

final report

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Prepared by: MLA

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Technical support on the application of < 82°C water for knife and equipment sterilisation

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1 Background

For many decades it has been the practice in abattoirs for operators to clean their knives between carcases, and when necessary, by dipping them in baths ("sterilisers") containing water no cooler than 82°C.

In 2003 MLA commissioned Food Science Australia (FSA) to investigate firstly, whether there was any scientific basis for 82°C and secondly, whether alternative cleaning procedures were possible using water cooler than 82°C. After an exhaustive literature review the researchers were unable to find any scientific evidence for the selection of 82°C. They also demonstrated that temperatures cooler than 82°C could be used providing knives were immersed for longer than the momentary dip currently used.

In 2004 MLA worked with M.C. Herd Pty Ltd of Geelong to validate an alternative system based on operators using two knives which were immersed in a water bath at 60°C for a longer immersion time. At Herd's the alternative system gives a hygienic outcome at least the equivalent to that of 82°C and there are cost savings:

- Reduced energy use at 60°C
- Reduced impact of injuries at 60°C

The work done at M.C. Herd was fully considered by Meat Standards Committee (MSC) which agreed, based on an assessment made by Food Science Australia (FSA), that the proposal submitted by MC Herd Pty Ltd demonstrates equivalence to the outcome achieved using hot water at a temperature of 82°C for the sanitisation of knives as required by the Australian Standard for the Hygienic Production and Transportation of Meat and Meat Products for Human Consumption.

MSC also agreed that all jurisdictions would approve future proposals for the use of hot water at a temperature cooler than 82°C on the following basis:

a. subject to a scientific assessment provided by FSA confirming equivalence of the proposal to use hot water at a temperature of 82°C for the sanitisation of knives, pending development of a model for time-temperature equivalence;

or

b. following development and approval of the model, verification of the use of the model by meat processing establishments to demonstrate equivalence;

and

c. approval of an arrangement that demonstrates the capacity of the meat processing facility to operate in accordance with the proposal submitted to FSA subject to importing country requirements.

In late-2006, trials conducted at three very small plants (VSPs) further demonstrated the equivalence of lower temperature water for knife sanitising and showed how this alternative procedure can be implemented in a small operation. With scientific advice from Food Science Australia, the controlling authorities in three states approved the use of water cooler than 82°C following assessment of the way knives are used in each operation. The controlling authorities referred to the FSA model (see below) and did not require further microbiological validation.

In 2007, FSA completed development of a model for assessing the effectiveness of using hot water cooler than 82°C for sanitising knives. The model has been presented to MSC and can now be used in demonstrating equivalence.

Meat Standards Committee, at a meeting in June 2007, agreed that all jurisdictions would approve future proposals for the use of hot water at a temperature cooler than 82°C on the following basis:

a. verification of the use of the model by meat processing establishments to demonstrate equivalence;

and

b. approval of an arrangement that demonstrates the capacity of the meat processing facility to operate in accordance with the proposal submitted to the controlling authority;

and

c. subject to importing country requirements.

In order to help you decide whether such an approved arrangement will be valuable to your company, we have published this short booklet to present:

- The scientific basis for using alternative knife cleaning systems.
- Technical aspects of putting this into practice.
- An explanation of the model you can use to validate your alternative process.
- The type of information regulatory authorities will need to consider your alternative procedure.

2 The science underpinning knife cleaning

The Australian Standard (4696:2007) specifies that facilities for cleaning and sanitising implements be provided with an adequate supply of hot potable water at no less than 82°C or receive an equivalent method of sanitising. Traditionally, the practice has been for all meat processors to provide water at 82°C or warmer.

Not only are the origins of 82°C/180°F unclear, it is customary when temperatures are specified e.g. 65°C (cooking), 72°C (pasteurisation), and 121°C (sterilisation e.g. canning) to specify an effective treatment time. In the case of knife dipping no treatment time is stipulated and no reason is given for using water of 82°C/180°F.

Publications earlier than 1960 refer to a number of equipment cleaning procedures e.g. water at140°F for one minute or at 130°F for 5 minutes. In their search for the scientific basis for 82°C water use, Food Science Australia (FSA) staff contacted their colleagues at the United States Department of Agriculture. It is believed that, in the 1950s, Dr Sloan, working for the USDA Agricultural Research Service (ARS) investigated methods of sterilising carcass-splitting saws. Sloan found that dipping the carcass splitting saws in 180°F water effectively killed sufficient numbers of organisms to satisfy regulatory requirements.

Eventually 180°F water became the standard for all slaughter floor operations, with 82°C its metric equivalent. Another explanation is that the 180°F requirement was based on the heat resistance of the bacterium which caused tuberculosis, *Mycobacterium tuberculosis*, an important target organism in milk and other foods back in the 1950s.

So there appears to be no clear scientific basis for the historical international focus on 82°C/180°F as a disinfection temperature. Momentary exposure to 82°C is not sufficient to ensure a bactericidal (killing) effect (for example, brief immersion of knives at 82°C is ineffective in killing all salmonellas). The effectiveness of a "dip" into 82°C water depends on a number of variables:

Thermal inertia of the equipment prevents surfaces attaining the water temperature until several seconds have elapsed.

When fats or proteins are present on a stainless steel plate, immersion at 82°C for as long as 10 seconds will not give large reductions in bacterial contamination i.e. surfaces must be washed first to get a suitable reduction.

Hot water at 82°C will "fix" (glue) proteins onto the surface of the knife leading to entrapment of bacteria and ineffective cleaning.

3 Trials undertaken by Food Science Australia

Meat & Livestock Australia (MLA) commissioned FSA to examine the potential benefits of using knife cleaning treatments other than 82°C. The researchers noted numerous benefits:

- Reduced risk of operator injury (scalds etc)
- Reduced hot water consumption, particularly by knife sterilisers
- · Less water, particularly hot water, going to effluent ponds
- Savings in energy costs for heating
- Reduced fogging and condensation
- Potential reduction in maintenance requirements

3.1 Laboratory trials

FSA researchers based their work on the fact that the lethal effect of hot water is a function of temperature and contact time – the longer the contact time, the lower the temperature which will be effective. They did a series of preliminary experiments in the laboratory where they immersed knives for different temperatures and times and measured the effectiveness of the treatment on killing bacteria on the knife. They also coated knives with meat and with fat to simulate abattoir conditions. They found that 72°C/15 seconds or 75°C/10 seconds were equally effective as 82°C/10 seconds.

3.2 In-plant trials

Trials were undertaken in an establishment at stations where operators had access to free-flowing handwash water in order to pre-wash knives before immersion. The stations chosen were the first (hind) leg stand, heads dressing, evisceration and fat trimming. Data loggers were used to monitor steriliser temperature, which was maintained at no cooler than 72°C. The researchers counted bacteria on knives before pre-washing, after pre-washing and after immersion in the steriliser. The effectiveness of pre-washing and hot water immersion can be seen from Figures 1 and 2.

The total viable count (TVC) on knives in use varied along the beef processing line, being higher at earlier operations. From Fig 1 it can be seen that pre-washing in hand wash water had a great effect, particularly on knives bearing high bacterial loadings (1st leg and head stations) and immersion in 72°C water for 15 seconds further reduced loadings to insignificant levels.

Levels of E. coli on knives in normal use were also monitored (Fig 2). Pre-washing was again extremely effective at removing the organism with immersion at 72°C for 15 seconds completing the elimination of E. coli except at the 1st leg station where E. coli survived the knife cleaning process.

The researchers concluded that pre-washing in hand-wash water then holding knives at lower temperatures for sufficient time was just as effective as dipping in 82°C water. This finding led to further work in the abattoir.



Figure 1: Efficacy of reducing microorganisms (TVC) by pre-washing naturally -contaminated knives in handwash water then disinfecting them in hot water sterilisers at 72°C for 15 s



Figure 2: Efficacy of reducing E. coli by pre-washing naturally-contaminated knives in hand-wash water then disinfecting them in hot water sterilisers at 72°C for 15 s

4 Establishing an alternative knife cleaning procedure in an abattoir

In 2004-05 M. C. Herd used the findings of the FSA research as a basis for establishing an alternative procedure for knife cleaning. The company's QA staff, assisted by MLA, carried out microbiological analysis to determine whether immersing in 60°C water for sufficient time could lead to an outcome equivalent to momentary immersion in 82°C water.

The company proposed a two-step trial to the controlling authority:

Step 1: Undertake a line survey to establish baseline contamination loadings on knives after 82°C treatment

Step 2: Establish whether water cooler than 82°C could produce an equivalent outcome by extending the immersion time

A baseline was established for the microbiological quality of knives after cleaning by the industry standard of momentary dipping in 82°C water in "sterilisers".

An equivalent procedure was trialled using a two-knife system of knife cleaning at 60°C in which one knife remained in the "steriliser" for the period while the other knife was used on the carcase. The operator exchanged knives between carcases. It should be emphasised that, during the trial, the company exercised extreme caution and ensured that, after knives had been tested by the researchers, they also received a final 82°C dip before being used on product.

Testing was carried out at each station on the beef and mutton floors (Fig 3). On the beef floor (Fig 4) prevalence of *E. coli* was similar after cleaning for both cleaning systems (10% for 82°C versus 9.5% for 60°C) and the mean TVC was lower using 2 knives and 60°C water (158/cm² versus 60/cm²).

On the mutton floor (Fig 5) mean TVCs were almost identical for the two knife cleaning systems. Although, prevalence of *E. coli* was slightly higher using the 2-knife system at 60°C (22% versus 19%) the difference is not significant statistically.

The results of these trials were published in the International Journal of Food Microbiology.1

The company used the microbiological comparison of the current versus the alternative procedure to demonstrate the equivalence of their alternative procedure, and this was the basis for a successful application to the Meat Standards Committee.



Figure 3: Location of stations at which knives were tested



Figure 4: Bacterial loadings on cleaned knives in use on the beef floor after 82°C and 60°C treatment. Mean total viable counts/cm² are in the top chart and the prevalence of E. coli on the lower chart



Figure 5: Bacterial loadings on cleaned knives in use on the mutton floor after 82°C and 60°C treatment. Mean total viable counts/cm² are in the top chart and the prevalence of E. coli on the lower chart

5 Trials conducted in Very Small Plants (VSPs)

A project was conducted to validate alternative time and temperature regimes for effective knife sterilisation in smaller meat processing plants. Observations in very small plants (VSPs) indicated operators typically carried out many operations between knife cleaning and intuitively it appeared VSPs might benefit from a 2-knife system since the residence time of the knife would be at least 30 seconds.

VSPs in Tasmania, South Australia and Queensland were visited to assess the process and possibility of running a suitable experimental trial. One VSP was selected in each State and data collected in October-November 2006.

Methodology

Alternative procedures tested involved comparing steriliser water temperature around 60°C for longer contact time of the knife in the steriliser unit using a two-knife system with the current procedure in which knives were dipped momentarily in 82°C water.

Five knives were tested at each station by the same sponge method as was used in the earlier plant trial. Samples were transported to a NATA accredited laboratory for testing.

Estimates of the time taken to perform slaughter floor procedures and the residence time of knives in each steriliser were made.

Results

Tables 1 and 2 summarise the results for knives for *E. coli* and TVC in all three establishments, for beef, sheep and overall.

Table 1: Number of positive E. coli samples ≥ 0.25 cfu/cm² divided by the total number of knives sampled on beef and sheep slaughter & dressing floors

	Beef		Sheep	
	≥82°C	≥60°C	≥82°C	≥60°C
Queensland	6/25*	0/25	1/25	1/25
Tasmania	10/20	9/20	3/20	1/20
Sth Australia	3/25	6/25	10/20	2/20
Total	19/70	15/70	14/65*	4/65

*Significant P<0.05 (Chi-square test)

	Beef		Sheep	
	≥82°C	≥60°C	≥82°C	≥60°C
Queensland	1.49	1.46	1.33	1.51
Tasmania	2.99*	2.26	1.85	1.84
Sth Australia	1.34	1.32	1.76	1.67
Overall	1.87 (n = 70)	1.64 (n = 70)	1.63 (n = 70)	1.66 (n = 70)

Table 2: Mean log₁₀ TVC counts for knives on beef and sheep slaughter & dressing floors

* Significant ≤P 0.05 2-tail t-Test

Residence times of knives in the sterilisers (i.e. time for procedures to be performed) ranged from 5 seconds to 5 minutes, with most procedures taking 30 to 60 seconds.

Findings & Conclusions

The main findings and conclusions were:

- *E. coli* was present less often on knives cleaned using the alternative procedure.
- Total bacterial loadings were similar for the two methods.
- Residence times for knives varied widely according to the unit operations being undertaken but were always of the order of 30 seconds.
- The core findings of previous plant trials were confirmed in all three sites and for the overall data set, namely that the alternative procedures were equivalent to the current method.

6 The model for validating your alternative process

Following the decision by MSC to support approval of applications which validated the alternative using a model, MLA engaged FSA researchers to further develop such a model.

The concept behind the model was to determine, for the various temperature and time regimes likely to be used in practice, what the respective reductions in *E. coli* would be. The advantages for having an *E. coli* reduction model for knife sanitation such as this are that it:

- Focuses industry on reducing target organisms all the way along the chain
- Introduces time parameters for the first time
- Quantifies the reductions levels likely to be achieved.
- Allows for approvals of alternative processes without having to disrupt regular operations, or conduct extensive microbiological testing.
- Provides a consistent tool for industry and regulators to work with.

How contaminated are knives?

To gauge the total inactivation needed by the knife cleaning process the contamination levels of hides, hands and "dirty" knives (knives which had completed their task) were noted.

Research in New Zealand abattoirs (Table 3) was used to determine the likely levels of contamination.

Table 3: Contamination levels determined by previous studies ^{2 and 3}

	Log TVC/cm ²	Log E.coli/cm ²
Sheep Carcases		
Long, dirty fleece	5.2	1.5
Knife blade after work	5.0	nd*
Hand	5.0	nd*
Beef Carcases		
Beef hide	4.5-5.1	1.7-2.2
Knife blade after work	3.6	nd*
Hand	4.7	nd*

*nd = not done

The work of Bell and co-workers established that a 2-log reduction of *E. coli* during knife cleaning would be required. This is equivalent to having 99 cells killed out of 100, should they be present.

 ² Bell, R. 1997. Distribution and sources of microbial contamination on beef carcasses. J. appl. Microbiol. 82: 292-300.
³ Bell, R., Hathaway, S., 1996. The hygienic efficiency of conventional and inverted lamb dressing systems. Journal of Applied Bacteriology 81: 225-234.

Methodology for developing the model

Meat patties inoculated with known levels of *E. coli* were used to provide high starting counts (minimum of 4 logs) for the experiment.

By drawing the knife through the meat patty both sides of the blade were coated with the meat/culture mixture, to give a similar bacterial loading on both sides of the blade.

One side of the knife was then swabbed with using the sponge method prior to the experimental treatment, and the other side was sponged after the treatment. Treatment combinations tested were:

- Time: 1, 5, 10, 20, 30, 45, 60 seconds
- Temperature: 60, 65, 70, 75, 80, 82°C

All combinations were tested in triplicate and all combinations were repeated with and without a 1 second 40°C pre-rinse.

Results

Tables 4 and 5 summarise the percentage reductions in *E. coli* on the knife blade for the different temperature and immersion time combinations without and with a pre-rinse, respectively.

All combinations of temperature and time which gave a 2-log (99%) reduction in *E. coli* are highlighted in green. As shown in Table 5, a pre-rinse in the process greatly increases the reduction of all temperature and time combinations.

Tables 4 and 5 therefore form the basis of the model required by MSC and can now be used to assess the likely reduction in *E. coli* for the process used in a meat processing establishment. Table 6 interprets Tables 4 and 5 to indicate the minimum temperature that should be used for observed minimum immersion times.

Finally, a key part of the application for an alternative to 82°C water are the procedures to be used in the approved arrangement, accompanied by an assessment of the time and a description of whether there is a pre-rinse used at each step. This will enable the controlling authority to make assessment of the process described against the model (i.e. Tables 4 and 5).

Table 4: Percentage reduction in E. coli population on knife blade following immersion for different periods of time in water at different temperatures

Immersion	Temperature (°C)					
time (sec)	60	65	70	75	80	82
1	66.1	76.7	<1	49.4	98.6	92.2
5	96.0	97.8	97.8	97.2	99.96	>99.99
10	98.0	96.5	99.98	>99.99	>99.99	>99.99
20	97.2	99.7	99.6	>99.99	>99.99	>99.99
30	97.2	99.6	99.96	>99.99	>99.99	>99.99
45	98.3	99.9	>99.99	>99.99	>99.99	>99.99
60	98.5	99.98	>99.99	>99.99	>99.99	>99.99

Immersion			Temperature	(°C)		
time (sec)	60	65	70	75	80	82
1	98.5	98.8	98.5	99.7	99.7	99.95
5	98.4	93.6	99.95	99.93	99.94	>99.99
10	98.5	98.4	99.93	>99.99	>99.99	>99.99
20	99.7	99.8	>99.99	>99.99	>99.99	>99.99
30	99.6	99.9	>99.99	>99.99	>99.99	>99.99
45	99.98	99.93	>99.99	>99.99	>99.99	>99.99
60	96.1	99.99	>99.99	>99.99	>99.99	>99.99

Table 5: Percentage reduction in E. coli population on knife blade following immersion for different periods of time in water at different temperatures, after a pre-rinse in 40°C running water

Table 6: Minimum water temperatures for knife sanitation, according to the minimum observed immersion time of knives during routine operation

Observed minimum immersion time (sec)	Immersion temperature without pre-rinsing (°C)	Immersion temperature following a pre-rinse in 40°C running water (°C)
1	82	75
5	80	70
10	70	70
20 or more	65	60

7 The task for establishments wanting to go forward

The trials described here involved companies sampling every knife several times, leading to the accumulation of a large volume of data. The process was expensive in terms of resources needed for the testing program and for the application to MSC.

However, the accumulation of data from a number of processors and the FSA research developing the model (Tables 4 and 5) function as a broad validation for knife cleaning at temperatures cooler than 82°C. This works in the same way as milk pasteurisation; no one needs to validate that the time and temperature in their pasteuriser is effective against pathogens in milk, they only need to show that the time and temperature meets prescribed values.

The model plus supporting evidence can now be used by other establishments going forward provided temperature and time combinations at the steriliser in the proposed alternative procedure are similar to those already researched and the immersion times and pre-rinse procedures can be confirmed and maintained. Of course, you will need to verify steriliser temperatures with a thermometer or data logger but the methodology used by FSA and in the abattoir trials form the basis of a template for intending establishments.

An intending establishment should be able to negotiate a protocol with their Controlling Authority which involves far less testing, if any, than that done by the abattoirs in the 2004-05 and 2006 trials.

Now that there is a model available for predicting the likely reduction in *E. coli*, any company seeking to apply for an alternative to 82°C in its approved arrangement should document the time the knives are to be immersed at the alternative steriliser temperature and whether there is a pre-rinse or not at the respective stations along the slaughter line. This prevents the need for microbiological testing.

If testing is thought to be necessary it should be based on "worst cases" such as on knives used for hide incision or which have short residence times in the steriliser.

The amount of testing that you need to do depends on the level of confidence you and your regulator need with the process. But, if you have a slow chain speed, use a pre-rinse, and want to use water at 70°C, then both you and your regulator will be able to consult Tables 4 and 5 (or Table 6) with confidence. You might only need to collect a small amount of micro data at one or two steps to satisfy your Controlling Authority.

Table 7 below provides a simple example of how an establishment could document the time and pre-rinse procedures in their application to the Controlling Authority.

Obviously, the more stations and sterilisers there are the more detailed Table 6 needs to be to reflect the operations at that establishment. Once you have validated the process in your establishment the Controlling Authority will also have requirements for how you operate on a day-to-day basis.

Because there is a smaller margin for mistakes with a lower temperature process, your Controlling Authority may:

- require you to check times and temperatures more carefully,
- be more vigilant to stop the process if there is a low temperature or major time difference; and
- say how you will deal with any affected product.

Of course, microbiological testing may be required by the controlling authority if there are certain stations which do not meet the reduction targets set i.e. don't achieve the predicted log reductions set out in the model.

Steriliser Temperature		65°C at all stations	
Station	Operations	Minimum available time for knife cleaning	Pre-rinse (Yes or No)
Stunning and sticking	After stunning the operator sticks the animal before shackling and hoisting. The operator rinses the knife and puts it in the steriliser after sticking.	45 seconds	Yes
Y cut	Three operators perform all forequarter operations, making the Y cut, freeing the legs and shoulders. The operators rinse and sterilise their knives between bodies.	30 seconds	Yes
Brisket and pelt clearing	The operator makes a midline spear cut then clears the brisket. The weasand is freed and tied. The pelt is cleared. Two knives are used. They are changed after making the spear cut.	30 seconds	Yes
Evisceration	The operator rings the bung, pushes the rectum into the cavity and removes gonads The abdomen is opened with a reverse cut and gut removed to a trolley. Viscera are removed for inspection. The brisket is split by knifing. Two knives are used. They are changed after removal of the gonads and again after viscera removal.	20 seconds	Yes

Table 7: Template for estimating times and use of pre-rinsing of knives in a slaughter and dressing process

8 Regulatory points to consider

Processors regulated by a State jurisdiction (Controlling Authority) should be able to implement an approved arrangement after submitting validation/verification data to their Controlling Authority demonstrating equivalence with the 82°C procedure using the approach outlined above.

Processors controlled by AQIS may be required to undertake additional work and it may be necessary to gain the agreement of an importing country before being able to implement the alternative procedure. Countries importing Australian product may have prescriptive rules about temperatures for steriliser water and it may not be possible to expect approval. For export-registered premises the following questions need to be answered when considering a trial:

- For which countries are you listed?
- Do you need the product to be acceptable for all countries or only for some?
- Do those countries have requirements covering this aspect of the process?
- Do those countries allow an alternative procedure?
- Do those countries require or expect that AQIS will give approval or will they need to give approval themselves?

The answers to these questions will determine whether AQIS needs to notify or request permission from another country to accept product from the trial of the alternative procedure.

Australia has been at the forefront of supporting outcomes-based regulation internationally and also requiring that processes are demonstrated to produce a safe product. Nevertheless, establishments have to consider disposition of product from a trial:

- Can you send product processed during the trial to market/s that will accept it?
- Can the product be diverted for other uses?
- Can product be held until lot-by-lot approval is given by an AQIS ATM based on agreed standards?

There are a number of steps which you need to plan with your Controlling Authority:

- Notify the authority of your intention to apply for an alternative arrangement.
- Provide a clear summary of what you intend to do e.g. along the lines shown in Table 6.
- Provide scientific backing for your proposal (e.g. the FSA report on knife sanitising).
- Comparing the procedures for time, temperature and pre-rinse to the model shown in Tables 4 and 5
- Deciding with the authority on the type and amount of in-plant testing required, if any (e.g. for procedures where the knife has less than 2 seconds duration in the steriliser at lower temperatures).
- Undertaking any trials in the way that you have agreed with your regulator.
- Make arrangements for holding carcases/product processed during the trial until microbiology results are available, if applicable.
- Present results for review by FSA and/or sign-off by your Controlling Authority.

9 Support for your intended alternative procedure

Meat & Livestock Australia (MLA) is able to assist with your alternative procedure by providing advice and support as you go through the steps for regulatory approval.

If you're interested in progressing an alternative procedure for knife and equipment sanitising please contact:

lan Jenson (02 9463 9264)