SAMPLING PLANS TO MONITOR MICROBIAL QUALITY OF COMMERCIAL BEEF CARCASSES Sampling : Excision samples (12.5 cm<sup>2</sup>) were shaved off as thin as possible with sterile intruments, wrapped RaITRAD .9 Service Qualité des Viandes, INSTITUT DE L'ELEVAGE, 14310 VILLERS-BOCAGE, France and and antipues ne sont bas on TVG 5 P (FOURNAUD and al. 1973) after incubation for 5 days at 22°C. For Pseudomonas (PS) counts, say RAMMUS

In a first trial, several sites on commercial beef fore quarters were sampled. It was found that the restriction of sampling to 3 defined carcass areas (outside of the neck, central position of shoulder, forerib) and a single sample (by bulking the sites) was acceptable for microbial evaluation of the whole quarter. In a second trial, the bacteriological quality of batches of commercial carcasses was assessed by sampling around 50 quarters per batch. Precision of sampling plans versus the number of sampled quarters was then established.

# INTRODUCTION

It is now well established that the slaughtering of the animal is an important source of contamination during meat processing (INGRAM and ROBERTS, 1976 ; FOURNAUD et al, 1978 ; ROBERTS, 1980 ; EUSTACE, 1981 ; NORTJE and NAUDE, 1981). As visual control is obviously not sufficient to garantee good hygienic quality of carcasses (EUSTACE, 1981; ROBERTS and al, 1984 ; STOLLE, 1988), it is necessary to count number of bacteria on carcasses during their production and distribution. The problem of adequate sampling for bacteriological purposes have been discussed by INGRAM and ROBERTS, 1976 ; ROBERTS et al, 1980 ; STOLLE, 1988). Number and distribution of bacteria on beef carcasses at the end of the slaughterline or 24 hours later have been reported in many studies (INGRAM and ROBERTS, 1976 ; EUSTACE, 1981 ; ROBERTS ans al, 1984 ; STOLLE, 1988). However, only few bacteriological data have been published on commercial carcasses (ROBERTS et al, 1980). Furthermore, in practice, time and effort required for microbiological testing do that it is necessary to sample a limited, but representative, number of carcasses. The aim of this study was to examine microbial quality of a <sup>substantial</sup> number of commercial carcasses obtained from different abattoirs, and to propose a microbiological control system of such carcasses, in order to the possible application of quality control rejection procedure of batches of carcasses.

As proposed by others, we choose to seek Total Viable Counts (TVC), micro-organisms capable of growing at refrigeration temperature (Pseudomonas, PS) and indicator of faecal pollution (Coliforms, CF). As sampling method we choose excision rather than a swabbing method that is not sufficiently reproductible (ROBERTS et al, 1984). A satisfactory sampling scheme <sup>needs</sup> to answer to : 1) Which parts of a carcass to examine ? 2) On a particular batch, how many carcasses to sample ?

# MATERIALS AND METHODS

# 1 - Experimental design

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Two consecutive trials (I and II) were performed.

Trial I : In the purpose to choose which part of a carcass to examine, 24 fore quarters coming from 3 different abattoirs were sampled as they were delivered (i.e. from 1 day to 5 days after slaughter). On each carcass, 20 defined sites were sampled (figure 1).





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Trial IL : A total of 869 commercial fore quarters were sampled at 3 different sites (see results trial I). Those quarters were : taken in 18 lorries including french and foreign deliveries, so that between 40 and 60 quarters were examinated in each lorry. Each quarter was identified according to its position in the lorry (front or back, middle or side).

shoulder and foreitb), and a single sample (by bulking the 3 sites before bacteriological analysis) was acceptable for the

### 2 - Bacteriological assessment

Sampling : Excision samples (12.5 cm<sup>2</sup>) were shaved off as thin as possible with sterile intruments, wrapped in sterile bag and frozen counting. The samples were blended (stomacher 80) for 1.5 mn in 50 ml of 0.1% peptone solution. Decimal dilutions were prepared and 1 ml samples were plated on media and incubated. Total Viable Counts (TVC) were determined on TVG 5 P (FOURNAUD and al, 1973) after incubation for 5 days at 22°C. For Pseudomonas (PS) counts, samples were incubated for 2 days at 22°C, using Pseudomonas Agar Base (Oxoïd) and Pseudomonas C.F.C. Supplement (Oxoïd). Coliforms (CF) were monitored on desoxycholate Agar (Difco) after incubation for 1 day at 44°C.

All counts were transformed to logarithms and expressed as log <sup>10</sup> CFU (Colony Forming Unit) per cm<sup>2</sup> of beef sample.

### **RESULTS AND DISCUSSION**

## 1 - First trial : Pattern of fore quarter bacterial contamination.

For each quarter, mean values for TVC (calculated from number of bacteria on the 20 sites), the maximum and minimum values and the sites at which they were obtained are shown in table 1. As previously observed by others (INGRAM and ROBERTS, 1976 ; ROBERTS et al, 1980, 1984 ; JOHANSON et al, 1983 ; STOLLE, 1988), we found high variation in microbial load from one quarter to another. Average contamination ranged from 3.3 log to 5.3 log. Distribution of bacteria on surface quarter also shown high variation. Indeed, it is common for the maximum and minimum counts on a quarter to differ by as much as 4 logarithmic units. Furthermore, as found by others (INGRAM and ROBERTS, 1976; ROBERTS et al, 1980, 1984 ; STOLLE, 1988), the most contaminated sites, that are particularly interesting for quality control, were not the same for different quarters, and the same was true for the least contaminated sites (table 1). Maximum counts were found on sites 2, 3, 18 (brisket), site 6 (short ribs), sites 11, 12, 19 (shoulder and fore shank), sites 14, 15, 16 (neck) and site 20 (fore rib, medial). Those data suggest that the distribution of bacteria is not sufficiently systematic to warrant sampling only one site. Consequently, several sites must be sampled, although as proposed by ROBERTS et al (1980), the samples from one carcass may be bulked before bacteriological analysis.

TABLE 1 : Diffe	rences in bacter	ial contamination	on (local viable	Counts, 102	g 10 / CIII2)
at different sites of	on 24 fore quart	ters			
Г	Minimur	n value	Maximum	value	

	Minimu	m value	Maximu			
N° of quarters	Number of Site (1) bacteria		Number of bacteria	Site (1)	Mean value	
quarters  1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23	bacteria 2,81 2,66 1,20 2,53 0,48 1,08 0,60 1,78 2,30 1,38 0,60 0,48 1,51 1,08 1,60 0,60 0,48 1,51 1,08 1,60 0,60 0,48 1,30 1,08 2,56 2,20 2,38 2,72	15 17 7 17 17 7 6 7 6 7 7 6 17 9 17 7 10 6 7 7 17 8 7 7	bacteria 6,75 6,26 6,41 6,12 5,12 4,62 4,20 4,83 4,98 4,51 4,08 4,51 4,26 4,56 4,56 4,51 4,78 5,20 4,30 5,34 5,00 4,56 6,16	$\begin{array}{c} 12\\ 11\\ 20\\ 16\\ 6\\ 19\\ 16\\ 20\\ 18\\ 3\\ 3\\ 3\\ 3\\ 16\\ 14\\ 16\\ 16\\ 16\\ 15\\ 2\\ 20\\ 16\\ 15\\ 20\\ \end{array}$	value           5,62           5,32           5,15           5,21           4,47           3,78           3,48           3,77           4,13           3,74           3,74           3,79           3,54           3,77           3,45           3,81           4,44           4,24           3,92           5,26	
24	1,30	1	4,80	5	5,00	



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FIGURE 2 : Correlation between contamination of sites 11, 16, 20, and contamination of all sites (1-20) with TVC as bacteriological index

(1) For names of sites, see fig 1

In order to select the subset of sites that most characterized the bacteriological quality of a quarter, correlation betwee subsets of "dirty sites" and mean of all sites (used as the bacteriological index of the whole quarter) was studied. Whe subsets included only 2 sites, the highest correlation was observed with sites 11 and 20 (r = 0.85, results not shown). Nov when subset included 3 sites : sites 11, 16, 20, coefficient correlation was up to 0.90 (r = 0.93, figure 2). With additional sites it was not possible to obtain a coefficient of correlation significantly higher than observed with 3 sites (results not shown) Those results showed that the restriction of sampling to 3 defined carcass areas (outside of the neck, central position of shoulder and forerib), and a single sample (by bulking the 3 sites before bacteriological analysis) was acceptable for the bacteriological evaluation of the whole quarter when TVC was used as bacteriological index.

With Pseudomonas (PS) or Coliforms (CF) as criterion adopted to judge hygienic quality, high carcass to carcass variations and site to site variation were also observed (results not shown). Correlation of PS and CF to TVC was not very high (PS : r=0.51; CF : r=0.80 ; n=480), suggesting that number and distribution of those specific organisms did not reflect the behaviour of Total Viable Counts. However, the restriction of sampling to sites 11, 16, 20 seems also acceptable for the bacteriological evaluations of the whole carcass (figures 3 and 4). Finally, it can be concluded that those 3 dirty sites are suitable to judge numbers of TVC, PS or CF on a quarter. This method was used in trial II to assess microbial quality of a substantial number of commercial carcasses (see below).



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## <sup>2</sup> - Second trial : Microbial quality of batches of commercial quarters.

Bacteriological quality of 18 batches of quarters was assessed by sampling around 50 quarters per batch (see methods). Results are tabulated in table 2. On any particular batch, the range of counts between quarters was very large. In the same batch, maximum and minimum counts for TVC, PS and CF were generally as follow : TVC and PS : from less than 2-3 log to 6-7 log ; CF : from less than 1 log to 2-3 log.

LORRY	Number of quarters	Total Viable Counts (TVC)		Pseudomonas (PS)			(CF)			
		Min	Max	Mean± std. dev.	Min	Max	Mean ± std. dev.	Min	Max	Mean ± std. dev.
A	60	2,94	5,64	$3,92 \pm 0,65$	1,48	3,75	2,20 ± 0,67	0,48	3,51	1,01 ± 0,6
B	60	3,38	7,82	$4,60 \pm 0,78$	1,48	5,15	$2,42 \pm 0,81$	0,48	3,56	$0,84 \pm 0,6$
C	60	2,66	5,30	$3,78 \pm 0,54$	1,48	4,58	$2,44 \pm 1,02$	0,48	2,26	$0,74 \pm 0,4$
D	60	3,20	5,73	$4,45 \pm 0,69$	1,48	5,45	$2,16 \pm 0,80$	0,48	3,08	$0,77 \pm 0,5$
E	60	3,15	5,64	4,13 ± 0,47	1,48	4,92	$3,02 \pm 0,63$	0,48	3,02	$0,90 \pm 0,5$
F	60	3,11	5,48	4,01 ± 0,55	1,48	4,15	$2,10 \pm 0,63$	0,48	2,60	$0,84 \pm 0,5$
G	60	3,08	5,13	$3,80 \pm 0,50$	1,48	3,00	$1,61 \pm 0,34$	0,48	2,51	$0,60 \pm 0,3$
H	40	3,00	5,58	$4,26 \pm 0,48$	1,48	3,46	$2,33 \pm 0,59$	0,48	2,53	$1,28 \pm 0,6$
I	41	3,26	6,94	4,13 ± 0,69	1,48	4,33	$2,26 \pm 0,78$	0,48	2,41	$0,77 \pm 0,5$
J	41	3,16	5,20	4,05 ± 0,56	1,48	5,05	$3,35 \pm 0,80$	0,48	2,26	$0,82 \pm 0,4$
K	39	2,88	5,58	4,13 ± 0,65	1,48	4,15	$2,11 \pm 0,79$	0,48	2,15	$0,72 \pm 0,4$
L	44	3,57	6,27	4,73 ± 0,71	1,48	6,04	$3,74 \pm 1,11$	0,48	3,73	$0,99 \pm 0,9$
M	40	3,32	7,09	4,73 ± 0,82	1,90	6,20	4,09 ± 1,24	0,48	2,58	$0,73 \pm 0,4$
N	42	2,72	6,18	4,41 ± 0,85	1,48	6,20	4,05 ± 1,27	0,48	1,08	$0,49 \pm 0,0$
0	42	3,26	5,38	4,17 ± 0,48	1,48	5,26	$2,82 \pm 0,89$	0,48	1,72	$0,55 \pm 0,2$
P	40	3,98	5,72	4,74 ± 0,43	1,48	5,86	$3,63 \pm 1,08$	0,48	1,90	$0,62 \pm 0,3$
Q	40	3,30	6,68	4,93 ± 0,66	2,08	6,51	4,33 ± 0,97	0,48	1,72	$0,65 \pm 0,3$
R	40	2,81	5,08	3,97 ± 0,58	2,45	5,41	4,03 ± 0,80	0,48	1,30	$0,51 \pm 0,1$

TABLE 2 : Bacterial contamination (TVC, PS and CF) of 18 batches of fore quarters

The reason for such extreme differences is not understood and isolated occurrences may probably be responsible (INGRAM and ROBERTS, 1976). Identification of each quarter according to its position in the lorry (see methods) showed that this factor did not generally matter. However, in some cases, and especially with PS as bacteriological index, differences were observed between quarters in the front and the back of the lorry (figure 5). Those data agreed with the fact that, in such lorry days of slaughter were not the same for all quarters. For example, in lorry C (figure 5), quarters situated at the back were most freshly-slaughtered than quarters situated at the front. In such lorry, the distribution of PS bacterial counts was not "normal" (results not shown). Consequently, PS average contamination could not be estimated by sampling a limited number of carcasses. With CF as bacteriological index, and whatever the batch, the assumption of log normality was not valid.





FIGURE 5 : Bacteriological quality of commercial quarters : differences in Pseudomonas counts (log10/cm2) from the front and the back of the lorry

FIGURE 6 : Estimation of TVC average contamination of a batch of quarters by sampling a limited number of quarters

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So, as found for PS bacterial counts, CF average contamination could not be estimated. In the opposite, TVC bacterial counts were normally distributed and TVC average contamination could be estimated by sampling some quarters. Precision on average contamination was 1.2 log when only one carcass was sampled, 0.8 log for 4 sampled carcasses and 0.3 log for 12 sampled carcasses (Figure 6). As the average contamination varied a little bit from batch to batch (in the present study) average counts range from 3.8 logs to 4.9 logs, table 2), it can be advanced that sampling plans with only one or two quarters are not suitable to estimate average quality of a particular batch.

### CONCLUSION

Bacteria are scattered ununiformly over the surface of the carcass. However, as previously proposed by others, present study shows that the restriction of sampling to "dirty" sites and a single analysis (by bulking sites before bacteriological analysis) is acceptable for microbial evaluation of the whole quarter. Consequently, hygienic quality of individual quarter can be easily estimated. In contrast, for microbial evaluation of a batch of quarters, there are 2 basic difficulties : 1) a large number of quarters must be sampled since carcass to carcass variation is very high ; 2) In some cases, average contamination could not be estimated since the assumption of log normality is not valid, except when TVC is used as bacterial index.

### REFERENCES

EUSTACE I.J., 1981. Food Technol. Aust., 33, 28-32. FOURNAUD J., SALE P., VALIN C., 1973. 19ème Réunion Européenne des Chercheurs en Viande, PARIS, 1, 291-313. FOURNAUD J., GRAFFINO G., ROSSET R., JACQUE R., 1978. Ind. Aliment. et Agricol., 95, 273-282. INGRAM M., ROBERTS T.A., 1976. Roy. Soc. Hith. J., 96, 270-276. JOHANSON L., UNDERDAL B., GROSLAND K., WHELEHAN O.P., ROBERTS T.A., 1983. Acta Vet. Scand., 24, 1-13. NORTJE G.L., NAUDE R.T., 1981. J. Food Protect., 44, 355-358. ROBERTS T.A., 1980. Roy. Soc. Hith. J., 100, 3-9. ROBERTS T.A., MacFIE H.J.H., HUDSON W.R., 1980. J. Hyg. (Camb.), 85, 371-380. ROBERTS T.A., MacFIE H.J.H., HUDSON W.R., 1980. J. Hyg. (Camb.), 85, 371-380. ROBERTS T.A., JLEISTNER L., GEHRA H., GLEDEL J., FOURNAUD J., 1984. Meat Sci., 11, 191-205. STOLLE F.A., 1988. Meat Sci., 22, 203-211.

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