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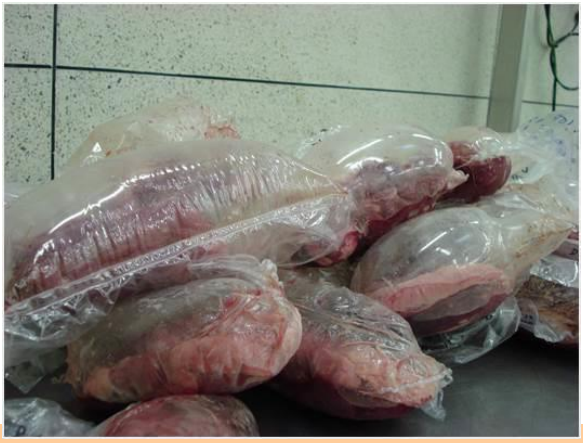
Packaging and chilling technologies to enhance meat quality and safety



Key external stakeholders:
Irish beef farmers, beef processors, FSAI, DAFM, public health personnel, epidemiologists and scientists interested in beef microbiology, food safety and spoilage.

Practical implications for stakeholders:

- Hot/warm boning promotes blown pack spoilage and the survival of key pathogens (*Salmonella* and *E. coli* O157) on beef carcasses during chilling. ComBase software may be used to accurately predicted *Pseudomonas* spp. and *Br. thermosphacta* growth on beef carcasses and primals. A real-time PCR technology was developed that can low levels of blown pack spoilage Clostridium spp. (*C. estertheticum, C. gasigenes* and *C. ruminantium)* on equipment and meat samples.

Main results:

The main results were:

- Bacterial counts on beef primals increased to 6-7 $\log_{10}$ cfu cm^{-2} after 6 weeks chilled storage.
- Significantly higher TEC, *Pseudomonas* spp. and *Br. thermosphacta* counts were observed on cold boned primals *versus* hot boned samples.
- BPS pack distension or bursting occurred considerably sooner in hot boned product.
- Any decrease in pathogenic bacteria during beef chilling may be significantly less for hot boned beef depending on the bacterial strain.

Opportunity / Benefit:

This project characterised beef carcass chilling in terms of the physical parameters (temperature, relative humidity, pH and aw) and microbiology (total viable count (TVC) mesophilic (TVCm, 30°C) and psychrophilic (TVCp, 6°C), total *Enterobacteriacae* counts (TEC), *Pseudomonas* spp., *Clostridia* spp., Lactic acid bacteria (LAB) and *Brochotrix thermosphacta*). The data generated showed that significantly (P<0.05) higher TVC, LAB and *Clostridium spp*. concentrations were obtained on hot boned beef and that BPS pack distension or bursting occurred considerably sooner in hot boned product. Thus the beef sector should carefully review these findings before considering using hot boning as an alternative to current practices. This project also developed and validated a set of real-time PCR assays, capable of detecting 4-5 *C. estertheticum,* 2 *C. gasigenes* and 8 *C. ruminantium* spores per ml/cm^2 and transferred this technology to the Irish beef industry via the 'Blown Pack Spoilage' testing service at Ashtown.

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Collaborating Institutions:

University College Dublin (Professor Seamus Fanning & Professor Paul Whyte) and University College Cork (Professor Joseph Kerry).

Teagasc project team: Dr. Declan Bolton (PI)

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1. Project background:

Hot/warm boning offers significant cost saving opportunities for the Irish beef industry. While the improvements in meat quality and yield have been scientifically proven, information on the microbiological aspects of this technology is lacking. The elevated storage temperatures encountered during hot/warm boning could support the growth of spoilage and pathogenic bacteria. Moreover, microbial contamination has long been recognised as a primary source of spoilage and reduction of meat quality. Blown pack spoilage (BPS) is of particular concern as hot/warm boning could further exacerbate an already serious issue.

2. Questions addressed by the project:

- The lack of data on the microbiology of beef carcasses during chilling.
- Information on the microbiology of hot/warm boning versus conventional chilling, specifically; does hot/warm boning promote the growth of spoilage and pathogenic bacteria on beef?
- The current inability to detect low concentrations of BPS spores.

3. The experimental studies:

In a commercial abattoir, total viable count (TVC), total *Enterobacteriacae* count (TEC), *Pseudomonas* spp., lactic acid bacteria (LAB), *Brochotrix thermosphacta* and *Clostridum* spp. were monitored on beef carcasses (n=30) and primals (n=105) during chilled storage using EC Decision 2001/471/EC and ISO sampling/laboratory procedures. The surface and/or core temperature, pH and water activity (a_w) were also recorded.

To investigate hot versus cold boning on the risk of bacterial growth and BPS, primals were prepared from M*. Longissimus thoracis et lumborum*, M. *psoas major*, M. *quadriceps* and M. *semitendinosus* muscles from cold and hot boned carcasses, vacuum-packaged and stored for 42 or 100 days at 2°C and 7°C. Storage temperature, carcass or primal surface temperature, pH and a_w were monitored. Samples were taken periodically and tested for total viable count mesophilic (TVCm), TVC psychrophilic (TVCp), total *Enterobacteriacae* count (TEC), *Pseudomonas* spp., lactic acid bacteria (LAB), *Clostridium* spp. and *Brochothrix thermosphacta.* A fifth primal, M. *biceps femoris*, was used to examine the impact of hot boning on blown pack spoilage (BPS).

To investigate the fate of *Salmonella* Typhimurium and *Escherichia coli* O157 on hot boned versus conventionally chilled beef, samples were individually inoculated with *S.* Typhimurium ATCC 14028, *S.* Typhimurium 844, *E. coli* O157 EDL 933 or *E. coli* T13. Half the samples were subject to the same time-temperature chilling profile used for conventionally chilling beef carcasses while the other half was subject to hot boned conditions. Duplicate samples were removed periodically and tested for TVC, TEC, *Salmonella* and/or *E. coli* O157. pH and aw were also monitored.

Finally, a set of real-time PCR assays for the detection of *C. estertheticum, C. gasigenes* and *C. ruminantium*, the causative agents of blown pack spoilage (BPS) in vacuum packaged beef, was developed. Robust validation of the sensitivity and specificity was carried out in the three matrices (beef meat drip, wet environmental swabs and dry environmental swabs) as encountered in our testing laboratory and against *Clostridium* strains (n= 76) and non-*Clostridium* strains (n=36).

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4. Main results:

Over the course of the experiment the surface temperature decreased from 37°C to 0°C, pH from 7.07 to 5.65 and a_w from 0.97 to 0.93. *Clostridium* (1.89 $\log_{10}$ cfu/cm^2) and *Pseudomonas* spp. (2.12 $\log_{10}$ cfu/cm^2) were initially the most prevalent bacteria on carcasses and primals, respectively. The shortest mean generation time (G) was observed on carcasses with *Br. thermosphacta* (20.3h) and on primals with LAB (G = 68.8h) and *Clostridium* spp. (G = 67h). The observed *Pseudomonas* spp. and *Br. thermosphacta* growth was more or less within the range of predictions of Combase. In contrast, the FSSP completely overestimated the growth of LAB. This study contributes to the very limited microbiological data on beef carcasses and primals during chilling. Primal counts increased to 6-7 $\log_{10}$ cfu cm^{-2} after 6 weeks. Significantly (P<0.05) higher TEC, *Pseudomonas* spp. and *Br. thermosphacta* counts were observed on cold boned primals *versus* hot boned samples. In contrast, significantly (P<0.05) higher TVC, LAB and *Clostridium spp*. concentrations were obtained on hot boned beef. Moreover, BPS pack distension or bursting occurred considerably sooner in hot boned product.

The surface pH (5.5) and a_w (0.95 to 0.97) were stable. *S.* Typhimurium and *E. coli* O157 counts, which decreased by up to 1.0 and 1.5 $\log_{10}$ cfu cm^{-2}, respectively, were statistically similar (P > 0.05), regardless of the chilling regime applied, with the exception of *E. coli* O157 EDL 933, where the counts on hot boned beef were significantly (P < 0.05) higher. It was concluded that any decrease in pathogenic bacteria during beef chilling may be significantly (P < 0.05) less for hot boned beef depending on the bacterial strain.

It was possible to detect 4-5 spores per ml for *C. estertheticum,* 2 spores per ml for *C. gasigenes* and 8 spores per ml for *C. ruminantium,* without the need for enrichment of the samples. This high sensitivity is particularly important for the beef sector, not just for testing spoiled product but also in the early detection of contaminated beef and in validation of sporicidal cleaning procedures for critical pieces of equipment such as the vacuum packaging machine, which have the potential to contaminate large volumes of product.

5. Opportunity/Benefit:

This project provided data on the microbiology of beef carcasses during chilling and an evaluation of hot/warm boning which should inform the Irish beef industry when considering applying this technology. The real time PCR assay will allow the beef industry to, for the first time, evaluate the sporicidal cleaning and disinfection procedures currently applied and to revise these if necessary.

6. Dissemination:

We organised a Beef Industry Workshop (Packaging and Chilling Technologies to Enhance Meat Quality and Safety; FIRM 11/F/033) on Tuesday 6th December 2016, Training and Conference Centre, Teagasc Food Research Centre, Ashtown, Dublin 15. The findings of this project were also disseminated through oral and poster presentation at several conferences including;

- Rachel Reid, Roland Lindqvist, Seamus Fanning, Paul Whyte, Joe Kerry and Declan Bolton (2015). The microbiology of beef chilling: do the observations fit the predictions?, Poster 189, Abstract book page 111, poster presented at the EFSA Expo conference 'Shaping the future of food safety together', Milan, Italy 14th to 16th October 2015.

- Rachel Reid, S. Fanning, P. Whyte, J. Kerry and D.J. Bolton (2016) An investigation of the time to onset of blown pack spoilage in hot and conventionally boned beef. Poster

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presentation at the FOOD MICRO International Conference, University College Dublin, 19th to 22nd July 2016, Abstract book, page 440.

- Rachel Reid, Roland Lindqvist, Seamus Fanning, Paul Whyte, Joe Kerry and Declan Bolton (2016). The microbiology of beef chilling: do the observations fit the predictions?, Poster 214, Abstract paper no. 914, poster presented at the 18th World Congress of Food, Science and Technology (IUFoST International 2016), RDS, Dublin, 21st-25th August 2016., Abstract Book, page 904.

And through peer review publications;

- Reid, R., Fanning, S., Whyte, P., Kerry, J., Lindqvist, R., Yu, Z. and Bolton, D. J. (2017) The microbiology of beef carcasses and primals during chilling and commercial storage. Food Microbiology, 61, 50-57.

- Rachel R., Fanning, S., Whyte, P., Kerry, J., Bolton, D. J. (2017). The effects of hot versus cold boning of beef carcasses on bacterial growth and the risk of blown pack spoilage. Meat Science, 125, 46-52.

- Rachel R., Fanning, S., Whyte, P., Kerry, J., Bolton, D. J. (2017). An investigation of the effect of rapid slurry chilling on blown pack spoilage of vacuum packaged beef primals. Letters in Applied Microbiology, 64, 2, 177-181.
- Rachel R., Fanning, S., Whyte, P., Kerry, J., Bolton, D. J. (2017). The fate of *Salmonella* Typhimurium and *Escherichia coli* O157 on hot boned versus conventionally chilled beef. Meat Science, 126, 50-54.
- Rachel R.,, McCabe, E., Fanning, S., Whyte, P., Kerry, J., Bolton, D. J. (2017). Real-time PCR methods for the detection of blown pack spoilage causing Clostridium species; *C. estertheticum*, *C. gasigenes* and *C. ruminantium*. Meat Science, 133:56-60.

7. Compiled by: Dr. Declan Bolton

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