



FOOD STANDARDS
Australia New Zealand
Te Mana Kounga Kai – Ahitereiria me Aotearoa

A Review of Sulphites in Raw Meat Sausages

FINAL REPORT

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Abbreviations

ADI	Acceptable Daily Intake
AFGC	Australian Food and Grocery Council
AFSCoE	Australian Food Safety Centre of Excellence
APC	Aerobic Plate Count
CP	Control Point
CCP	Critical Control Point
CL	Chemical Lean
DC	Distribution Centre
ESAM	<i>Escherichia coli</i> and <i>Salmonella</i> Monitoring
FSANZ	Food Standards Australia New Zealand
FSIS	Food Safety and Inspection Service
HACCP	Hazard Analysis and Critical Control Points
HUS	Haemolytic Uraemic Syndrome
MAP	Modified Atmosphere Packaging
MLA	Meat and Livestock Australia
RTE	Ready-to-Eat
SARDI	South Australian Research and Development Institute
TVC	Total Viable Count
USDA	United States Department of Agriculture
VSP	Very Small Plants

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Executive Summary

Traditionally, sausage manufacture was a means by which retail butchers utilised surface trim from beef, pig and mutton carcasses, grinding it and filling into natural casings made from intestines. Today, the trade continues in much the same way; around 3,000 retail butchers build up surface meat in the 'trim bucket' which, along with other cuts, are ground and filled into synthetic casings.

In recent decades, in order to service supermarket trade, a small number of high-volume manufacturers have installed equipment to manufacture the approximately 1 million kg/week required by this sector of the sausage market.

Little is known of the microbiology of surface trim but it thought, intuitively, to be inferior to trim which comes from internal portions, for which there is ample data from three national baseline studies plus ESAM (*Escherichia coli* and *Salmonella* Monitoring) testing of beef trim from export-registered plants. These data indicate that frozen boneless beef and pork trim is in the range $10^{1-2}/g$.

For the microbiology of finished sausages, data exist from two sources: the Western Australian Health Department and a recent study undertaken under the aegis of the Australian Food and Grocery Council. These data indicate that sausages leave the processing plant with a total bacterial count between $10^4/g$ and $10^5/g$.

Supply chains vary widely between trade in urban and local/remote communities. The latter is based on sausages manufactured at butcher shops, while in cities and towns supermarkets, specialty shops and delis are the major suppliers.

Shelf-life requirements also vary between these two supply chains. Butcher shops require only a few days (one to five) shelf-life while high-volume suppliers require up to 18 days to include manufacture, transport to the supermarket distribution centre (DC), storage, retail and home use.

Transport from high-volume manufacturer to supermarket DC is onerous, involving journeys of two to three days e.g. Sydney-Perth and Sydney-Darwin. Supermarkets typically require 75% of the shelf-life remaining when stock is delivered to the DC. Supermarkets have a policy to discount sausages as the use-by date (packed on date +14 days) is approached.

In other countries e.g. the UK and Ireland, the trade is similar to that in Australia – centralised packing from a small number of high-volume producers to supermarkets, together with 'family butcher' trade. In North America, breakfast sausages are also manufactured though with more reliance on supermarket than retail butcher trade.

While in Europe sulphite is allowed up to 450 mg/kg, its use in North America has been banned for several decades. In the latter, distances between manufacturer and supermarket can be as long as in Australia and the trade is maintained despite a shelf-life of 12 days being imposed. To achieve marketing and shelf-life needs, use is made of modified atmosphere packaging.

A term of reference for this study is to consider the potential impacts of lowering levels of sulphite in raw sausages. Two scenarios are canvassed: reduction of in-going sulphite to 200 mg/kg and its complete elimination from sausage formulations.

Removing or reducing sulphite from sausage formulations may have little effect on small butcher shops as butchers are able to manufacture almost to order.

By contrast, for medium-volume manufacturers, any reduction would be onerous and will require changes to either formulation and/or packaging. Unfortunately this sector of the supply market may not have the resources to undertake the R&D needed to maintain their customer base, which could be lost to high-volume suppliers.

For sausages produced by high-volume manufacturers, reduction to 200 mg/kg would effectively halve shelf-life to around nine days, while completely eliminating sulphite would give no more than four days before the bacterial loading becomes excessive. It should be emphasised that sulphite-free sausages may quickly (two days) take on a grey appearance, further limiting their acceptability.

Maintaining supply of low- or no-sulphite sausages to major supermarkets will involve significant R&D by the small number of manufacturers which currently service this market, with a proportion of the cost passed on to consumers. Most likely solutions involve use of antimicrobial ingredients such as lactate and diacetate, coupled with modified atmosphere packaging. Improvements to temperature control during production, storage at manufacturer, transport to DC, storage at DC, transport to retail store, storage back-of-house and display may also be effective in extending shelf-life.

The economic impact of manufacturing low- or no-sulphite sausages is difficult to ascertain because companies have different business models. To properly evaluate any change to sulphite use will involve individual enterprises embarking on in-depth analysis of their businesses practices and those of their suppliers.

1 Introduction

Sulphite has been used as a preservative for millennia – ancient Egyptians and Romans used the fumes of burning sulphur as an anti-microbial agent for wine-making. Raw meat sausages date from several centuries BC but it was in Roman times that they became extremely popular, particularly at festivals. Because sausages were associated with Roman orgies their consumption was banned by Emperor Constantine. In those times, sausages were made from pork, beef and blood stuffed into a casing made from intestines, a *modus operandi* which persisted until the middle of the 20th century.

Raw sausages as eaten in Australia and New Zealand are based on the British ‘breakfast’ sausage. Traditionally these were made from surface trimmings of carcasses boned at butcher shops, the high bacterial level of which gave sausages a poor public health reputation. An old saying that the law is a bit like sausage making ‘*don’t look too closely at what goes into it*’ typified the raw materials that were used.

The use of sulphur dioxide, usually added as sodium metabisulphite, was indispensable for obtaining sufficient shelf-life for the British sausage. The mechanism by which sulphite extends the shelf-life of raw meat sausages was established by Dyett and Shelley (1962, 1966) who showed that it was active against Gram-negative bacteria in prolonging their lag phase. Sulphite addition changes the microflora of refrigerated meat, being a selective agent for Gram-positive bacteria and yeasts.

As will be seen from Section 2 of this report, despite the use of sulphite, *Salmonella* was often isolated from raw sausages in the UK and the product was regularly associated with food poisonings (Mattick *et al.*, 2002). While it is generally accepted that in countries like the UK, Australia, New Zealand, Ireland, Canada and the USA sausages are eaten after cooking, it is doubtful whether all pathogens, even the relatively heat-labile Gram-negative pathogen are all inactivated by normal cooking methods. In the UK, Mattick *et al.* (2002) found that *Salmonella* survived frying, grilling and barbecuing in sausages which appeared fully cooked.

In Australia, over the period January 2001-December 2006 there were five outbreaks of food poisoning in which sausages were implicated (OzFoodNet, unpublished data, 2007). As indicated in Table 1.1, *Salmonella* Typhimurium was responsible for three of the five outbreaks; the other two were of unknown aetiology. Of these five outbreaks, two were directly linked to sausage consumption; the other three outbreaks were highly likely to be due to sausages. One outbreak was likely to be due to cross-contamination of bakery products with sausage meat through the use of a common piping bag. The remaining two outbreaks had unknown food vehicles, but implicated a variety of foods of which sausages and sausage rolls were identified food items.

While sulphites have long had a history of safe use in meat products, exposure has been linked with the exacerbation of asthmatic and other respiratory conditions in some sensitive individuals. In the USA, use of sulphites has long been banned in meats and in fresh fruit and vegetables. In Australia, New Zealand, the UK and Ireland, however, sulphite can still be used in raw sausages.

Due to concerns over exceedance of the Acceptable Daily Intake (ADI) for sulphites in foods, particularly for children, Food Standards Australia New Zealand (FSANZ) has requested information on sulphites in raw sausages.

Table 1.1: Outbreaks associated with sausages, Australia, January 2001 to December 2006 (n=5).

Year	State	Setting	Ill	Hospitalised	Food vehicle	Aetiology
2001	VIC	Private	14	3	Suspected beef products	<i>S. Typhimurium</i> 170
2002	NSW	Take-away	4	0	Chips, fried eggs and sausages	Unknown
2002	SA	Bakery	22	7	Bakery cakes and buns (suspected cross-contamination with sausage meat from a piping bag)	<i>S. Typhimurium</i> 99
2005	VIC	Private	13	0	Unknown	<i>S. Typhimurium</i> 12
2005	WA	Restaurant	15	0	Unknown	Unknown

The terms of reference (TORs) of the study are to identify whether and which good hygienic practices would replace the use of sulphites and what the financial costs of using alternatives would impose on industry. Specific TORs are to:

- 1 Review recently generated industry data on the effects of sulphite levels on the shelf life of sausages and the residual levels of sulphites present in cooked sausages, and the subsequent industry conclusions and recommendations;
- 2 Review the supply chain distribution and the necessary product shelf life requirements for sausages in urban and remote regions in Australia and New Zealand;
- 3 Examine and review the current storage, transport and retail practices for processed meat in Australia and New Zealand, with a particular focus on raw meat sausages;
- 4 Identify changes in the manufacturing processes of sausages including control of meat quality, hygiene of production plants etc. that may have a bearing on levels of preservative required in products;
- 5 Consider potential impacts of lowering levels of sulphites on processed meat industry sector with a particular focus on raw meat sausages;
- 6 Examine and review the processed meat industry in other countries (e.g. USA and Europe), with an emphasis on quantitative data where possible. This review should be focused predominantly on sausages with information provided in relation to:
 - manufacturing processes, including preservatives use, in-coming meat quality and shelf-life issues;
 - production processes, including production plant hygiene; and
 - distribution, storage, transport and retail of processed meat products; and
- 7 Liaise with Authority staff as required.

Following the Introduction, the present study responds to the terms of reference in the order listed above, with one exception, the report by Australian Food and Grocery Council (AFGC) on industry-generated data (*Residual Sulphites in Sausages*). The study was well designed and the data generated were both unique and current allowing us to use them at several points in our study (Anon., 2007). We wish to acknowledge that an experimental study, using company laboratories in three States, can present great difficulties. Company laboratories

function to do routine testing, often with resources which are intended primarily for the daily flow of technical information required. It is also difficult for laboratory staff to incorporate the demands of a research study into their daily work schedule. Given these difficulties, we acknowledge the many people who designed, managed and undertook the AFGC-industry work.

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2 The Manufacture of Raw Meat Sausages

2.1 Process

A typical process for both large manufacturers and retail butcher shops is presented in Figure 2.1. There are significant differences between the meat raw materials and processes used by large and small manufacturers.

2.1.1 Using frozen trim

Cartons of frozen trim form a significant component of sausage meat used by large manufacturers. Blocks of beef, pork or mutton are tempered in a chiller overnight to soften slightly. After de-cartoning, each block is passed through a chipper which produces small flakes of frozen meat. This stage is undertaken only by larger manufacturers.

2.1.2 Comminution

Chilled or flaked, frozen meat is ground in a mincer. The degree of comminution varies according to sausage type with texture ranging from coarse to a paste-like consistency. Some operations use a combination of grinding and bowl chopping to produce a sausage emulsion.

2.1.3 Ingredient addition

Ingredients include water, wheat flour, rice flour, mineral salt, sulphite, dextrose, spices and flavours. Large manufacturers make batches of the order of 200 kg based on trim (at least 140 kg), water (40 kg) and premix containing some or all of the ingredients listed previously. Premix is purchased so that one bag is used for each batch. Large manufacturers have combined grinder/blenders in which premix and water are mixed with ground meat by a ribbon blender.

Small manufacturers make batches of the order of 20-50 kg and weigh out premix to give the correct level of ingredients, including sulphite, in the finished batter. On a small scale, meat, water and premix may be mixed in a small ribbon blender.

2.1.4 Filling

At the manufacturing level the entire batch (200 kg) is hoisted and inverted above a hopper which services the filler. Sausage emulsion under vacuum is extruded from the filling orifice into collagen casings and the end of each sausage is linked.

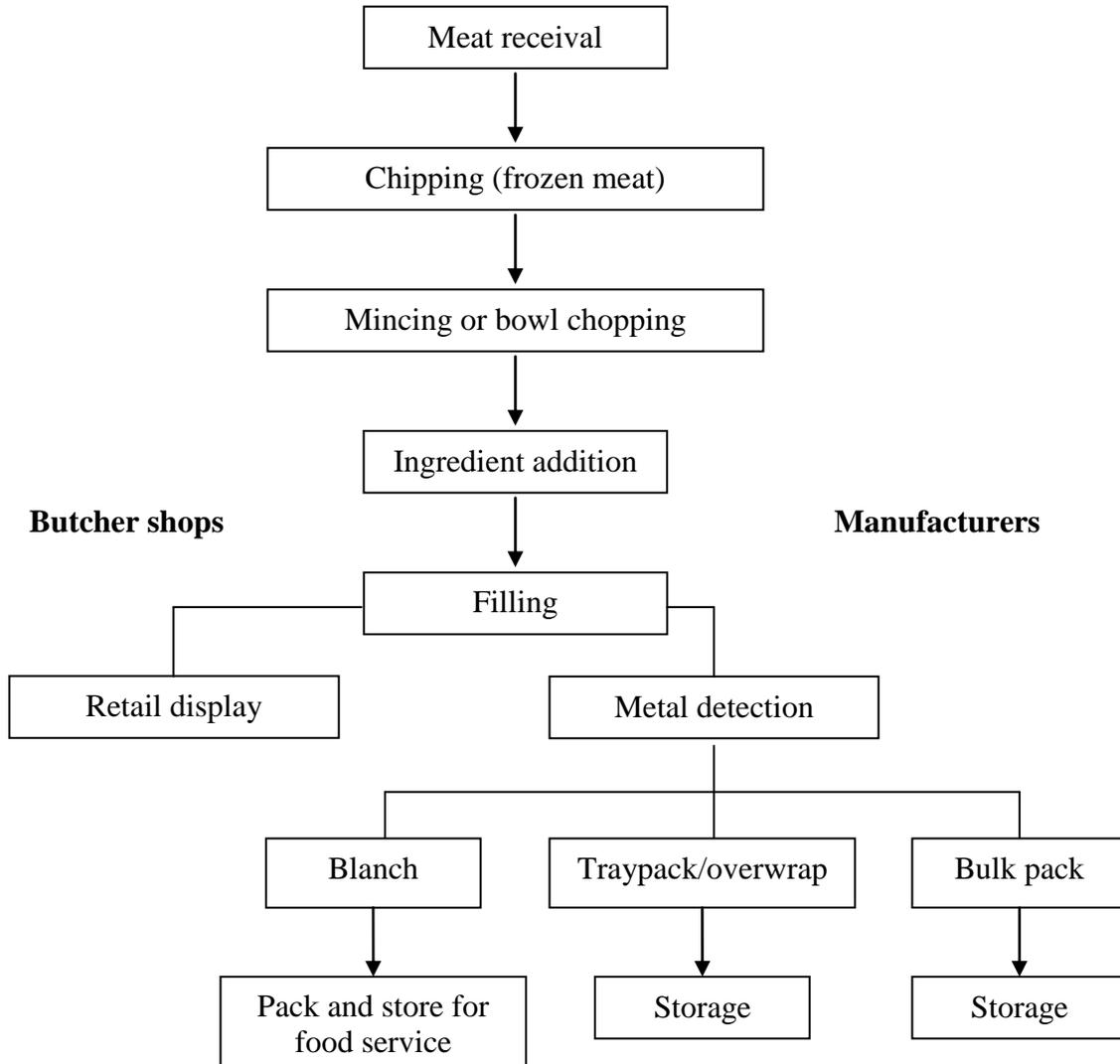
In small operations, sausage batter is fed into the filler chamber and pressure applied to extrude it into the casing (also usually collagen). The butcher links sausages into clusters.

2.1.5 Blanching

Blanching is becoming increasingly common in the supply of food service operations. This is done either by hanging the sausage links in a steam cabinet or by immersing in water at 75-80°C for a short time. Both these processes give a surface heat treatment and set the core of

the sausage. Blanching does not cook the sausage and the product cannot be labelled or marketed as cooked sausage. Blanching is not a Critical Control Point (CCP) and the final product still requires thorough cooking to ensure safety.

Figure 2.1: Outline process flow diagram for fresh sausage



2.1.6 Packaging

At large manufacturers, sausages are packed in two formats: bulk packs and tray packs. Bulk packs are of the order of 5 kg and are intended for sale in delicatessens. Sausages are packed into a gas-impermeable bag and flushed with a mixture of nitrogen, oxygen and carbon dioxide. Bags are packed in cartons.

The base of each tray pack is filled manually with sausage and conveyed to an overwrapper which cases the tray in gas-permeable film. The tray is weighed and labelled with a use-by date 14 days after packing. The pack contains an ingredients panel and nutritional information. Trays are packed in cartons.

Butchers store sausages in the chill store overnight and in the retail cabinet during retail hours. Sausages are weighed by hand and sold to order.

2.1.7 Storage

Manufacturers store cartoned product on pallets in the holding chiller which operates between close to zero and 5°C depending on location in the chiller and time of day (chillers cool down when closed at night). Product is conveyed to the supermarket distribution centre (DC) on a daily basis either by the supermarket's or the manufacturer's transport.

Retail butchers store sausages in the holding chiller which cycles between close to zero and 10°C, depending on location and time of day.

2.1.8 Control Points

Irrespective of the scale of production, the manufacture of fresh sausages has the following Control Points (CPs):

- Ingredient addition (sulphite)
- Storage temperatures of finished products.

Based on the Codex definition of Critical Control Point (CCP) as "... a stage, process which prevents, eliminates or reduces the hazard to an acceptable level" it is neither sulphite addition nor storage temperature that qualifies as a CCP. However, some company HACCP plans may give both stages CCP status.

For some asthmatics, sulphite is a hazard and the product must be labelled to prevent accidental consumption. While most people who are sensitive to sulphite are well aware that it is allowed as an additive for fresh sausage, correct labelling on packs and retail displays is necessary to prevent the hazard which could stem from accidental consumption.

An upper limit of 5°C is regulated for all storage, distribution and retailing phases (see Section 4). Chill storage prevents growth of *S. aureus*, *Salmonella* and pathogenic *E. coli* but does not prevent growth of *L. monocytogenes*.

2.2 Factors Affecting Microbiological Levels of Raw Sausages

2.2.1 Raw meat quality

It has long been known that the primary determinant of microbiological quality of raw sausages is the microbiological loading of the trimmings used for manufacture. Surkiewicz *et al.* (1972) surveyed beef trimmings and raw sausages at the production level in the USA and established a clear relationship. Sausages made from trimmings with an aerobic plate count (APC) <10⁵/g had counts around 2 x 10⁵/g 75% of the time and <5 x 10⁵/g 96% of the time. By contrast, sausages made from trimmings with APC >10⁵/g had counts >2 x 10⁵/g 87% of the time and >5 x 10⁵/g 49% of the time.

2.2.2 Sulphite addition

Sulphite has a marked effect on the microbiological quality of raw sausages by inhibiting growth of psychrotrophic, Gram-negative bacteria which cause spoilage of sausages through putrefaction (Stiles, 1994).

2.3 Raw Materials Available to Australian Manufacturers

In Australia, large manufacturers use meat trimmed from carcasses to a specific fat content e.g. 70 CL (70% chemical lean). This trim, in the case of sausages manufactured for Australia's large supermarket companies, comes predominantly from the carcasses owned by those companies. In some cases sausage manufacturing is on the same site as the slaughter establishment and its integrated boning room. Trim may also be purchased in carton form, both chilled and frozen.

The microbiology of beef trim used for fresh sausage manufacture has been characterised by Phillips *et al.* (2006, 2007) both for Total Viable Count (TVC) and indicator organisms in frozen trim (Table 2.1) and for pathogens in chilled ground trim (Table 2.2).

Table 2.1: Microbiological quality of frozen beef trim (Phillips *et al.*, 2006)

	TVC ^b	<i>E. coli</i> ^c	<i>Enterobacteriaceae</i> ^c	Coagulase +ve staphylococci ^c
Prevalence (%) ^a	82.5	1.8	7.1	20.3
Mean log ₁₀ cfu/g	1.28	1.46	1.30	0.80
Median	1.32	1.30	1.18	0.70
Standard deviation	0.83	0.78	0.59	0.32
90 th percentile	2.30	2.67	2.01	1.00
95 th percentile	2.71	2.79	2.62	1.74
99 th percentile	3.54	2.79	2.95	2.24
Maximum	5.49	2.79	2.95	2.32

^a Limit of detection 5 cfu/g

^b TVC, counts are given as log₁₀ cfu/g. All counts were incremented by one cfu/g before log transformation.

^c Coliform, *E. coli*, *Enterobacteriaceae* and staphylococci counts per unit area are given as log₁₀ cfu/g of positive samples only.

Table 2.2: Prevalence of pathogens in frozen beef trim and chilled ground beef (Phillips *et al.*, 2006; 2007)

	Positive/Total (%)	
	Frozen boneless	Chilled ground
<i>Salmonella</i>	1/1082 (0.1)	4/360 (1.1)
<i>Escherichia coli</i> O157	0/1082	1/357 (0.3)
<i>Campylobacter</i>	0/1082	0/91
<i>Clostridium perfringens</i>	Not done	0/94

From Tables 2.1 and 2.2 it can be seen that trim for beef sausage used by manufacturers has a mean TVC of log 1.28/g with low prevalence of indicator organisms and pathogens. It should also be emphasised that beef trim is not released for sausage manufacture until the lot has cleared the *E. coli* O157:H7 screen test.

The microbiological profile of pork carcasses is also well characterised for export establishments, many of which supply large manufacturers. Total bacterial loadings and

prevalence of *E. coli* on pig carcasses are presented in Table 2.3 and for pork portions in Table 2.4.

Table 2.3. Total viable count (TVC) and *E. coli* on Australian chilled skin-on pig carcasses in the export sector (Source AQIS 2004-2006)

	TVC ^(a)			<i>E. coli</i> ^(a)		
	2004	2005	2006	2004	2005	2006
n	2354	1921	2089	3883	3729	3860
Prevalence (%)	-	-	-	7.49	7.32	7.07
Mean log ₁₀ cfu/cm ²	1.90	1.81	1.18	-0.45	-0.51	-0.54
Median	1.88	1.82	1.26	NA	NA	NA
90 th percentile	2.83	2.62	2.32	-	-	-
95 th percentile	3.04	2.91	2.54	-1.08	-1.08	-1.08
99 th percentile	3.32	3.32	3.27	0.26	0.33	0.12
Maximum	4.00	4.86	5.00	2.90	1.66	2.77

^a Limit of detection 0.08 cfu/cm²

Table 2.4: *E. coli* and Total Viable Count of finished pork products (Reyes-Veliz, 2005)

Product	TVC	<i>E. coli</i>
	Mean log/cm ²	Positive/Total (mean log of positives/cm ²)
Tenderloin (n=100)	2.11	30/100 (-0.21)
Belly (n=5)	2.01	1/5 (-0.90)
Jowl (n=5)	1.99	4/5 (0.11)
Loin (n=5)	2.53	5/5 (-0.55)
Leg skin-off (n=5)	1.35	0/5
Leg skin-on (n=5)	2.72	5/5 (-0.36)
Shrink-wrapped primal (n=3)	2.65	0/3
Shrink-wrapped shoulder roll (n=3)	2.11	3/3 (0.34)

^a Mean log of positives/cm²

Taken together, the data in Tables 2.3 and 2.4 indicate a slightly higher bacterial loading in raw materials than beef trimmings; prevalence of *E. coli* is also higher in raw materials.

Data on loadings of pathogens on pig carcasses are presented in Table 2.5; the prevalences are similar to those found in beef trim.

Table 2.5: Prevalence of major foodborne pathogens on 680 pig carcasses in Australia (Coates et al., 1997)

Pathogen	Number (%) Positive
<i>Salmonella</i> spp.	7 (1.0)
<i>Yersinia enterocolitica</i>	1 (0.15)
<i>Staphylococcus aureus</i>	97 (14.9)
<i>Campylobacter coli/jejuni</i>	0 (<0.5)
<i>Listeria monocytogenes</i>	0 (<0.5)
<i>Escherichia coli</i> O157	0 (<0.5)

From Tables 2.1-2.5 it can be seen that, in the manufacturing sector, raw meat ingredients potentially enable manufacture of fresh sausage with a TVC of the order of 10^3 /g.

2.4 Raw Materials for Retail Butchers

Typically retail butchers receive carcasses on one or two days each week from a local domestic abattoir or, in remote areas from ‘Very Small Plants’ (VSPs). Formerly termed ‘slaughterhouses’, VSPs slaughter, by definition, a maximum of 150 cattle units/week, a ‘cattle unit’ being either eight sheep, six calves, six pigs, eight goats or six deer. In butcher shops it is common practice to ‘hang’ carcasses for three to four days in a holding chiller, a process which butchers believe improves eating quality. During this period in chill storage the bacterial loading on the surface increases by a quantum dependent on the storage temperature. Typical microbial loadings of beef carcasses (chilled overnight) produced at four domestic abattoirs are presented in Table 2.6 from which it can be seen that both mean TVC and prevalence of *E. coli* are substantially higher than on carcasses from larger (and usually export-registered) abattoirs which supply the large sausage manufacturers (see Tables 2.1-2.5).

Table 2.6: Mean \log_{10} TVC counts and *E. coli* prevalence for beef carcasses produced at domestic abattoirs (Anon. 2007a)

	TVC	<i>E. coli</i>
Queensland	2.3	2/10 ^a
Tasmania	2.7	8/10
South Australia	1.6	1/10
Victoria	2.1	2/25

^a Positive/Total samples

After hanging carcasses for three to four days the bacterial level will be of the order of 1 log unit higher at the beginning of carcass boning. Retail butchers bone carcasses into primals and sub-primal cuts. As surface fat is excised during trimming it is accumulated in a ‘trim bucket’, to which is added lower value meat for subsequent grinding as patties or sausage meat. The high ratio of surface meat means the TVC/g of meat used as sausage trim in the retail sector is likely to be substantially higher than that in the manufacturing sector. There are no published data in Australia to inform on microbiological levels of retail trim.

2.5 Microbiological Profile of Raw Sausages at the Production Level

2.5.1 Microbial ecology of raw sausages

At manufacture, raw sausages have a pH between 5.4 and 6.0, depending on the pH of the meat ingredients used, together with the proportion of added fat, the pH of which is higher than that of lean meat (Anon., 2007b). Since sausage premix contains cereal, sodium metabisulphite and sodium chloride, the water activity is reduced from that of raw meat to around 0.97-0.98 (Anon., 2007b). During refrigerated storage the predominant microflora becomes Gram-positive fermentative bacteria and yeasts (Dowdell and Board, 1971; Dalton *et al.*, 1984). However, because of the buffering capacity of meat and the sodium content of the product, pH does not alter during refrigerated storage.

The microbial ecology of raw sausages is growth-permissive for Gram-negative pathogens though their growth is prevented by refrigerated storage. The relatively benign conditions of pH and water activity are not sufficient to cause inactivation of pathogens. Because it is psychrotrophic, *L. monocytogenes* can grow during refrigerated storage.

2.5.2 Overseas historical data

In the study of Surkiewicz *et al.* (1972), 29% of sausages had total bacterial counts greater than 5×10^5 /g; 50% between 1×10^5 /g and 5×10^5 /g and 20% $<10^5$ /g; *Salmonella* was isolated from 28% of samples of beef trim and finished sausages. Surkiewicz *et al.* (1972) commented that at retail the median total bacterial count was 10^6 /g with some sausages having $>10^8$ /g; shelf-lives were estimated at three to seven days.

In Canada, Farber *et al.* (1988) analysed 62 lots of raw (breakfast-type) sausages from 55 manufacturers, taking samples at the plant level. Geometric means were 4.5×10^5 /g for TVC (maximum was $>10^8$ /g), *E. coli* was 7.3×10^1 /g and *S. aureus* was 3.6×10^2 /g; *Salmonella* was detected in 9/61 (14.8%) of samples.

With the implementation of HACCP-based food safety systems it is generally accepted that there have been significant improvements in the microbiological quality of raw meat during recent decades. In the UK, surveys have been conducted on prevalence of *Salmonella* over the period 1969-2004 which reflect changes in meat processing. Roberts *et al.* (1975) analysed 3,309 samples from manufacturers finding *Salmonella* in 435 (29.7%). In 1981 the prevalence was 12% (Anon., 1981) and in 1989 it was 9.8% with samples from retail butchers being 2.6% and branded products being 14.3% (Anon., 1989). In 1995, Nichols and de Louvois surveyed raw sausages and found *Salmonella* in 136/738 (17%) of samples. In 2002, Mattick *et al.*, isolated *Salmonella* from 14/162 (8.6%) of sausage samples and in 2004, Broughton *et al.* isolated the pathogen from 27/921(2.9%) of sausages in Ireland.

2.5.3 Australian data

Two datasets are available for raw sausages at production in Australia: routine surveillance carried out by the Western Australian Health Department (1997-2005) and a recent survey undertaken on behalf of the Australian Food and Grocery Council (AFGC).

The Western Australian Health Department data were gathered as part of a routine surveillance testing program and involved testing samples of sausage obtained at the production plant. The data, summarised in Tables 2.8-2.10, involve around 150 analyses for

indicator organisms and pathogens over the period 1997-2005: *E. coli* was present in 94/155 (60.6%) of samples with the mean log of positive samples being 0.72/g.

Table 2.8: Mean log TVC (cfu/g) of freshly-made raw sausages in Western Australia

Year	Number of samples	Mean	Median	75 th percentile	Maximum	Standard Deviation
1997	9	5.73	5.15	6.36	8.15	1.35
1998	21	5.41	5.58	6.08	8.53	1.39
1999	21	5.25	5.18	6.15	8.08	1.40
2000	21	4.52	4.61	5.00	5.46	0.60
2001	30	4.66	4.48	5.03	7.40	0.79
2002	31	5.80	5.69	7.44	8.42	1.70
2003	10	4.81	4.87	5.61	6.08	0.92
2004	1	3.89	NA	NA	NA	NA
2005	3	3.71	3.60	3.77	3.94	0.20
Overall	147	5.13	4.92	5.72	8.53	1.32

As seen from Table 2.8, mean TVC over the period was higher than that obtained in a 2007 survey (Table 2.11). Although there is limited information from 2003 onwards, the sampling period between 1997 and 2002 shows that the levels of TVC were similar and have not changed significantly over time.

Table 2.9: Prevalence (in 25 g samples) of pathogens in freshly-made raw sausages in Western Australia

Pathogen	Prevalence (%)
<i>Salmonella</i>	4/153 (2.6%)
<i>Campylobacter</i>	1/46 (2.2%)
<i>Listeria</i>	89/142 (62.7%)

While *Salmonella* and *Campylobacter* were at a prevalence which might be expected from processed pork and beef products, that of *Listeria* spp was higher. Serovars of *Listeria* present in samples, presented in Table 2.10, indicate *L. monocytogenes* in 30/142 (21%) of samples. This is a surprisingly high prevalence as Vanderlinde *et al.* (1998) isolated *L. monocytogenes* from 4/190 (2.1%) of beef carcasses and Coates *et al.* (1997) did not isolate the pathogen from 680 pig carcasses.

Table 2.10: *Listeria* serovars of pathogens in freshly-made raw sausages in Western Australia

	Number of isolations
<i>L. innocua</i>	48
<i>L. monocytogenes</i>	30
<i>L. welshimeri</i>	7

In 2007, a survey of the microbiology of raw sausages was undertaken by three large Australian manufacturers with the primary aim of informing on shelf-life and sulphite level (Anon., 2007b). The dataset is not large and involves only fourteen samples from two manufacturers. Microbiological data for raw sausage at production are summarised in Table 2.11. Of major impact in shelf-life considerations is that the mean log TVC of freshly-made sausages is 4.4/g, allowing a 2.5 log increase during the shelf-life while still remaining $<10^7$ /g (see Section 3.3.1). Five of six samples were positive for *E. coli* with a maximum count of 90/g. One sample was positive for *S. aureus* at a count of 2.6×10^3 /g.

Table 2.11: Microbiological profile of freshly-made raw sausages (Anon. 2007b)

Mean log TVC/g	<i>E. coli</i> Positive/total (mean log of positives)	<i>S. aureus</i> Positive/total	<i>L. monocytogenes</i> Positive/total
4.4	5/6 (0.9)	1/6	0/6

It is instructive to compare the mean log TVC for freshly-prepared sausages (Table 2.11) with that of in-going beef trim (Table 2.1) where there is a 2-3 log increase during grinding and mixing.

2.6 Microbiological Profile of Raw Sausages at the Production Level

In late-2007 a baseline survey of beef sausages at retail was undertaken in the capital cities of each mainland state. Sample numbers were equated with population densities in each city and retail mode (supermarkets 80%, butcher shops 20%). Summary statistics of Total Viable Counts of sausages from both retail modes are presented in Table 2.12.

Table 2.12: Total Viable Counts (\log_{10} cfu/g) of beef sausages in butcher shops and supermarkets

	Butcher	Supermarket
n	10	43
Minimum	4.97	3.00
25 th percentile	5.47	3.30
Median	6.01	3.95
Mean	5.96	4.27
75 th percentile	6.30	4.68
Maximum	7.15	7.63
SD	0.66	1.26

Mean TVC was significantly higher in sausages manufactured and purchased at butcher shops than those sold at supermarkets ($p < 0.001$), the mean \log_{10} difference being 1.69 cfu/g. Prevalence of *E. coli* was significantly higher ($p = 0.01$) on sausages purchased at butcher shops (6/10) compared with supermarkets (7/43: 16%). A more detailed report is presented as Appendix 1.

2.7 Key Findings

1. Raw sausages are made by two distinct methods in Australia. In butchers shops, surface trim and other cuts from carcasses are used in a 'traditional' method, while high-volume manufacturers use mainly cartoned trim (chilled and frozen) of specific fat content.
2. Little is known of the microbiology of surface trim but it thought, intuitively, to be inferior to trim which comes from internal portions, for which there is ample data from three national baseline studies plus ESAM testing of beef trim from export-registered plants.
3. Data from two sources: the Western Australian Health Department and a recent study undertaken under the aegis of the Australian Food and Grocery Council indicate that sausages leave the processing plant with a total bacterial count between 10^4 /g and 10^5 /g.
4. The microbial ecology of raw sausages (pH around 6.0 and water activity 0.97-0.98) allow survival and growth of all pathogens.
5. Growth of Gram-negative pathogens and *Staphylococcus aureus* is prevented at refrigeration temperatures, though *L. monocytogenes*, which is psychrotrophic, can grow.

2.8 Data Gaps

- 1 Microbiological profile of sausages from butcher shops and medium sized manufacturers.
- 2 Pathogen prevalence in raw sausages.

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3 Supply Chain for Raw Meat Sausages

In both Australia and New Zealand, the industry has three supply chains:

- Local supply
- National supermarket supply
- Supply to food service operations.

In Australia, raw sausages are produced by 8-10 large manufacturers (see Section 2) for the major supermarket chains and food service operations. A similar number of large manufacturers supply the same markets in New Zealand (Rob Archibald personal communication). In Australia, around 3,000 very small manufacturers, almost all located back of house at retail butcher shops are an alternative source of manufacture. Table 3.1 contains an estimate of the number and location of butcher shops in each state (Australian Meat Industry Council, personal communication). For the purposes of the present report it is assumed that every butcher shop manufactures raw sausages.

Table 3.1: Location of butcher shops in Australia

State	Estimated Number
Queensland	650
NSW	850
Vic	950
SA	230
WA	250
Tasmania	70
Total	3000

In Australia, anecdotal information from major supermarket companies suggests that production is seasonally-based with consumption increasing in the warmer months to more than 1.5 million kg/week of fresh sausage. For the 3,000 smaller producers, production can be made almost 'to order' as demand dictates. In retail butcher shops, sausages are manufactured on two to six days/week. There are no production data available from this sector of the industry but if it is assumed conservatively that each retail butcher manufactures 100 kg/week, the sector accounts for 300,000 kg/week. Taken together, Australian production is estimated at around 1.8 million kg/week (93,600 t/annum).

Data are also collected by Meat and Livestock Australia (MLA) in monthly market surveys which ask respondents to state the number of serves of a particular commodity they have consumed in the last seven days. For raw sausages, over the period January 2006-October 2007, MLA data estimate 730 million serves of sausages with a beef:pork ratio of 53:47 (Anon., 2008). If it is assumed that the mass of each serving was 150 g, around 109,000 t of sausages were consumed in Australia in 2007. Given similar consumption patterns between Australia and New Zealand it would seem reasonable, on a proportional population basis, to assume around 22,000 t of sausages were consumed in New Zealand

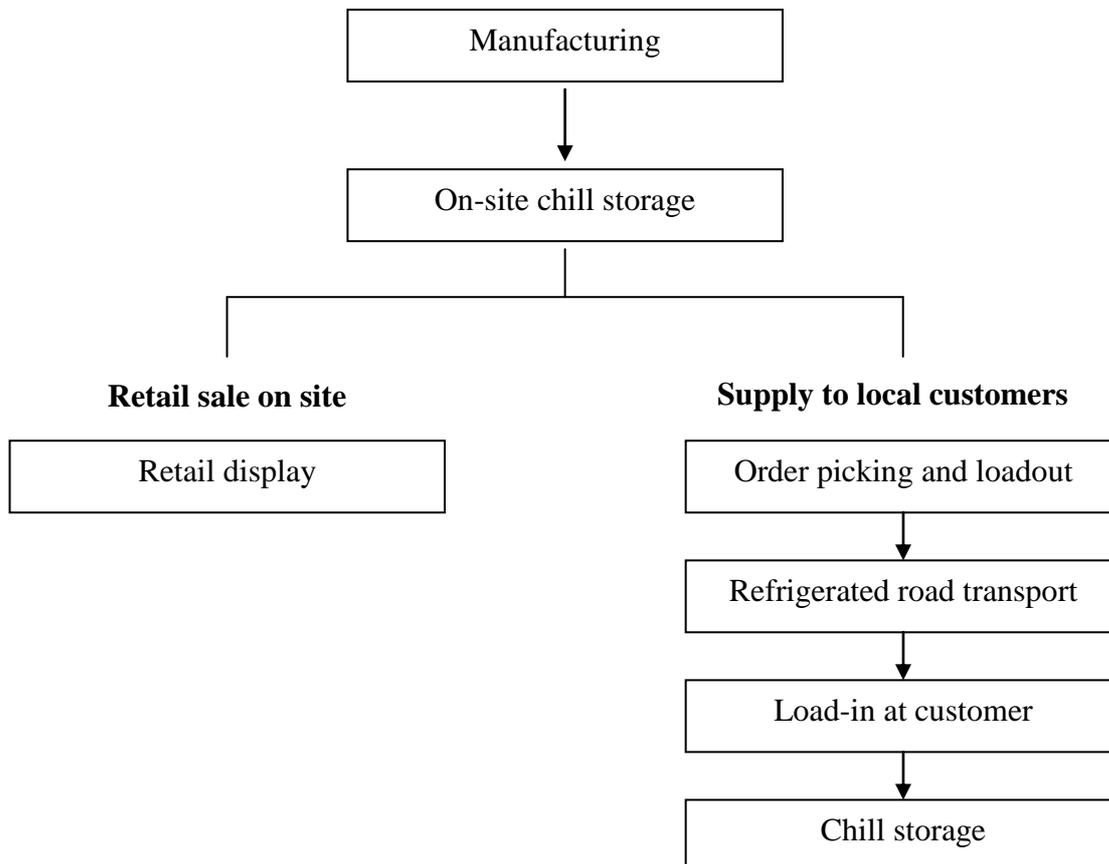
In both Australia and New Zealand, supermarket companies receive sausages from a small number of high-volume manufacturers which are shipped to their distribution centres (DC).

In both countries, high-volume manufacturers also supply the food service sector. Both New Zealand and Australia also have small retail butcher shops each of which manufactures sausages for local consumption.

3.1 Local Supply

Local supply is carried out by butcher shops of differing size. Small butcher shops typically manufacture twice a week and produce two batches for sale, each around 20-25 kg. Larger butcher shops typically manufacture 50-100 kg a week for retailing front-of-house in their own shop and also for wholesaling to local customers such as the hospital or school. There are a small number of medium-sized manufacturers which produce in the order of 3 t of sausages each week and supply independent supermarkets and local institutions (hospitals etc). Typical distribution chains are presented in Figure 3.1.

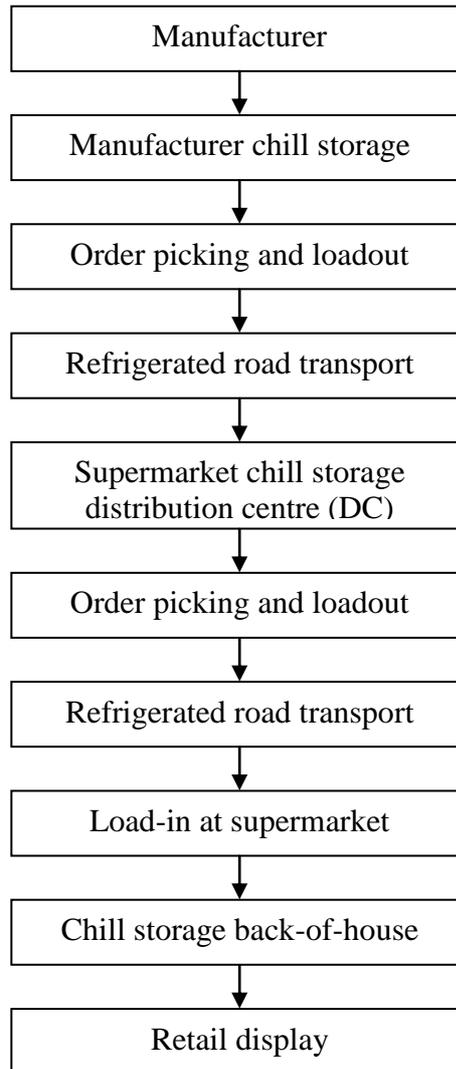
Figure 3.1: Supply chain for fresh sausage used by local suppliers



3.2 National Supply Chains

Supermarket chains purchase fresh sausages in a chilled format from dedicated suppliers according to Figure 3.2.

Figure 3.2: Supply chain for fresh sausage used by supermarket companies



3.3 Shelf-Life Requirements

3.3.1 Supermarket

Each supermarket chain requires their manufacturers to use a shelf-life of 14 days from the date sausages are packed, so-called 'Packed-on plus 14'. To allow for weekends and for shipping, manufacturers aim to achieve a shelf-life of at least 18 days. Supermarkets also impose specifications for end-of-shelf-life microbiology with a typical specification

stipulating TVC no more than $10^7/g$ and *E. coli* no more than 10/g at Day 18. Each supplier is required to 'validate' that their process can meet the specification and to verify production by regular testing.

Typically, supermarkets require at least 75% of the shelf-life in their DCs and retail stores i.e. product should be delivered to the DC no later than three days after production. In Australia this requirement is onerous when product is transported between Sydney and Perth, or between Brisbane and Cairns.

3.3.2 Local and remote supply requirements

Shelf-life requirements for locally-supplied sausages are much less onerous because the supply chains are either non-existent (over-the-counter retail sale) or very short (local wholesale delivery).

3.4 Key Findings

1. Supply chains vary widely between trade in urban and local/remote communities.
2. In Australia and New Zealand, the latter are supplied by butcher shops and small independent supermarkets.
3. In cities and towns supermarkets, specialty shops and delis are major suppliers.
4. Shelf-life requirements also vary between these two supply chains.
5. Butcher shops require only a few days (one to five) shelf-life and can make to order when required.
6. High-volume suppliers require up to 18 days allowing for manufacture, transport to the supermarket DC, storage, retail and home use.

3.4 Data Gaps

1. Production volumes in each of the three sectors; butcher shops, medium and high-volume manufacturers.
2. Sulphite levels in each sector.

References

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4 Storage and Retail of Raw Meat Sausages in Australia and New Zealand

Fresh sausages are considered as a potentially hazardous food and as such their storage is governed by *Food Standards Code Standard 3.2.2* which stipulates they must be stored at no warmer than 5°C at premises and during transport. This requirement applies to all processed and manufactured meats, including raw sausages.

4.1 Refrigerated Transport

For transport to and from distribution centres, refrigerated vehicles are used with sufficient refrigeration capacity to maintain product temperature in accordance with Standard 3.2.2. In fact, certain areas of the load may become chilled to cooler than 5°C with the possibility of some parts reaching zero. In Australia, this is especially important for trans-continental loads such as Sydney-Perth and Sydney-Darwin, where product is under refrigeration for around 48 hours. In New Zealand it is believed that each of the supermarket companies has suppliers and a distribution centre in each island, leading to distribution times of no more than one day (Rob Archibald personal communication).

For local transport, *AS 4696:2007* (Australian Standard for the Hygienic Production and Transportation of Meat and Meat Products for Human Consumption) states that “*The meat carrying compartments of meat transport vehicles are adequately insulated and supplied with operating refrigeration equipment*” (Clause 25.9). A refrigerated vehicle is used in all states except New South Wales where a non-refrigerated vehicle may be used for small consignments over short journeys during which product temperature will not become warmer than 5°C at the site of microbiological concern, which is the surface of sausage (Meat Standards Committee February 17, 2006). The temperature of incoming stock is required to conform with *AS 4694:2007* and be no warmer than 5°C at the site of microbiological concern. Verification of receipt (incoming) temperatures is required as part of an enterprise’s food safety plan.

4.2 Chill Storage

Large chill stores at manufacturing plants, distribution centres and supermarkets, together with small chillers at butcher shops, generally have an air temperature no warmer than 5°C and when closed overnight, will cool to 2-3°C.

4.3 Retail Storage

4.3.1 Supermarkets

Fresh sausages are generally stored on retail display shelves, where an air curtain maintains product temperature, or in refrigerated chambers. There are no data which describe temperatures in retail cabinets of fresh sausages *per se*. However, during the summer of 2005-06 a survey of the temperature of poultry meats (whole birds and portions) at the retail

level was undertaken in South Australia (Pointon *et al.*, in preparation). The surface temperature of each sample was scanned repeatedly before being removed from the retail display and the most common temperature recorded. Since sausages and poultry are stored in identical display cabinets the data have utility for the present report and are presented in Tables 4.1 and 4.2. Mean temperature of product in South Australian supermarkets was 2.1°C.

Table 4.1: Temperature profile of chicken meat on retail shelves in supermarkets in South Australia (n=141)

Temperature (°C)	South Australia
Minimum	-4.8
Mean	2.1
Median	2
90 th percentile	4.6
99 th percentile	7.5
Maximum	8.0

4.3.2 Butcher shops

As for supermarkets, temperature measurements of chicken meats were collected from butcher shops in the study by Pointon *et al.* (in preparation). In South Australia, the mean product temperature in butcher shops was 3.8°C.

Table 4.2: Temperature profile of chicken meat on retail shelves in butcher shops in South Australia (n=14)

Temperature (°C)	South Australia
Minimum	-1.8
Mean	3.8
Median	4.2
90 th percentile	6.2
99 th percentile	6.5
Maximum	6.5

4.4 Retail of Fresh Sausages

4.4.1 Supermarkets

Labelled traypacks are stored back-of-house until filled onto shelves. For supermarkets sausage retailing is much less flexible because sales cannot be completely predicted. An important part of supermarket retailing is to give the appearance of full shelves. This means, on occasion, product will need to be discounted on special as a prelude to ‘shrinking’ unsold stock which has passed its use-by date.

4.4.2 Delis

Delicatessens and specialist food shops retail raw sausages either as their own brand or under a manufacturer's brand. Typically, bulk packs containing 5 kg sausages are received and packed in-store in tray packs and overwraps as required. This allows the bulk pack to be stored back-of-house at a cooler temperature than in lighted retail displays. Manufacturers may supply labels which are affixed during packing in-store.

4.3.3 Butcher shops

For sausage manufacture in retail butcher shops there is great flexibility. A new batch can be made very quickly in response to sales and orders. Butchers keep cartons of frozen trim to augment trim from their own carcasses and most butchers make sausages at least twice weekly. Shelf-life need never be a problem and butchers rarely 'shrink' (discard) sausages.

4.5 Key Findings

1. In Australia, transport from high-volume manufacturers to supermarket DCs is onerous, involving journeys of two to three days e.g. Sydney-Perth and Sydney-Darwin.
2. In New Zealand, distribution times are much shorter (typically less than 24 hours) because supermarket chains have suppliers and DCs in each island.
3. Supermarkets typically require 75% of the shelf-life remaining when stock is delivered to the DC.
4. Supermarkets have a policy of discounting sausages as the use-by date (packed on date +14 days) approaches.
5. Butcher shops have no such requirements and can almost make to order.

4.6 Data Gaps

1. Temperature:time data for product through the production, distribution and retailing phases.

References

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5 Changes to Manufacturing which will allow Reduction in Sulphite Levels

There are numerous ingredient and process changes in sausage manufacture which have been shown to be more effective than sulphite addition. Each will be evaluated in terms of the following criteria, all of which influence shelf-life, including microbiological and sensorial attributes:

- Providing an 18-day shelf-life at temperatures used in the commercial cold chain.
- Conforming with a maximum total viable count of 10^7 /g at 18 days.
- Preserving the bloom (red colour) needed in pork sausages.
- Avoiding oxidative changes.

In other words, any change in formulation or manufacture should not compromise either microbiological or sensory aspects of shelf-life. For some supermarkets this may involve manufacturing at more sites which will increase the company's carbon footprint, a factor which is becoming important to some consumers.

As seen in Section 2.5, the mean TVC for freshly-made sausages at two major Australian manufacturers was $\log 4.4$ /g which allows an increase of 2.5 log units to remain below the specification of 10^7 /g at end of shelf-life. These key points ($\log 4.4$ /g at manufacture and $\log 7$ /g at end of shelf-life) will be used to evaluate the effectiveness of modifications to formulation or manufacturing process.

5.1 Formulation Changes

A number of studies assessing the effect of various alternative antimicrobials are summarised in this section.

5.1.1 Bacteriocins

Bacteriocins, produced as excretory products by certain lactic acid bacteria, have anti-bacterial properties, in some cases against a wide spectrum of bacteria. Three bacteriocins, nisin, lactacin and carnocin have been evaluated as replacers for sulphite in raw sausage.

Scannell *et al.* (1997) used nisin alone and in combination with sodium lactate and sodium citrate, as ingredients in pork sausage stored at 4°C for 10 days. Sodium lactate (2%) combined with nisin (500 IU g⁻¹) was superior to 450 mg/kg sulphite (the final APC was 1.5×10^7 /g compared with 1×10^8 /g. However, since no data were presented on initial APC it is not possible to evaluate whether an 18 day shelf-life is possible using this combination.

Scannell *et al.* (2000) further evaluated lactacin alone and in combination with lactate, citrate and nisin. Lactacin plus citrate allowed a 2.5 log increase and lactacin plus lactate a 1.6 log increase when used in sausages stored at 4°C for 14 days.

Roller *et al.* (2002) employed carnocin as an ingredient in UK pork sausages; both alone and in combination with chitosan, carnocin was no more effective than sulphite and counts exceeded 10^7 /g within four days at 7°C.

5.1.2 Chitosan

Chitosan is a form of chitin, a polysaccharide found in the shells of crustaceans, which has attracted interest as an anti-microbial agent against a range of foodborne bacteria. Its role as a meat preservative has been reviewed by Darmadji and Izumimoto (1994). Roller *et al.* (2002) found chitosan with low sulphite (170 mg/kg) extremely effective at maintaining shelf-life with 10^7 /g being attained after 18 days at 4°C, composed largely of Gram-positive organisms. In terms of sensory attributes chitosan plus low sulphite was superior to other treatments and to a sulphite control. It should be noted however, that chitosan/sulphite sausages had a much lower (10^3 /g) initial count than those with other treatments (10^4 - 10^5 /g).

5.1.3 Lactate

Potassium lactate has been used for some years as an ingredient in cured, cooked meats which are destined to be sliced and eaten without further heating (RTE meats). Potassium lactate in raw sausages has been evaluated by Seyfert *et al.* (2006) where sausages held in lighted retail displays at around 4.3°C for 10 days showed a 1.8 log increase to log 5.9/g. It is likely that an 18-day shelf-life would be achieved with back-of-house storage plus one to two days in retail display. The researchers used retail display to accentuate any colour loss and lipid oxidation, both of which proceed more rapidly in lighted conditions. Sausages with lactate had good colour retention without significant oxidation.

5.2 Packaging Changes

Changing the gas mix in the space around products, modified atmosphere packaging (MAP), has been used since the 1930s when the Australian and New Zealand meat exporters packed carcasses in containers with elevated carbon dioxide levels. More recently, retail-ready meats have been packed in MA with a substantial headspace to give a gas:product ratio of at least 2:1. Modified atmosphere packaging is widely used for processed meats but has not been used for raw sausages, probably due to the relatively high cost of packaging compared with the low value of fresh sausages.

5.2.1 Vacuum packing

The efficacy of vacuum packing of fresh sausages was demonstrated by Adams *et al.* (1987) in the UK. In aerobic packs, sausages reached 10^7 /g in four days at 6°C and in 10 days in vacuum packs. One major manufacturer in Australia uses this vacuum-packing mainly for high-value sausages sold in delis and specialist shops, where the shelf-life is relatively short. The cost of the container, rigid plastic base and impermeable top web, is greater than the overwrap/polystyrene tray used for lower-value sausages.

5.2.2 MAP

There have been several studies in which raw sausages were stored under modified atmospheres. In Italy, Tremonte *et al.* (2005) packed sulphite-free pork sausages in film with low gas permeability, removed air by vacuum packing and introduced modified atmospheres. A gas mix of CO₂:O₂ (40:60) resulted in a total bacterial count of 10^2 /g after 10 days storage at 4°C, falling from an initial 10^4 /g; the colour was good.

In Spain, Martinez *et al.* (2006) packed sulphite-free pork sausages in a mix of CO₂:O₂:N₂ (20:0:80) and included an oxygen scavenger in the pack. After 20 days at 2°C the total count had risen from log 4.8/g to log 7/g. The pork sausages had good colour using this gas mix.

In the USA, Laury and Sebranek (2007) stored sulphite-free sausages at 2-4°C/21 days in an atmosphere of CO₂:CO (99.6:0.4) during which time the total count rose from log 3.5/g to log 7/g. The sausages had good colour but purge (fluid loss into the pack) was more pronounced than in controls with low CO₂. The authors recommended trialling gas mixtures with lower proportions of carbon dioxide.

5.3 Storage Temperature Changes

There is no published information on temperatures used in the manufacturing, storage, transport and retailing continuum. It is usually stated, anecdotally, that the cold chain maintains temperatures between close to zero and +5°C. Until evidence is obtained of temperature:time relationships it is not possible to assess whether there is potential for improved temperature storage. It is clear, however, that temperature:time regimes are a major influence on shelf-life and managing this regime at seven sites: production, storage at manufacturer, transport to DC, storage at DC, transport to retail store, storage back-of-house and display requires management by numerous parties.

5.4 Raw Material Changes

In a recent study (Anon., 2007), the mean total count on freshly-produced sausages was log 4.4/g with counts ranging from log 3.8/g to log 5.3/g. Compared with levels for boneless beef (see Section 2.3) determined in the 3rd national baseline study, of mean TVC log 1.3/g and 90th percentile of log 2.3/g (Phillips *et al.*, 2006), the increase of 2-3 log units during grinding and mixing indicates potential for improvement at the production level.

5.5 Blanching

Blanching of raw sausages is increasingly being used for the food service trade because:

- Blanching pasteurises the product so that a high proportion of spoilage and pathogenic bacteria are killed. Food safety and shelf life are improved.
- Blanched sausages can be vacuum packed, which protects them physically and further extends the shelf life.

Blanching and vacuum packaging extends the shelf-life, reduces preparation time at the food service venue and may also reduce pathogen loads.

5.6 Freezing

Freezing and frozen storage of raw sausages has not achieved significant volume, possibly because chilled sausages have filled the market niche.

5.7 Key Findings

1. A single processing step by itself may not provide a viable option as a sulphite substitute.
2. Several process modifications, in combination as detailed, appear potentially useful.
3. Significant R&D by manufacturers will be required to assess various options and their overall cost-effectiveness compared with sulphite addition.
4. If sulphite is reduced or removed from sausage formulations it need have little effect on trade from butcher shops since batches can be made according to sales of a particular line.
5. For both medium and high-volume manufacturers any reduction in sulphite will be onerous and will require changes to either formulation and/or packaging.
6. Various anti-microbial agents have been shown to be capable of replacing sulphite in sausages but some (e.g. nisin) are expensive.
7. Lactate is currently used by smallgoods manufacturers for controlling *L. monocytogenes* in RTE meats and is relatively cheap.
8. Packaging retail-ready overwrapped packs in a master carton with modified atmosphere may also be effective in extending shelf-life of low-sulphite or sulphite-free sausages.

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6 Potential Impact of Lowering Sulphite Levels in Raw Meat Sausages

In this section the impact of reducing sulphite levels will be considered separately for four categories of operation:

- Retail butcher
- Medium-sized operations
- Sausages for food service
- High-volume operations servicing supermarkets.

Two levels of reduction will be considered:

- Reduction of 60% to 200 mg/kg ingoing
- Complete removal of sulphite.

The latter is included because both Australian supermarket chains stated that some consumers request reduction or elimination of sulphite where possible.

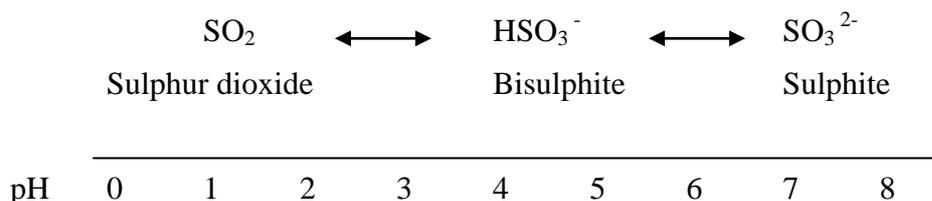
6.1 Effects of Sulphite Addition

6.1.1 Antimicrobial activity of sulphite

Sulphur dioxide (SO₂) has been used for several decades as a means of preserving raw sausages to obtain a marketable shelf-life. The necessity for its use was based primarily on the poor microbiological quality of beef trimmings used in sausage manufacture. Surveys of retail raw sausages in several countries pointed to extremely high total bacterial loadings. At retail for example, in the UK, Adams *et al.* (1987) found counts between 10⁵-10⁷/g; in the USA, Surkiewicz *et al.* (1972) determined a median retail count of 10⁶/g; in NZ, counts ranged between 10⁴ and 10⁷/g (Sumner *et al.*, 1979) while at production level in Canada the geometric mean was 4.5 x 10⁵/g (Farber *et al.*, 1988). It is instructive that, in 1988, the Institute of Food Science and Technology (UK) published a monograph *Preservatives in Food* (Anon., 1998) which expressed the view that, because of the trend towards centralised production and longer supermarket shelf-life, preservatives were *needed* in a range of foods.

Sulphur dioxide (SO₂) is a colourless gas with a suffocating odour. It is applied to foods and beverages as a liquefied gas or, more usually, in the form of sulphite, bisulphite or metabisulphite salts. When sodium metabisulphite (Na₂S₂O₅) is added during sausage manufacture it dissociates into a number of moieties, depending on pH.

As pH falls the proportion of sulphite ions is decreased and the proportion of sulphur dioxide increased:



Therefore, at the pH of raw sausages (around pH 6), metabisulphite ($S_2O_5^{2-}$) dissociates mainly to bisulphite (HSO_3^-) and also to sulphite (SO_3^{2-}), with sulphur dioxide being an unlikely product (Banks and Board, 1982).

The mechanism by which sulphur dioxide has its preservative effect is the inhibition of the aerobic, psychrotrophic, Gram-negative bacteria which are associated with spoilage of refrigerated red meats. Inhibition of Gram-negatives leads to a microbiota, fermentative in nature and dominated by Gram-positive bacteria and yeasts (Roller *et al.*, 2002). Fermentative growth is known to produce acetaldehyde which binds with sulphite and reduces antimicrobial activity (Banks *et al.*, 1987).

In research undertaken under the auspices of the Australian Food and Grocery Council (AFGC) in 2007 the effect of different sulphite levels on microbial growth of raw sausages was studied (Anon., 2007). The data, summarised in Figure 6.1, indicate that, in sulphite-free sausages, the critical level associated with incipient spoilage ($10^7/g$), was reached in four days at 4°C from a starting level of approximately $10^5/g$, a 2-log increase in four days. As the in-going level of sulphite was increased, shelf-life was extended: six days at 120 mg/kg, nine days at 200 mg/kg, 11 days at 350 mg/kg and 18 days at 500 and 600 mg/kg.

6.1.2 Sensory attributes

In pork sausages, sulphites promote the retention of the red ‘bloom’ which is a positive attribute for consumers. In the AFGC study (Anon., 2007) ‘colour’ and ‘saleability’ were assessed by methods not disclosed. However, for the purposes of the present report the assessments of Anon. (2007) will be accepted. Apart from sulphite-free sausages, both sensory attributes closely followed microbial increase, with product becoming unacceptable around the same time that total bacterial counts reached $10^7/g$.

6.1.3 Sulphite levels during storage

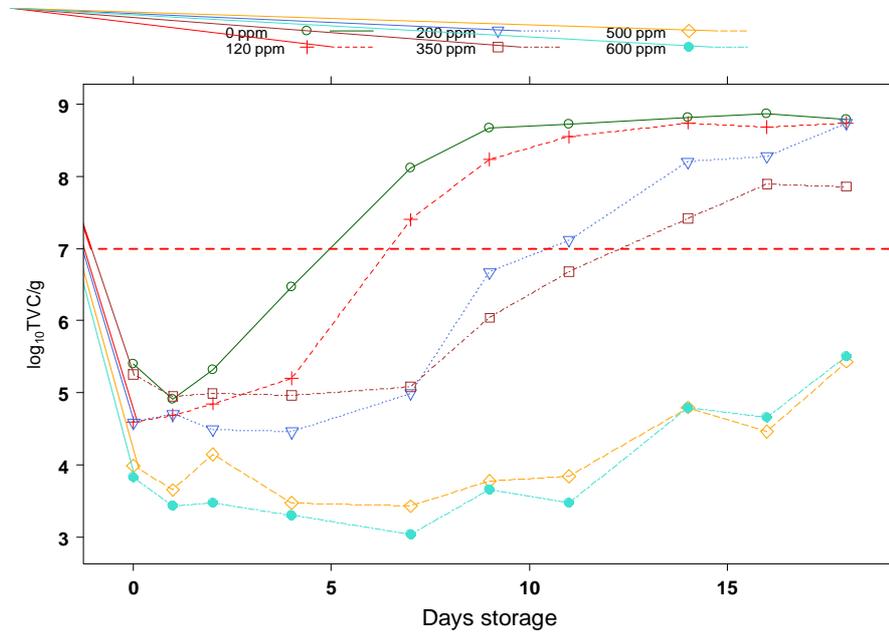
It is known that sulphite levels fall during storage (Banks and Board, 1982). In the AFGC study, sulphite levels were determined throughout an 18-day storage period at 4°C (Figure 6.2). From Figure 6.1 it is clear from Day 0 determinations that sulphite, irrespective of level of addition, exhibited an initial reduction from the declared in-going concentration. Thereafter, throughout the storage period, sulphite levels fell gradually and at the day when microbial count reached $10^7/g$, ranged from 40 mg/kg (ingoing 120 mg/kg) to 250 mg/kg (ingoing 600 mg/kg). The study indicates that around 200 mg/kg residual sulphite is needed for shelf-life to attain that required in the supermarket cold chain.

6.1.4 Sulphite in raw sausages – what is being measured?

Banks and Board (1982) state that, in sausages, HSO_3^- and SO_3^{2-} are lost by oxidation and that they combine irreversibly with pyruvate, aldehydes and ketones and also form addition compounds with glucose and maltose, all of which are present in significant quantities in raw sausages. Once bound, antimicrobial activity is lost and it is presumed that preservation is dependent on presence of unbound HSO_3^- and SO_3^{2-} , which are loosely termed ‘free sulphite’. In the AFGC study the method is not disclosed but it is believed the Monier-Williams, distil and titrate method was used. This method was used to determine only ‘total sulphite’, which is a combination of ‘free’ and ‘bound’ sulphite. However, the study did not measure free sulphite, which is relevant from the shelf-life and allergen viewpoint.

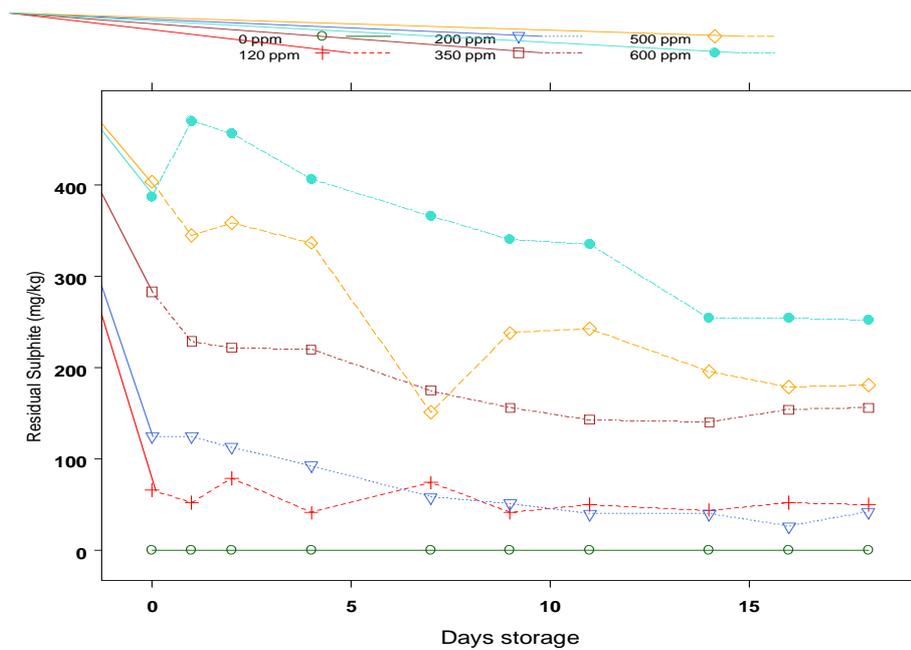
It is against this background of sulphite as a preservative and an allergen that the impact of reduced sulphite levels in sausage meat is considered.

Figure 6.1: Effect of sulphite level on microbial growth in sausages stored in aerobic packaging at 4°C (Anon., 2007)¹



¹ Batches were manufactured at different times, from different raw meat ingredients which influences variability of initial bacterial loadings; data represent a single analysis on each sampling occasion

Figure 6.2: Effect of in-going sulphite level on residual sulphite in sausages stored at 4°C (Anon., 2007)



6.2 Impact on Retail Butchers

Retail butchers use predominantly surface trim from carcasses which may be four to five days old. If 200 mg/kg sulphite were allowed it is possible that there would be no impact on retail trade. If sulphite were eliminated, for retail butchers who manufacture three to five times a week, there need be no real impact other than to inform customers of the need to refrigerate the product and to consume within two days. However, for those who make once or twice a week there would be an impact in that sausages would need to be made in smaller batches and more frequently.

6.3 Impact on Medium-Sized Manufacturers

Medium sized manufacturers use a combination of surface trim and beef trim purchased to provide correct fat content of finished sausages. By using sulphite such operations can guarantee a five-day shelf-life. By manufacturing each week day it is unlikely that product will spoil in the normal retail chain or in the wholesale chain to schools, institutions and independent supermarkets. The process is relatively inflexible; carcasses are delivered usually twice weekly and trim is produced as these are boned for primal and subprimal cuts. Carcasses are usually three to four days old when boned to yield surface trim, which may be held over the weekend before comminuting into sausage batter.

The only improvement to the process would be to keep product cooler during comminuting by addition of flake ice and/or dry ice. This would add to the cost of production and may not be sufficient to extend shelf-life. It follows that significant changes would be needed to the formulation or packaging to guarantee shelf-life at its current level. These modifications are described in Section 6.5 and individual manufacturers will need to ascertain which modification(s) will be required.

6.4 Impact on Suppliers of Sausages for Food Service

An intervention, blanching (see Section 5.5), is already used for supplying ‘raw’ sausages for the food service market, though this is mainly intended to facilitate the cooking and serving process, rather than for shelf-life. The technology could be effective in tandem with freezing (blanch/freeze) though there would be a cost impost brought about by additional processes: freezing, storage, thawing. It may also be necessary to alter formulations to minimise the effects of fat oxidation during frozen storage.

6.5 Impact on High-Volume Manufacturers

6.5.1 Reduction to 200 mg/kg ingoing

A reduction to 200 mg/kg has been chosen arbitrarily because it:

- Represents a significant (60%) reduction in ingoing sulphite level.
- Allows some shelf-life extension, to around nine days, based on the shelf-life study.
- Has a low residual sulphite level at the end of shelf-life (around 40 mg/kg after cooking).

For high-volume sausage manufacturers and their supermarket customers the effect of removing 60% of the sulphite from the formulation would be onerous, reducing shelf-life to an estimated nine days. If no changes were made to either the formulation or the manufacturing process the trade would be sustained only by:

- Manufacture of smaller batches
- Increased frequency of distribution from DC to supermarket
- More frequent stocking of shelves
- Increased discounting as stock approaches end of life
- Increased shrinkage.

Estimating the costs flowing from each of these elements would involve in-depth knowledge of various businesses and is beyond the scope of the present study.

6.5.2 Sulphite-free sausage

The concept of sulphite-free sausages is an attractive proposition for supermarket companies since it aligns with what a significant proportion of consumers would regard as a positive. It goes without saying, however, that it would not be possible to market fresh sausages using current formulations, production and packaging methods. This is clear from both the sensory and microbiological data which showed sulphite-free sausages stored at 4°C to have unacceptable colour and ‘saleability’ on Day 2 while, Day 4 TVC approached 10⁷/g. Options for achieving shelf-life are presented below, adoption of any of which will involve significant R&D.

(i) Replacing sulphite with lactate

While there are numerous antimicrobial ingredients (Section 5.1) which have been shown to prolong the shelf-life of raw sausages to that achieved by sulphite at 500 mg/kg, bacteriocins and chitosan are ‘new’ to the industry and expensive. By contrast, the use of lactate alone or in combination with diacetate is already routinely used by all large smallgoods manufacturers in the control of growth of *L. monocytogenes* in cooked, ready-to-eat meats. There are a number of commercial blends of lactate and/or diacetate available and the system has been subjected to challenge testing by the Australian Food Safety Centre of Excellence (AFSCoE) and published by Mellefont and Ross (2007). Although the use of lactate/diacetate in raw sausages is an extension of existing commercial practice, there may well be the need for R&D on formulation as lactates/diacetates impart an acidic flavour note.

(ii) Using modified atmospheres

Storage and distribution in the USA relies heavily on modified atmosphere packaging of retail-ready packs in master cartons with high oxygen (for meat colour) and carbon dioxide (for inhibition of Gram-negative bacteria). Technology for gas flushing master packs is well-established e.g. ‘Snorkel’ equipment which removes air and flushes gas mix throughout the pack. Modified atmosphere packaging will impose additional cost.

(iii) Improved process control

Since the time to end of shelf-life is primarily influenced by the microbiology of ingredients plus control of temperature throughout production, storage and distribution, there is a need for improved process and storage control at each level of the commercial chain. Managing temperature:time regimes at all stages in the chain: production, storage at manufacturer, transport to DC, storage at DC, transport to retail store, storage back-of-house and display requires control by numerous parties. Through-chain management is never easy, especially where different companies or divisions within a company share responsibility.

6.6 Financial Aspects of Sulphite Reduction/Elimination

6.6.1 Butcher shops

The impact of withdrawing sulphite completely from sausage formulations would be confined to retail butchers who make batches only once or twice a week. For this group there would be the need to make smaller batches, more frequently. If a batch of sausages takes 30 minutes to manufacture this cost would need to be apportioned through the total value of all product made during the week.

6.6.2 Medium-sized operations

Because medium-sized manufacturers supply institutions and independent supermarkets they lack control over product handling, potentially by hundreds of end-users (consumers and institution staff). At present, the risk of product failure (spoilage) is reduced by use of sulphite, which may be as high as 700-800 mg/kg in-going (Anonymous, personal communication) and is depleted to around 500 mg/kg within the first 24 hours.

For medium-sized operations reduction to 200 mg/kg sulphite would effectively be the same as complete elimination, as the risk of product failure is tangible. Medium-sized operations would be required to undertake similar changes to high-volume manufacturers without the benefit of economy of scale. It is possible that their market niches may be lost to high-volume manufacturers.

6.6.3 High-volume manufacturers

(i) Reduction to 200 mg/kg sulphite

Assuming reduction of in-going sulphite to 200 mg/kg with a likely shelf-life of nine days and no changes made to formulation or manufacturing process the trade would be maintained only by one or more of the following strategies (listed previously in Section 6.5.1):

- Manufacture of smaller batches
- Increased frequency of distribution from DC to supermarket
- More frequent stocking of shelves
- Increased discounting as stock approaches end of life
- Increased shrinkage.

Estimating the costs flowing from each of these elements requires in-depth knowledge of various business models. For example, in Australia, supermarket company A has six suppliers (one in each state) plus one DC in each state while supermarket company B has two suppliers nationally and DCs in each state. Clearly these companies have very different business models and reducing sulphite levels would apply differently to each of them. Each is also concerned with their current carbon footprint, which would increase in response to handling smaller batches since trucks would make more journeys.

(ii) Elimination of sulphite

High-volume manufacturers would be required to undertake significant R&D to provide 18-days shelf-life to the supermarkets. The costs of this phase are a matter of conjecture but are likely to be in the hundreds of thousands of dollars for each manufacturer. Any likely solution will involve changes to product formulations plus changes to packaging. Financial analysis will involve both current (baseline) costs of production and packaging compared with those of the outcomes of R&D by each manufacturer. It may be that some manufacturers will not be able to accommodate their customers and the recent rationalisation of the smallgoods industry will continue.

One possible solution, described for North America (see Section 7.2) involves modified atmosphere packaging of master cartons and temperature:time control sufficient to provide one week in-store shelf-life. It should be emphasised, however, that lactate use at the slaughter floor, boning room (fabrication) levels has become widespread in the USA and it may be that lactate use has already been factored in to some extent.

Given the foregoing and the lack of any cost disclosure by any manufacturer or supermarket chain it is not possible to proceed with a financial analysis.

6.6.4 Institutional and food service

For manufacturers which supply the food service sector there is added risk because of the possibility of temperature:time abuse, especially at venues which lack full-scale preparation and temperature control facilities. It may be that manufacturers would see post-blanch freezing as the only solution. Added costs therefore include:

- Packaging in liners and cartons
- Freezing
- Transport to frozen storage
- Frozen storage
- Thawing.

It is likely that food service companies would pass on increased costs to customers since there is little consumer knowledge of costs and no means of comparison as there is at retail.

6.7 Key Findings

1. Butcher shops will likely have little impact from reduction or elimination of sulphite.

2. Butchers who manufacture only once or twice weekly will likely need to make more batches, which will add to the cost.
3. Medium-sized manufacturers lack flexibility and technical resources necessary to accommodate reductions to in-going sulphite levels.
4. This sector is vulnerable to temperature abuse by end-users.
5. For suppliers to the food service sector, freezing may be the only alternative to accommodate reduced sulphite levels.
6. For high-volume manufacturers, reducing or eliminating sulphite from sausages will require large-scale changes to formulation and packaging methods.
7. Significant R&D by high-volume manufacturers will result in cost to be passed on to consumers.
8. A number of key findings under Section 5.7 also apply.
9. Response by supermarkets and high-volume manufacturers will vary depending on their current and preferred responsive business models.
10. Financial analysis will be company-specific.

6.8 Data Gaps

1. A financial analysis will require a large volume of information from key players (manufacturers and supermarkets).
2. Currently this information is not forthcoming.

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7 Processed Meats in Europe and North America

There are significant differences in how processed meats are regulated and manufactured in overseas countries. There is also evidence that microbiological profiles of meat raw materials differ. Some of these differences have relevance for the present consideration and are detailed later in this section.

7.1 Europe

In many ways European countries regulate processed meats in general and raw sausages in particular, in a similar way to Australia. Exceptions are RTE meats where *L. monocytogenes* in some countries is not regulated by a zero tolerance approach as it is in Australia and New Zealand. Germany and Denmark, for example, have a risk-based approach to RTE meats, allowing up to 100 cfu/g for products which support the growth of *L. monocytogenes* but which have short shelf-lives.

7.1.1 Manufacturing processes

In terms of raw sausages, generally known as ‘breakfast sausages’, sulphite is allowed up to 450 mg/kg. In many respects the industry is similar to that of Australia and New Zealand, being based on supermarkets supplied by a small number of high-volume manufacturers together with a large number of low-volume butcher shops. Raw sausages manufactured in the UK and Ireland are formulated in a similar way to those in Australia and New Zealand. Shelf-life requires sulphite to augment temperature control through the marketing cold chain. There are no UK statistics available on incoming meat quality and for the purposes of the present report, it is assumed that this is not significantly different from that in Australia or New Zealand.

7.1.2 Production and plant hygiene

At both high-volume and butcher shops, production is identical to that in Australasia, using the same equipment and the same unit operations as set out in Figure 2.1, with large manufacturers using a mixture of frozen and chilled trim specifically cut for sausage manufacture or retail butchers using surface trim. It is likely that hygiene at both types of operation is also similar to that of Australasian plants.

7.1.3 Distribution storage, transport and retailing

For all practical purposes the systems of distribution, storage, transport and retailing are essentially similar in Europe compared with Australasia.

7.2 North America

For control of *L. monocytogenes* in RTE meats, the USA introduced an interim rule (Anon., 2003) which was risk-based and which grouped establishments into three categories.

Category 1 includes establishments which embrace two risk mitigation strategies: addition of antimicrobial ingredients such as lactate and diacetate plus in-pack pasteurisation. Category 2 establishments use only one of these mitigations while Category 3 rely solely on hygiene measures and product testing. The regulatory response is to test more stringently in establishments which are not Category 1.

7.2.2 Manufacturing processes

In terms of raw sausages, sulphite has not been allowed for many years, despite which there are occasional recalls of sausages containing sulphite. For example, a recent Recall Release (FSIS-RC-037-2007) cites the recall of around 8,000 t of gourmet sausages from a producer in Buffalo, New York. Levels of sulphite are not disclosed in the recall notice but it is noted that all products contained wine which may have been the source of the sulphite contamination.

The industry in North America is predominantly from high-volume manufacturers with small butcher shops being less prominent than in Australia and New Zealand.

In North America, sausage manufacture is predominantly from chilled trim packed into combo bins which hold around 900 kg. The combo bin as a unit presents significant differences from Australia and New Zealand where the carton is the unit for packaging and transport. Combo bins are shipped both intra- and interstate for further processing, most notably for grinding into hamburger patties, but also for sausage making.

7.2.2 Production and plant hygiene

Three datasets on raw materials used in the USA for sausage manufacture are available, two on beef trimmings and one on pork trimmings. Scanga *et al.* (2000) surveyed beef trimmings destined for grinding from eight commercial packing and grinding facilities across the USA. Samples were withdrawn from combo bins by drilling into the core at five points. Samples from frozen cartons were taken by drilling meat shavings from the block; a proportion of the frozen samples were imported beef trimmings. Summary data, which provide a microbiological profile of beef trim used for grinding in the USA, are presented in Table 7.1.

Table 7.1: Microbiological profile of chilled and frozen beef trimmings in USA (Scanga *et al.*, 2000)

	Chilled trimmings	Frozen trimmings
APC (mean log cfu/g)	3.3	2.9
<i>E. coli</i> (mean log cfu/g)	1.1	1.1
<i>S. aureus</i> (mean log cfu/g)	1.0	1.0
<i>L. monocytogenes</i> prevalence	1.6	6.3
<i>Salmonella</i> prevalence	3.1	Not detected

The data indicate a slightly higher total bacterial loading in chilled product compared with frozen product, with the former having a higher prevalence of *Salmonella* and the latter of *L. monocytogenes*. However, an important aspect of using chilled trimmings in combo bins is that they may be shipped to distant grinding plants. The effect of shipping was determined by

Scanga *et al.* (2000) by sampling combo bins over the period between production and four days storage (Table 7.2).

Table 7.2: Aerobic Plate and *E. coli* count on chilled beef trimmings over four days following production (Scanga *et al.*, 2000)

	Age of product (days)			
	0	1-2	3-4	>4
APC (mean log cfu/g)	2.7	3.4	3.9	3.4
<i>E. coli</i> (mean log cfu/g)	1.0	1.1	1.3	1.0

It is clear that the total bacterial loading can vary by the order of 1 log unit in chill product, depending on the period between fabrication and grinding.

In a second dataset, Bosilevac *et al.* (2007) compared the microbiological profile of frozen beef trim from three countries which export to the USA (Australia, New Zealand and Uruguay) with that of chilled trim produced domestically. The findings for indicator organisms and for pathogens are presented in Tables 7.3 and 7.4, respectively, with data for New Zealand and Uruguay included for completeness.

Table 7.3: Microbiological profile of indicator organisms in beef trimmings destined for ground beef (Bosilevac *et al.*, 2007)

	Mean log cfu/g	Prevalence (%)		
	APC	<i>Enterobacteriaceae</i>	<i>E. coli</i>	<i>S. aureus</i>
Australia	1.6	8.2	1.0	4.0
New Zealand	2.2	9.0	0.5	8.2
Uruguay	2.8	31.3	9.5	29.5
USA	2.5	37.8	7.2	4.2

As indicated in Table 7.3, microbiological levels of indicator organisms were, in general, substantially lower than those of the USA. Prevalence of pathogens was invariably lower in Australian beef trimmings compared with USA beef trimmings (Table 7.4). This was especially so when positive isolations of Shiga-toxic *E. coli*, in general and HUS serotypes, in particular, are compared.

Table 7.4: Prevalence of pathogens in beef trimmings destined for ground beef (Bosilevac *et al.*, 2007)

	Prevalence (%)			Number of positives	
	<i>Salmonella</i>	<i>Campylobacter</i>	<i>L. monocytogenes</i>	STEC	HUS serotypes
Australia	0	0	2.0	9	0
New Zealand	0.4	0.5	2.3	4	2
Uruguay	0.4	0.4	24	40	6
USA	0.8	1.3	5.0	28	5

From the study, undertaken by USDA researchers, it is clear that beef trimmings imported from Australia have a significantly superior microbiological profile compared with those generated domestically in the USA. The USDA researchers commented: “Overall the results provide objective evidence that standards of hygiene during the slaughter and processing of beef in Australia continue to be very high.”

It is appropriate to consider product from export-registered abattoirs in Australia because product from this sector of the industry predominates in trimming supply for high-volume manufacturers.

The microbiology of ground pork and pork sausages was surveyed by Duffy *et al.* (2001) both at the fabrication and retail levels in the USA. The data are summarised in Table 7.5.

Table 7.5 Mean log₁₀ TVC and *E. coli*/gram in retail level pork/pork sausage (Duffy *et al.*, 2001)

	Number of samples	Mean log ₁₀ TVC/g	Mean log ₁₀ <i>E. coli</i> /g
Freshly-ground	40	3.0	1.0
Pre-packaged	96	3.8	1.0
Store-ground	96	5.6	1.0

7.2.3 Distribution storage, transport and retailing

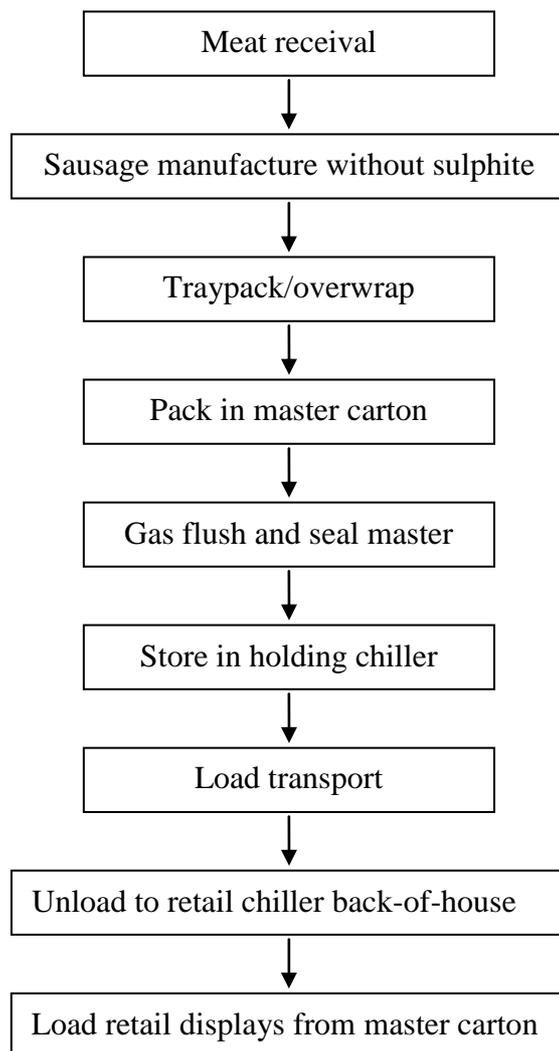
Shipping distances and times in North America can be of the same order as the longest in Australia and obtaining sufficient shelf-life without the use of any form of preservative requires alternative means of inhibiting spoilage bacteria. A typical process is described in Figure 7.1, based on personal communication with Dr. Colin Gill, Agriculture Canada.

Sausages are made without sulphite in a manner similar to that in Australia and packed in trays which are overwrapped in gas-permeable film. Traypacks are then packed into a gas-impermeable bag within a master carton. The bag is vacuumised and then gas flushed with a mix of CO₂:O₂:N₂ (20:60:20). The high carbon dioxide level inhibits growth of Gram-negative spoilers without causing excessive purge and the high oxygen level promotes retention of the red colour (bloom) needed for pork sausages; nitrogen is a filler.

The product has a shelf-life in the cold chain of 12 days and packs are labelled both with packed-on and use-by dates. Date labelling indicates the journey of 3,000 km from production plant to supermarket was achieved two days after packing, allowing up to 10 days for retailing.

The concept of centralised packaging of retail meats is especially well developed in North America, particularly in conjunction with MAP. For a review of the concept see Tewari *et al.* (1999).

Figure 7.1: Packaging and shipping of raw sausage in USA



7.3 Key Findings

1. Other countries e.g. the UK and Ireland, have a trade similar to that of Australia and New Zealand.
2. Centralised packing from a small number of high-volume suppliers services the supermarket trade.
3. The ‘family butcher’ trade in UK is also basically similar to that in Australia and New Zealand.
4. In North America, breakfast sausages are also manufactured, though with more reliance on supermarket than retail butcher trade.
5. In Europe sulphite is allowed up to 450 mg/kg.
6. In North America sulphite has been banned for several decades.

7. In North America, distances between manufacturer and supermarket can be as long as in Australia.
8. A shelf-life of 12 days is required.
9. To achieve marketing and shelf-life of sulphite-free sausages, use is made of modified atmosphere packaging.

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Appendix 1: Microbiological Baseline Survey of Beef Sausages

Objective

Determine the microbiological status of beef sausages in retail trade in Australia.

Background

No survey of the microbiological status of beef sausages has been published for Australian product. Hence, a survey of beef sausages retailed by supermarkets and retail butcher shops in each state capital city was undertaken in late-2007.

Materials and Methods

Sampling Protocol

A total of 53 beef sausages were sampled from retail outlets in capital cities around Australia (Table A.1). Sample numbers were proportionally allocated based on the population estimate of individuals in the five largest capital cities obtained from the 2006 Census (ABS website). Samples were obtained from both supermarkets (80%) and butcher shops (20%). Prior to sampling the major supermarket chains in each city were identified and the supermarket samples distributed amongst the various outlets. Coles and Woolworths/Safeway as the largest chains made up 50% of the supermarket sample.

Table A.1: Total number of beef sausage samples estimated and the actual number collected in each City.

Capital City	Population Estimate	Estimated	Actual
Adelaide	1,105,839	5	5
Sydney	4,119,190	17	19
Melbourne	3,592,591	15	15
Brisbane	1,763,131	7	7
Perth	1,445,078	6	7
TOTAL		50	53

Transport of samples to the laboratory

Samples purchased were kept in the original packaging, placed in insulated containers with ice bricks and transported overnight to the laboratory for testing within 24 hours. At the laboratory, samples were refrigerated at 4°C until testing commenced.

Microbiological testing

The external surfaces of the sausage skins were decontaminated with alcohol wipes. A sterile scalpel was used to cut open the skin and the sausage meat was removed using a sterilised spoon.

From each sample approximately 25 g of beef sausage meat was weighed into a sterile stomacher bag. A decimal dilution was prepared by adding 225 mL of Peptone Saline Solution (PSS) and homogenising for two minutes using a stomacher (IUL Instruments, Barcelona, Spain). Serial 10 fold dilutions were prepared in 9 mL volumes of PSS (Media Production Unit, The University of Melbourne).

Aliquots of 1 mL from each serial dilution were inoculated on to either Petrifilm™ Aerobic Plate Count Plates or Petrifilm™ *E. coli*/Coliform Count Plates (3M Corporation, St Paul, Minnesota) and incubated at 35°C for two days. Selected samples were also tested for Total Plate Count at 25°C for 96 hours. Colonies were identified and counted as per the manufacturers' instructions.

Statistical Analysis

The data for log₁₀ Total Viable Count (TVC) was analysed by analysis of variance (ANOVA). The aim of this test was to determine if there was a significant difference in the mean log₁₀ TVC between butcher shops and supermarkets. The possible variability that could exist between cities was accounted for in the model.

Fishers Exact Test was used to test for differences in the prevalence of *E. coli* in sausage samples from supermarkets and butcher shops.

Results

Days from purchase date to 'use by' date

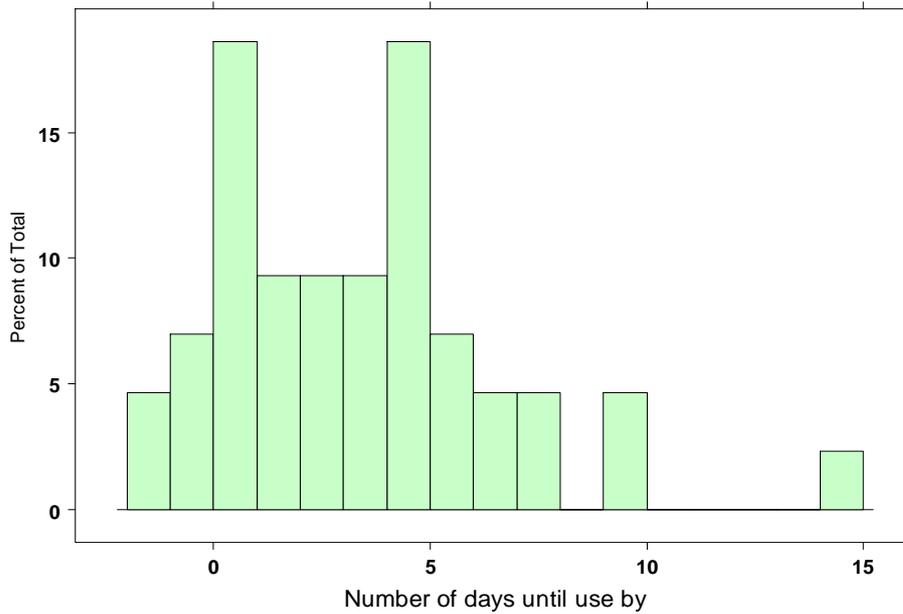
Use-by data were obtained from the labels of supermarket product. From Table A.2 and Figure A.1 it can be seen that, on average, sausages had four days remaining before the use-by was reached. On two samples the use-by date had been exceeded.

Table A.2: Summary of statistics for number of days to use-by date for beef sausages

Sausage	N	Min	1 st Qu	Median	Mean	3 rd Qu	Max	SD
Beef	43	-2.00	1.00	4.00	3.81	5.00	15.00	3.3

Use by dates only available from Supermarkets

Figure A.1: distribution of days until use by for each product type from supermarkets



Product Hygiene Indicators

Total Viable Counts (TVCs) are presented in Figure A.2 and Table A.3 from which it can be seen that samples obtained from butcher shops had on average significantly higher TVC (cfu/g) than those from supermarkets ($p < 0.0001$; mean \log_{10} difference of 1.69).

Figure A.2: Boxplot of \log_{10} TVC (cfu/g) for beef sausages in butcher shops and supermarkets

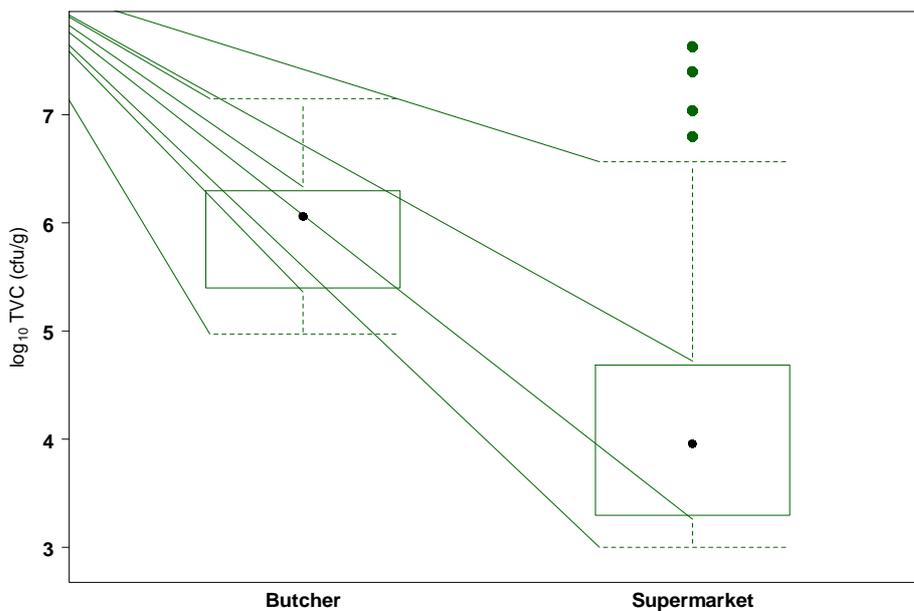


Table A.3: Summary of statistics for log₁₀ TVC (cfu/g) for beef sausages in butcher shops and supermarkets

	Butcher	Supermarket
N	10	43
Min	4.97	3.00
1 st Qu	5.47	3.30
Median	6.01	3.95
Mean	5.96	4.27
3 rd Qu	6.30	4.68
Max	7.15	7.63
SD	0.66	1.26

E. coli was significantly more prevalent in butcher shops compared with supermarkets (p=0.01). Given the small number of samples from butcher shops, this difference should be treated with caution.

Table A.4: Prevalence of *E. coli* found in beef sausages at butcher shops and supermarkets

Outlet	Prevalence
Butcher	6/10 (60%)
Supermarket	7/43 (16%)