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# Use of predictive microbiology in meat hygiene regulatory activity

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**Abstract**

New Zealand is a supplier of refrigerated raw meat to world markets. To maintain this supply, from regulatory and commercial perspectives, production standards need to deliver products that are both hygienically adequate and commercially viable. A dynamic Temperature Function Integration (TFI) model, as a form of predictive microbiology, was used jointly by regulators and processors to develop justifiable criteria for the management of refrigeration during the production of hot and warm-boned meat, the post-slaughter handling of ovine carcasses and the handling of offals. Current processes operating according to accepted standards for Good Manufacturing Practice (GMP) were quantified in terms of TFI. The hygienic adequacy of new processes were similarly determined using the TFI model and compared to relevant GMP standards. From a regulatory perspective, the dynamic TFI model has provided a rapid and cost effective method of quantifying a temperature dependent process in terms of the potential for microbial proliferation. It has also produced a method for determining parameters for new or intended processes by comparing the potential for microbial proliferation with previously validated outputs, and has complemented traditional quantitative microbiology to provide a rapid, cost effective method of verifying that a process is performing according to design parameters. However, it could not be used to validate standards for processing in the absence of existing standards for GMP, or in the absence of microbial standards previously established using the principles of risk assessment. 

*Keywords:* Microbiology; Predictive microbiology; Regulation; Temperature; Time

## 1. Introduction

New Zealand annually exports approximately 700,000 tonnes of frozen or chilled lamb, mutton, veal, beef or venison as whole or part carcasses, bone-in or bone-out cuts and offals (NZMPB, 1994). To satisfy the regulatory and commercial conditions for the entry of these products into a wide range of world market places, it is essential that these products possess attributes of hygiene and utility.

The hygiene of products, and a considerable proportion of the utility of products, can be quantified by their microbiological profile (Brown, 1982; ICMSF, 1980). From New Zealand's perspective, this profile will be a direct consequence of the manner in which the animals were slaughtered and dressed, and the physical conditions that prevail during the post-slaughter period until products are subjected to conditions of preservation.

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From a regulatory perspective, the physical conditions during the period immediately after slaughter that provide a window for maximum mesophile growth has been the subject of considerable interest to the Ministry of Agriculture (MAF) in carrying out its' regulatory responsibility to the hygiene of processing.

During the mid-1980's, The Meat Industry Research Institute of New Zealand, Inc. (MIRINZ) developed a practical method for determining the potential *Escherichia coli* proliferation that might occur during the cooling of meat by integrating the temperature history with the growth characteristics of the bacteria (Gill et al., 1985).

The method, Temperature Function Integration (TFI), was utilised in 1988 by MAF, MIRINZ and industry to develop a set of criteria for the hot boning of carcasses, i.e., boning directly after slaughter (MAF, 1991). The exercise was later extended to develop criteria for the warm boning of beef but, unfortunately, could not be applied to warm boning of ovines, simply because the current EC conditions for deviating from the cold cutting requirements differ substantially from those that apply for beef.

During the development of the TFI method, MIRINZ proposed a set of criteria for the cooling of offals (Gill and Harrison, 1985; Gill and Phillips, 1984). These criteria are still accepted as alternatives to current regulatory standards (MAF, 1991). In 1990, MAF, MIRINZ and industry again utilised the MIRINZ TFI method to develop and promulgate a set of criteria for the post-slaughter handling of ovine carcasses (MAF, 1991).

In applying the MIRINZ TFI method to the development of processing criteria, similar processes operating according to standards for Good Manufacturing Practice have been used as reference points. The range of TFI outputs were derived from existing processes of carcass cooling to a deep shoulder temperature of 10°C, with boning and subsequent cooling of the meat to 7°C. A criterion for the boning of meat whilst hot (hot boning) and the subsequent cooling of the meat was based on the range of values obtained from studying this 'cold boning' process. The criterion states that the TFI values of hot boning processes are not expected to exceed a mean of 7, an 80th percentile of 10 and a maximum of 14 where a TFI unit equals one generation of *E. coli* growth (Reichel et al., 1991). The EC 7°C deep meat temperature within 48 h of slaughter was used as the accepted GMP reference when developing the criteria for warm boning of beef.

By way of illustration, this paper discusses how TFI was applied in the development of criteria for the post-slaughter handling of ovine carcasses.

## 2. Predisposing factors

The 1990 commercial specifications for New Zealand export lamb to a substantial number of markets included standards for tenderness. The procedures that would satisfy these standards for tenderness required carcasses, after conditioning, to be aged by holding them after slaughter at temperatures not less than 6°C, for not less than 8 h. Although procedures for conditioning and aging lamb before freezing were available to the industry (Anon., 1973), this process required dedicated facilities and involved holding carcasses in a controlled environment for up to 96 h after slaughter.

Whilst the overall number of carcasses that were produced according to this specification during the 1970's was quite large, routine production would not have exceeded 5% of the lamb kill. However, with the introduction of the 1990 specifications greater than 45% of all export lamb carcasses would now require aging. In order to process this number of carcasses on a routine basis, substantial modification in the function of all current facilities used for holding carcasses immediately after slaughter would be required.

Routinely, the function of the post-slaughter holding facilities had been to assemble carcasses according to grades and hold them for a minimum of 90 min to comply with procedures for conditioning, branding and wrapping before being transferred to freezers or chillers. Many of these existing facilities had little or no temperature control, and the prospect of extending the holding period of carcasses in these facilities to comply with the procedures for aging presented a risk of bacterial proliferation.

The objective in developing a set of criteria for the post-slaughter handling of carcasses was to establish a range of times and temperatures at which carcasses might be held, that would satisfy the commercial

requirements for aging and not present a microbiological hazard.

## 3. Methods

Five time/temperature schedules (see Table 1) were selected as being representative of the current meat industry capability for commercial post-slaughter operations and which were capable of aging carcasses to a tenderness acceptable under the 1990 commercial specifications (Dr. B. Chrystall, MIRINZ, pers. comm., 1989).

Temperature histories were derived for the deep leg and leg surface of a PM grade lamb carcass using finite element models for dealing with regular shapes (Pham, 1988). The PM grade is a medium fat lamb in the weight range of 13.3–17.1 kg with a GR fat measurement of over 7 mm and up to and including 12 mm, where the GR is a fat content assessment based on measurement of the total tissue depth over the 12th rib at a point 11 cm from the mid-line of the carcass (Anon., 1995). From these derived temperature histories, carcass cooling curves were constructed for the first phase of cooling for each of 5 different biphasic cooling processes using air temperatures of 25, 20, 18, 15, and 10°C for 4, 6, 8, 12 and 18 h respectively.

An air temperature of 7°C was maintained in each process for the second phase of cooling. In all processes, the air flows were assumed to be 0.2 and 1.0 m/s for the first and second phases, respectively.

Temperature function integration was applied to the calculated temperature histories of the 5 different processes to determine the potential *E. coli* growth on the leg surface of each carcass.

Table 1
Number of predicted generations of *Escherichia coli* observed during aging of ovine carcasses using commercially acceptable time/temperature regimes

| Temperature (°C) | Time (h) | Generations/24 h |
|---|---|---|
| 25 | 4 | 9.8 |
| 20 | 6 | 10.3 |
| 18 | 8 | 10.7 |
| 15 | 12 | 10.5 |
| 10 | 18 | 7.6 |

## 4. Results

The cooling and potential *E. coli* growth curves for each of the 5 different processes are shown in Table 1 and Fig. 1.

For all processes, the calculated *E. coli* growth on the leg surface did not exceed 10.7 generations (range of 7.6–10.7). The process operating at a phase one temperature/time combination of 10°C/18 h resulted in a potential *E. coli* growth of 7.6 generations.

In each of the processes, the deep leg temperature was reduced to 7°C in less than 24 h (range of 14 to 22.5) of the carcass leaving the slaughterfloor. The deep leg temperatures of carcasses subjected to the higher temperature but shorter time combinations (20°C/6 h and 25°C/4 h) were reduced to 7°C sooner (14 h) than when the lower phase one temperature/longer time combinations were used.

## 5. Validation

The majority of lamb carcasses are currently being processed according to the 1990 specifications for aging (K. O'Grady, New Zealand Meat Producers Board, pers. comm.). As such, the microbiological consequences of this process may be assessed from a recent survey and compared with similar data from surveys of the previous (1973) conditioning and aging process.

Microbiological monitoring of carcasses in New Zealand meat works has been routinely performed since the early 1970's. Criteria, based on the Aerobic Plate Count incubated at 37°C ($APC_{37}$) have been specified for quality control of meat processing (Nottingham, 1974). For the period 1972 to 1977, the results of routine testing of the conditioning and aging process were collected and summarised in a report prepared as a background for a FAO/WHO working group on microbiological criteria for foods (Nottingham, 1979).

More recently, a survey of the results of routine microbiological testing of carcasses for APC's incubated at 25 to 30°C ($APC_{30}$) for the years 1993 to 1995 was done and the results, largely from aged lamb, provided a useful basis of comparison between the two different aging processes (Armitage, 1995).

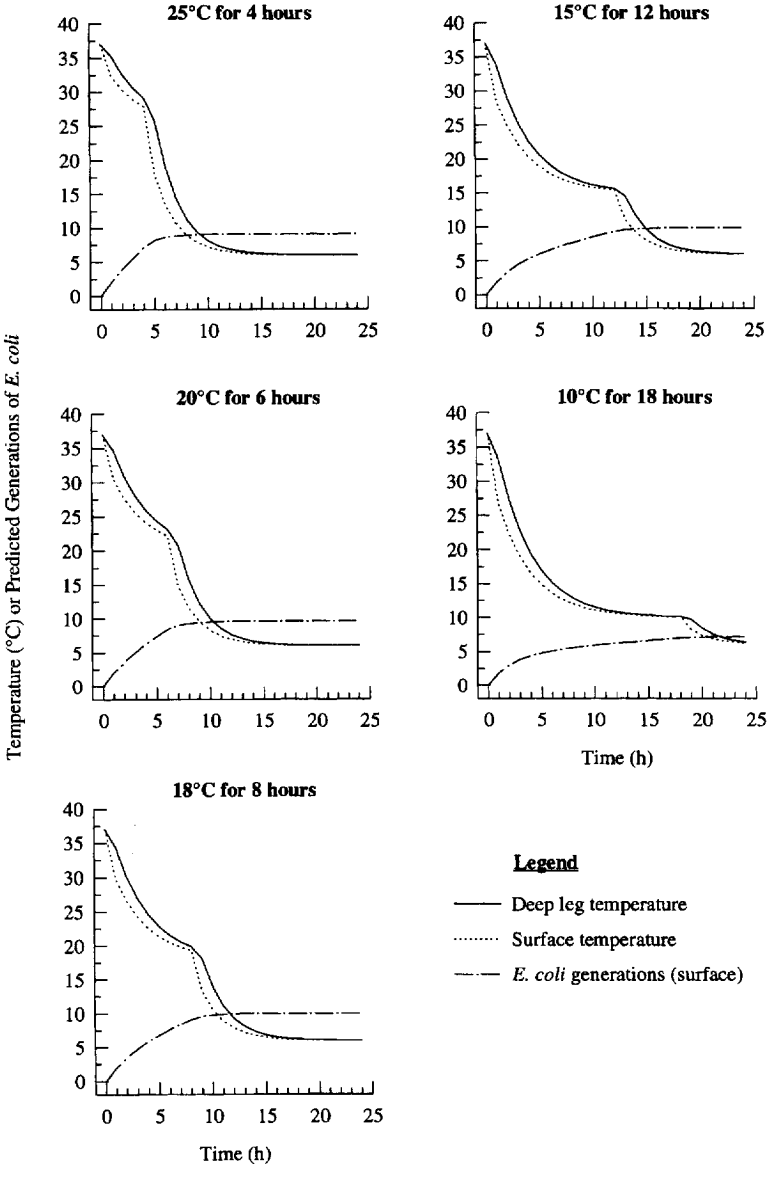


Fig. 1. Cooling profile and predicted *Escherichia coli* growth curves for ovine carcasses during aging.

The comparative results are presented in Table 2 and Fig. 2 Fig. 3. The microbiological consequence of aging, i.e., the increase in bacterial growth, resulted in a 151% increase (2.5 times the initial) in the mean $\log_{10} APC_{37}$ after processing according to the 1973 criteria, compared to a 38% (1.4 times the initial) increase in $\log_{10} APC_{30}$ when processing according to the 1990 criteria.

The 1990 process criteria appeared to have effected an improvement in the microbiological consequence of aging, with a 72% reduction in the mean counts ($APC_{37}$ vs. $APC_{30}$) compared to the 1973 process criteria. The actual difference might in fact be greater due to the lower plate incubation temperature now being routinely used in the industry and a factor in the 1995 survey. It should also be noted that there was a 50% reduction in the mean count on carcasses prior to aging.

Table 2
Comparison of 1979 and 1995 surveys of surface aerobic plate counts from ovine carcasses

| | | Carcasses after slaughter ($\log_{10}$) | Carcasses after aging ($\log_{10}$) | Mean change (%) |
|---|---|---|---|---|
| 1979 Survey ($APC_{37}$) | mean | 3.51 | 3.91 | 151% |
| | SD | 0.82 | 1.12 | |
| 1995 Survey ($APC_{25-30}$) | mean | 3.21 | 3.35 | 38% |
| | SD | 0.78 | 0.97 | |
| | % change | − 50% | − 72% | |

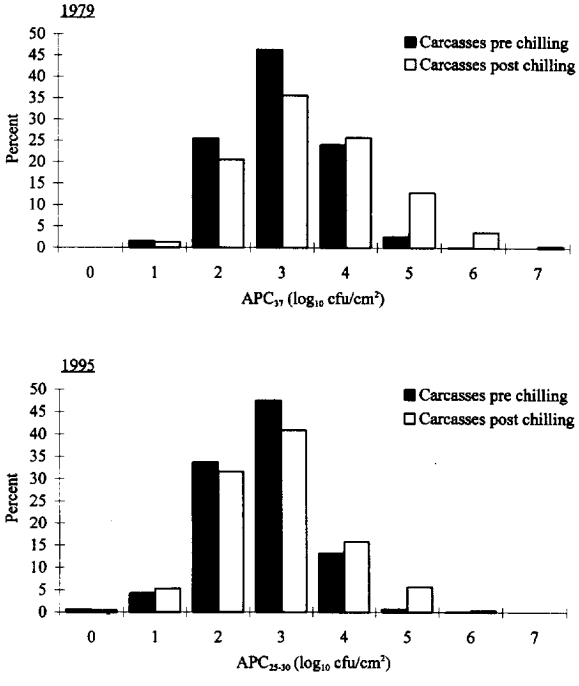


Fig. 2. Comparative distribution of bacterial counts on the surface of ovine carcasses: 1979 ($APC_{37}$) and 1995 ($APC_{25-30}$).

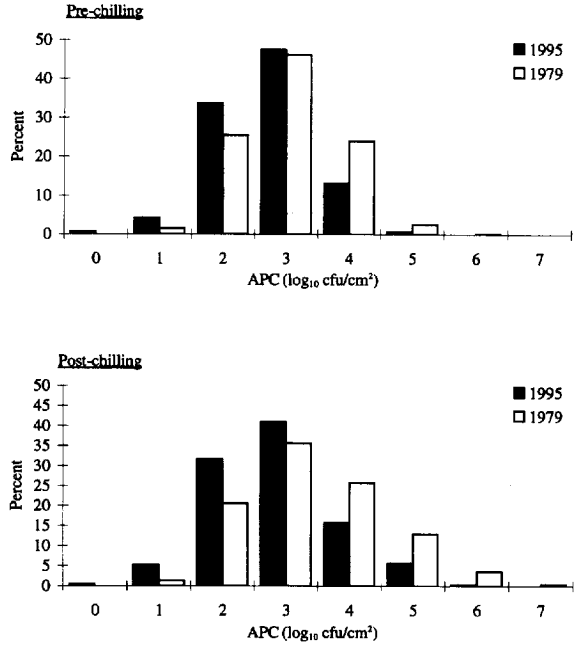


Fig. 3. Comparative distribution of bacterial counts on the surface of ovine carcasses pre- and post-chilling.

## 6. Discussion

### 6.1. Indexing utility of TFI

It has been customary for regulations to prescribe parameters for processing that may have been derived empirically. The exercise conducted by MAF, MIRINZ and industry, using TFI, determined that a set of proposed processing parameters that may have appeared to contradict hygienic principles could in fact have a microbiological consequence equivalent to an existing process.

The post-slaughter handling of ovine carcasses demonstrated that cooling processes employing relatively high temperature/time combinations in phase one of the process ( > 10°C/18 h) could be expected to reduce the deep leg temperature of the carcasses to 7°C more rapidly than the lower temperature/time combinations. All of the temperature/time combinations were capable of producing deep meat temperatures that would satisfy New Zealand and EC standards for carcasses intended for cutting the day after slaughter, and the four warmer temperature/time combinations would satisfy the cooling criteria specified by the EC for warm boning of ovines (7°C in 20 h).

The calculated potential *E. coli* proliferation for all

of the ovine processes were within the recommended guidelines for beef cooling, where the deep temperature of carcasses is to be reduced to 7°C or less within 48 h of leaving the slaughterfloor (Gill et al., 1991).

Therefore, based on the calculated temperature histories and the subsequent calculation of bacterial growth, the five carcass cooling processes were expected to have very similar hygienic consequences, and the calculated potential *E. coli* growth for the warmer temperature/shorter time combinations of phase one of the cooling processes were equivalent to a potential increase in bacterial numbers of 3 $\log_{10}$, i.e., 10 generations or doublings.

TFI, in this instance, was used to index processing parameters between different interventions in carcass cooling processes, and within a range of operating parameters for one process, to establish an equivalent outcome measured in TFI terms.

### 6.2. Actual growth versus index value

The TFI unit of measurement is one generation of *E. coli* growth. When TFI was applied to the cooling performance of different carcass cooling processes, a value of 10 generations (3 $\log_{10}$ potential growth) appeared to be common to both between and within process variations. However, the results from the 1995 survey did not demonstrate the 3 $\log_{10}$ increase in the aerobic plate count that might have been inferred by the TFI model.

This difference between the actual bacterial count and the index value may have resulted from an accumulation of several, but independently conservative elements that could affect the modelling process.

- An overall increase in mean APC's was observed although it did not exceed one generation. However, in contrast, at the 99.9th percentile ( + 3SD) the increase in APC's was larger and represented approximately 2.25 generations of growth.
- Whilst it might be possible for *E. coli* to increase by a factor of 3 $\log_{10}$ (say $-2$ to $+1$ $\log_{10}$) without a detectable change in the APC count, the process was not designed to select for mesophiles, i.e., the temperature parameters would be more likely to have potentially promoted growth of psychrotrophic bacteria. The APC results do not reflect any change in the composition of the flora that may have occurred during chilling.
- The temperature history was calculated for a PM grade of lamb. This type of lamb is moderately heavy with a heavy fat cover and represents approximately 20% of the total lamb kill. Sixty percent of New Zealand lambs are lighter or have less fat cover and would, therefore, be expected to cool more rapidly than the PM grade, with a consequential reduction in the rate of bacterial proliferation.
- Temperature function integration uses a model for *E. coli* growth that is only limited by temperature. The growth model makes no allowances for a reduction in available water that could be expected to occur as the surface of the carcass dries during cooling. Because the numerical increase in *E. coli* is dependent on moisture, the actual increase must be expected to be less than the predicted increase if any degree of surface drying takes place.
- The temperature/time schedules that suggested a potential 3 $\log_{10}$ *E. coli* proliferation reflected physical conditions that might occur during the warmest months of the year. The results of the 1995 survey represent two years production and include periods of the year where ambient temperatures were considerably less than the temperatures used in the TFI calculations.
- The TFI calculations must be considered to be conservative in that whilst good agreement can be demonstrated between observed and predicted values in vitro, in practice the observed value is frequently less than the predicted value (Gill and Harrison, 1985). In carcass cooling studies where the surfaces of microbial concern were uncovered, the correlation between predicted and observed *E. coli* counts was poor and numerous counts extending through the 1 $\log_{10}$ range were observed for a given predicted value (Phillips et al., 1996).
- Except for temperature, all other characteristics that could be expected to favour the growth of *E. coli* are also assumed to be present, including *E. coli* having a selective advantage in the presence of competing organisms.

The quantitative microbiological results from the 1995 survey demonstrated that the 1990 process criteria for the post-slaughter handling of ovine

carcasses results in carcasses that are no less hygienic than those produced from the previously accepted (1973) process.

## 7. Conclusions

Temperature function integration has provided a rapid, cost effective method of quantifying a temperature dependent process in terms of the potential for microbial proliferation. TFI, in its application to developing the post-slaughter handling of ovine carcasses, facilitated the determination of process parameters for a new process by comparing the potential for microbial proliferation with previously validated outputs.

However, the predictive model used in this instance could not take into account the numerous factors that influence microbial growth on foods, and thus could not be relied on to validate a process outcome in the absence of quantitative microbiology.

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