Results for today; Ideas for tomorrow

Predictive microbiology for comparing the performance of carcase chilling patterns

In the previous newsletter, 4/04 August 2004, we discussed various approaches that might be considered for validating carcase chilling, including the use of published literature, undertaking a program of microbial testing and using predictive microbiology. This newsletter focuses on the predictive microbiology approach. It outlines how models can be useful in various steps of HACCP, and describes how they can be used to assess the likely effects of changes to chilling procedures. It also explains some recently completed work on the water activity at the surface of carcases during chilling and how this might affect the modelling of microbial growth.

Predictive microbiology is a tool that can be used to evaluate the microbiological consequences of different food processing and handling procedures. Models allow environmental factors that can be measured reliably (e.g. temperature, pH, water activity) to become proxies for microbiological tests. Models that consistently predict microbial growth on carcase and meat surfaces are particularly useful in situations—like carcase chilling—where microbiological testing is unreliable because *E. coli* or other bacteria of concern are rarely detected.

Models are useful in HACCP for various steps, including:

Hazard analysis: models (particularly growth/no growth) can be used to show which microbial hazards will grow on carcases and meat products; and, if so, how fast they will grow, thereby helping to characterise the importance of the hazard;

Identification of CCPs: by defining the process in terms of parameters such as temperature, water activity and pH, it is possible to identify steps at which significant growth (or death) occurs, and whether critical control can be achieved and maintained;

Specification of limits: 'what if' scenarios can be performed for different chilling procedures, to see if alterations will allow new hazards to emerge or increase the risk of an existing hazard;

Specification of corrective action: if a loss of control occurs at a CCP, the change in microbial numbers associated with the process deviation can be quantified, and appropriate corrective steps specified.

Predicting the growth of *E. coli* on meat was undertaken by the CSIRO in the early 1980s and this work formed the basis for the first AQIS hot-boning meat orders. More recently, Dr Tom Ross and his colleagues at the University of Tasmania developed a model specifically for predicting the growth of *E. coli* on meat under Australian commercial practice. They have validated that the model reliably predicts the effects of temperature, pH, water activity and lactic acid concentration on the growth of *E. coli*. The model also takes into account the period of time (lag time) before *E. coli* that are transferred to a newly exposed carcase surface, begin to grow. At present the algorithm does not account for reductions in numbers of bacteria during chilling. The significance of this will be mentioned shortly.

Before we consider the specific ways that predictive microbiology can be applied, it is worth reviewing the behaviour of bacteria at chilled temperatures higher than 7°C.

Chilling carcases: effects on bacteria

Temperature has a major influence on whether bacteria present on meat can increase in numbers. The growth of pathogenic bacteria, such as *Salmonella* and certain strains of *E. coli*, on the surface of a beef carcase is also influenced by the water activity (a_w) of the surface and by the pH.

It is generally accepted that cooling meat to, and holding it at, 7°C or below will assure no growth of *E. coli*, *Salmonella* or several other pathogens, even on carcases held over weekends. Published literature is available to validate that 8°C is the minimum temperature for their growth on meat in practical situations.

The literature also supports the viewpoint that while chill temperatures slightly higher than 8°C may allow some growth of

pathogens, the rate of growth is very slow. For example, under ideal conditions of nutrient and water supply (as in laboratory growth media), *E. coli* O157:H7 and other *E. coli* may double in numbers in less than half an hour at 35–40°C, but may take up to 10 hours to double at 10°C. Importantly they will also take much longer to adapt to the new conditions at the lower temperature, and it may be over 50 hours before they even begin to grow. In contrast, the lag at 37°C could be as little as 90 minutes. If nutrient and water availability are less than optimal at 10°C, then the lag time will be even longer and the rate of growth, once growth begins, will be slower.

Whether temperatures above 7°C are safe, therefore, depends on the length of time involved and the specific conditions applying. For beef sides or smallstock carcasses chilled overnight, boned, packaged, then promptly chilled or frozen, there will probably be no detectable growth of *E. coli* if the carcase holding temperature is 10°C instead of 7°C. Whether there is growth on carcases held over weekends at 10°C and boned on Mondays will depend on the relative humidity in the chiller, whether carcases need to be rewarmed, and whether some of the bacteria have been injured by chilling or other interventions.

Various studies in Australia and overseas have shown that numbers of *E. coli* decline during chilling of carcases. Reductions of two logs (100 fold or 99%) have been reported, but it appears that reductions of around 0.5 logs (or three-fold) commonly occur. There may be circumstances when reductions are smaller; however, there is clear evidence that effective chilling generally does more than prevent increases in numbers of pathogens: it decreases numbers. It has been suggested that both reduced water activity at the surface and 'cold shock' may contribute to the inactivation effect of chilling. The available predictive equations do not yet account for reductions in water activity during chilling, although it may be possible to modify them to do so in the near future. The fact that reductions in numbers are likely to occur means that predictions of increases—using the current models that do not account for such reductions—are likely to be quite conservative in that they will be over-estimates.

How does predictive modelling work?

Microbial growth models employ complex equations that are developed from actual test results obtained under a range of conditions. They are then validated under the conditions where they are to be used. In the case of carcase chilling, where temperature, pH, water activity and other relevant variables change during the chilling cycle, a spreadsheet developed from the UTAS model takes inputs of temperature and other variables at short intervals (15 minute intervals are commonly used) and computes values at the end of chilling that are predictions of the likely growth of the microorganism in question.

In the 'new EMOs', the log value of the predicted growth is termed the refrigeration index (RI). The acceptable criteria for RIs are that the average RI for the chilling process is no more than 1.5; 80% of RIs are no more than 2.0; and no RI is more than 2.5. In this context, the chilling process is regarded as including carcase chilling plus cooling

Regulatory requirements for chilling

Australian Standard 4696-2002 'Australian standard for the hygienic production and transportation of meat and meat products for human consumption' includes requirements for cooling carcases and carcase parts. It states that chilling and freezing applied to carcases and carcase parts must meet either: requirements that are specified in the standard; or alternative time and temperature controls.

The defined requirements are that within 24 hours of slaughter, the refrigeration must be capable of cooling carcases, sides, quarters and bone-in major cuts to 7°C on all their surfaces, and for any other carcase part to 5°C at the site of microbiological concern. In the case of cartoned meat, the site of microbiological concern is the thermal centre of the carton rather than the surface.

Alternatively, the refrigeration must achieve time and temperature controls that are specified in an arrangement that is approved by the appropriate regulatory authority. The Export Control (Meat and Meat Products) Orders 2004 ('new EMOs') also provide for approved arrangements that specify alternatives to chilling to 7°C or 5°C for carcase parts.

There are various reasons why plants might wish to gain regulatory approval to introduce carcase and/or carton-chilling procedures other than those specified in section 11.6 of the standard. The most likely one is to reduce the OH&S difficulties associated with boning carcases with hard fat. It is sometimes necessary to rewarm beef carcases with heavy fat cover for a short time, in order to soften the subcutaneous fat before boners and slicers are able to process them safely.

Rewarming or any departure from the AS 4696 requirement to cool carcases and meat to 7°C, and maintain them at or below 7°C, requires an alternative arrangement.

cartons of product below the minimum temperature for growth of *E. coli*—which is 8°C.

Through predictive modelling of the growth of *E. coli*, it is possible to understand the relative effects of carcase surface temperature, water activity (a_w) and chilled holding time on the ability of *E. coli* to grow under different chilling regimes. Comparisons can be made using several different surface-cooling profiles and water-activity profiles. For example, it is relatively easy to consider changes in the following situations: overnight chill, weekend chill, and re-warming before boning.

Where to measure

For the predictions to be reliable, the input variables must reflect what is actually happening on the carcase surface during chilling. We recommend that probes for measuring surface temperature of carcases be located at the point end of the brisket or on the outside of the neck. Although the surface over the butt will cool more slowly than over the brisket, neck or some other surfaces (see Figure 1), not only is it difficult to access, it is consistently drier and, as will be explained

shortly, growth of bacteria is likely to be slower over the butt at any given point in time.

The surface temperature should be recorded using a temperature logger with a thin probe (approx. 3 mm diameter) threaded under the surface tissue of the carcase so that the sensing tip is no more than 1 mm under the surface (Figure 2).

The surface water activity (a_w) is a measure of surface dryness of carcases. The water activity of pure water is 1.000; that of fresh meat is usually taken as 0.993–0.995. As a_w falls, bacteria find it more difficult to grow. During the fast-chill phase, a_w of surfaces of beef carcases regularly falls to 0.980 or below, with some surfaces—including the surface over the butt—frequently falling to 0.960 or lower. At these a_w levels and, in the temperature range to which the surfaces have fallen by this time (near 10°C), the rates of growth of *E. coli* and other pathogens and spoilage bacteria are known to be greatly reduced.

Predictive microbiology models, in which the decline in a_w can be accounted for, show that the potential growth of *E. coli* is generally less over the butt than over the neck or brisket, even though the rate of cooling might be slower. This is because the water activity falls further over the butt (as stated above).

With the progression to the holding phase of chilling, the temperature and RH of the air rise and the air velocity generally falls. As a result of this, the surface a_w of carcases rises to 0.980 or a little higher after
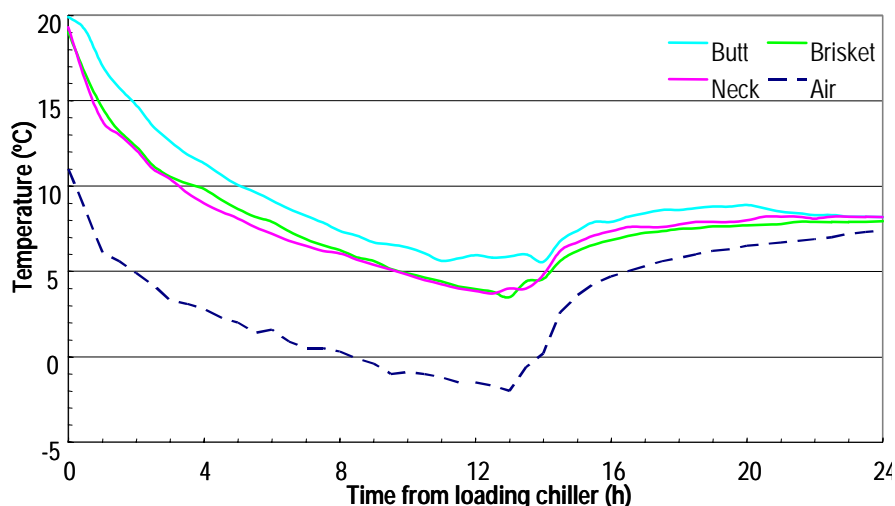

Figure 1. Typical beef carcase surface and air temperatures during chilling

Figure 2. Measurement of carcase surface temperature

around 14 hours of chilling. From limited data, it seems that it will gradually rise further to 0.985–0.990 after 24 h. It is possible to incorporate the surface a_w into the UTAS model at short intervals (e.g. of 15 minutes), in the same way as temperature. Currently, though, there isn't a means of conveniently logging a_w throughout a chill. For the spreadsheet, a constant a_w is used.

However, there is a good relationship between a_w and the average relative humidity (RH) during the first several hours of chilling. The RH of the chiller air influences the a_w. The drier the air, the lower will be the water activity. RH varies from chiller to chiller, depending on the specifications of the refrigeration equipment. It also varies during the course of overnight chilling of carcases, and is at its lowest a few hours after commencement of chilling, during the initial fast-chill phase. Once chilling progresses to the holding phase, both the temperature and the RH of the air rise (Figure 3). In the case of the brisket (which normally has the highest a_w at any given time during chilling), the a_w will fall to 0.980 or lower, provided that the RH is kept below 87% for around 4 hours or longer. It is hoped that ongoing investigations will lead to a_w and perhaps meat pH being included as variables.

Examples of alternatives

Predictive modelling provides a convenient way of determining the significance of modifications to a chilling procedure—rewarming carcases for a period of two or three hours to reduce the problem of hard fat, for example. The likelihood of growth of *E. coli* will depend on the state of the cells at the start of rewarming, the actual duration of rewarming, and the temperature reached; but the effect on the refrigeration index can be shown to be around 0.4 units. Similarly, the effect of holding carcases for all, or part, of a weekend at a temperature slightly higher than 7°C can be predicted. Predictions of *E. coli* growth made recently using temperature and water activity histories obtained some years ago indicate that beef sides in those chillers can be held at 10°C over a weekend, but if there needs to be a period of rewarming prior to boning, the holding temperature should be lowered slightly.

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Limitations

There are some limitations of predictive microbiology that need to be considered. The models cannot be extrapolated outside the ranges (for temperature, a_w, pH etc.) over which they were derived. Predictions outside the experimental ranges are usually inaccurate and may sometimes be nonsensical. As mentioned earlier, the models may predict growth rates that are faster than observed rates, particularly if they are developed from results of tests conducted mainly in laboratory media. This makes them fail-safe, but they may be overly conservative. You should have evidence that the model you select has been appropriately validated for meat surfaces.

In December 2003, FSIS issued Notice 50–03 that provides information about microbial pathogen computer modelling. The notice also contains guidance material about the role and limitations of modelling programs. You should be aware of the FSIS position that it is not yet possible to rely solely upon a predictive modelling program.

As stated earlier, rewarming or any departure from the AS 4696 requirement to cool carcasses and meat to and maintain them at, or below, 7°C requires an alternate arrangement. The predictive microbiology approach is a good way to assess and validate alternative procedures that might be adopted in approved arrangements.
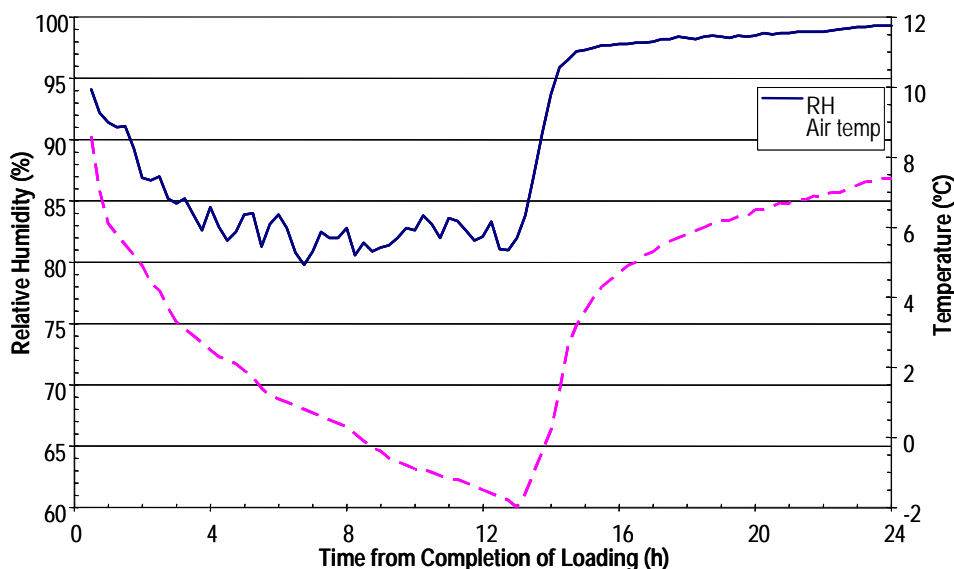

Figure 3. Typical record of relative humidity and air temperature

Further reading

Ross, T. (1999) Predictive Microbiology for the Meat Industry. Meat and Livestock Australia.

Sumner, J., Krist, K. (2002) The use of predictive microbiology by the Australian meat industry. Journal of Food Protection 73: 363–366.