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An Efficient Sampling Technique Used To Detect Four Foodborne Pathogens on Pork and Beef Carcasses in Nine Belgian Abattoirs

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ABSTRACT

The method presented in this paper should prove useful in assessing the effectiveness of HACCP plans developed in slaughterhouses. Samples were collected by swabbing well-defined areas of pork and beef carcasses with sterile gauze. Between 160 and 420 half-carcasses were swabbed in each of nine pork or beef slaughterhouses. Swabs from five carcasses were placed in the same sterile Stomacher bag, constituting a single composite sample. Standard or validated analytical methods were used to isolate and characterize four foodbome pathogens. *Salmonella* spp., *Listeria monocytogenes, Campylobacter* spp., and verocytotoxin-producing *E. coli* were detected, respectively, in 27, 2, 2, and 14% of the pork samples and 0, 22. 10, and 5% of the beef samples. Of the 10 samples positive for *E. coli* O157, only one yielded an isolate confirmed to be entcrohemorrhagic. Since *Salmonella* spp. appear as the main contaminant of pork (27%) and *L. monocytogenes* as the main contaminant of beef (22%), any slaughterhouse sampling plan should include testing for the former in the case of pork carcasses and for the latter in the case of beef carcasses. One should also test regularly for the presence of *E. coli* O157 and *Campylobacter* spp. in pork and beef abattoirs. The method presented here is an easy way to assess the contamination rate of carcasses at the end of the slaughtering process.

Contamination of carcass surfaces by foodbome pathogens *(Salmonella* spp., *Listeria monocytogenes, Campylobacter* spp., and verocytotoxin-producing *Escherichia coli* O157) is a major public health problem. Foodborne illness, after effects, and deaths have been linked to ground beef. Many outbreaks have occurred because of these microorganisms. Bean and Griffin *(6)* reported that in the United States *Salmonella* spp. account for 48% of all beef-related outbreaks. *E. coli* O157:H7, furthermore, has been increasingly implicated in foodborne diseases, many outbreaks having occurred in the United States, the United Kingdom, Canada, and Japan. These outbreaks have been linked to the consumption of beef products or products cross-contaminated by bovine feces *(33, 20, 7, 16, 40, 39). L. monocytogenes* is also sporadically responsible for human cases of foodbome disease, but no epidemiological link has been established between the consumption of fresh meat and human listeriosis *(8, 39).* Campylobacteriosis is a major public health concern for infants and young adults during summertime *(12, 11).* It is the major cause of foodbome disease in the United States and Western Europe. Few campylobacteriosis outbreaks have been linked to the consumption of fresh red meat, but processed meat products have caused several recent outbreaks due to *L monocytogenes* or *Campylobacter* spp. These meat products were probably contaminated by the environment by ingredients, or through primary contamination of the meat.

Sources of these microorganisms on carcasses are multiple. A healthy animal may harbor pathogenic bacteria on its hide, hair, and hooves, in its intestinal tract, and around the lymph nodes *(5, 18, 29, 19, 14).* For beef, the primary source is the hair and hide, which contaminate the carcass during removal *(14).* These parts of the animals are exposed to dust, dirt, and feces *(5, 14).* Diseased animals may also reach the stunning step of the slaughtering process. Generally, the internal surfaces of the carcasses are sterile, and transfer of bacteria results from dressing and skinning defects occurring during the slaughtering process *(14).* Food handlers are a common source of pathogens. Workers in the food industry may indeed harbor and excrete (as healthy carriers) *Salmonella* spp., *Campylobacter* spp., enteropathogenic strains of *E. coli,* and *L. monocytogenes (13, 9, 21).*

In order to manage and control the microbiological hazards associated with meat products, it is essential to reduce, via a hazard analysis critical control point (HACCP) plan, any surface contamination occurring at the slaughtering step. In the European Community, Directive 93/43/CEE proposes the implementation of HACCP principles in the food industry. The World Congress on Meat and Poultry Inspection has proposed a program to lower contamination during animal production (preharvest pathogen reduction).

It is therefore very important to find a system for monitoring the surface contamination of carcasses in order to assess good manufacturing and hygiene practices and also assess HACCP plans. To achieve this, operators need an easy method for assessing the microbiological quality of carcass surfaces so as to supervise the process and detect deviations at the critical control points. These checks must be done in the field by operators in the food industry and by inspection services. They can also provide an objective basis for comparing the surface contamination of carcasses in different slaughterhouses.

The severity of illnesses caused by enterohemorrhagic *E. coli* O157, furthermore, makes it necessary to determine the origin of such contamination. The Belgian Sanitel system makes it possible to follow any bovine from birth to the slaughterhouse. Samples must thus be identified with the carcass number and, in the case of *E. coli* O157, sampling should be done on the rearing or fattening farm.

Different methods have been proposed for monitoring the microbiological quality of meat products *(25),* but they are intended for assessing the microbiological contamination of carcasses at reception level, in butcher's shops, or in the meat product industry. Few methods have been proposed for evaluating contamination at the slaughterhouse before the boning and cutting steps.

The aims of our study were (1) to establish a reliable and easy method for monitoring the microbiological quality of carcass surfaces at the end of the slaughtering process, before boning and cutting, and (2) to assess the prevalence of carcass surface contamination in nine Belgian slaughterhouses.

MATERIALS AND METHODS

Bacteriological sampling. Five beef slaughterhouses and four pork slaughterhouses distributed throughout Belgium and representative of Belgian carcass production were selected for this study. From January to March 1996 between 160 and 420 half carcasses were swabbed per slaughterhouse (40 to 120 per contaminant sought) (Table 1). The same operator collected all samples in all slaughterhouses. In each abattoir sampling was done in one morning or one afternoon on the day the animals were slaughtered, just after the chilling chock.

Four zones were delineated on the half carcasses. Figures 1 and 2 depict these zones, totaling in area about 1.200 cm^2 per half pork carcass and 1,500 cm^2 per half beef carcass. These zones were swabbed with sterile gauze, with use of a sterile glove. The gauze was wiped firmly over the sampling areas. The gauzes from five carcasses were placed in a same sterile Stomacher bag, constituting a single composite sample. This increased the total swabbed area per sample to 6,000 cm^2 for pork carcasses and 7,500 cm^2 for beef carcasses. The identification numbers of the carcasses were recorded. The samples were placed immediately in a refrigerated box and transferred in the box to the food laboratory where they were processed the same day.

TABLE 1. *Numbers of half carcasses sampled for each pathogen*

	Number of half carcasses swabbed				
	Total samples per abattoir	*Salmonella* spp.	*L monocytogenes*	*Campylobacter* spp.	*E. coli* O157
Pork carcasses					
Slaughterhouse 1	320	80	80	80	80
Slaughterhouse 2	240	60	60	60	60
Slaughterhouse 3	240	60	60	60	60
Slaughterhouse 4	180	45	45	45	45
Total	980	245	245	245	245
Beef carcasses					
Slaughterhouse 5	420	120	60	120	120
Slaughterhouse 6	240	60	60	60	60
Slaughterhouse 7	180	45	45	45	45
Slaughterhouse 8	160	40	40	40	40
Slaughterhouse 9	175	45	40	45	45
Total	1,180	310	250	310	310

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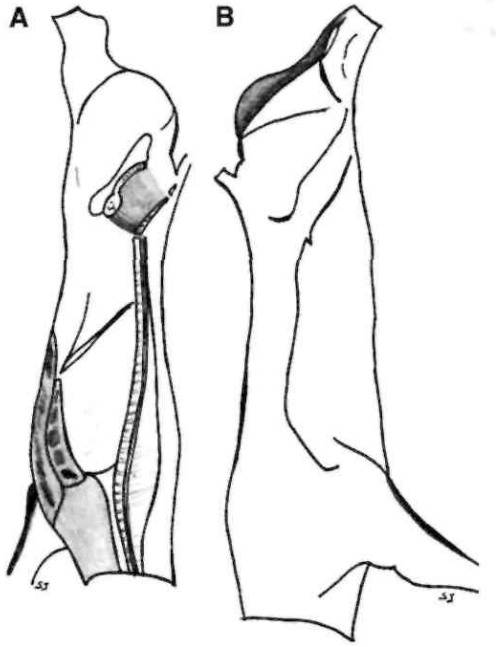
FIGURE 1. *Sampled areas on beef carcasses (in black): internal (a) and external (b).*



Microbiological analyses. French standard analytical methods were used for *Salmonella* spp. (NF V 08-052 *(2)* and *L monocytogenes* (NF V 08-055) *(3).* The ISO method 10272 was used for *Campylobacter* spp. *(4)*.

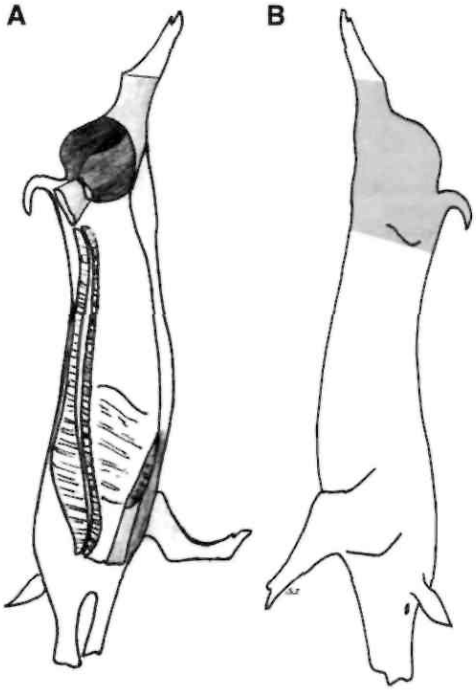
For detection of *Salmonella* spp., 120 ml of buffered peptone water (BPW; Oxoid CM 509 B, Unipath Ltd., Basingstoke, UK) was added to the five gauzes in the Stomacher bag. After mixing for 1 min, this preenrichment suspension was incubated at 37°C for 16 to 20 h. Selective enrichment was done by transferring 0.1 ml of the BPW suspension to 10 ml of Rappaport-Vassiliadis (RV) broth (Oxoid CM 669 B) and 2 ml to 20 ml of selenite cystine (SC) broth (Oxoid CM 699 B) supplemented with sodium biselenite (Oxoid LP 121 A). The RV suspension was incubated at 42°C for 18 to 24 h and the SC suspension at 37°C for 18 to 24 h. Isolation and identification were performed by plating on xylose lysine desoxy-cholate (XLD) agar (Oxoid CM 469 B) and modified brillant green agar (BGAM; Oxoid CM 329 B), which were incubated for 18 to 24 h at 37°C. One suspected colony on each medium was transferred onto triple sugar iron (TSI) agar (Oxoid CM 277 B) for identification. Several confirmation tests were also performed: catalase, cytochrome oxidase, multivalent serum (OMA-OMB) agglutination, and API 20E (BioMérieux. Marcy l'Etoile, France).

For isolation of *L monocytogenes,* 120 ml of half Fraser broth (obtained by adding 8.88 ml of Half Fraser Selective Supplement [Oxoid FR 166 G] to 1 liter of Fraser Base Broth [Oxoid CM 895 B]) was added to the five gauzes in the Stomacher bag (primary enrichment) and the bag contents were mixed for 1 min. After incubation at 30°C for 18 to 24 h, 0.1 ml was transferred to 10 ml Fraser broth (obtained by adding 10 ml of Fraser Selective Supplement [Oxoid FR 156 E] to 1 liter of Fraser Base Broth [Oxoid CM 895 B] for secondary enrichment. The resulting suspension was incubated at 37°C. Isolation was done by plating on Oxford agar (Oxoid CM 856 B) from the primary enrichment broth after a 24-h incubation and from the secondary enrichment broth after incubation for 24 and 48 h. Two confirmation tests were performed on five suspected colonies: catalase and CAMP tests (on Trypticase soy agar with 5% sheep blood [Becton Dickinson, 38240 Meylan, France; 4354053]). Hemolytic colonies were confirmed as *L monocytogenes* with API Listeria (BioMérieux).

To detect *Campylobacter* spp., 120 ml of Preston broth (Nutrient Broth no. 2 [Oxoid CM 67 B] with three supplements: Campylobacter Selective Supplement [Oxoid SR 117 E], Laked Horse Blood [Oxoid SR 48 C], and Campylobacter Growth Supplement [Oxoid SR 84 E]) was added to the five gauzes in the Stomacher bag. After mixing for 1 min, the bags were placed for 18 to 24 h at 42°C in an anaerobic jar and the normal atmosphere was replaced with a microaerobic atmosphere of 5% 0_2, 10% $C0_2$, and 85% N_2 by volume. After incubation, surface plating was performed on Skirrow agar (Blood Agar Base no. 2 [Oxoid CM 271 B] with two supplements: Campylobacter Selective Supplement [Oxoid SR 69 E] and laked horse blood) and Karmali agar

(Campylobacter Agar Base [Oxoid CM 935 B] supplemented with Campylobacter Selective Supplement [Oxoid SR 167 E]). The two media were incubated at 42°C in a microaerobic jar (Campy Gen, [Oxoid CN 35] containing 5% 0_2, 10% $C0_2$, and 85% N_2). The media were observed after 48 h, 72 h, and 5 days, respectively. On one suspected colony obtained on isolation medium, a Gram staining was performed, and the morphology and color were assessed. Confirmation of the suspected colony was done by plating on Trypticase soy agar with 5% sheep blood and incubating at 42°C under microaerobic conditions for 24 to 48 h. Several confirmation tests were performed: catalase, cytochrome oxidase, and API Campy (BioMérieux). Verocytotoxin-producing *E. coli* (serotype O157) was identified by the VIDAS ECO method (BioMérieux). After enrichment for 18 h, colonies were isolated on sorbitol MacConkey agar or with the help of the *E. coli* Petrifiim kit (Laboratories 3M Santé, Malakoff, France). Biochemical characteristics were demonstrated on API 20E (BioMérieux), sorbitol MacConkey agar (Unipath), and Rapid E. coli medium (Sanofi Diagnostic Pasteur, Paris, France). Virulence characteristics were assayed by the Belgian Reference Laboratory (Denis Pierard, AZ-VUB. Belgium). PCR was used to detect the *eae* gene and to determine verocytotoxin type.

FIGURE 2. *Sampled areas on pork carcasses (in black): internal (a) and external (b).*



RESULTS

Swabs from five half carcasses constituting each sample, 49 pork samples and 62 beef samples were tested for the presence of *Salmonella* spp., *Campylobacter* spp., and *E. coli* O157. Forty-nine pork and 50 beef samples were examined for the presence of *L. monocytogenes.*

All pork slaughterhouses combined, the average percentage of pork samples positive for *Salmonella* spp. was 27% (Table 2), with little variability between slaughterhouses (range: 19 to 33%). No *Salmonella* sp. was isolated from any of the beef samples by the standard analytical methods used (Table 2).

In two pork slaughterhouses, no *Listeria* sp. contamination was detected (Table 3). All relevant slaughterhouses combined. *Listeria* sp. contamination was detected in 10% of the pork samples and 54% of the beef samples. In slaughterhouses 4, 8, and 9, no *L monocytogenes* was isolated, and beef slaughterhouses varied considerably as to the proportion of samples found to be contaminated by this pathogen (8 to 75%). In abattoir 6, all isolated *Listeria* spp. were confirmed as *L. monocytogenes.* The *Listeria* contamination rate in our samples may be underestimated, because the method used (NF V 08-055) revealed numerous contaminating florae (especially *Micrococcus* spp.) on Oxford agar.

No *Campylobacter* was isolated from the swabs collected in five slaughterhouses (three pork and two beef slaughterhouses; Table 4). On the average, *Campylobacter*-contaminated samples represented 2% of the pork samples and 10% of the beef samples (Table 4).

As regards *E. coli* O157, 14% of the pork samples (7 samples) and 5% of the beef samples (3 samples) tested positive with the VIDAS kit (Table 5). Only 3 of these 10 positive samples yielded isolates. Two isolates were obtained from pork samples and one from a beef sample. All three isolates were nonmotile. They were characterized by the Belgian Reference Laboratory. Only the beef sample isolate harbored the *eae* gene, produced human-pathogenic verocytotoxins, was enterohemolysin-positive and β-glucuroni-dase-negative, and failed to ferment sorbitol (Table 5).

The average sample contamination rates for all four foodborne pathogens are presented together in Table 6. On the surface of pork carcasses the main contaminant is *Salmonella* spp. (27%); on beef carcasses it is *L.monocytogenes* (22%).

DISCUSSION

To get as total a picture as possible of carcass contamination, several areas were swabbed on each carcass. Roberts et al. *(34)* recommended sampling at three or four sites on carcasses, because contamination appears to vary considerably between different sites, the brisket exhibiting a particularly high contamination level. Since each of our samples comprised swabs from five half carcasses, the contamination rates obtained in this work are only approximate estimates of the prevalence of carcass contamination by the organisms sought (since one positive sample may represent between one and five contaminated half carcasses). The advantage of this approach was to keep down the cost of the microbiological analyses while ensuring a large total swabbed area per sample. The method appears reproducible. Variability due to sampling by different workers *(34)* was avoided by having the same operator swab the carcasses in all slaughterhouses.

In two separate studies of beef carcasses preinoculated or not with bovine feces, Dorsa et al. *(15)* compared various sampling techniques in terms of recovery (colony-forming units retrieved per unit area). The swabbing materials compared were cheesecloth, sponge, and cotton-tipped wooden swabs in the study of uninoculated carcasses and cheesecloth, griddle screen, and 3M mesh in the study of preinoculated ones. In both studies excision gave the best results, but swabbing with the most abrasive materials (cheesecloth for uninoculated carcasses, griddle screen and 3M mesh for artificially contaminated ones) approached the effectiveness of excision. Dorsa et al. premoistened their cheesecloth and sponge swabs just before sampling, while to make sampling easier the gauze swabs used here were not premoistened. To compensate for the possibility that pre-moistening might improve recovery, a larger area was swabbed here than in the Dorsa et al. study.

Anderson et al. *(1)* reported a swabbing-to-excision recovery ratio ranging from 6 to 16% for nonwashed carcasses and from 1 to 22% for washed, sanitized carcasses. Nortje et al. *(30)* reported a correlation coefficient of 0.49 between recovery after swabbing and recovery after excision.

Thus swabbing clearly is less effective than excision, but results depend on the swabbing technique adopted and can approach those obtained by excision. Excision, furthermore, is an impractical and destructive method not routinely applicable in a commercial slaughterhouse. In the present study, the aim was not to count colony-forming units per unit area but to estimate the proportion of carcasses contaminated with certain pathogens. The proposed method is simple and nondestructive. It allows sampling of a large area per carcass and of many carcasses in a reasonable time. Its results are in keeping with published figures, and it successfully revealed a presumably minor contaminant never detected previously on a beef carcass in Belgium (see below).

***Salmonella* spp.** In this study, contamination of pork samples by *Salmonella* spp. averaged 27%. This figure is in keeping with the prevalence of *Salmonella* sp. contamination of pork carcass surfaces reported by other authors, ranging from 10 to 20% *(17, 27, 37),* especially since our use of composite rather than individual samples probably leads to somewhat overestimating the proportion of contaminated half carcasses. In 1994, Denys (personal communication) obtained a contamination rate of 4.6% on swabs from pork carcasses and 6.4% in minced meat made from pork pieces. The lowest contamination rate observed in the present study (19% of our pork swab samples) was obtained in pork slaughterhouse 1, whose workers split the carcass manually with the use of an axe. The relevance of this fact, however, requires confirmation through further investigation.

L. monocytogenes. The beef slaughterhouses in this study were found to vary considerably as to the degree of contamination by this species. This may reflect the diversity of types of slaughtered animals. Slaughterhouse 6

principally slaughters dairy cows, whose diet consists mainly of silage. One author has emphasized the role of silages (especially poorly made silages) in the development of animal listeriosis or of a carrier state in animal tissues *(24).* Other studies appear to corroborate the difference between pork and beef carcasses as regards this contaminant; Mc-Clain and Lee *(28)* observed a contamination rate of 70% in ground beef, 43% in pork sausages, and 48% in poultry.

Regarding contamination of pork carcasses by *L. monocytogenes,* Buncic *(10)* demonstrated a carrier state in pigs, observing a prevalence of 3% in feces and 45% in tonsils. The same author found *L. monocytogenes* prevalence rates of 29% in swabs from retropharyngeal nodes of cattle, 19% in cattle fecal samples, 19% in raw dry sausages, 21% in vacuum-packaged smoked sausages, and 69% in ground meat (mixed pork and beef). The sausages and ground meat were collected in supermarkets and butcher's shops in Yugoslavia.

The use of the more specific Palcam medium instead of Oxford medium for agar plate isolation might allow detection of more *Listeria*-positive samples.

***Campylobacter* spp.** Our data concerning carcass contamination by *Campylobacter* spp. appear in keeping with other studies. Various authors have found a higher rate of surface contamination on beef and lamb carcasses than on pork carcasses *(36, 26).* This might reflect the more marked surface dehydration of pork carcasses *(23, 31).* Studies have demonstrated that poultry products are the main source of human campylobacteriosis, followed by beef and lamb products. Pork products are thus not a major source of human campylobacteriosis. According to the estimates of various authors, the rate of contamination of red meats at retail level is 0.2 to 5% *(22, 38, 36).* Stern et al. *(35)* have demonstrated that freezing storage has a deleterious effect on *Campylobacter* spp.

***E. coli* O157.** Only three isolates of *E. coli* serotype O157 were isolated from the 10 samples detected by the enzyme immunoassay, perhaps due to the long enrichment period (18 hours) before isolation on agar plates. A shorter enrichment period followed by immunomagnetic separation might improve the isolation rate of pathogenic strains of *E. coli.*

The verocytotoxin-producing *E. coli* isolated in this study is the first isolated from a foodstuff in Belgium to display all the characteristics of a human-pathogenic isolate. Never before in Belgium had a verocytotoxin-producing *E. coli* been isolated from a beef carcass. This should alert the official bodies in charge of monitoring public health and prompt them to set up meat contamination surveillance programs. The risk of hemorrhagic colitis or HUS (hemorrhagic uremic syndrome) exists in Belgium. The cause of infection could be undercooking of bovine meat or the consumption of raw meat. Following isolation of the pathogenic strain by the Belgian Reference Laboratory, a survey was conducted under the supervision of Dr. Dufey (Ministry of Agriculture) at the farm where all five animals contributing to the positive sample were reared. This was possible thanks to the Belgian Sanitel system, through which any bovine can be tracked from birth to the slaughterhouse. Animals are marked with two ear numbers, one at birth and one 30 days later. The Belgian Federations download to computers all changes affecting the animals (herd disease status, marketing, shipping, transport to other farms, transport to the slaughterhouse, etc.). This system makes it possible to know the history of all cattle in Belgium. In the present case the farm of origin was identified the same day the Belgian Reference Laboratory confirmed the virulence characteristics of the isolate. The herd was composed of dairy cows, yielding cows, and fattening bulls. Other animals were also present: goats, ducks, cats, dogs, and poultry. A potentially serious hazard appeared in the fact that the children on the farm usually consumed raw milk produced there. Raw milk, furthermore, was delivered from the farm directly to consumers.

Unfortunately, the VIDAS method revealed no verocytotoxin-producing *E. coli* (after overnight enrichment in BPW broth) in 12 pools of 50 fecal samples collected on the farm of origin only 7 h after confirmation. The incriminated animals might have been cross-contaminated during transport, or the pathogenic strain might have been present on the equipment of the slaughterhouse where it was isolated. The Sanitel system made it possible to rule out any risk of contamination on the farm in a very short time, so that the farm may continue its activity. Further investigation is needed, and the Ministry of Public Health has established a surveillance program to check for the presence of enterohem-orrhagic *E. coli* (EHEC) in minced beef produced and marketed in Belgium. To date, no EHEC strain of the O157 serotype has been isolated in Belgian ground meat (more than 1,000 samples) by immunomagnetic separation with Dynabeads (Dynal A.S., Oslo, Norway), EIA testing (VIDAS Eco, BioMérieux, and EHEK-TEK, Organon Technica, Oslo, Norway), and PCR *(32).*

In conclusion, the sampling method presented here requires only a minimal amount of material (only gloves, Stomacher bags, and sterile gauzes). It allows detection of infrequent contaminants *(E. coli* O157, *Campylobacter* spp.) and yields results in accordance with current contamination levels for *Salmonella* spp. and

L. monocytogenes. Random variation was eliminated in this study by the great number of carcasses swabbed. In high-output slaughterhouses, a sampling program should be designed to choose randomly the carcasses to be swabbed.

Slaughterhouses must aim to ensure lower contamination rates than observed, on the average, here. Inspection departments should establish a systematic surveillance plan of this type. Our method will make it possible to compare different slaughterhouses on an objective basis, to know the current level of carcass contamination, and to monitor its evolution.

Since *Salmonella* spp. appear as the major contaminant on pork carcasses and *L. monocytogenes* on beef carcasses, any slaughterhouse sampling plan should include isolation of the former where pigs are slaughtered and of the latter in beef abattoirs. In both pork and beef slaughterhouses, moreover, it would be worth checking regularly for *Campylobacter* spp. and *E. coli* O157.

TABLE 2. *Surface contamination by* Salmonella *spp.: percentage of samples contaminated*

	Number of half carcasses assayed	Total number of samples	Positive samples (number)	Positive samples (percentage)
Pork carcasses				
Slaughterhouse 1	80	16	3	19%
Slaughterhouse 2	60	12	4	33%
Slaughterhouse 3	60	12	3	25%
Slaughterhouse 4	45	9	3	33%
Total	245	49	13	27%
Beef carcasses				
Slaughterhouse 5	120	24	0	0%
Slaughterhouse 6	60	12	0	0%
Slaughterhouse 7	45	9	0	0%
Slaughterhouse 8	40	8	0	0%
Slaughterhouse 9	45	9	0	0%
Total	310	62	0	0%

TABLE 3. *Surface contamination by L. monocytogenes: percentage of samples contaminated*

	Number of half carcasses swabbed	Total number of samples	Samples positive for *Listeria* spp.	Samples positive for *L. monocytogenes*
Pork carcasses				
Slaughterhouse 1	80	16	2 (12%)	1 (6%)
Slaughterhouse 2	60	12	0	0
Slaughterhouse 3	60	12	0	0
Slaughterhouse 4	45	9	3 (33%)	0
Total	245	49	5 (10%)	1 (2%)
Beef carcasses				
Slaughterhouse 5	60	12	5 (42%)	1 (8%)
Slaughterhouse 6	60	12	9 (75%)	9 (75%)
Slaughterhouse 7	45	9	3 (33%)	1 (11%)
Slaughterhouse 8	40	9	4 (44%)	0
Slaughterhouse 9	40	8	6 (75%)	0
Total	250	50	27 (54%)	11 (22%)

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TABLE 4. *Surface contamination by* Campylobacter *spp.: percentage of samples contaminated*

	Number of half carcasses assayed	Total number of samples	Positive samples (number)	Positive samples (percentage)
Pork carcasses				
Slaughterhouse 1	80	16	1	6%
Slaughterhouse 2	60	12	0	0%
Slaughterhouse 3	60	12	0	0%
Slaughterhouse 4	45	9	0	0%
Total	245	49	1	2%
Beef carcasses				
Slaughterhouse 5	120	24	2	8%
Slaughterhouse 6	60	12	2	17%
Slaughterhouse 7	45	9	0	0%
Slaughterhouse 8	40	8	0	0%
Slaughterhouse 9	45	9	2	22%
Total	310	62	6	10%

TABLE 5. *Surface contamination by* E. coli O*157: percentage of samples contaminate*

	Number of half carcasses assayed	Total number of samples	Number of O157 VIDAS positive samples (%)	Characteristics of identified serotypes
Pork carcasses				
Slaughterhouse 1	80	16	2 (12%)	2 *E. coli* O157:H^-, VT^-,eae^-, β-glu^+, $sorb^+$, $enth^{-a}$
Slaughterhouse 2	60	12	2 (17%)	NI[b]
Slaughterhouse 3	60	12	0	NI
Slaughterhouse 4	45	9	3 (33%)	NI
Total	245	49	7(14%)	
Beef carcasses				
Slaughterhouse 5	120	24	1 (4%)	*E. coli* O157:H^-, $VT1^+$ VT2vh-a^+, eae^+, β-glu^-, $sorb^-$, $enth^{+\,c}$
Slaughterhouse 6	60	12	0	
Slaughterhouse 7	45	9	1 (11%)	NI
Slaughterhouse 8	40	8	0	
Slaughterhouse 9	45	9	1 (11%)	NI
Total	310	62	3 (5%)	

[a] Two strains of *E. coli* O157:H^-, negative for verocytotoxin gene by PCR, negative for *eae* gene by PCR, positive for β-glucuronidase, positive for fermentation of sorbitol, negative for enterohemolysin activity.

[b] No strain isolated.

[c] One strain of *E. coli* O157:H^-, positive for presence of type 1 verocytotoxin gene by PCR, positive for presence of type 2 verocytotoxin gene human variant *a* by PCR, positive for presence of *eae* gene by PCR. negative for β-glucuronidase. negative for fermentation of sorbitol, positive for enterohemolysin activity.

TABLE 6. *Average sample contamination rates for four pork and five beef slaughterhouses*

	Pork slaughterhouses, percentage of samples contaminated	Beef slaughterhouses. percentage of samples contaminated
Salmonella spp.	27%	0%
L. monocytogenes	2%	22%
Campylobacter spp.	2%	10%
E. coli O157[a]	14%	5%

[a] Among the 11 positive samples, only one strain was isolated and confirmed as enterohemorrhagic.

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REFERENCES

1. Anderson. M. E., H. E. Huff. H. D. Naumann. R. T. Marshall. J. Damare. R. Johnston, and M. Pratt. 1987. Evaluation of swab and tissue excision methods for recovering microorganisms from washed and sanitized beef carcasses. J. Food Prot. 50:714-743.

2. Anonymous. 1993. Microbiologie alimentaire—méthode de routine pour la recherche des *Salmonella.* NF V 08-052. AFNOR, France.

3. Anonymous. 1993. Microbiologie alimentaire—recherche de *Listeria monocytogenes*—méthode de routine. NF V 08-055. AFNOR, France.

4. Anonymous. 1996. Microbiologie des aliments—méthode horizontale pour la recherche de *Campylobacter* thermotolérants. NF ISO 10272. V 08-026. AFNOR. France.

5. Ayres, J. C. 1955. Microbiological implications in the handling, slaughtering, and dressing of meat animals. Adv. Food Res. 6:109-161.

6. Bean, N. H.. and P. M. Griffin. 1990. Foodbome disease outbreaks in the United States, 1973-1987: pathogens, vehicles, and trends. J. Food Prot. 53:804-817.

7. Belongia, E. A., K. L. MacDonald, G. L. Parham, K. E. White, J. A. Korlath, M. N. Lobato, S. M. Strand, K. A. Casale, and M. T. Osterholm. 1991. An outbreak of *Escherichia coli* O157:H7 colitis associated with consumption of precooked meat patties. J. Infect. Dis. 164:338-343.

8. Bracken. R. E. 1988. Presence and persistence of *Listeria monocytogenes* in food and water. Food. Technol. 42:162-164.

9. Buchwald. D. S., and M. J. Blaser. 1984. A review of human salmonellosis: ∏. Duration of excretion following infection with nontyphi *Salmonella.* Rev. Infect. Dis. 6:345-356.

10. Buncic, S. 1991. The incidence of *Listeria monocytogenes* in slaughtered animals, in meat, and in meat products in Yugoslavia. Int. J. Food Microbiol. 12:173-180.

11. Butzler, J. P., H. Goosens, and Y. Glupczynski. 1992. *Helicobacter pylori, Campylobacter* and other bacterial infections of the gastrointestinal tract. Current Opinion in Infect. Dis. 5:80-87.

12. Butzler, J. P., and M. B. Skirrow. 1979. Campylobacter enteritis. Baillieres Clin. Gastroenterol. 8:737-765.

13. Cohen, D. I.. T. M. Rouach, and M. Rogol. 1984. *Campylobacter* enteritis outbreak in a military base in Israel. Isr. J. Med. Sci. 20:216-218.

14. Dickson. J. S., and M. E. Anderson. 1992. Microbiological decontamination of food animal carcasses by washing and sanitizing systems: a review. J. Food Prot. 55:133-140.

15. Dorsa. W. J., C. N. Cutter, and G. R. Siragusa. 1996. Evaluation of six sampling methods for recovery of bacteria from beef carcass surfaces. Lett. Appl. Microbiol. 22:39-41.

16. Doyle. M. P. 1991. *Escherichia coli* O157:H7 and its significance in foods. Int. J. Food Microbiol. 12:289-302.

17. Duitschaever. C. L., and C. Buteau. 1979. Incidence of *Salmonella* in pork and poultry products. J. Food Prot. 42:662-663.

18. Gill. C. O., and K. G. Newton. 1978. The ecology of bacterial spoilage of fresh meat at chill temperature. Meat Sci. 2:207-217.

19. Grau. F. H. 1987. Prevention of microbial contamination in the export beef abattoir, p. 221-233. *In* F. J. Smulders (éd.). Elimination of pathogenic organisms from meat and poultry. Elsevier Science Publishing Co.. Amsterdam. The Netherlands.

20. Griffin. P. M., S. M. Ostroff, R. V. Tauxe, K. D. Greene, J. G. Wells, J. H. Lewis, and P. A. Blake. 1988. Illnesses associated with *Escherichia coli* 0157:H7 infections. Ann. Intern. Med. 109:705-712.

21. Griffiths. M. W. 1989. *Listeria monocytogenes:* its importance in the dairy industry. J. Sci. Food. Agric. 47:133-158.

22. Harris. N. V., D. Thompson, D. C. Martin, and C. M. Nolan. 1986. A survey of *Campylobacter* and other bacterial contaminants of premar-ket chicken and retail poultry and meats. King County. Washington. Am. J. Public Health 76:401-406.

23. Hudson. W. R., and T. A. Roberts. 1982. The occurrence of *Campylobacter jejuni* on commercial red meat carcasses from an abattoir, p. 273. *In* D. G. Newell (éd.). *Campylobacter:* epidemiology, pathogenesis, biochemistry. MTP Press. Lancaster, England.

24. ICMSF. 1996. Micro-organisms in foods 5. Microbiological specifications of food pathogens. Blackie Academic and Professional, London. England.

25. Jouve. J.-L. 1996. Viandes et produits carnés, p. 201-335. *In* CNERNA-CNRS (éd.). La qualité microbiologique des aliments: maîtrise et critères. Polytecnica. Paris. France.

26. Lammerding. A. M., M. M. Garcia, E. D. Mann, Y. Robinson, W. J. Dorward, R. B. Truscott,. and F. Tittiger. 1988. Prevalence of *Salmonella* and thermophilic *Campylobacter* in fresh pork, beef, veal and poultry in Canada. J. Food Prot. 51:47-52.

27. Mafu. A. A., R. Higgins, M. Nadeau, and G. Cousineau. 1989. The incidence of *Salmonella. Campylobacter* and *Yersinia enterocolitica* in swine carcasses and the slaughterhouse environment. J. Food Prot. 52:642-645.

28. McClain, D., and W. H. Lee. 1987. Isolation and identification of *Listeria monocytogenes* from meat. *In* USDA. FSIS. Microbiol. Div., Lab. Comm. no. 57 (9 September).

29. McMeekin, T. A. 1982. Microbial spoilage of meats, p. 1-40. *In* R. Davies (éd.). Developments in food microbiology, vol. 1. Applied Science Publishing. London.

30. Nortje. G. L., E. Swanepoel, R. T. Naude, W. H. Holzapfel, and P. L. Sleyn. 1982. Evaluation of three carcass surface microbial sampling techniques. J. Food Prot. 45:1016-1017.

31. Oosterom, J. 1980. Het voorkomen van *Campylobacter fetus* subspecies *jejuni* bij normale slachtvarkens. Tijdschr. Diergeneesk. 105:49-50.

32. Pierard. D., L. Van Damme, D. Stevens, L. Moriau, and S. Lauwers. 1994. Detection of verocytotoxin-producing *Escherichia coli* in meat in Belgium, p. 77-80. *In* M. A. Karmali and A. G. Goglio (éd.). Recent advances in verocytotoxin-producing *Escherichia coli* infections. Elsevier Science Publishing Co.. Amsterdam.

33. Riley. L. W. 1987. The epidemiologic, clinical, and microbiologic features of hemorrhagic colitis. Annu. Rev. Microbiol. 41:383-407.

34. Roberts. T. A., W. R. Hudson, O. P. Whelehan, B. Simonsen. K. Olgaard, H. Labots, J. M. Snijders, J. Van Hoof, J. Debevere, J. F. Dempster, J. Devereux, L. Leistner, H. Gehra, J. Gledel, and J. Fournaud. 1984. Number and distribution of bacteria on some beef carcasses at selected abattoirs in some member states of the European Communities. Meat Sci. 11:191-205.

35. Stern. N. J., S. S. Green, N. Thaker, D. J. Krout, and J. Chiu. 1984. Recovery of *Campylobacter jejuni* from fresh and frozen meat and poultry collected at slaughter. J. Food Prot. 47:372-374.

36. Stem. N. J., M. P. Hernandez, L. Blankenship, K. E. Deibel, S. Doores, M. P. Doyle, H. Ng, M. D. Pierson, J. N. Sofos, W. H. Sveum, and D. C. Westhoff. 1985. Prevalence and distribution *of Campylobacter jejuni* and *Campylobacter coli* in retail meats. J. Food Prot. 48:595-599.

37. Swaminathan. B., M. A. B. Link, and J. C. Ayres. 1978. Incidence of salmonellae in raw meat and poultry samples in retail stores. J. Food Prot. 41:518-520.

38. Turnbull, P. C. B., and P. Rose. 1982. *Campylobacter jejuni* and *Salmonella* in raw red meats. A Public Health Laboratory Service survey. J. Hyg. 88:29-37.

39. U.S. Department of Agriculture. 1995. Pathogen reduction; hazard analysis critical control point (HACCP) systems—generic HACCP for raw beef. U.S. Department of Commerce. Washington, D.C.

40. Wells. J. G., L. D. Shipman, K. D. Greene, E. G. Sowers, J. H. Greene, D. N. Cameron, F. P. Downes, M. L. Martin, P. M. Griffin, S. M. Ostroff, M. E. Potter, R. V. Tauxe, and I. K. Wachmuth. 1991. Isolation of *Escherichia coli* serotype O157:H7 and other shiga-likc-toxin-producing *Escherichia coli* from dairy cattle. J. Clin. Microbiol. 29:985-989.