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Microbial safety of meat in the European Union

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Abstract

The two most frequently reported zoonotic diseases in humans in the EU in 2005 were *Campylobacter* and *Salmonella* infections with incidences of 51.6 and 38.2 cases per 100,000 population, respectively. Reported human infections caused by *Yersinia* spp., Verocytotox-igenic *Escherichia coli*, and *Listeria monocytogenes* had comparably lower incidences of 2.6, 1.2 and 0.3 cases per 100,000 population, respectively. Meat and meat products are important sources for these infections but knowledge on exactly how important they are compared with other types of food, drinking water and environmental exposure is quite limited. Occurrences of zoonotic pathogens in raw meat are variable, although most often are between 1% and 10%, depending on the organism, geographical factors, farming and/or meat production practices, etc.

Zoonotic pathogens in meat have to be controlled through a complete, continuous farm-to-fork system. It is of utmost importance to control faecal contamination of carcasses through efficient HACCP-based process hygiene management systems. © 2007 Elsevier Ltd. All rights reserved.

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1. Introduction

Zoonoses are diseases or infections, which are transmissible from animals to humans. Zoonotic agents reportedly affected over 387,000 persons in the EU in 2005; these diseases can be acquired directly from animals but are most often acquired through ingestion of contaminated foods. The two most frequently reported zoonotic diseases in humans in the EU in 2005 were Campylobacter and Salmonella infections with incidences of 51.6 and 38.2 cases per 100,000 population, respectively (EFSA, 2006b). Reported human infections caused by Yersinia spp, Verocytotoxigenic Escherichia coli (VTEC), and Listeria monocytogenes had comparably lower incidences of 2.6, 1.2 and 0.3 cases per 100,000 population, respectively. Nevertheless, these diseases may be more severe, at least in at-risk sub-populations, such as VTEC infections in children or

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L. monocytogenes infections in immunocompromised individuals with rather high fatality.

Although various foods can serve as sources of foodborne illness, meat and meat products are important sources of human infections with Salmonella spp., Campylobacter jejunilcoli, Yersinia enterocolitica, VTEC and, to some extent, L. monocytogenes. All these foodborne pathogens can be harbored in the gastrointestinal tract of food-producing animals. The most frequent chain of events leading to meat borne illness involves food animals as healthy carriers of the pathogens; these organisms are subsequently transferred to humans through production, handling and consumption of meat and meat products. Occurrences of Salmonella spp., C. jejunilcoli, Y. enterocolitica and VTEC in fresh red meat are variable, although most often are between 1% and 10%, depending on a range of factors including the organism, geographical factors, farming and/or meat production practices.

Zoonotic pathogens in foods including meats have to be controlled through a complete, continuous farm-to-fork system and should take into account not only the risks,

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but also technical possibilities, consumers' attitude and behaviors, and cost-benefit analysis. However, some aspects of the control system are pathogen-specific. Thus some pathogens in meats (e.g. *Salmonella* spp., *Campylobacter* spp., *Y. enterocolitica* and VTEC are most efficiently controlled by the main interventions applied in the primary production combined with optimization of the slaughter hygiene. For some others, such as more environmentally ubiquitous *L. monocytogenes*, the main control measures are focused on later stages of the meat chain.

The present paper is not an exhaustive review of meat borne pathogens but gives an overview of the main microbial risks associated with the meat chain using examples of selected bacterial pathogens and production processes.

2. Main microbial foodborne infections in Europe

The two most frequently reported zoonotic diseases in humans in the EU in 2005 were *Campylobacter* and *Salmonella* infections with incidences of 51.6 and 38.2 cases per 100,000 population, respectively (EFSA, 2006b).

Campylobacteriosis in humans is caused by thermophilic *Campylobacter* spp. with *C. jejuni* followed by *C. coli* being the most common. The most common symptoms of campylobacteriosis include diarrhoea often bloody, abdominal pain, fever headache and nausea. Usually, infections are self-limiting and last a few days but complications such as arthritis and neurological disorders occur occasionally. In 2005, a total of 197,363 cases of campylobacteriosis were reported by 22 EU Member states (MS). The EU incidence of 51.6 per 100.000 population makes campylobacteriosis the most frequently reported zoonotic disease in EU. The community incidence increased by 7.8% compared to 2004, but no common trend within the MS was evident.

Human salmonellosis is usually characterized by fever, diarrhoea, abdominal pain and nausea. Symptoms are often mild and most infections are self-limiting within a few days. Occasionally the infection may be more serious with severe dehydration and even death. Salmonellosis has also been associated with chronic sequelae like arthritis. A total of 176,395 cases of human salmonellosis were reported by 24 MS in 2005. Even though seven MS reported a slight increase in cases, an overall decrease of 9.5% in the incidence was observed compared with 2004.

Yersiniosis caused by *Y. enterocolitica* affects mainly young children and symptoms are dominated by diarrhoea. In older children and adults abdominal pain and fever may be predominant. Complications such as joint pains or spread of bacteria to the blood stream can occur. In 2005 a total of 9630 cases of human yersiniosis were reported by 21 MS. The total number of cases reported within EU has decreased slightly from 2002 to 2005. In 2005 the EU incidence was 2.6 per 100,000 population.

Verocytotoxigenic *E. coli* (VTEC) are a group of *E. coli* bacteria that are characterized by their ability to produce verocytotoxin (VT). Human pathogenic VTEC usually har-

bor additional virulence factors important for the development of disease in man. A large number of different E. coli serotypes include VT-producing strains, but the majority of reported human VTEC infections are associated with a small number of O:H serotypes. VTEC O157 is the most frequently reported to be associated with human disease. The symptoms associated with VTEC in humans are mild to bloody diarrhoea often accompanied by severe abdominal cramps. VTEC infections can also result in haemolytic uraemic syndrome (HUS). HUS develops in up to 10% of patients infected with VTEC O157 and is the leading cause of renal failure in young children. In 2005, a total of 3,314 human cases were reported from 18 MS. The overall incidence in the EU was 1.2 per 100,000 population. However, because large differences in the VTEC diagnostic practices exist, trend analysis and between-country comparisons are difficult.

Listeriosis is a disease caused by infection with *L. monocytogenes*. Symptoms of listeriosis may range from mild flu-like symptoms and diarrhoea to life threatening forms characterized by septicaemia and meningitis. In pregnant women, the infection may spread to the foetus and result in abortion or birth of a child with septicaemia. Human listeriosis is rare but the disease is severe. Old and immuno-compromised persons are those most often affected. In 2005, 1,439 cases of listeriosis were reported in the EU. The overall incidence was 0.3 cases per 100.000 population.

3. The contribution of meats as sources of foodborne infections

Although it is clear that meat or meat products can be implicated in all of the above zoonotic infections in humans, understanding in quantitative terms of the importance of meat and meat products compared with other types of food, drinking water and environmental exposure is quite limited. Efforts to quantify the (relative) importance of specific food sources and animal reservoirs for human cases of foodborne illness have been named "human illness attribution". Several human illness attribution approaches, and related data, are currently used worldwide (Batz et al., 2005) including: (a) microbial subtyping, (b) analysis of outbreak data, and (c) exposure assessment.

3.1. Microbial sub-typing

Microbial sub-typing involves characterisation of the pathogen by different pheno- or genotypic typing methods (e.g. sero-typing, phage-typing, antimicrobial susceptibility testing, pulsed-field gel electrophoresis and sequence-based sub-typing). The Danish Zoonosis Centre has, over the past decade, produced annual estimates of the number of human *Salmonella* infections attributable to the various food animal sources based on a model using microbial sub-typing results (Hald, Vose, Wegener, & Koupeev, 2004; Anonymous, 2005b).

During this period, the validity of the estimates produced by the attribution model has been improved considerably. The Salmonella surveillance programmes have been gradually extended, resulting in more abundant data. Estimates of the number of human Salmonella infections attributable to the various food animal sources in 2005 in Denmark are indicated in Fig. 1. It appears that meat was implicated in between 20% and 50% of all human cases of human salmonellosis. The single most important type of meat was domestically produced pork (9-15.7%) and imported chicken (8.6-13.4%). Nevertheless, at the EU-level, available data are still insufficient for proper understanding and quantification of the role of meat in foodborne disease, so development of methods for source attribution and their implementation in individual countries need to be treated as a priority.

3.2. Analysis of outbreaks data

The importance of meat in foodborne outbreaks of salmonellosis and campylobacteriosis in the EU is illustrated by data shown in Tables 1 and 2, respectively. It appears (Table 1) that eggs and products thereof comprise the most important single food that contributes to outbreak related human salmonellosis (11%) while poultry and pork meat contributes to 2.7% and 1.5%, respectively. Salmonella was the most frequently reported organism involved in foodborne outbreaks in 2005 and 25,760 cases of human salmonellosis are reported to be related to outbreaks. In contrast, there were only 2478 outbreak-related cases of campylobacteriosis. Thus, the majority of reported human cases of campylobacteriosis were sporadic cases. Poultry meats make up the most important single food type contributing to outbreak-related human campylobacteriosis (Table 2).



Fig. 1. Estimated sources of 1775 cases of human salmonellosis in Denmark 2005 (adapted from Annual Report on Zoonoses in Denmark 2005 (Anonymous, 2005b)).

Table 1

Relative relevance of meat in foodborne outbreaks of salmonellosis in the EU (adapted from EFSA, 2006b)

Sources	Persons ^a infected (%) in outbreaks in 2005
Meats	
Poultry meats	2.7
Porcine meats	1.5
Bovine meats	0.3
Eggs and products thereof	11
Other ("unknown", seafood, fruit/ vegetables, etc.)	75

^a A total of 25,760 persons in 2005.

Table 2

Relative relevance of meat in foodborne outbreaks of campylobacteriosis in the EU (adapted from EFSA, 2006b)

Sources	Persons ^a infected (%) in outbreaks in 2005
Meats	
Poultry meats	13.0
Bovine meats	_
Other meats ("unspecified")	0.5
Eggs and products thereof	0.2
Water	28.7
Other ("unknown", fruit/vegetables, not reported)	57.6

^a A total of 2478 persons in 2005.

Foods are often implicated as a vehicle in *E. coli* O157 outbreaks (Smith, Willshaw, Cheasty, & O'Brien, 2001; Smith, 2004; Rangel, Sparling, Crowe, Griffin, & Swerdlow, 2005). In 2005 within the EU, eighteen outbreaks due to Verotoxigenic *E. coli* (VTEC) involving 180 cases were recorded. One outbreak involving 69 cases occurred in France and was linked to consumption of minced bovine meat. Other sources included cheeses and fresh produce (EFSA, 2006b).

Examples of red meat-borne outbreaks of *E. coli* O157 that have occurred worldwide are shown in Table 3. Although many cases have been caused by consumption of ground beef (minces/burgers/patties), other meat products including fermented sausages and dried venison were also involved.

When considering sources of infection in the main examples of reported outbreaks of human listeriosis (EFSA, 1999; Ryser, 1999, chap. 10; Farber & Peterkin, 2000), it appears that processed Ready-To-Eat (RTE) meat products are commonly involved. Examples of RTE meat products involved in outbreaks of human listeriosis are shown in Table 4.

3.3. Exposure assessment

3.3.1. Pre-harvest phase

3.3.1.1. Occurrence of foodborne pathogens in animals. The original sources of foodborne pathogens that presently

Table 3Examples of meats as sources in E. coli O157 outbreaks

Outbreaks		
Implicated meats	Cases (deaths)	Reference
Ground beef	26	Wells et al. (1983)
	21	Wells et al. (1983)
	34 (4)	Ryan et al. (1986)
	51	Pavia et al. (1990)
	54	Belongia et al. (1991)
	732 (4)	Meng et al. (2001)
		Bell et al. (1994)
		CDCP (1993)
	22 (1)	CDSC (1999)
	8 (1)	CDSC (2000)
	19	CDCP (2003)
	28	Barrett et al. (1992)
Beef tacos	13	Conway (1995)
Beef ("seeme rolle")	11	Werber et al. (2002)
Beef (roast)	65	CDCP (1990)
Cooked meat	496 (20)	Pennington (1998)
	30	Rajpura et al. (2003)
Meat balls, coleslaw	13	Meng et al. (2001)
Raw, fermented sausages	19	CDCP (1994)
	39	Williams et al. (2000)
	150	Health Canada
		(2000)
	28 (3)	Hjertquist et al.
		(2002)
Sausages (mortadella and teewurst)	28 (3)	Ammon et al. (1999)
Venison jerky	11	Keene et al. (1997)

Table 4

Examples of meats as sources of *Listeria monocytogenes* infections (adapted from Buncic and Avery, 2004)

Meat types involved	Country (year)	Number of deaths
Red meats		
Pate (outbreaks)	UK (1987–1989)	94 (at least)
	Australia (1990)	6
	France (1993)	12
Hot dogs (outbreak)	USA (1998– 1999)	>10
Pork tongue in aspic (outbreak)	France (1992)	85
Sausages (sporadic cases)	USA (1989)	Not known
	Sweden (1993)	0
Poultry meats		
Turkey frankfurters (sporadic case)	USA (1985)	0
Cooked chicken (sporadic case)	England	1

cause most human meat-borne bacterial diseases are farm animals that show no symptoms of illness but which faecally excrete the pathogens. Recently reported data on the occurrence of the main microbial foodborne pathogens in farm animals in the EU (EFSA, 2006a, 2006b) are indicated in Table 5.

Among *Campylobacter* spp. in animals, the most commonly isolated in cattle was *C. jejuni*, whilst it was *C. coli*

Table	5	
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Occurrences of microbial foodborne pathogens in meat farm animals in the EU (adapted from EFSA, 2006a, 2006b)

Pathogen	Reported occurrence (%)						
(Year)	Cattle ^a	Pigs ^a	Sheep/goats ^a	Poultry ^b			
Campylobacter							
2005	0.3-46.9	24.7-85.4	DNA/I	0.2-85.2			
2004	0.1-64.2	0.4–79.6	DNA/I	3.1–91.0			
Salmonella spp.							
2005	0-6.7	0-60.0	DNA/I	0 - 18.2			
2004	0.1–1.5	0.4–29.4	DNA/I	1-23.4			
VTEC							
2005	0-21.6	0-9.2	0–11.8 (FD)	DNA/I			
2004	1-24.1	4.9-8.6	1.1–9.4	DNA/I			
Listeria							
2005	DNA/I	DNA/I	DNA/I	DNA/I			
2004	DNA/I	DNA/I	DNA/I	DNA/I			
Yersinia							
2005	1.0	0.7	0.6	DNA/I			
	(Germany)	(Germany)	(Germany)				
2004	0.4–17.6	0.9–10.4	0.1–0.2	DNA/I			

^a Herds.

^b Flocks; FD, few data; DNA/I, data not available or insufficient.

in pigs. Salmonella was reported in all farm animals, but was found most frequently in poultry. In red meat animals, Salmonella was most frequently found in pigs, followed by cattle. VTEC in general, as well as VTEC O157, were most frequently found in cattle confirming their role as the main reservoir of the pathogen. Nevertheless, due to the lack of further genotypic and virulence characterization data for the isolates, their public health relevance is unclear. Reported data on *L. monocytogenes* in animals was scarce, and some of them relate actually to isolates from clinical cases – occurring primarily in small and large ruminants – rather than from monitoring. With Yersinia, the dominant type found in animals was *Y. enterocolitica* O3.

3.3.1.2. Factors affecting the occurrence in animals and related controls. These enteric pathogens from animals can be further spread in the meat chain by a variety of routes. To minimize their further transference to post-farm phases of the food chain, it is necessary to understand the epidemiology of pathogens at the farm level. Although in-depth consideration of the risk factors and the controls on farm would need to be both pathogen- and animal species-specific, many aspects are common amongst, and applicable to, all the pathogens dealt with in this paper. Due to space limitations, they will be only generally indicated here.

Animal feeds/diet. The main examples of contaminated feed being important source of foodborne pathogens include, but are not limited to, Salmonella spp. in poultry and pigs. In such cases, protein components of the feed are of main concern. "Exotic" strains of Salmonella spp. associated with purchased feed are often transient, whilst "local", well-established strains of Salmonella spp. are usually the most persistent at the farm. However, feeds also can be contaminated with pathogens excreted by vermin (rodents, birds). On the other hand, there have been claims that some types of diet can affect the occurrence and/or levels of shedding of the pathogens by farm animals, but their relevance is still unclear. For example, the long-lasting debate whether shedding of *E. coli* O157 is higher in grain-fed or hay-fed cattle is still unresolved (Doyle & Erickson, 2006). Overall, it is very difficult to compare the effects of particular diets on pathogens' shedding between different studies due to influences of other animaland/or farm-related variable factors acting simultaneously. With respect to animal feed-based control measures to reduce faecal shedding of pathogens on farms, following approaches have attracted significant attention:

- Feed can be treated to control pathogens. This includes fermentation, e.g. of liquid feeds for pigs so to reduce risk of *Salmonella* infection or acidification by acidulants, as well as heat treatments.
- Probiotics can be incorporated in the diet. This is based on feeding with viable microorganisms antagonistic toward pathogens via either modifying environmental factors in the gut or producing antimicrobial compounds.
- Competitive exclusion concept can be applied, primarily in monogastric animals. This involves feeding with complex mixtures of bacteria that reduce attachment of pathogens to the gut mucosa. For example, colonization with *Salmonella* spp. in intensively reared chicks can be inhibited by feeding with gut content of mature hens.

Stress. The normal, balanced gut microflora in animals provides reasonable protection against colonization with pathogens, e.g. Salmonella spp. Stress in animals can disturb this status, weaken the immune responsiveness, and cause an increase in shedding of pathogens. Hence, stress management is a relevant aspect of pathogen control. Some stressors occur "naturally", e.g. parturition and weaning, whilst others occur due to poor animal husbandry, e.g. inadequate housing, sudden changes in diet and rough handling.

Biosecurity. An important source of foodborne pathogens on farm is newly introduced infected animals. Furthermore, spread of pathogens can occur between distant pens within, or indeed between, farms via a range of vectors including vermin, wild animals, farm staff/visitors and farm equipment.

Animal husbandry. Intensive indoor farming (i.e. group housing) with animals being in close proximity generally results in increased horizontal transmission of pathogens, compared to outdoor farming. The transmission routes include aerosols, frequent physical contacts with contaminated environmental surfaces or contaminated animal coats in a contained space, and social exchanges (licking/ grooming). All water drinkers on farms, used by more than one animal, can serve as a source of animal infections and re-infections. In the farm environment, pathogens can survive for extended periods, from days to months, in/on various substrates: faeces, soil, water and building materials (Hutchison, Nicholson, Smith, Keevil, & Moore, 2000). As found with a range of substrates, pathogens survive better under dirty/humid/cold than under clean/dry/warm environmental conditions (Small, Reid, & Buncic, 2003) although significant strain-related variability can exist, as shown with *E. coli* O157 (Avery & Buncic, 2003). Overall, application of good husbandry practices including effective cleaning/sanitation regimes are essential tools in pathogen control on farm.

Land management: Spreading untreated abattoir- and/or farm wastes (manure, slurry) containing enteric pathogens as fertilizers on pasture or agricultural land for crop production can mediate further infections or re-infections of animals with pathogens through contaminated grazing, harvested feed or water supply (Pepperell, Massanet-Nicolau, Allen, & Buncic, 2003; Hutchison, Walters, Avery, & Moore, 2004). Related control measures include appropriate storage (composting) of manure resulting in "auto-sterilisation" through generation of heat (55–60 °C) and "lagoon" treatment of effluents before their application to agricultural land (Hutchison et al., 2000).

Vaccination: Vaccination of animals, particularly when combined with other measures implemented further along the food chain, can be an efficient strategy for pathogen reduction. For example, in the UK, vaccination of poultry against *Salmonella* contributed to significant reduction of the pathogen in poultry meat (EFSA, 2004). Nevertheless, for other pathogens, such as *E. coli* O157 or *Campylobacter*, vaccines are being researched but effective ones are not yet practically available.

Transport and lairaging: The transport-lairage (TL) phase usually increases the occurrence and/or levels of foodborne pathogens in animals (Berends, Urling, Snijders, & Van Knapen, 1996; Fravalo, Rose, Eveno, Salvat, & et Madec, 1999). Generally, when presented for slaughter, the pathogen-positive animals can be comprised of: (a) the initial, on-farm infected faecal shedders; (b) those which became faecal shedders during the TL due to reactivation of on-farm latent infection; (c) newly infected shedders; and (d) those with the surface (skin) crosscontaminated from either faecal shedders or non-shedders. Microbial cross-contamination during TL phase occurs via animal-to-animal and/or animal-to contaminated surfacesto animal routes; the latter can mediate between-batches cross-contamination (Collis et al., 2004). An additional meat safety concern is that naturally-occurring pathogens, such as Salmonella or E. coli O157, can persist on related surfaces even after routine sanitation (Swanenburg, Urlings, Keuzenkamp, & Snijders, 2001; Small et al., 2003; Tutenel, Pierard, Van Hoof, & De Zutter, 2003). The main factors contributing to the increasing occurrence of pathogens in animals during TL phase include mixing of animals of different origin, stress, extended TL duration, and dirtiness of transport vehicles and lairage pens.

Understandably, pathogen controls in the TL phase are focused on effective measures to prevent/minimize these contributing factors.

3.3.2. Harvest phase

3.3.2.1. Occurrence of foodborne pathogens inlon raw meats. Recently reported data on the occurrence of the main microbial foodborne pathogens in raw meats in the EU are indicated in Table 6.

With Campylobacter, it is important to note that its reported occurrence in red meats (up to few percents) was drastically lower than in raw broiler meat (up to 66%) although the occurrence in red meat animals - particularly pigs (up to 85% herds) – was not that much dissimilar to poultry (up to 85% flocks) (Table 5). The reasons for that discrepancy are not entirely clarified, although they probably include comparably less faecal contamination occurring in red meat abattoir operations and more extensive dying-off of the pathogen on drier surfaces of red meat carcasses. However, in case of Salmonella, no such discrepancy was observed. Although pork was the most frequently contaminated among red meats, Salmonella occurrence in both pigs and pork was lower than the occurrence in broilers and broiler meat. The red meat most frequently contaminated with VTEC (including O157) was beef, which is in accordance with the highest VTEC occurrence in cattle (Table 5). Reported data on occurrence of L. monocytogenes in raw read meats was scarce, because monitoring of this pathogen is focused primarily in ready-to-eat (RTE) foods including RTE meats. With Yersinia, pork was more frequently contaminated than beef, although the ranges of overall Yersinia incidence in pigs and cattle appear comparable (Table 5). It is important to note that the occurrence

Table 6

Occurrences	of	micro	obial	foc	odborne	pathogens	in	raw	meats	in	the	EU
(adapted from	m]	EFSA	, 200	6a,	2006b)							

Pathogen (Year)	Reported occurrence (%) in raw meat					
	Beef	Pork	Sheep/goat	Poultry		
Campylobacter						
2005	0-2.1	0-0.5	DNA/I	3.1-66.4		
2004	0.8 - 2.9	1.1 - 5.0	DNA/I	2.2-77.0		
Salmonella spp.						
2005	0–8.3 ^a	0–18 ^a	DNA/I	3.9-18.5		
2004	$0.3 - 7.2^{a}$	1.2–12.7 ^a	DNA/I	3.9-18.5		
VTEC						
2005	$1-7.1^{a}$	0-6.2	0 (FD)	DNA/I		
2004	$1 - 38.2^{a}$	0.7-5.4	0.8% (FD)	DNA/I		
Listeria						
2004	DNA/I	DNA/I	DNA/I	DNA/I		
2005	DNA/I	DNA/I	DNA/I	DNA/I		
Yersinia						
2005	0-4.4	0-16.7	DNA/I	DNA/I		
2004	3.2–7	1.6-10.4	DNA/I	DNA/I		

FD, Few data; DNA/I, Data not available or insufficient. ^a Including minced meat. of human pathogenic *Yersinia* serotypes is more relevant than the overall *Yersinia* occurrence, and that pigs and pork are considered as the primary source of those serotypes.

When analyzing the EU foodborne pathogens' monitoring data in raw meats, it is very difficult to interpret, and compare, differences in reported occurrences of pathogens even between same pathogen-same meat species combinations. The reasons include, among numerous others, lack of specifying how the sampling points were related to the stages of the production processes. It is known that the occurrences of any microbial pathogen in raw meat production can significantly vary both between individual operators and between the main manufacturing stages of the same operator, as illustrated via the example of *E. coli* O157 in raw beef production (Table 7). Obviously, this aspect should be taken into account when designing related monitoring plans.

3.3.2.2. Factors affecting carcass meat contamination and related controls. The general mechanisms of meat contamination in abattoir operations are the same with all enteric pathogens and all meat animal species. The primary sources of microbial contamination of both the slaughterline environment and the carcass meat are the hair/skin/ feathers of animals, the alimentary tract (i.e. gut content; faeces), the nasopharyngeal cavities and the external portion of the urogenital tract. To reduce slaughterline contamination from incoming animals, the so-called "logistic" slaughter approach – slaughtering pathogen-free animals before pathogen-carrying animals - can be used (Swanenburg, van der Wolf, Urlings, Snijders, & van Knapen, 2001). Once the slaughterline environment becomes contaminated, "secondary" sources of carcass contamination include aerosols, the contaminated surfaces and equipment/tools on the slaughterline, in the chiller and in the boning area. In addition, meat handlers including meat inspectors may serve as the contamination source.

Both the relative relevance of individual sources for, and the overall extent of, microbial contamination of raw meat, are highly dependent on the technology and the level of the abattoir process hygiene. Therefore, to ensure raw meat safety, the approach of choice is to ensure adequate process hygiene. Understandably, fundamental differences exist

Table 7

A	summary	of	published	occur	rences	of	Е.	coli	015	7	in	raw	beef
pro	oduction (a	dap	ted from	Buncic	et al.,	2004	4; A	very	and	Bu	inci	ic, 20	05)

Stage of the production process	Median occurrence % (range)
Hides at skinning	23.6 (0-56.0)
Faeces at evisceration	9.5 (0-27.8)
Carcass on slaughterline (not decontaminated)	8.9 (1.1–43.4)
Final carcass (decontaminated and/or chilled)	2.5 (0-3.2)
Raw cut beef	3.8 (0-36.0)

between abattoirs' technologies used for different red meat animal species (cattle, sheep, pigs, poultry). Furthermore, variations in technology and the process hygiene can be marked even between abattoirs slaughtering the same species. Therefore, mandatory process hygiene management system, based on Hazard Analysis and Critical Control Points (HACCP) plans, has to be tailored for each abattoir individually. Because it is not possible to analyze in-depth between-abattoir differences in this paper, only typical steps that are most important for both meat contamination and its control (so-called Critical Control Points; CCPs) for cattle/sheep abattoirs are mentioned here (Table 8).

Generic CCPs in cattle/sheep abattoirs are similar, whilst in pig and poultry abattoirs they include some others, such as scalding. In cutting plants and re-packaging centres, generic CCPs include receipt of meat, pre-cut inspection, chill storage and dispatch-transport (if under the operator's control).

For the verification of the effectiveness of HACCPbased system in abattoirs, microbiological testing of carcasses is commonly used. This is usually done by determining whether counts of general hygiene indicator organisms on carcasses are within given acceptable ranges. The indicator organisms include "aerobic colony count" and Enterobacteriaceae count in the EU (Anonymous, 2005a), or E. coli count in the USA (Anonymous, 2002). In addition, carcasses are tested for presence of Salmonella to determine whether its prevalence exceeds given acceptable values, for example ≤ 2 positives/50 carcasses of cattle and $\leq 5/50$ of pigs in the EU (Anonymous, 2005a), or $\leq 1/82$ of steers/heifers and $\leq 2/58$ of cows/bulls in the USA (Anonymous, 2002). Should the results of these microbiological tests be unsatisfactory as a trend, the process hygiene is considered not to be under effective control so the meat safety risks are unacceptably high. Such a

situation requires thorough review/revision of the HACCP-based system at the abattoir.

However, it should be kept in mind that, even in best abattoirs, total prevention of microbial contamination of all carcasses – hence total absence of microbial foodborne pathogens – is unachievable under commercial conditions.

3.3.2.3. Factors affecting cut raw meat contamination and related controls. After chilling, carcasses are cut into different parts. Meat cutting and deboning operations involve relatively intensive manipulation and handling of meat which markedly increases the microbial risks due to: (a) microbial cross-contamination via hands and utensils (knives, saws, conveyers, etc.); and (b) transfer of bacteria from the meat surface to the internal parts. Fresh meat can be ground and sold as such. Although minced meats are commonly cooked before consumption, they may be eaten raw in some cultures. Furthermore, minced meats can be used for meat preparations containing additives (salts, spices), e.g. in case of hamburgers/meat patties intended to be cooked before consumption.

Ensuring the microbial safety of raw non-carcass meats is, similar to carcasses, based on application of proper process hygiene. It is managed through a HACCP-based system. In the EU, verification of the hygienic functioning of the manufacturing process for minced meat/meat preparations is done through their microbiological testing to determine whether process hygiene indicator organisms ("aerobic colony count" and/or *E. coli* count) are within given acceptable ranges (Anonymous, 2005a). On the other hand, raw minced meat/meat preparations placed on the market must meet the applicable EU food safety criteria: absence of *Salmonella* in 10 and 25 g if intended to be eaten cooked or raw, respectively (Anonymous, 2005a).

Table 8

Examples of generic CCPs in HACCP for cattle/sheep abattoir operations (adapted from Buncic, 2006)

1 0		1 , ,	
Critical control points (CCPs) ^a	Critical limits	Monitoring	Corrective actions
(A) Acceptance of animals	Defined cleanliness score	Visual, every animal	Rejection; cleaning
(B) Hide decontamination (example: with a sanitizer)	Defined sanitizerConcentration, contact duration, temperature.	Defined regime of checking the sanitizer for the critical limits	Re-processing carcasses
(A) De-hiding	 (a) No visible contamination; or (b) % contamin. rate (c) Sterilisers 82 °C 	(a) Visual, every carcass;(b) Computerised push-button	Trimming; retraining; fixing/ replacing equipment
(A) Evisceration	Same as de-hiding	Same as de-hiding	Same as de-hiding
(A) Splitting (spinal cord removal)	No residual tissue	Visual, every carcass	Same as de-hiding
(B) Carcass decontamination (example: with hot water)	75–85 °C 0–15 Pa 5–12 s	Water temperature, continuous	Re-processing carcasses
(A) Chilling	\geq 7 °C, air humidity, velocity, spacing	(a) Instrumental; (b) Visual	Reject; retraining; fixing/

^a So-called "HACCP with interventions" system typically used in USA includes both A and B types of CCPs; so-called "HACCP without interventions" system typically used in the EU includes only A types of CCPs.

3.3.3. Post-harvest phase

3.3.3.1. Occurrence of foodborne pathogens on processed and readv-to-eat (RTE) meats: Example of Listeria monocvtogenes. Cases of human listeriosis are most often caused by RTE food products containing high numbers of L. monocytogenes. Most frequently, the processing environment is a source of contamination of RTE food products with L. monocytogenes. The prevalence of L. monocytogenes in RTE food products in the EU, based on almost 50,000 samples tested, is shown in Table 9. The pathogen was found in 2.7% of the RTE meat products. Comparably, the prevalence of L. monocytogenes in RTE fishery products was higher (7.5%) whilst in cheeses, it was lower (0.6%). It is generally accepted that RTE foods containing >100 cfu L. monocytogenes per gram pose a much higher risk compared with RTE foods containing lower numbers. Amongst the RTE meats indicated in Table 9, up to 3.1%were contaminated with L. monocytogenes numbers above the $100 \, \text{cfu/g}$ value.

3.3.3.2. Factors affecting occurrence in processed/RTE meats and related controls. Further processing of meat. Generally, meat processing techniques can involve various treatments including salting and/or curing based on the addition of salt (sodium chloride) alone or together with other additives (e.g. sodium nitrite, potassium nitrate or their combination), smoking, drying, fermentation, and/or heat treatment. These treatments can be used in various combinations to produce a very large number of different types of meat products in different countries; due to their large numbers, it is not possible to consider various meat products individually in this paper. From the microbial meat safety perspective, processed meat products could be divided into following global groups: (a) products that received a bactericidal step, i.e. in which pathogens are expected to be eliminated (mainly cooked); (b) products that did not receive a bactericidal step and in which pathogens can survive but cannot multiply under expected storage conditions; and (c) products that did not receive a bactericidal step and in which pathogens can survive and can multiply under expected storage conditions.

Meats at retail level. At the retail level, meats and meat products are further extensively handled, including slicing

Table 9

Occurrence of *Listeria monocytogenes* in RTE-products in EU in 2005 (EFSA, 2006b)

Ready-to-eat foods (RTE)		Occurrence (%)
RTE fruits and vegetables		0.8
RTE fishery products		7.5
RTE dairy products	Cheeses	0.6
	Other than cheeses	0.8
RTE meat products ^a	Overall	2.7
-	Bovine	0.7-5.3
	Porcine	0-26.5
	Poultry	0-3.1

^a From 0% to 3.1% of the products contained > 100 cfu *L. monocytogenes*/g.

into individual parts (e.g. ham, sausages, pâtés) and packaging, both of which can lead to cross-contamination. Based on an EU project, the retail-level issues have been recently summarized in the form of brief guidelines for the operators (Bolton & Maunsell, 2004b).

Meats at catering-consumer level. Food safety problems associated with microbial foodborne pathogens in meats at catering and consumer levels are similar; they relate to final preparation of food for consumption. Based on an EU project, the catering-level issues have been recently summarized in the form of brief guidelines for the operators (Bolton & Maunsell, 2004a). At consumer level, epidemiological data from Europe (Tirado & Schmidt, 2000), North America, Australia, and New Zealand indicate that substantial proportions of foodborne disease can be attributed to food preparation practices used in the domestic environment. The main risk factors include:

- (a) cross-contamination from raw to cooked foods via refrigerators; contaminated hands, cutting boards and kitchen towels;
- (b) inadequate refrigeration;
- (c) improper cooking; and
- (d) inadequate post-cooking handling including slow cooling and/or re-contamination.

However, quantitative contributions of these factors, or their combinations, specifically to red meat-borne infections have yet to be determined. A more general overview of factors contributing to red-meat-borne outbreaks in England and Wales (Smerdon, Adak, O'Brien, Gilespie, & Reacher, 2001) indicated that inappropriate storage was implicated in 32%, inadequate heat treatment in 26% and cross-contamination (most commonly, raw-to-cooked) in 25% of those.

3.4. The events leading to a foodborne outbreak: Example of a VTEC outbreak from fermented sausage

In early 2006, a foodborne outbreak caused by E. coli O103:H25 occurred in Norway. The outbreak affected 18 people, mostly young children, 10 of whom developed haemolytic uraemic syndrome (HUS), and one of them died (Anonymous, 2006). Based on the results of a case-control study on 6 cases and 18 controls, the National Institute of Public Health concluded that minced meat of a specific brand was the most likely cause of the outbreak (Schimmer et al., 2006). Further investigations showed that the outbreak was not caused from minced meat but from a raw, cured sausage product from producer A (Schimmer, 2006). Several lots of the incriminated sausages were sampled and E. coli O103 was isolated from unopened packages of three cured sausage products produced in the same facility. The product's isolates and the patients' isolates had identical multi-locus variable number of tandem repeats analysis (MLVA) profiles. The E. coli O103 outbreak strain was also isolated from a batch of mutton at

a production plant of the producer A. This type of mutton was one of the ingredients in the incriminated sausages. This case-story is not unique either for this outbreak or for the *E. coli* O103 serotype, as outbreaks of *E. coli* O157:H7 infections from raw, cured sausage have been reported earlier (Williams et al., 2000). These reinforce the questions of critical points for both the contamination and its control in the production of such meat products.

The raw materials for the cold-smoked fermented mutton sausages from the Norwegian outbreak included sheep meat, pig meat and fat, as well as blood from cattle. The raw materials were minced at temperatures between -4 and -6 °C. A lactic acid bacteria starter culture was added to speed up the fermentation, and carbohydrates and blood were added to enhance the growth/activity of the starter. The sausage batter was stuffed into casings and subjected to fermentation for a week. The fermentation period included a two-days cold-smoking period starting at around 20 °C and finishing at 16 °C. In the product, a pH drop to between 4 and 4.5 normally occurs within the first 2–3 days of fermentation. Following the fermentation, the sausages were subjected to drying/ripening for 2 weeks at 14-15 °C and at 75% relative air humidity. At the end of production process, normally, the water activity (a_w) is around 0.82 and the product contains around 5-6% salt.

The production process is characterized by not including any bactericidal step (e.g. heat-treatment) efficient in killing pathogenic microorganisms. The main intrinsic antimicrobial factors in the final product (pH 4–4.5 and a_w 0.82) are considered to be sufficient to prevent growth of pathogens such as VTEC. The production records from the producer A, from the time of production of the incriminated sausages, did not show any failures and the main process parameters (time-temperature, pH and a_w) did not differ from the values recorded during other periods. Generally, the pH and the $a_{\rm w}$ in the incriminated sausages are even lower (so presumably safer) than what is considered normal for several other salami types. Nevertheless, it is generally considered that intrinsic factors typical for most fermented sausages can prevent the growth of the main pathogens, but their ability to inactivate the pathogens if initially present in the batter – is limited. The latter is of particular relevance if the pathogens have low "infectious dose", such as VTEC has. Overall, the Norwegian outbreak indicated that the production process was not able to inactive the VTEC that were present in the raw meat. Whether this outbreak E. coli O103:H25 strain has particular characteristics such as higher resistance to the intrinsic factors acting in the sausage, and/or possesses particular virulence genes, and/or was present in higher initial counts, remains to be found. Although no particular single causative factor in the outbreak could be identified, an inquiry pointed out that the slaughter hygiene at the slaughterhouses supplying producer A with raw meat for sausage production was inadequate (Anonymous, 2006).

The lesson to be learnt from the outbreak is that hygienic slaughter of animals and microbiological quality of the raw ingredients is of the utmost importance especially when used for production of raw meat products including uncooked, fermented sausages. Following the outbreak, raw meat used for production of the sausages by producer A is tested for *E. coli*, and if numbers exceed 10 cfu/g the meat is not used in the production of these cured meat products. In addition, both the raw sausage batter and the final products are tested for *E. coli* O103 and *E. coli* O157 and they are discarded if these organisms are found.

To prevent foodborne diseases from salami and other similar raw meat products, better understanding of the ability of individual processes to inactivate/reduce identified hazards like VTEC, as well as further quantitative information on the inactivation mechanisms involved, are necessary. Understandably, appropriate requirements for the microbial quality of the raw material also must be established for such production processes.

4. Concluding remarks

The two most frequently reported zoonotic diseases in humans in the EU in 2005 were *Campylobacter* and *Salmonella* infections. Comparably fewer incidences of human infections caused by *Yersinia* spp., Verotoxigenic *E. coli*, and *L. monocytogenes* were reported, but infections with these organisms, especially *L. monocytogenes* and VTEC, may be much more severe in certain risk groups. Meat and meat products are important sources for these infections but knowledge comparing the importance of meats with other sources – food, drinking water and environmental exposure – is limited. Occurrences of zoonotic pathogens in raw meat are variable, although most often are between 1% and 10%, depending on the organisms, geographical factors, farming and/or meat production practices.

Zoonotic pathogens in meat have to be controlled through a complete, continuous farm-to-fork system. It is of utmost importance to control direct and indirect faecal contamination of carcasses through efficient HACCPbased process hygiene management systems.

There are many routes by which the zoonotic pathogens can reach consumers via meats including consumption of contaminated, uncooked or improperly cooked RTE product and cross-contamination from raw to RTE foods. Better knowledge on the relative importance of these different routes is needed. For that, both epidemiological and microbiological approaches as well as risk assessments of specific pathogens in specific foods need to be applied. Such knowledge is important to tailor and optimise the risk management strategies and activities.

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