MICROBIOLOGICAL GUIDELINES FOR MEAT

T.A. ROBERTS

Meat Research Institute, Langford, Bristol, BS18 7DY, England.

Much has been said and written over the past few years by those apparently wishing to impose microbiological standards. Reasonable persons have made unreasonable proposals, even to the extent of seeming to have forgotten the original purpose of standards and the basic foundations on which they may be established.

A "standard" is a part of law or regulation, while a "guideline" is similar, but not enforceable by law, and intended to improve hygiene. We shall not be concerned with "specifications" which are simply commercial agreements between manufacturers and buyers.

Standards should be established in the manner laid down in WHO Expert Committee Technical Report No.543 (1974) and guidelines warrant exactly similar consideration.

a) they should be based on factual studies, to determine good manufacturing practice, to minimise health risks or to measure keeping quality; (factual studies related to health risks exist almost nowhere).

b) they should be attainable under commerce with good manufacturing practice.

c) the methods of examination should be simple and inexpensive.

d) details of methods, sampling, examining and reporting should be laid down and recorded.

e) tolerance levels should be written into the standard/guideline to allow for sampling variation, errors of method etc.

Standards and guidelines might therefore achieve similar ends : the reduction, even virtual elimination, of food-borne disease; the improvement of hygiene; and improved shelf-life and quality by encouraging the reduction of total bacterial numbers. The preceding seems to imply that good hygiene is equated with low numbers of bacteria, which are necessarily safe and desirable. This is, of course, not always true. Low numbers of bacteria may be dangerous, and large numbers are normal in some meats which have impeccable health records (e.g. bacon, vacuum-packed fresh beef, fermented sausages).

If we accept that circumstances dictate that guidelines are desirable, how should they be set?

1. Which microbes to seek:

Several different groups of bacteria are commonly suggested:

Salmonella
E. coli I
presumptive coliforms
Enterobacteriaceae
enterococci ("faecal streptococci")
Clostridium perfringens
"total viable count" (standard plate count)

Consider first the elimination of diseases. Each year in the UK some food-poisoning outbreaks, usually caused by *Salmonella* and *Cl.perfringens*, are attributed to red-meat and red-meat products. The causative organisms are assumed to be present initially on the meat and to have multiplied under inadequate storage conditions, although this is unsupported by good evidence. An unknown number of outbreaks will have resulted from cross-contamination during handling.

If the eradication of all meat-based salmonellosis could be achieved by rejection of meat containing *Salmonella* such extreme measures might be justified, but what are the chances of finding *Salmonella* on meat? Examining carcasses to find *Salmonella* is very unrewarding because they are so rare e.g. in one commercial abattoir monitored periodically over four years, a total of 879 carcasses were examined by methods known to recover *Salmonella* efficiently, but only 12 were found (Table 1). Examination of meat later in the distribution chain might result in more isolations.

Such rare occurrence tempts one to monitor for *Salmonella* by counting other bacteria present more often e.g. *E.coli I*, "coliforms", *Enterobacteriaceae*. This seems reasonable on the carcass because many, although not all, originate in the gut and the presence of large numbers of coliforms indicate increased likelihood of *Salmonella* occurring. However, nowhere are data adequate to justify such conclusions for meat. There is no constant ratio of *E.coli* or coliforms or *Enterobacteriaceae* to *Salmonella*, and Salmonella may be found on clean or dirty carcasses. Later in the distribution chain matters become even worse because some coliforms and *Enterobacteriaceae* are able to grow below 5°C, which Salmonella are not. In fact I have seen data, referring to a variety of meats over several years sampled just before manufacture, which indicate clearly that there is no correlation between the detection of *E.coli* and *Salmonella*, or indeed the presence of *Salmonella* and the "total viable count".

In summary, the only way to monitor meat for Salmonella seems to be to test for it, and with reasonable

manufacturing practice such as in the UK, isolations will be relatively few, and the total cost high. (The exception in the UK is chicken, when Salmonella can be demonstrated readily. The health hazard appears low if cooking is adequate but chicken might be a source of cross-contamination).

There is some hope that the occurrence of Salmonella on meat can be reduced by better recognition and control before slaughter of "carrier" animals excreting Salmonella but showing no symptoms of disease. This, too, is likely to be a costly exercise.

"Hygienic practice" is frequently monitored by counting E.coli, coliforms, or Enterobacteriaceae, probably by analogy with monitoring water supplies. However, members of the coliform group are distributed widely in meats and many are able to grow on meat stored under refrigeration, making them a less useful indicator than in water where they die rapidly, and if detected indicate recent faecal contamination. Similarly the enterococci are widespread in nature, and their presence in fresh meat does not indicate faecal contamination. Finally "total viable count" could be monitored in many instances to give some indication of hygienic practice, and also the shelf-life of the meat. "Total viable count" is not related to the incidence of Salmonella, and is probably better considered, in those products where it is relevant, a monitor of potential shelf-life.

Clostridium perfringens is ubiquitous and present in the gut of every animal. It can almost always be detected in the musculature of carcasses if sufficiently large samples (ca 100g) are examined. No method is known of producing carcasses free from clostridia. Some reduction in numbers may occur during the chill storage of meat but after ten year's experience we expect to find Cl.perfringens often in meat samples, albeit in low numbers (<1/g) and requiring enrichment. If this is the norm there is no reason to examine meat for low numbers of Cl.perfringens, and only large numbers, detectable by plating methods, need be a cause for concern.

2. Problems arising from choice of microbe.

Many methodological problems arise as a result of the choice or microbe, but will be considered only briefly. Selective media frequently pose problems of reproducibility (e.g. media containing bile salts), even toxicity (e.g. selenite), and may not be sufficiently selective or diagnostic (e.g. most media for enterococci). "Most Probable Number" methods are statistically unreliable and should never be used for standards or even guidelines. Some media require judgement to be made (e.g. colony form, colour - often critically dependent on incubation conditions).

If the "total viable count" is to be used the medium is more readily standardised, but agreement on incubation temperature is rare. Should 35°C be used to include coliforms or 20 - 25°C to assess spoilage potential? Counts incubated at 35° are generally lower than at 25°, but there is little difference at incubation temperatures between 22.5° and 27.5° when examining meat stored under refrigeration.

3. How important is the method?

In an ideal situation everyone should use exactly the same method. However, developments occur usually aiming to save time and/or money.

The traditional pour plate is slow and expensive. Spread plates are faster, and considerably cheaper because dilutions can be spread on quarters of the medium and this method has now been adopted by the ISO. Other methods might also be permitted, provided results can be expressed uniformly e.g. bacteria/cm^2 or/g : e.g. Colworth 'Droplette', loop-tile method (used at MRI), spiral plate. All these are as accurate as traditional methods but there is great reluctance to permit their use.

There is no general agreement on any of the above points.

4. Carcass meat as an example:

Application of guidelines to meat implies examination of carcasses and/or cuts. Have we sufficient knowledge to support such a scheme? Carcasses will be used as an example to illustrate the problems, which are discussed in more detail in Ingram & Roberts (1976).

4.1. When to sample:

4.1.1. immediately after slaughter to assess slaughter hygiene or practice
4.1.2. after chilling to assess in addition the effects of chilling
4.1.3. during distribution to include further handling and temperature control
4.1.4. at retail to include all the above
4.1.5. concentrate on the final product, cuts of meat or minced (ground) meat as has been done in N.America
4.1.6. at the point of consumption, to take into account cross contamination at all stages.

4.2. How to sample

Examination of the carcass or cuts raises additional problems of how to remove a sample for examination : by excision, scraping, swabbing, adhesion or washing. Each of these methods has its advocates, but few comparisons have been made and no one has yet made adequate comparisons between methods on the recovery of microbes from surfaces of fat, lean and cut muscle.

4.3. Where to sample

Similarly on carcasses there is no agreement whether to sample one large area, or several areas and pool samples, or several areas separately. We examined 10-13 areas per carcass systematically on pigs, bovine and lamb carcasses and showed that in one abattoir there were no systematically clean sites or "average" sites, but the heavily contaminated sites were consistently so. However this site, the

dirtiest on the carcass, will differ with differing slaughter practice. We then made comparisons between large numbers of carcasses sampling this known dirty site by double swabbing (simply because samples were taken in a commercial abattoir and damage to the carcass surface was to be avoided).

4.4. Carcass to carcass variation: about 125 carcasses were sampled on each of 4 consecutive days at two abattoirs of high standard, one killing only pigs and the other only bovines. At both abattoirs on any day the range of counts between carcasses was very large, from less than 100 to more than 100,000/cm^2 (Table 2). The reasons for such extreme differences are not understood, but clearly require elucidation if the bacterial count on carcasses is to be controlled rather than be an almost random occurrence.

4.5. Day to day variation was small compared with that between carcasses.

4.6. Abattoir to abattoir variation: Six abattoirs were visited on 4-6 occasions sampling 25 lamb carcasses on each visit. The abattoirs represented the range for visually excellent to visually indifferent. The data showed a smaller difference than anticipated between the bacterial count on lamb carcasses from the excellent and other abattoirs. The difference between visits to any one abattoir was as great as the difference between abattoirs suggesting that changes in slaughter method and in hygienic practice are doing little to improve the bacteriological condition of carcasses (Table 3).

4.7. Country to country variation: Attempts to compare data already published or kindly supplied by workers in different countries were largely thwarted by differences in sampling and counting methods. Given a uniform method it would be necessary to examine many abattoirs in a particular country, on many days over a long period, and a substantial number of carcasses on each occasion, to get a valid measure of the state of that country's meat production. At present few, if any, European countries have such data.

5. A method of comparison

Many observations have apparently been made to little practical purpose for lack of a satisfactory basis for comparison. If bacterial counts from a large number of carcasses are expressed as logarithms and grouped at 0.5 $\log_{10}$ intervals in a frequency distribution that distribution is essentially "normal". This has occurred with counts on bovines, lamb and pig carcasses from several slaughterhouses. Since this distribution is normal the carcass-to-carcass variation may be represented by the standard deviation about the mean (logarithmic) count. Moreover the chances of an inspector entering an abattoir and selecting at random an 'average' carcass as his sample can be assessed. From data accumulated there is roughly a 2 out of 3 chance that the counts from such a sample would not differ from the average by more than ten-fold and therefore only a 1 in 6 chance of detecting "dirty" carcasses. Plainly examination of samples from one or two carcasses is likely to give a highly unreliable estimate of average quality. Comparison of such normally distributed data is simplified by the probit transformation, comparing slopes and displacements of the resulting straight lines (Ingram & Roberts, 1976). Carcasses have been dealt with in some detail to illustrate the problems of accumulating and expressing data. In addition to total viable counts, we have counted coliforms and Enterobacteriaceae on those same sites. Either may be used to classify carcasses in principle, but both counts are frequently very low on bovines and lambs, and are not recommended for that reason. Counts of coliforms or Enterobacteriaceae simply select different carcasses than the total viable count, as acceptable or unacceptable.

From our data it is plainly inappropriate to suggest the same standard/guideline for beef and lamb carcasses, although this has already been done by the ICMSF. They, too, suggest data accumulation in the frequency distribution illustrated above before selecting their limits "m" marginally acceptable, and "M" unacceptable. Regrettably no advice is offered on how these values should relate to the frequency distribution, and their selection still seems arbitrary judgement. No matter what values are chosen, it seems foolish to reject some proportion of a commodity as valuable as meat is today unless the reason is especially good, and one must question whether a high "total viable count", provided the meat is not spoiled, is an adequate reason. The meat will keep less well but may or may not have been subjected to poor hygienic practices, and may or may not be more likely to carry Salmonella. One could equally question whether the demonstration of Salmonella need necessarily be adequate reason. Throughout we have been considering testing a very small fraction of a lot, perhaps 10-1000g for Salmonella, but effectively a fraction of a gram for the total viable count, and applying the result to the lot. If the values are above a standard we are planning to reprocess or destroy that lot. Such action is frighteningly expensive and we must ask very seriously whether it is worthwhile when we realize that the standard is an arbitrary number. To base the standard on fact, a representative number of samples must be examined over a realistic period of time, and one wonders whether even in the case of ground beef whether sufficient samples have been examined to substantiate the choice of standards/guidelines currently being suggested. It is a formidable task to examine representative samples from 50-100 producers and several times that number of retailers. The cardinal rules are that standards shall be relevant, attainable with good manufacturing practice, and based upon an adequate sample.

Meat production, distribution and retailing could be a cleaner operation, and possibly give a marginally safer product, but the method of examination need not be precise. Much could be achieved with simpler methods than those being proposed. Though these simpler methods might seriously underestimate the true bacteriological condition of the product improvements could still be effected.

After relating a series of incidents where regulatory authories in the USA obtained false positive results (for Salmonella, staphylococcal enterotoxin, Cl.botulinum toxin and B.cereus) Professor E.M. Foster concluded that these errors, shrugged off by the agencies, but economically devastating for the companies concerned, were due to "ignorance and carelessness" and that "bacteriological analysis of food continues to suffer.....from the employment of incompetent people using inadequate methods to do the unnecessary".

While this should not happen with guidelines, I join him in making a plea, in devising them, for a little common sense. I am not against guidelines in principle. Meat distribution and retailing could be a cleaner operation

and possibly give a safer product but let us not put ourselves in the position of having to reject without good reason a proportion of a valuable product in short supply, particularly when the industry is struggling against inflationary production costs and could ill afford further added financial burden.

References

Ingram, M. & Roberts, T.A. (1976). The microbiology of the red meat carcass and the slaughterhouse. Roy. Soc. Hlth. J., (in press).

Foster, E.M. (1975). Interpretation of analytical results for bacterial standards enforcements. Ass. Fd Drug Off. U.S.A. Quart. Bull., 38(4) 267-276.

Table 1. Occurrence of salmonella on carcass meat in one commercial abattoir in the U.K.

	beef	lamb	pig	
1969-70	-	-	8/51	(7. S.typhimurium, 2 S.albany)
1970-71	*1/126 (S.typhimurium)	-	3/149	(1 S.heidelberg, 1 S.panama)
1971-72	0/96	0/60	0/128	
1972-73	0/55	0/112	0/102	

* 1/126 = of 126 carcasses examined, salmonella was detected on one.

Table 2. Day to day variation in bacterial contamination (log_{10}/cm^2) of carcasses in one abattoir ($100\ cm^2$ samples at 'most contaminated site': flank for beef, cheek for pigs).

Beef day	n	Max.	Min.	Mean	Std. dev.
1	125	5.3	1.4	3.4 ±	0.6
2	125	4.7	1.8	3.4 ±	0.7
3	125	4.7	1.8	3.1 ±	0.6
4	100	4.0	1.5	2.8 ±	0.6
Overall	475	5.3	1.4	3.2 ±	0.6
Pigs					
1	125	4.7	2.1	3.3 ±	0.5
2	125	3.6	1.7	2.5 ±	0.4
3	125	4.6	1.6	2.7 ±	0.5
4	125	3.7	1.9	2.9 ±	0.4
Overall	500	4.7	1.6	2.9 ±	0.5

Table 3. Variation from abattoir to abattoir (A-F*) in bacterial counts (25°C) /cl^2 on lamb carcasses. (25 carcasses per visit, 1 50 cm^2 groin sample/carcass).

Visits	A	B	C	D	E	F
1	5.1	4.7	4.4^{b}	4.4	3.7	3.5
2	4.6^{a}	4.5	4.8	4.2^{e}	4.2^{f}	3.7^{g}
3	4.5^{a}	4.9	4.5^{b}	4.7	4.4^{f}	3.8^{g}
4	4.7^{a}	4.1	4.1^{cd}	4.2^{e}	4.5^{f}	4.4^{h}
5			4.4^{bc}			4.2^{h}
6			4.1^{d}			
mean	4.7	4.5	4.4	4.4	4.2	3.9

* Visually worst A and C
" intermediate B and D
" best E and F (both excellent)

a-h Within any column, mean values with the same superscript were not significantly different at the 5% level.