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Research Note

Microbiological Sampling of Carcasses by Excision or Swabbing with Three Types of Sponge or Gauze

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ABSTRACT

Fifty-five bovine, 50 equine, 60 ovine, and 50 porcine carcasses were sampled in a slaughterhouse in eastern Spain. Two samples were taken from each carcass, one using the excision method and the other using the swabbing method. Four different materials were used for swabbing: cellulose, polyurethane, or viscose sponges, and medical gauze. Samples were collected at the end of the process by four different people before the carcasses were taken to the cooler. The samples were examined for total viable bacteria counts (TVCs) and *Enterobacteriaceae* counts (ECs). The mean TVC for all species sampled by excision was 4.50 log CFU/cm^2, which was significantly higher than the 3.53 log CFU/cm^2 obtained by swabbing. The TVCs obtained using gauze and the cellulose and polyurethane sponges were significantly higher ($P < 0.05$) than the corresponding TVCs obtained using viscose sponges. Animal species, the person who collected the samples, and microbiological load also had a significant effect on TVC. ECs were obtained from 82.8% of excision samples, from larger percentages of samples obtained using cellulose or polyurethane sponges or gauze swabs, but from smaller percentages of samples obtained using viscose sponges. The *Enterobacteriaceae* load significantly influenced the EC. In contrast, animal species and the person who collected the samples had no significant effect. The cellulose sponge, polyurethane sponge, and gauze gave high mean log counts of aerobic bacteria and *Enterobacteriaceae,* which makes these swab types suitable for use in slaughterhouses for the purpose of assessing production process hygiene.

In the European Union, slaughterhouse operators are required to carry out weekly microbiological tests on samples taken from carcass surfaces to objectively assess production process hygiene levels (Commission Regulation [EC] No 2073/2005) *(8).* Microbiological determinations require total viable bacteria counts (TVCs), *Enterobacteriaceae* counts (ECs), and the presence or absence of *Salmonella*. The methods prescribed for sampling carcass surfaces are described in ISO 17604 (International Organization for Standardization, Geneva, Switzerland). The Commission regulation stipulates that any of the methods described in ISO 17604 can be used for the TVC and EC, although the reference method is the excision method. By contrast, *Salmonella* must be identified using the abrasive sponge sampling method. Therefore, all slaughterhouses in the European Union must use two methods (excision and sponge swabbing) to sample carcass surfaces, making sample collection more complicated and time-consuming, doubling the materials necessary, and possibly interfering in the production chain. For these reasons, a simplified carcass sampling method is needed.

For counts of mesophilic bacteria and *Enterobacteriaceae,* sampling with the sponge swabbing method is permitted, but only when there is a close correlation with the destructive method *(15)* and criteria for bacterial contamination have been established that can be compared with the results from the excision method. Thus, a method is acceptable only when a large proportion of bacteria present in the sample are recovered *(7, 21).* It is necessary to know the bacterial recovery achieved with an alternative method under evaluation and how that recovery is correlated with the recovery achieved with the excision method to duly evaluate the results of the alternative method.

The excision method recovers significantly more bacteria from the surface of the carcasses than do other nondestructive methods *(7, 11, 12),* which is why the excision method is the reference method against which all other methods are evaluated *(24).* However, certain pathogenic microorganisms such as *Escherichia coli* O157 and *Salmonella* generally have low prevalence and a heterogeneous distribution on the carcass surface *(1).* Thus, the small size of the sampling area used in the excision method, usually about 20 cm^2 per carcass *(15, 19, 22, 24),* is an important limitation when studying these microorganisms. In contrast, the nondestructive methods obtain a sample from a large area, 400 cm^2 or more *(10, 15, 17, 19, 21, 22, 24),* so these methods would be suitable for studying less prevalent microorganisms.

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Various materials are used with nondestructive collection methods. Several authors have used cellulose acetate sponges *(13, 22)*, polyurethane sponges *(3, 4, 22, 25–27)*, medical gauze bandages *(2, 11–13, 17, 18)*, cotton swabs *(6, 7, 14, 15, 20, 24, 28, 29)*, and metallic materials *(24)*. In general, microorganism recovery seems to be higher when abrasive materials are used *(7, 13)*.

The effect of microbial load on microorganism recovery from carcass surfaces has been studied only with the swab method at an experimental level *(6)* and with the double swab in slaughterhouses under normal working conditions *(19)*. However, no studies have been done on the possible effect that the bacterial load may have on microorganism recovery using the most widely used swabs (cellulose, polyurethane, and gauze).

The carcass species also seems to influence the percentage of bacteria recovered *(16)*. Ingram and Roberts *(16)* obtained bacterial recovery rates with swabbing ranging from 1 to 24% for fresh beef carcasses, 27 to 52% for fresh mutton, and 13 to 67% for fresh pork compared with rates obtained from excised and blended samples. Pressure in the application of the swab, time of swabbing, and operator-related differences also can influence the effectiveness of bacterial removal by swabbing *(5)*.

The aim of this work is to study the effect of carcass surface microbial load on microorganism recovery (mesophilic bacteria and *Enterobacteriaceae*) using four different types of swabs. We also evaluated the effects of animal species (cattle, horses, sheep, and pigs), type of swab, and the person performing the sampling on microorganism recovery.

MATERIALS AND METHODS

Animal species examined. Fifty-five bovine, 50 equine, 60 ovine, and 50 porcine carcasses were sampled in a slaughterhouse in eastern Spain. The slaughterhouse had three slaughter lines (cattle plus horses, sheep, and pigs).

Sampling methods. Two samples were taken from every carcass, one by excision and another by a type of swab. The total of 430 samples were analyzed: 215 obtained by excision and 215 obtained by swabbing. Samples were collected at the end of processing, before the carcasses were taken to the chiller. The carcasses were selected randomly from the slaughter chain.

Eight areas were sampled from each carcass (four areas on the left half of the carcass and four on the right): four areas by the excision method and the other four by the swabbing method. The sampling areas were the rump (perineal area), flank, brisket, and neck for bovine, equine, and ovine carcasses and the ham (perineal area), loin, brisket, and jowl on porcine carcasses.

Sampling of near-consecutive carcasses was undertaken alternately using the excision method on one half of the carcass and swabbing on the other half. Thus, when the first carcass selected was sampled by excision on the right half and by swabbing on the left half, the next carcass was sampled by excision on the left half and by swabbing on the right half.

In each sampling session, five carcasses of the same species were sampled. Two people (A and M) took part in sampling by excision, and four people (A, C, M, and Z) took samples with swabs.

Excision involved removing a 5-cm^2 piece of tissue, with a maximum thickness of 3 mm, from the four zones on each carcass sampled, for a total of 20 cm^2 per carcass. A flame-sterilized stainless steel square tube impregnated with coloring (Allura red, E-129) was used to mark the area. The four tissue samples removed from each carcass were placed inside a sterile plastic container filled with 20 ml of peptone water (0.1% peptone, 0.85% NaCl).

Four different types of swabs were used for swabbing. The cellulose acetate sponge (Biotrace International, Barcelona, Spain) is marketed to sample carcass surfaces, whereas the other three types are marketed for other uses. The polyurethane sponge (Deliplus, Barcelona, Spain) and reinforced viscose sponge (Bosque Verde, Barcelona, Spain) were cut and then sterilized in an autoclave (121°C for 15 min) before use. Hydrophilic gauze (Gaspunt, Lleida, Spain) and cellulose acetate sponges are sold already sterilized. All swabs were 8 by 4 cm, except the gauze, which was 7 by 6 cm.

Each swab was placed inside a stomacher bag and moistened immediately before use with 20 ml of sterile peptone water. The swab was then drained into the bag so the liquid did not spill onto the carcass. An area of 100 cm^2 (50 cm^2 for sheep carcasses) was outlined by a plastic stencil, sanitized with 70% alcohol, and rubbed with approximately one-quarter of the swab surface area. Each of the four sampling areas was rubbed, applying as much pressure as possible, first vertically (20 times) and then horizontally (20 times). The total sampled surface area was 400 cm^2 (200 cm^2 for sheep carcasses) in each carcass. The sampling swab was then placed inside the stomacher bag containing 20 ml of peptone water to form a sample.

Immediately after the samples were placed inside their corresponding containers, both types of samples were shaken vigorously for 5 s to help the microorganisms move from the tissue and cotton fibers of the swab into the solution. Samples were held in a refrigerator at 4°C and transported to the laboratory in a cold box containing ice packs.

Microbiological examination of samples. TVCs and ECs were determined by standard plate count methods according to the criteria specified by ISO 4833:2003 and ISO 21528-2:2004, respectively. All analyses were conducted in the same laboratory. The excision samples (20 ml of peptone water with excised pieces of tissue) was placed inside a stomacher bag with 80 ml of peptone water and homogenized for 2 min in a stomacher (Lab Blender 400, Seward Medical, London, UK). Another 80 ml of peptone water was added to the stomacher bag containing the sampling swab plus 20 ml of peptone water, and this sample also was homogenized with the stomacher.

Each sample homogenate (excision or swabbing) was then diluted decimally in peptone water, and 1 ml aliquots were added to suitable petri dishes, reaching a 10^{-6} dilution (TVC) and a 10^{-4} dilution (EC). Samples were analyzed within 24 h of collection. The culture media were plate count agar (PCA; Microkit, Madrid, Spain) for TVCs and violet red brilliant green agar (Microkit) for ECs. PCA plates were incubated at 37°C for 48 h before colonies were counted. *Enterobacteriaceae* were incubated at 37°C for 24 h. *Enterobacteriaceae* presence was confirmed by oxidase testing and ability to metabolize glucose. The detection limit of the technique was 5 CFU/cm^2 for the samples taken by excision and 0.25 CFU/cm^2 for the samples taken by swabbing from bovine, equine, and porcine carcasses. The detection limit was 0.50 CFU/cm^2 for all ovine carcasses.

Data analysis. All bacterial counts were expressed as CFU per square centimeter of carcass surface and then log transformed. When no *Enterobacteriaceae* were detected, a value of half of the limit of detection was used for the calculations *(9, 15)*.

The Statistical Analysis System statistical package (SAS Institute, Cary, NC) was used for the analysis of variance (ANOVA) of bacterial counts following the general linear model procedure. For each carcass, the difference between log values was calculated (log CFU/cm^2 obtained by excision − log CFU/cm^2 obtained by swabbing). To analyze the effect of carcass species, person, and swab type on the TVCs, the carcasses were classified as one of three levels according to the TVC obtained by excision: TVC low (<4.0 log CFU/cm^2), medium (4.0 to 4.5 log CFU/cm^2), and high (>4.5 log CFU/cm^2).

To analyze the effect of carcass species, person, and swab type on the ECs, the carcasses were classified as one of three levels according to the EC obtained by excision: EC low (<1.6 log CFU/cm^2), medium (1.6 to 2.6 log CFU/cm^2), and high (>2.6 log CFU/cm^2). Only carcasses for which the EC exceeded 1 CFU/cm^2 were taken into account, with both the excision method and the swabbing method.

For comparison of the results obtained by excision or swabbing for different species or by different people, the data for viscose sponges have been excluded because the bacteria were recovered in substantially smaller numbers than those recovered with the other materials, and the inclusion of these data for obtaining the mean and standard deviation could be misleading. Significant differences were defined at the 95% level ($P < 0.05$).

RESULTS

Mean logarithmic counts according to carcass species, person, and swab type are shown in Table 1 (TVC) and Table 2 (EC). Viable bacteria were detected on every carcass by both excision (EX samples) and swabbing (SW samples). However, *Enterobacteriaceae* were detected in 82.8 and 82.8 of the EX and SW samples, respectively (cellulose, 81.8% of samples; gauze, 78.2% of samples; polyurethane, 83.6% of samples; viscose, 88.0% of samples).

Mean log TVC for all species sampled by excision was 4.50 log CFU/cm^2, which was significantly higher than the 3.53 log CFU/cm^2 obtained by swabbing. Most of the samples (63.7%) had TVCs of 4.1 to 5.0 log CFU/cm^2, and most of the carcasses (49.8%) had ECs of 1.1 to 2.5 log CFU/cm^2.

The percentage of bovine, ovine, and porcine carcasses in which *Enterobacteriaceae* were detected when the sample was taken with a cellulose sponge (67%) or a polyurethane sponge (93%) exceeded that obtained when sampling was done by excision (55%); the percentage of *Enterobacteriaceae*-positive porcine carcasses sampled with gauze was higher than that detected in by the excision method (92 and 18%, respectively).

Counts obtained from EX samples (TVC and EC) were significantly higher ($P < 0.05$) than those obtained with the four types of swabs studied. These differences were observed for the four animal species tested. For TVCs, the differences were significant for all four types of swabs, whereas for the ECs the differences were not always significant.

The difference between mean values of TVCs for EX and SW samples was 2.48 log CFU/cm^2. However, the type of swab, animal species, person collecting the sample, and bacterial load exerted a significant effect ($P < 0.05$) on the mean of log TVCs (Table 1). In contrast, no two-way interaction was significant. The differences between mean log TVCs from gauze, cellulose, and polyurethane swabs were low (0.46, 0.37, and 0.49 log CFU/cm^2, respectively) and significantly lower than that from viscose swabs (2.48 log CFU/cm^2). Among the first three types of swabs, the differences were not significant. For different species (excluding data for viscose sponges), the difference between mean log TVCs for bovine carcasses (0.61 log CFU/cm^2) was significantly higher ($P < 0.05$) than that for equine and ovine carcasses (0.25 and 0.34 log CFU/cm^2, respectively). However, with high TVCs (>4.5 log CFU/cm^2) in the EX samples, this difference for bovine carcasses was similar to that found for ovine carcasses. In contrast, after excluding data obtained with viscose sponges one of the people who collected samples (C) obtained significantly higher differences in mean TVCs (0.70 log CFU/cm^2) than were obtained by the other three individuals (A, 0.29; Z, 0.36; and M, 0.34 log CFU/cm^2). These results are similar for different microbial loads. Differences between mean TVCs recovered increased significantly ($P < 0.05$) as the microbial load increased (Table 1). Thus, when the mesophile count was high (>4.5 log CFU/cm^2) the difference between mean values was 1.40 log CFU/cm^2, whereas at intermediate EX counts (between 4.0 and 4.5 log CFU/cm^2), the difference was 0.70 log CFU/cm^2 and at low counts (<4.0 log CFU/cm^2) the difference decreased to 0.39 log CFU/cm^2.

The analysis of differences between mean ECs considered only those carcasses in which these microorganisms were detected at more than 1 CFU/cm^2 (127 carcasses). Thus, two-way interactions could not be included in the ANOVA model, and therefore the effect of the swab type, animal species, and person who collected the sample could not be studied with reference to *Enterobacteriaceae* load.

The mean EC for the SW samples was 1.22 log CFU/cm^2 (Table 2). The type of swab and the *Enterobacteriaceae* load significantly influenced the EC ($P < 0.05$). In contrast, animal species and the person who collected the sample (excluding data for viscose sponges) had no significant effect ($P > 0.05$). The differences in mean ECs obtained with a cellulose sponge, polyurethane sponge, and gauze (0.62, 0.80, and 0.97 log CFU/cm^2) were significantly lower than those obtained with the viscose sponge (1.67 CFU/cm^2).

When the level of *Enterobacteriaceae* was higher than 2.6 log CFU/cm^2, the difference between mean ECs was 1.31 CFU/cm^2, which is significantly lower ($P < 0.05$) than that observed at medium levels (between 1.6 and 2.6) and low levels (<1.6) of *Enterobacteriaceae*. Between these two levels, no differences were detected ($P > 0.05$).

DISCUSSION

Counts obtained from EX samples (TVCs and ECs) were significantly higher ($P < 0.05$) than those obtained with the four types of swabs studied. Our results are in accordance with those of Pearce and Bolton *(22)*, who obtained higher mesophile counts with the excision method than from samples collected with cellulose and polyurethane sponges. In contrast, in other studies the mesophile counts obtained with the excision method were similar to those

TABLE 1. *Total viable counts from carcasses obtained by the swabbing method (SW) compared with those obtained by the excision method (EX) according to microbial load (by EX), swab, carcass type, and person who collected the samples*[a]

	EX values <4.0 log CFU/cm²				EX values 4.0–4.5 log CFU/cm²				EX values >4.5 log CFU/cm²				Total (log CFU/cm²)			
SW sample	*n*	EX (log CFU/cm²)	SW (log CFU/cm²)	Diff[b]	*n*	EX (log CFU/cm²)	SW (log CFU/cm²)	Diff	*n*	EX (log CFU/cm²)	SW (log CFU/cm²)	Diff	*n*	EX (log CFU/cm²)	SW (log CFU/cm²)	Diff
Type of swab																
Cellulose	10	3.72 ± 0.23	3.62 ± 0.19	0.16 AX	25	4.26 ± 0.16	3.97 ± 0.23	0.23 BX	20	5.04 ± 0.28	4.24 ± 0.36	0.70 CX	55	4.44 ± 0.54	4.04 ± 0.38	0.37 X
Gauze	9	3.80 ± 0.15	3.70 ± 0.38	0.12 AX	20	4.26 ± 0.13	3.89 ± 0.44	0.45 BX	26	5.10 ± 0.52	4.22 ± 0.43	0.82 CX	55	4.58 ± 0.63	4.01 ± 0.47	0.46 X
Polyurethane	13	3.76 ± 0.21	3.71 ± 0.36	0.09 AX	19	4.21 ± 0.11	3.72 ± 0.52	0.46 BX	23	4.99 ± 0.36	4.07 ± 0.36	0.89 CX	55	4.43 ± 0.57	3.86 ± 0.45	0.49 X
Viscose	8	3.80 ± 0.20	2.15 ± 0.85	2.09 AY	10	4.25 ± 0.14	1.43 ± 1.56	2.74 BY	32	4.83 ± 0.23	2.22 ± 0.76	2.60 BY	50	4.55 ± 0.45	2.05 ± 1.01	2.48 Y
Carcass type[c]																
Bovine	9	3.77 ± 0.18	3.48 ± 0.37	0.41 AX	15	4.28 ± 0.11	3.60 ± 0.63	0.52 AX	21	5.23 ± 0.47	4.32 ± 0.45	0.91 BXZ	45	4.62 ± 0.71	3.91 ± 0.63	0.61 X
Equine	4	3.83 ± 0.15	3.96 ± 0.39	0.02 AY	12	4.27 ± 0.12	3.98 ± 0.28	0.35 BXY	19	4.98 ± 0.37	4.39 ± 0.32	0.52 BY	35	4.61 ± 0.51	4.20 ± 0.37	0.25 Y
Ovine	13	3.76 ± 0.23	3.70 ± 0.24	0.09 AY	29	4.23 ± 0.16	3.97 ± 0.39	0.25 AYZ	8	4.82 ± 0.25	4.16 ± 0.29	0.68 BXY	50	4.20 ± 0.39	3.93 ± 0.31	0.34 YZ
Porcine	6	3.71 ± 0.21	3.76 ± 0.17	0.03 AY	8	4.20 ± 0.11	3.84 ± 0.22	0.36 BXZ	21	5.01 ± 0.28	3.94 ± 0.31	1.09 CZ	35	4.60 ± 0.58	3.88 ± 0.28	0.49 XZ
Person[c]																
A	7	3.65 ± 0.18	3.56 ± 0.24	0.03 AX	14	4.21 ± 0.15	4.07 ± 0.29	0.19 AX	14	4.90 ± 0.27	4.21 ± 0.44	0.73 BX	35	4.37 ± 0.53	4.02 ± 0.42	0.29 X
C	3	3.90 ± 0.02	3.64 ± 0.32	0.50 AY	19	4.18 ± 0.12	3.57 ± 0.53	0.65 AY	19	5.33 ± 0.55	4.30 ± 0.46	0.95 BY	41	4.69 ± 0.71	3.91 ± 0.60	0.70 Y
M	8	3.70 ± 0.25	3.59 ± 0.40	0.06 AX	13	4.33 ± 0.13	3.91 ± 0.24	0.36 BX	18	4.97 ± 0.34	4.18 ± 0.32	0.74 CX	39	4.49 ± 0.56	3.97 ± 0.38	0.34 X
Z	14	3.82 ± 0.17	3.79 ± 0.29	0.03 AX	18	4.28 ± 0.11	4.00 ± 0.28	0.28 BX	18	4.93 ± 0.22	4.12 ± 0.37	0.77 CX	50	4.39 ± 0.48	3.99 ± 0.34	0.36 X
Total		3.77 ± 0.20	3.37 ± 0.77	0.39 A		4.24 ± 0.13	3.54 ± 1.04	0.70 B		4.98 ± 0.37	3.57 ± 1.07	1.40 C		4.50 ± 0.23	3.53 ± 0.49	0.97

[a] Values are mean ± standard deviation. *n*, number of carcasses.

[b] Diff, log CFU/cm² by EX − log CFU/cm² by SW. Within the same column, means with different letters (X, Y, or Z) are significantly different ($P < 0.05$). Within the same row, means with different letters (A, B, or C) are significantly different ($P < 0.05$).

[c] Data for viscose sponges have been excluded.

TABLE 2. Enterobacteriaceae *counts from carcasses obtained by the swabbing method (SW) compared with those obtained by the excision method (EX)*[a]

SW sample	n	*Enterobacteriaceae* count (log CFU/cm^2) EX	SW	Diff[b]
Type of swab				
Cellulose	39	1.95 ± 0.60	1.32 ± 0.61	0.61 A
Gauze	34	2.49 ± 0.94	1.52 ± 0.91	0.83 A
Polyurethane	32	2.09 ± 0.81	1.28 ± 0.76	0.83 A
Viscose	22	2.14 ± 0.73	0.46 ± 0.48	1.69 B
Carcass type[c]				
Bovine	30	2.33 ± 0.94	1.45 ± 0.94	0.82 A
Equine	25	2.37 ± 0.91	1.49 ± 0.72	0.84 A
Ovine	25	1.85 ± 0.57	1.24 ± 0.63	0.58 A
Porcine	25	2.10 ± 0.69	1.30 ± 0.72	0.73 A
Person[c]				
A	20	2.07 ± 0.77	1.38 ± 0.75	0.62 A
C	32	2.50 ± 0.97	1.52 ± 0.83	0.87 A
M	24	2.27 ± 0.78	1.31 ± 0.83	0.92 A
Z	29	1.79 ± 0.50	1.26 ± 0.65	0.52 A
Enterobacteriaceae load[c]				
High (>2.6)	27	3.25 ± 0.54	2.10 ± 0.77	0.94 A
Medium (1.6–2.6)	51	2.10 ± 0.29	1.21 ± 0.62	0.98 A
Low (<1.6)	27	1.21 ± 0.21	0.96 ± 0.49	0.30 B
Total		2.16 ± 0.33	1.22 ± 0.62	0.94

[a] Values are mean ± standard deviation. *n,* number of carcasses.
[b] Diff, log CFU/cm^2 by EX − log CFU/cm^2 by SW. Within the same sample group, values with different letters are significantly different ($P < 0.05$).
[c] Data for viscose sponges have been excluded.

obtained with cellulose sponges *(13),* polyurethane sponges *(3),* and gauze *(13),* whereas other workers obtained higher values with gauze *(19).* Pearce and Bolton *(22)* obtained higher ECs with the excision method than with cellulose and polyurethane sponges. In contrast, Byrne et al. *(3)* obtained similar ECs with the excision method and with polyurethane sponges, whereas Gill and Jones *(13)* obtained significantly lower counts of coliforms and *E. coli* in samples taken by excision than in those taken with a cellulose acetate sponge or gauze. These various results indicate that significant differences in bacterial recovery by the two methods (excision and swabbing) depend on a number of factors, including the type of swab used, whether the swabbed surface is fat or lean, whether the surface is skin (e.g., pork) or meat (e.g., beef), and whether the samples were collected immediately after processing or after a period of cold storage *(24).*

In general, we obtained less variation (standard deviation) in counts (TVCs and ECs) by swabbing (excluding viscose sponges) than with the excision methods, as other authors have found for mesophiles in samples collected with cellulose sponges *(13)* and gauze *(13, 18)* and for *Enterobacteriaceae* and *E. coli* in samples collected with gauze *(18).* This lower variability in the counts for SW samples seems to be due to the larger area per carcass that is sampled with swabs (400 cm^2; in sheep, 200 cm^2) compared with that sampled by excision (20 cm^2). This conclusion also was reached by Gill and Jones *(13),* who noted that the greater sampling area in carcasses sampled with gauze resulted in a tendency to lower the standard deviation of the counts.

The standard deviation in our study is lower than that published for other studies *(13, 18)* even though the average counts are higher in our study. This difference in standard deviations could be due to the fact that the samples in these other studies came from several slaughterhouses, which could account for increased variability in the counts. When the variability in bacterial counts is low, trends due to specific changes can be detected more easily, thereby facilitating hygiene improvements in the slaughter process *(18)* or, as in our work, facilitating assessment of the effects of different factors on the recovery of microorganisms from the surface of carcasses.

Several authors have noted that the mesophile counts obtained from samples taken with polyurethane sponges *(3, 22)* or with cellulose acetate sponges *(13)* are similar to those obtained from samples taken by excision. A common feature of these studies is that the mesophile counts were low (generally below 3.5 log CFU/cm^2). Our results are consistent with those of these studies; differences between mean TVCs for low bacterial loads (<4.0 log CFU/cm^2) were 0.16 and 0.09 log CFU cm^2 for the cellulose and polyurethane sponges, respectively.

The effect exerted by total microbial load on differences between mean log mesophiles counts was observed for all four kinds of swabs in the four species and the four people who collected the samples. This finding was described in a previous work evaluating the double swabbing method *(19).* The authors of that study suggested this phenomenon could be due to a rapid saturation of the swabs because of their small size. However, swab size cannot explain the fact that a lower percentage of bacteria was recovered when mesophile loads increased. Cenci-Goga et al. *(6),* in an in vitro study with bovine skin cuts inoculated with various suspensions of marker microorganisms (*E. coli, Enterococcus faecalis,* and *Staphylococcus aureus*), found that microorganism recovery rate obtained using wet-dry cotton swabs decreased as microbial load increased. These authors also found that for low levels of microorganisms (<3.20 log CFU/cm^2) counts obtained by the double swabbing method were similar to those obtained by excision. Our results are similar.

The percentage of bovine, ovine, and porcine carcasses in which *Enterobacteriaceae* were detected when the sample was taken with a cellulose sponge, gauze, or a polyurethane sponge exceeded that obtained when sampling was done by excision. These results agree with those of Pearce and Bolton *(22)* and Lindblad *(18).* The differences between means counts obtained with the nondestructive methods are probably due to the different sampling materials used because the sampled surface was the same in all the studies. According to Gill and Jones *(13),* the percentage of carcasses on which microorganisms of low prevalence (e.g., *Salmonella, E. coli,* and coliforms) were detected depended on the size of the sample area. Thus, in our study, it appears that if we increased the area sampled 10 times by using gauze (from 10 to 1,000 cm^2), the number of carcasses testing positive for *Enterobacteriaceae* also would increase.

In some carcasses, we found that the differences between mean TVCs and/or ECs were below 0 log CFU/cm^2, a result also obtained by other authors *(24)*. Thus, the count obtained by swabbing was higher than that obtained by excision, which is considered the reference method. This difference could be explained by the heterogeneous distribution of bacteria on the surface of the carcasses *(3, 23)* or by the larger sample area covered with abrasive swabs. Therefore, abrasive swabs may be more reliable for detecting low prevalence microorganisms because these swabs can be used to sample a larger area without causing damage to the carcass *(1)*.

On a practical level, the main problems encountered during sampling by excision were the dangers of using a sharp object (knife) and the drawback posed by dry heat sterilization (between carcasses) of the tweezers and the tip of the marker used to delineate the area to be sampled. Polyurethane and viscose sponges have the disadvantage of the format in which they are sold because they must be prepared before use (the sponge must be cut and sterilized). The only drawback to the hydrophilic gauze is that during sample collection these gauzes fold very easily. The only disadvantage of the cellulose acetate sponges is their higher cost compared with the other materials.

We conclude that sampling with cellulose acetate sponges, polyurethane sponges, and gauze results in large differences between mean log counts of mesophiles and *Enterobacteriaceae,* which makes these swabs suitable for use in slaughterhouses for assessment of production process hygiene. These sponges are excellent for sample collection when mesophile counts are lower than 3.5 to 4.0 log CFU/cm^2. Conversely, the viscose sponge is not recommended for sampling carcasses. Because of the significant impact that the animal species and the person collecting the sample has on the recovery of microorganisms, we recommend that the swabbing material be validated for used in every slaughterhouse for each animal species and each person collecting samples before the results obtained are compared with those obtained with the excision method.

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