Humans also can acquire infection by accidental ingestion of typical taeniid eggs, which are excreted in the faeces of infected dogs and foxes. When eggs are ingested by the intermediate hosts or by humans, the oncospheres liberated from the eggs migrate via the bloodstream to the liver, lungs and other tissues to develop hydatid cysts. Within the cyst brood capsules and protoscoleces develop. Each protoscolex is a potentially infective organism for canids.
- Indigenous unilocular hydatidosis in man has been sporadically reported in Belgium. Recommendations for basic risk-mitigation actions are destruction of contaminated viscera found at the slaughterhouse in order to avoid the infection of dogs.
- Echinococcus multilocularis is the agent of alveolar (multilocular) echinococcosis in humans. Alveolar echinococcosis in particular is of public health relevance as it is considered to be the most severe of all parasitic zoonoses since most untreated cases in humans are fatal. Foxes and dogs are the definitive hosts of this parasite and small rodents and voles the intermediate hosts. In the liver of rodents the invasive larval stage has a multi-compartimented appearance containing many protoscoleces. Ingestion of the eggs by humans can result in the development of invasive cysts in the liver. With regards to domestic animals, cats have been ruled out as hosts of E. multilocularis, since the parasite does not fully develop in their intestine.
- In Belgium, the percentage of infested foxes varies according to the region, with a decreasing rate from the South-East to the North-West. The endemic region is situated under the river Meuse, on the heights of the Ardennes. As the population of foxes increases in the last few years, the opportunity for contact between humans and this wild carnivore, even in urban areas, has consequently increased.

- Echinococcus in food animals
- Echinococcus in humans

- Possible risk factors include contact with dogs hunting for game, and ingestion of contaminated water or contaminated unwashed fresh products (in particular, raspberries and strawberries) and vegetables. Chewing grass is another practice to be associated with alveolar echinococcosis. Contamination of the hands during gardening, through contact with contaminated soil, may also carry some risk.
- Recommendations to improve the protection of public health are the use of good general hygiene practices such as washing fruit and vegetables before consumption, cooking berries or mushrooms (washing alone is not sufficient, neither does freezing at -18°C!), hand-washing after gardening and before the consumption of meals. Also hand-washing after contact with dogs, especially if they have direct contact with wildlife or if they live in areas where wildlife, in particular, foxes, rodents or voles, is abundant. Planned treatment of dogs with taenicides and subsequent hygienic disposal of their faeces in endemic areas is recommended.

Echinococcus in food animals

Surveillance programme and results

Post mortem macroscopic examination is done at the slaughterhouse in the Echinococcus domestic intermediate hosts: cattle, sheep, horses and pigs.

Whole carcasses or parts are rejected in case cysts are found.

Echinococcus in humans

In 2004, a serological study among 115 forest guards did not identify any suspect case of echinococcosis in this specific risk group.

One year later the National Reference Laboratory confirmed eight cases of hydatic echinococcosis. No case of alveolar echinococcosis was diagnosed.

In 2006, the National Reference Laboratory confirmed five cases of hydatic echinococcosis and one case of alveolar echinococcosis. This disease is probably under-diagnosed.

trends and sources

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cysticercosis

Cysticercosis

- Cysticercus bovis in muscular tissue of cattle is the larval stage of the tapeworm, Taenia saginata, a parasitic cestode of the human gut (taeniasis). The risk factor for bovine cysticercosis infection in cattle is the ingestion of feed contaminated with T. saginata eggs shed in human faeces. Cattle can become infected when grazing contaminated pastures in or around the farm. Free access of cattle to surface water, the flooding of pastures and the proximity of wastewater effluent have been identified as risk factors for bovine cysticercosis.

  Humans contaminate themselves by the ingestion of raw or undercooked beef containing the larval form (cysticerci). Usually the pathogenicity for humans is low. The tapeworm eggs contaminate the environment directly or through surface waters. Human carriers should be treated promptly. Strict rules for the hygienic disposal or sanitation of human faeces with a method that inactivates T. saginata eggs should be developed. The spreading of excrement on land is not allowed.

  Macroscopic examination is routinely done in adult cattle as well as in calves and sheep in the slaughterhouse. Serological examination is possible and confirmation of the lesions by PCR can be done. The introduction of serological techniques for the detection of cysticerci antigens in the serum of cattle should be developed. This would allow the detection of more cases than visual inspection of carcasses at the slaughterhouse, which has a low sensitivity.·

- Although Cysticercus ovis in sheep is not transmissible to humans, its presence causes total rejection of the carcass.·

- The Belgian pig population is virtually free from Cysticercus cellulosae. Taenia solium is not autochthonous in Belgium.

Cysticercosis in cattle

Post-mortem, macroscopic examination of carcasses is routinely done in the slaughterhouse. In 2006 496 181 adult cattle and 327 467 veal calves were tested.

Figures from the Federal Agency for the Safety of the Food Chain show that in 2006, 28 carcasses of adult cattle were rejected for generalised cysticercosis. In addition, the meat of 1 796 adult cattle was treated by a 10 days freezing before human consumption. No sheep were found to be infected.

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sarcosporidiosis and toxoplasmosis

Sarcosporidiosis and toxoplasmosis

The following species are of zoonotic importance: Sarcocystis bovihominis (man final host, bovine intermediate host), Sarcocystis suihominis (man final host, pig intermediate host) and Toxoplasma gondii (cat final host, man and most warm-blooded animals intermediate hosts).

Millions of oocysts from Toxoplasma gondii may be shed with the cat's faeces into the environment within the first two weeks after infection. These oocysts sporulate and are very resistant to environmental damage and can persist for several years. Oral ingestion of oocysts by a seronegative host leads to toxoplasmosis. The infection has an acute and a chronic phase. The latter characterised by the persistent presence of tissue cysts in the host (in muscle, brain, heart, …).

Man is infected with Sarcocystis spp by ingesting undercooked infected meat; infection with T. gondii occurs through ingestion of undercooked infected meat or upon accidental ingestion of sporulated oocysts from the environment.

Sarcocystis spp. infections are mostly asymptomatic but may cause mild a-specific gastrointestinal symptoms like nausea and diarrhoea. Most infections with T. gondii are asymptomatic, however mild (flu-like symptoms), moderate (lymphadenopathy, chronic fatigue) to severe disease (disseminated toxoplasmosis, encephalitis) may occur, the latter mainly in immunocompromised hosts. Moreover, when infection occurs in pregnant women, toxoplasmosis may cause abortion and congenital disorders. A percentage of congenitally infected children may develop symptomatic toxoplasmosis (e.g. ocular disease) between 1 to 14-year-old.

In the case of toxoplasmosis, the majority of adult persons have acquired immunity to re-infection but can remain carrier, while for human sarcosporidiosis there is no immunity development.

The majority of grazing animals are indiscernible carriers of tissue cysts.

- General overview
- Toxoplasmosis in humans
- Toxoplasmosis in animals

Toxoplasmosis in humans

General overview

Toxoplasma gondii is an obligate intracellular organism that can be found worldwide. The final hosts are the felidea (more commonly cats), humans and almost all warm-blooded animals are intermediate hosts. The sexual cycle takes place exclusively in the intestines of felidea. As a result millions of oocysts are shed into the environment with the cat's faeces within the first two weeks after infection. These oocysts sporulate and are very resistant to environmental damage and can persist for several years. Oral ingestion of oocysts by a seronegative host leads to toxoplasmosis. The infection has an acute and a chronic phase. The latter characterised by the persistent presence of tissue cysts in the host (in muscle, brain, heart, …). Carnivorous ingestion of infected tissues by a seronegative host (final or intermediate) will lead to development of the disease.

Sarcosporidiosis in animals

Surveillance programme in food animals

Carcasses are partially or entirely condemned when myositis eosinophilica (green colouring of the carcass) is seen. Myositis eosinophilica may be linked with sarcosporidiosis, although the association is not unequivocally proven.

Toxoplasmosis in humans

There is a whole battery of tests available to diagnose toxoplasmosis. As the disease is generally asymptomatic, diagnosis relies mostly on serological tests. In case of immunocompromised patients or congenital toxoplasmosis, more direct tests like PCR and bio-assay are needed to evaluate the gravity of the illness.

Only a very limited number of drugs may be used to control the infection: macrolides (spiramycine) and inhibitors of folate synthesis. In addition, these are only active on the free form of the parasite, not on the tissue cysts. The treatment takes a long time and is not without adverse effects. However, the effectiveness of antibiotic treatment in the case of congenital toxoplasmosis has been questioned. That is why preventive measures are very important for high-risk patients.

Efforts are made for primary prevention of toxoplasmosis during pregnancy. The mode of acquiring toxoplasmosis from meat, cat faeces and contaminated soil is so circumscribed that simple but effective measures should be recommended during pregnancy: regular hand-washing, especially after contact with cats, meat, soil and water. Freezing meat (at < -20°C for 48 hours) before consumption or adequate heating of meat during preparation are other effective measures.

Toxoplasmosis in animals

The majority of grazing animals are latent carriers of tissue cysts. There is a need for suitable microscopic, serological and molecular biological methods for both indirect and direct detection of T. gondii in animals and food. Serology based kits may detect infected animals. The presence of tissue cysts can be detected by PCR or bio-assay. Unfortunately, these tests are not routinely done and there is no data on the status of toxoplasmosis in the livestock in Belgium.

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avian influenza

Avian influenza

In 2003, the highly pathogenic avian influenza (HPAI) H5N1 strain, which started circulating in China in 1996, became endemic in poultry in several Asian countries. This unprecedented spread of HPAI was associated with a failure of surveillance and control measures in these countries, allowing the spread of the virus westwards since the summer of 2005, first in Europe and later in Africa. Another unprecedented feature of this HPAI H5N1 outbreak is its association with human disease and mortality. The total number of laboratory confirmed human cases since 2004 now reaches over 300, including almost 200 mortalities. The risk of generation of a new pandemic strain either by reassortment with circulating human influenza or by direct adaptation to humans is a considerable threat for public health. Unexpected infection of wild feline, cats and even dogs further illustrates unusual cross-species transmission of this H5N1 outbreak.

Due to the high contagiousness and the extreme severity of the disease, HPAI is the only "flu" of domestic animal considered as epizootic (i.e. former list A of the Office International des Epizooties), requiring drastic measures such as eradication for control. It has been estimated that hundreds million birds have been culled so far in attempt to control the spread of the Asiatic H5N1.

From fall 2005 to spring 2006, first HPAI H5N1 related mortalities were reported in Western Europe. This mortality was mainly reported in mute swans, but other waterfowl species were involved like mallards and common pochards, as well as raptors. Probably favoured by an exceptionally cold winter, this first incursion of H5N1 in Europe killed 741 wild birds between February and May 2006. Contrary to mortalities described in summer 2006 in Eastern Europe, these cases were independent of any poultry outbreak, suggesting that the birds were infected outside Europe and flew into our continent before dying. During the same period only 4 poultry holdings (one in France, in Germany, in Sweden and in Denmark) were affected, demonstrating the efficacy of the contingency plans in the EU Member States. Belgium, like neighbouring Netherlands and Luxemburg was not hit by H5N1, the closest case been recorded in Germany, at 200 km from the Belgian border.

- Monitoring of Avian influenza in 2006
- Avian influenza surveillance in humans

Monitoring of Avian influenza in 2006

Countries which are able to rapidly detect, contain and eradicate the disease based on a well-built surveillance system and control measures should continue with the stamping-out of infected flocks. The EU has established surveillance programs for AI in wild birds and poultry since 2003. In Belgium, like in other EU Member-States, an efficient monitoring plan has been implemented since autumn 2005 including 1) passive surveillance of dead birds, 2) active wild birds surveillance; 3) exclusion diagnosis in the professional sector (upon abnormal mortality rate or treatment set-up), and 4) increased serological surveillance in poultry (H5 and H7 specific Haemagglutination Inhibition tests). The monitoring is organised by the Federal Agency for the Security of the Food Chain in close coordination with the NRL (National Reference Laboratory) for Avian Influenza. The active wild bird surveillance is a close cooperation between the Royal Belgian Institute of Natural Science, the Veterinary Faculty of Liège and CODA-CERVA. All tests are realised at the NRL for Avian Influenza.

Passive surveillance of dead wild birds

Criteria for the passive monitoring and further analysis of dead birds and were related to the number of dead birds were determined, the finding place and the conditions in which the dead birds were found. During 2006, a total of 93 suspicions complying with these criteria (and corresponding to more than 500 wild birds) were analysed by Real Time RT-PCR and/or viral isolation, all with negative results. Between February and April 2006, the manifest increase of samples was a consequence of the positive cases observed in wild birds in Europe.

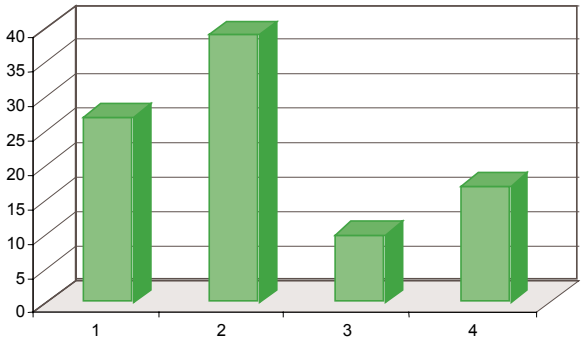

Figure 24. *Number of suspicion dossier / trimester of 2006*

Active surveillance of wild birds

A total of 2081 cloacal swabs were taken. From July 2006, oral swabs were additionally taken on a part of the birds, as it was found that H5N1 is mostly excreted by the respiratory track. The bird species were: mallard, common teal, Canada goose, Egyptian goose, coot, golden plover, lapwing, black-headed gull, herring gull, terns and raptors.

Sampling was organised in the whole territory of Belgium, but with a greater emphasis on areas where waterfowl density is the highest. Three groups were targeted: birds wintering in or migrating through Belgium and potentially originating from regions where H5N1 occurs, birds-eating raptors susceptible to be good indicators of virus contamination, and feral waterfowl representing a very important part of the biomass of Anatidae in Belgium, particularly during breeding season.

In total, 516 samples were taken from hunted mallards and teals and 2457 swabs were taken during ringing activities of other wild birds. No HP H5N1 was detected in wild birds during this active surveillance program, like in the other EU Member-States, but only low pathogenic avian influenza with an overall rate of about 1,8%.

Table 26. *List of wild birds species sampled for type A influenza viruses. Type column refers to birds category: M = migratory or wintering, R = bird-eating raptor (sampled during breeding season), F = feral species (mainly sampled during summer moult period).*

type	species	n sampled	cloacal	oral
R	Accipiter gentilis	44	44	
F	Alopochen aegyptiacus	35	35	
M	Anas acuta	32	32	
M	Anas crecca	7	7	
M	Anas platyrhynchos	280	280	23
M	Anas strepera	9	9	
M	Anser anser	4	4	4
M	Arenaria interpres	76	76	
M	Aythya ferina	39	39	3
M	Aythya fuligula	63	63	24
M	Aythya nyroca	1	1	
F	Branta canadensis	395	395	313
F	Cygnus olor	24	24	6
R	Falco peregrinus	23	23	
M	Fulica atra	150	150	3
M	Larus argentatus	156	156	
M	Larus fuscus	1	1	
M	Larus melanocephalus	2	2	
M	Larus michahellis	2	2	
M	Larus ridibundus	202	202	
M	Limosa laponica	1	1	
M	Numenius arquata	11	11	
M	Pluvialis apricaria	176	176	
M	Podiceps cristatus	1	1	
M	Sterna hirundo	142	142	
M	Sterna sandvicensis	37	37	
M	Sturnus vulgaris	2	2	
M	Tadorna tadorna	133	133	
M	Tringa totanus	1	1	
M	Turdus pilaris	1	1	
M	Vanellus vanellus	31	31	
	total	2081	2081	376

Surveillance of professional poultry flocks

In case of any abnormal symptom in a domesticated poultry flock, the owner had to inform his veterinarian who was obliged to examine clinical symptoms and evaluate a possible suspicion. In case of suspicion, samples are taken for further analysis. Since the summer of 2005, 550 possible cases were recorded and examined (figure). All results were negative. The peak of sampling was consistent with the increased passive surveillance of wild birds between February and May 2006.

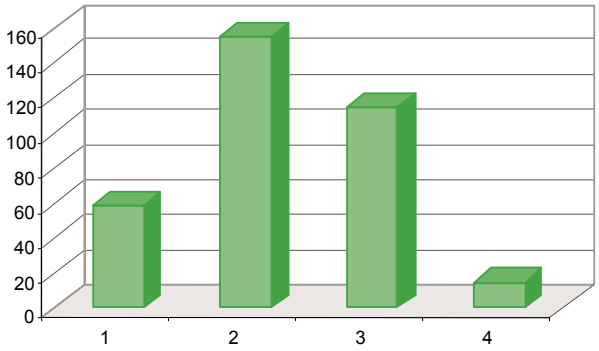

***Figure 25**. Number of exclusion diagnosis dossiers / trimester of 2006*

Vaccination of zoo birds

So far, three outbreaks have been documented in zoos in Asia (two in Hong Kong and one in Jakarta), indicating a possible threat for captive species. In case outbreaks would occur in or near zoos, possible eradication measures such as stamping out and pre-emptive culling of valuable and often endangered birds should be avoided by preventive measurements. Therefore, vaccination of zoo-birds was allowed by the European Commission, given certain provisions, at the 21st of October 2005 (2005/744/CE). After contacts between the government, zoo representatives and scientists in January 2006, zoos in Belgium could voluntary participate in a field trial with H5 inactivated vaccines (Ministerial decree 24/01/2006).

Birds were unequivocally identified by leg bands and/or by microchip and were vaccinated twice with 6 weeks interval. Official bio safety measurements such as keeping birds under cover or in confinement were followed in non-vaccinated birds as well as in vaccinated birds. During the trial, no vaccinated birds were transported outside the zoo and no birds were imported in the vaccinated flocks. Ten zoos participated in this field trial and 1175 birds were vaccinated.

Table 27. *Number of birds vaccinated against avian influenza*

Zoo	Number of birds involved
Antwerpen Zoo	65 (3 orders)
Planckendael Animal Park	111 (8 orders)
Le Monde Sauvage	102 (7 orders)
Vogelreservaat 't Zwin	251 (9 orders)
Parc Paradisio	122 (8 orders)
Plopsaland	47 (2 orders)
Sea Life Centre	10 (penguins)
Cracid Breeding and Conservation Centre	356 (2 orders)
Boudewijn Seapark	37 (6 orders)
Bellewaerde park	74 (5 orders)

The zoo veterinarians collected blood samples from a representative number of the bird group before the first vaccination and 4 weeks after the second vaccination. In total, 137 pre-vaccination sera and 125 post-vaccination sera four weeks after the boost vaccination were collected. Specific antibody titres against H5 and H7 subtypes were determined.

Before vaccination, 8% positive sera were found, mainly against H5 subtype. Cloacal swabs from these birds and from their group members were negative for viral isolation. Pre-vaccinal positive titres were mainly found in flocks of flamingos. All these flocks had access to outside ponds that could be visited by wild birds. The origin of the flocks was not always clear (include wild caught birds from Africa and North America and imports from other zoos), but most of them were established in the zoo before 1986, and with little exchange of birds after this date. These positive results are consistent with previous studies and indicate previous contact with H5 LPAIs.

After vaccination, 72% of the birds had titres higher or equal to 32; only 57% showed titres equal to or higher than 64. No significant difference was found between zoos. However, certain species seemed to have built up a high and presumably protective titre: galliformes, anseriformes, flamingos, gruiformes and ciconiformes. Bird species that responded badly to vaccination against avian influenza were birds of prey, pelicaniformes en ratites.

Avian influenza surveillance in humans

The surveillance of suspected cases of a virus infection by Influenza A/H5N1 is based on a standard operational procedure as made available for all clinicians (http://www.influenza.be/nl/document/Procedure_H5N1_voor_artsen_NL.pdf)

In 2006, 29 cases have been registered, and among these 10 were classified 'No case', 16 'Suspect', 2 'Possible' and 1 'Probable'.

The latter patient was a foreigner presented at the emergency department of a Brussels hospital on 13 January 2006. The man suffered from high fever, muscle pain, general discomfort, cough, nasal discharge and sore throat. He had visited poultry farms in the eastern province of Van, Turkey, from 9 to 12 January. According to the above mentioned SOP for the management of a potential case of human A/H5N1, the hospital reported the case to the Health Inspectorate of Brussels and the Scientific Institute of Public Health (IPH) in Brussels. After epidemiological evaluation, the patient was classified as a probable case. The laboratory results (rapid enzyme immunoassay test and RNA-based real time PCR tests [typing A and subtyping H5] and four nested RT-PCR tests [typing A and B, subtyping H5, subtyping H3 and H1, subtyping N1 and N2]) allowed investigators to discard the possibility of an A/H5N1 infection.

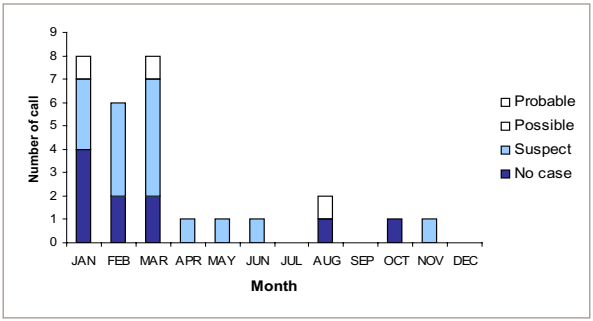

Figure 26. *Number of calls for suspicion of virus infection by Influenza A/H5N1, received at the guard duty of the department "Epidemiology", represented per month and classified by case definition, over the period January – December 2006, in Belgium*

Table 28. *Results of lab-tests carried out on 19 samples of suspect cases for virus infection by Influenza A/H5N1, represented per case definition, over the period January – December 2006, in Belgium.*

		Case definition			
Lab results		Suspect	Possible	Probable	Total
A	H1N1	2	1	0	3
	H3N2	0	0	1	1
B		1	0	0	1
Negative		13	1	0	14
Total		16	2	1	19

Table 29*. Distribution of the suspected cases of virus infection by Influenza A/H5N1, per destination, over the period January – December 2006, Belgium*

Country	Frequency	Lab results
Myanmar	1	
China	2	H1N1 (1)
China/Indonesia	2	H1N1 (2)
Indonesia/Bali	1	
Israel/Palestine	1	
Thailand	5	B (1)
Turkey	3	H3N2 (1)
Vietnam	2	
Vietnam/Cambodia	1	
No	1	
Total	19	

The surveillance system for the suspected cases of virus infection by Influenza A/H5N1 was effective. The family doctors had a good knowledge of the procedure, and they followed it up very well.

In 2006, no human case of virus infection by Influenza A/H5N1 has been identified at all.

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rabies

Rabies

Rabies is a zoonotic viral disease caused by Lyssaviruses and present in domestic and wild carnivores and bats all over the world. The animal reservoir are carnivores and bats. Other animals may be infected also, but do not play a role in the maintenance of the disease.

The Lyssavirus genus, within the Rhabdoviridae family, is sub-divided into several genotypes based on RNA sequencing:

- genotype 1 – 'Classic' rabies virus, worldwide spread
- genotype 2 – Lagos bat virus, Africa
- genotype 3 – Mokola virus, Africa
- genotype 4 – Duvenhage virus, Africa
- genotype 5 – European bat lyssavirus 1 (EBLV-1), Europe
- genotype 6 – European bat lyssavirus 2 (EBLV-2), Europe
- genotype 7 – Australian bat lyssavirus, Australia.

'Classic' rabies virus (RABV), genotype 1, causes an acute viral encephalomyelitis of warm blooded animals (e.g. foxes, dogs, cats, wildlife) and humans.

Rabies is transmitted to other animals and humans through close contacts with saliva from infected animals, especially via bites or scratches, or less frequently via licks on injured skin or on mucous membranes. The incubation period is usually from 4 to 8 weeks, but may range from 10 days to as long as one year or more. Once symptoms of the disease develop, rabies is fatal to both animals and humans. In humans, initial symptoms may include anxiety, headaches and fever. In a later stade, the effects of the encephalitis intensify. The inability to swallow liquids has given the disease the name of hydrophobia. Respiratory failure finally leads to death. Therefore it is important for any person who has been bitten by a 'suspected' animal (abnormal behaviour) to seek medical attention and start the necessary treatment consisting of wound treatment, passive immunization and vaccination.

• Rabies in animals

Some people may die despite post-exposure treatment using modern vaccines and/or rabies immunoglobulins. Pre-exposure vaccination should be offered to persons at risk, such as laboratory workers, veterinarians, animal handlers, international travellers. Currently available vaccines are safe and effective against both the classic rabies virus and the bat lyssaviruses.

Lyssaviruses and rabies in European bat species

Over one thousand species of bats are known world-wide. Bats are listed as endangered and protected animals across Europe. Rabies that may be detected in bats in some European countries is caused by two independent Lyssa virus genotypes 5 and 6 (EBL-1 and EBL-2) that are related to the Classical rabies virus. Some but not all the bat species carry the viruses. Bat rabies is a public health concern: after infection e.g. due to a bat bite, the disease is fatal in humans. Post-exposure vaccination and treatment following a bat bite or after exposure to bats is highly recommended. Education and recommendations should be given to travellers in order to reduce the risk of infection. Although dogs represent a more serious threat in many countries, the risk of rabies infection by bat bites should not be underestimated.

In July 2001, Belgium has obtained the official status of rabies-free country according to the OIE guidelines and the WHO recommendations. No indigenous cases of human rabies have been reported since 1923 although imported cases are diagnosed from time to time.

Rabies in animals

Surveillance programme and methods used

Food animals with nervous symptoms are suspect for rabies and therefore should be notified to the Federal Agency for the Safety of the Food chain. Affected animals are killed and their brain is examined by immunofluorescence and virus cultivation in neuroblastoma cells at the National Reference Laboratory. The remaining nervous tissue of rabies-negative animals is afterwards transmitted to the National Reference Laboratory for TSE diagnosis.

Wildlife found dead or shot is transferred to the clinical veterinary laboratories for autopsy. In case of suspected behaviour or lesions, brain samples are examined at the National Reference Laboratory.

Vaccination policy

Vaccine baits (Raboral, Rhône-Mérieux) were dispersed for the vaccination of foxes. In April and October 2003, a zone of approximately 1 800 km2 along the German border was covered by spreading 32 000 baits by means of a helicopter (17.78 baits per km2). Since there were no more cases of rabies for the last years, vaccination of foxes by baits was stopped by the end of 2003.

In the south of the country, below the rivers Sambre and Meuse, vaccination of dogs is compulsory.

Epidemiological investigations and results of 2006 surveillance

Passive surveillance of rabies

A total of 488 brain samples were examined for rabies virus at the National Reference Laboratory. The majority of samples originated from wildlife (n=94) especially foxes, deer (n=62), cattle (n=191) and sheep and goats (n=92). Twenty-one dead-found bats were also examined. The high number for cattle and small ruminants is the consequence of the surveillance system for transmissible spongiform encephalopathy (TSE) in these species: all suspected cases were first examined for rabies. Rabies must be considered in the differential diagnosis of TSE, although the course of the disease is usually shorter.

None of the samples was found positive. Since the last indigenously acquired case of rabies occurred in Belgium in a bovine in July 1999, the country is officially free of Classic rabies.

Surveillance of wildlife

Wildlife found dead or shot for signs of illness and/or agressivity are necropsied by the network of wildlife surveillance. In addition, brain samples are transmitted to the National Reference Laboratory. In 2006, the network has transmitted 94 samples of wild animals (foxes, wild cervids, badgers, mink and raccoon) to the National Reference Laboratory. All cases were negative (immunofluorescence and virus cultivation).

Seroprevalence of bat lyssaviruses

A preliminary study was undertaken to estimate the seroprevalence of EBL-1 and -2 in Belgian bats. Antibodies against EBL-1 were found in blood of 9 out of 58 bats captured in the South of Belgium. No antibodies against EBL-2 were found. Bats appeared in good health, indicating that EBL-1 circulates in Belgian bats without causing lethal disease.

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hantaviruses

Hantaviruses

Wild (or laboratory) rodents are the reservoir for hantaviruses worldwide; humans are accidental hosts. The infection is chronic and apparently asymptomatic in host animals. A hantavirus serotype is hosted by a specific rodent species. According to the infectious agent and its region, hanta-viral diseases present with different level of severity, from mild infections to severe hemorrhagic fever with renal syndrome (HFRS). HFRS shows as an acute onset of fever, lower back pain, hemorrhagic manifestations and renal involvement. Hantavirus pulmonary syndrome (HPS) was also described as an infection predominantly involving the respiratory system. Outbreaks of HFRS and HPS are generally observed during years with dense rodent populations resulting from favourable climatic and environmental conditions and when this population is heavily infected by the virus. Human activities, such as rodent trapping, farming, cleaning rodent-infested areas, camping and hunting, are also associated with- the occurrence of hantavirus disease.

Hantavirus is excreted through urine, faeces or saliva of rodents. The transmission of hantaviruses to humans mainly occurs via inhalation of infected excretions. Person-to-person transmission is rare. The virus can survive hours or days in the environment.

Strategies to prevent hanta-viral infections consist in controlling rodents in and around the houses, and cleaning houses with bleach. Preventive measures in endemic areas rely essentially on information campaigns and rodent control.

- Cases of Hantaviruses — data

Cases of Hantaviruses — data

In 2006, the Belgian Sentinel Laboratory Network and the Reference Laboratory reported 163 cases of hantavirus. This report indicates a decrease of cases as compared to 2005 (Figure 28).

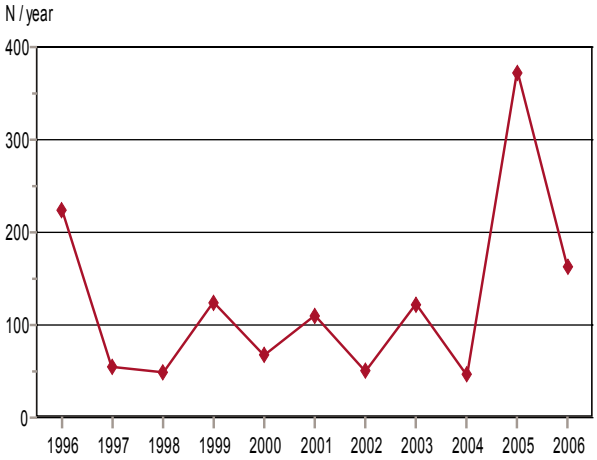

Figure 27. *Yearly number of Hantavirus infections, 1996 –2006*
Sources: Belgian Sentinel Laboratory Network and Reference Laboratory

Classically, hantavirus infections in Belgium display a seasonal peak in spring and summer and a periodic resurgence every 2 to 3 years. High seasonal peaks were reported in Belgium during the springs-summers of 1996, 1999, 2001, 2003 and specially 2005.

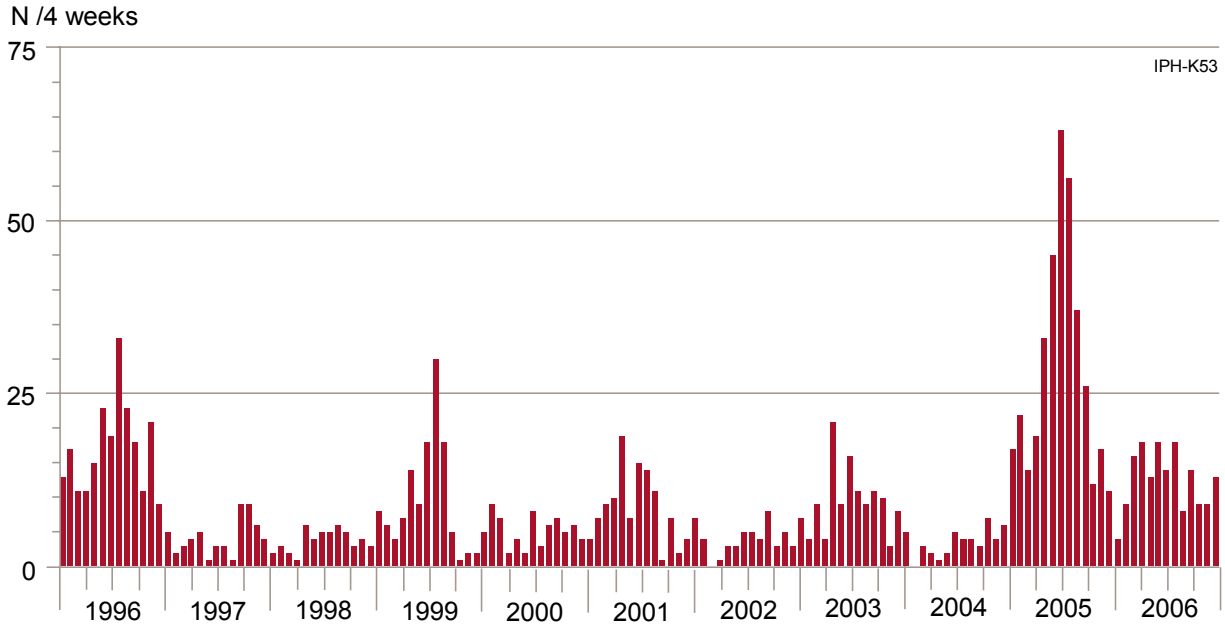

Figure 28. *Distribution of Hantavirus infections (N/4 weeks), 1996 –2006 Sources: Belgian Laboratory Sentinel Network and Reference Laboratory*

Among the cases reported in 2006, 83% (n=121) resided in Wallonia, 12% (n=18) in Flanders and 5% (n=4) in Brussels. The highest incidence rates are reported in the districts of Liège (n=26), Thuin (n=23), Philippeville (n=16) and Neufchâteau (n=15). Most of these areas are known to be endemic for the disease, but cases in the district of Liège are only reported from 2003 on.

In 2006, the majority of cases are adults over 19 years (80%) and 64% are males.

Part of the increase observed in 2005 could be due to a greater awareness among health professionals and to a higher hantavirus testing. However, under-diagnosing of hantavirus infections remains a problem.

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transmissible spongioform encephalopathy

Transmissible Spongioform Encephalopathy

Transmissible spongiform encephalopathies (TSEs) known as prion diseases, are caused by an infectious agent, whose molecular properties have not been fully determined. The animal TSEs include the archetype – scrapie in domestic sheep and goats – and animal diseases much more recently recognized, including transmissible mink encephalopathy (TME) and feline spongiform encephalopathy (FSE), chronic wasting disease (CWD) of deer and elk, and bovine spongiform encephalopathy (BSE).

Transmissible Spongioform Encephalopathy

BSE became a notifiable disease in Belgium in 1990. In the beginning of 2001, the 'passive' surveillance including the herd slaughter and compensation policy that started in 1997 was supplemented with an 'active' surveillance based on EU Regulation (EC) N° 999/2001 controlling slaughtered animals and the fallen stock.

The national reference laboratory uses 5 tests for diagnosis, i.e. the 'rapid' ELISA test , histopathology, immunohistochemistry, electronmicroscopic detection of scrapie associated fibrils (SAFs) and western blotting. All 19 private laboratories (primary 'active' screening) and the NRL are accredited (ISO 17025:2005) and the whole epidemiological surveillance is coordinated by the Federal Agency for the Safety of the Food Chain.

***Table 30**. Number of animals controlled in Belgium (2001-2006)*

Year		Slaughterhouse	Suspected Animals: Herd screening / farm, slaughter, autopsies	Fallen stock
2001	Cattle	360 948	3 522 / 379	13 060
	Small ruminants	0	11 / 45	0
2002	Cattle	410 379	3 277 / 377	36 386
	Small ruminants	2 195	428 / 85	780
2003	Cattle	357 398	1 126 / 250	33 691
	Small ruminants	2 447	205 / 52	499
2004	Cattle	358 120	172 / 254	35 322
	Small ruminants	39	333 / 170	1 650
2005	Cattle	325 302	15 / 234	41 729
	Small ruminants	703	8 / 86	1 588
2006	Cattle	320 541	8 / 185	44 066
	Small ruminants	8 076	81 / 90	3 064
Total	Cattle	2 132 688	8 120 / 1 707	204 254
	Small ruminants	13 460	1 066 / 528	7 581

Table 31. *Positive TSE cases in cattle and sheep in Belgium (First case – 2005)*

Year	Cattle	Sheep (primary outbreaks)
1992	0	1 (First case) / 5C
1993	0	0
1994	0	0
1995	0	0
1996	0	0
1997	1 (First case) / C	2 / C
1998	6 / C	8 / 3C – 5Sc
1999	3 / C	11 / 2C – 9Sc
2000	9 / C	0
2001	46 / 28S – 10C – 7F – 1Sc	0
2002	38 / 17S – 5C – 16F	25 (1 atypical case) / 1S - 2C - 2F – 20Sc
2003	15 / 10S – 5F	2 / F
2004	11 / 6S – 3C – 2F	11 (1 atypical case) / 1S – 3F – 7Sc
2005	2 / 1S – 1C	2 (2 atypical cases) / F
2006	2 / 1S – 1F	3 (3 atypical cases) / 2F – 1S
Total	133 (63 slaughterhouse / 38 clinical cases / 31 fallen stock / 1 second case in a farm)	69 (3 slaughterhouse / 14 clinical cases / 11 fallen stock / 41 Sc)

S = slaughterhouse control / C = suspected clinical / F = fallen stock / Sc = additional case in a herd

Laboratory and epidemiologic studies provided strong circumstantial evidence for a causal link between vCJD and the BSE epizootic in cattle with the most likely route of primary human infection being through dietary exposure to highly infected bovine tissues.

On 28th of January 2005, the European Commission confirmed the first known naturally occurring case of the BSE agent in a goat, slaughtered in France in 2002. Previously, sheep and goats had only been infected experimentally. No other goats from the same herd were demonstrated to have a BSE infection or to show any signs of BSE disease, and none of the animals entered either the food or feed chain. This incident was therefore not considered to represent a risk to public health. The infected goat was born in 2000. A ban on feeding meat and bone meal (MBM) to ruminants (i.e cattle, sheep and goats) is in place since 1994; this was extended to all farmed animals in 2001. Goats in the European Union generally only live for a few years, which means that the majority of goats in the EU today were born after the total feed ban was put in place. Nevertheless, in response to this case of confirmed natural BSE infection in a goat, the Commission proposed to improve vigilance for such incidents by increasing BSE testing of goats, and has set a target of 200 000 healthy goats to be tested in the European Union.

TSE Road map

On the 15th of July 2005, the European Commission published a TSE Road map. This document contains future goals in BSE policy on, among others, the definition and removal of Specified Risk Material (SRM), the feed ban and the age of testing. In fact, we have come to the stage that amendments of certain measures could be envisaged without endangering the health of the consumer or the policy of eradicating BSE, provided that the positive trend continues and scientific conditions are in place. Indeed different indicators already suggest a favourable trend in the BSE epidemic and a clear improvement of the situation in recent years due to the risk reducing measures in place. There is a significant overall decrease in the number of cases of the disease across the EU (see fig 30).

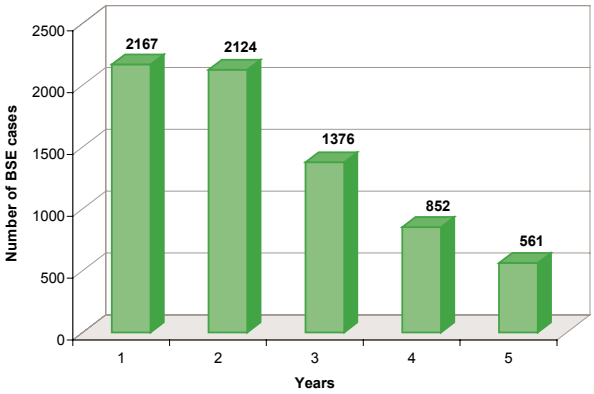

Figure 29. *Positive BSE cases in Europe (2001 – 2005)*

In the road map the EC also proposed, due to the considerable economic consequences in the United States and Canada, to set up a TSE survey on cervids in Europe. Based on that proposition, the NRL started with a screening of cervids in both the North and South part of Belgium. Based on these studies, a Bayesian framework was used for the estimation of the true prevalence of CWD in Belgium. The prevalence was estimated to have a median value of zero with a 95th percentile value of 0.00115 for the Southern and 0.0045 to 0.0049 for the Northern part respectively.

TSE in humans

After the description of variant CJD (vCJD) and the demonstration of a link with the epidemic of bovine spongiform encephalopathy, vigilance with regards to the incidence of both vCJD and sCJD was increased, leading to the implementation of the Belgian CJD surveillance network. The network is a collaborative study of the seven university centers of neurology/ neuropathology and the Institute of Public Health (IPH), which maintains the surveillance database. The university departments refer all patients with a clinical diagnosis of probable CJD to the IPH and eventually submit the final, if possible neuropathological, diagnosis to the database. The networks communicates incidence figures to the CJD Surveillance Center in Edinburgh and the European Center for Disease Control.

Sporadic Creutzfeldt-Jakob disease has a presumed incidence of 1.5 to 2.0 patients per million inhabitants per year (2005: n=20; 2006: n=16). The median age at death of CJD patients in Belgium has consistently been reported to be between 60 and 65 years of age although a broad range, between 16 and 90 year old, has been recognized. The clinical evolution of sCJD is variable but most commonly includes rapidly progressive dementia, motor disabilities, visual problems and eventually akinetic mutism, resulting in death within 1 year after onset. The definite diagnosis of CJD is based on the identification of the classical neuropathological triad: neuronal loss, gliosis and spongiform degeneration and recently by the identification of prion protein deposition in the brain. A patient is diagnosed with 'possible' CJD if in addition to a rapidly

progressive dementia with a duration of less than 2 years also 2 of the mutism and cerebellar signs are present. If periodic sharp wave complexes (PSWC) on the electroencephalogram (EEG) or the protein 14-3-3 test in cerebrospinal fluid (CSF) can be identified, patients are classified as 'probable' CJD.

A major development in the diagnosis of sCJD is the immunodetection of specific CSF biomarkers. Especially the detection of the 14-3-3 protein and tau protein are used as they are reported to be the most sensitive and specific biomarkers for sCJD. In addition, decreased levels of full-length amyloid-beta (Ab1-42) can also be found in sCJD.

The Born Bunge Institute (University of Antwerp) and the IPH database contains references to all patients in whom CJD was clinically suspected. This database can be used as an important fail-safe to identify patients not further investigated in one of the other university centers. In general, the autopsy rate of CJD patients in Belgium remains at 80%. The autopsies have been facilitated by financial support for the transport of patients to one of the reference centers. After autopsy, brain tissue is fixed in 10% formaldehyde and frozen samples are also obtained for subtype analyses and sequencing of the prion protein gene (PRNP). Since 1998, some 230 patients have been referred to the surveillance system. Until present, no variant CJD patients have been observed. No patients could reliably be associated with surgical procedures or injection of extracted human growth hormone or gonadotropin. We have identified two unrelated patients with an E200K mutation and 3 patients in a two separate families with an octapeptide repeat insertion in the PRNP gene. One patient was observed after treatment with a vCJD contaminated plasma derivative, but the causal relation remains questionable. At least 4 CJD patients had acted as blood donors at some point before developing the disease. The blood transfusion centers were notified of this occurrence.

We observed that Alzheimer's disease and dementia with Lewy bodies were the most frequent alternative diagnoses after neuropathological examination. The probable and definite sCJD patients had a median age at death of 66 years with a standard deviation (SD) of 10 years. The youngest CJD patient was 32 year at the time of demise, the oldest patient was 88. There was a small preponderance of males over females (1.2M:1F) although large year-to-year variations were observed. In our series, 86% of patients died within the first year after disease onset. All patients lived at least 1 month after disease onset with a median duration of 5.7 months (SD 5.6, range 1–38 months).

In most European countries an increased incidence of sCJD was reported, especially in patients between age 60 and 80 (age specific incidence between 5 and 6/106/year). This finding has been confirmed in our series although we obtained even higher incidence figures (±7.5/106/year) in this age group and also observed this increase in patients between 80 and 90 years (age specific incidence of 6.3/106/year). Most likely this increase is due to more accurate and vigilant surveillance of CJD. The Belgian CJD surveillance system, although voluntary, has identified one of the highest incidence figures in Europe. This is most likely due to a good coordination and the follow-up of the majority of the patients from clinical diagnosis to neuropathological confirmation.

Tables & figures

- Federal Agency for the Safety of the Food Chain (FAVV-AFSCA)
- Scientific Institute of Public Health (WIV-ISP)
- Veterinary and Agrochemical Research Centre (CODA-CERVA)