

# Consumer assessment of 'tenderstretched' loin steak

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# Introduction

'Tenderstretch' is the name used in Australia to describe a technique for reducing muscle shortening and consequent toughening of meat which can occur when carcasses or sides are cooled before rigor mortis sets in (Locker 1960; Herring et al. 1965a, b; Marsh and Leet 1966; Herring et al. 1967). Previous experiments involving mechanical and analytical taste panel measurements on both fresh and frozen tenderstretched meat in this laboratory (Bouton and Harris 1972; Bouton et al. 1973; Bouton et al. 1973) have shown that hanging a carcass from the aitch bone or sacro-sciatic ligament (tenderstretching) significantly improves the tenderness of the meat from important hindquarter muscles compared to that from a carcass hung in the conventional way from the Achilles tendon. The technique has been adopted in a few Queensland meatworks, but only for a small percentage of animals killed. The Queensland Department of Primary Industries uses a green roller brand to

identify tenderstretched meat. To date no confirmation of the improvement in tenderness has been obtained from consumers. This paper reports the results of a consumer comparison of conventional and tenderstretched loin steaks from steers reared in three different regions of New South Wales. A selection of 125 households in the Brisbane metropolitan area were given packages of both conventional and tenderstretched steaks to cook and eat in their homes as part of a normal meal. They were supplied with score sheets to record their assessment of the meat and the results were statistically analysed.

# Procedures

# Selection of panellists

A total of 125 families from 10 suburbs in the Brisbane metropolitan area took part in the experiment. Suburbs were chosen with the aid of a map of Brisbane, which had been prepared from the 1971 census (Cities



Fig. 1. Diagram showing position of cuts from which the steaks for the consumer trials were prepared. The commonly used names for these steaks are indicated.



Commission 1976), showing distribution patterns of the five socio-economic groups. Two suburbs were selected in each group, one north and one south of the Brisbane river, providing a total of 25 families in each of the five socio-economic groups. They were selected by interview, with the restriction that there was a minimum of two adults in the family who ate steak of grilling quality at least once per week.

# Preparation and distribution of samples

Meat for distribution to the consumers was kindly donated by the N.S.W. Department of Agriculture from an experiment being conducted on crossbred steers. The 120 animals involved were born and reared to weaning at Grafton Agricultural Research Station, and transferred to other locations in New South Wales for fattening. The animals were reared on pastures at Glen Innes, Trangie and Walwa, and slaughtered in commercial abattoirs at Tenterfield (slaughter 1, 41 animals), Mudgee (slaughter 2, 40 animals) and Wodonga (slaughter 3, 37 animals) respectively. Average ages of these groups of animals at slaughter were 21, 22 and 26 months and average carcass weights were 255, 245 and 243 kg respectively.

After slaughter, one side from each animal was hung from the Achilles tendon (normal) and the second side from the sacro-sciatic ligament (tenderstretched) in a chiller for a minimum of 24 h. At Tenterfield chillers were maintained at 2°C; at Mudgee the average temperature was 5°C and at Wodonga 0°C. Striploins and cube rolls were boned out from all sides and frozen before freighting to M.R.L. The striploins were divided into three portions (see Fig. 1). Six 2-cm thick steaks for consumers were sliced from each end portion by means of a band saw, and the central portion was used for laboratory measurements. Consumer steaks were also cut from the posterior end of each cube roll. The steaks were packaged on traypacks sealed with Cryovac D505 film with the aid of a Cryovac automatic packaging machine loaned by Coles New World Supermarkets at Cannon Hill and Sunnybank. All meat was kept frozen at -20°C until delivered to the consumer, so that any increase in tenderness caused by aging was minimized. Consumers were told that the steak was of grilling quality and had not been treated with chemicals in any way.

They were not told about the design of the experiment, or about tenderstretch meat.

Each consumer family received a total of six packs, each containing four to six steaks. From each slaughter they received a conventional and tenderstretch pack of the same cut (e.g. anterior end of striploin) from one animal. The conventional and tenderstretched samples were delivered 1 month apart. There was a gap of 6 weeks between slaughters 1 and 2, and a gap of 13 months between slaughters 2 and 3. Half the consumers received their conventional pack first and the others received their tenderstretch pack first. This delivery order was alternated for subsequent slaughters. Consumers were asked to keep the meat frozen until they were ready to use it, and to cook it within 2 weeks of delivery. Two types of score sheets were delivered with each package of meat. One was for the cook in the household to complete with comments on appearance of the raw meat and questions on cooking techniques (Fig. 2). Consumers were permitted to cook their first package of steaks by any method they chose, but instructions were included with subsequent packs to ensure that this method was used for all six packs. Score sheets were supplied for each of the tasters in the family to fill in before discussing their opinions with other members of the family. They were asked to rate their assessment of taste, tenderness, juiciness and acceptability of the meat (see Fig. 3). A stamped addressed envelope was supplied with the forms for consumers to post their completed score sheets back to the laboratory.

## Statistics

Analyses of variance were performed on the results for each slaughter by the Genstat statistical package on a Cyber 76 computer. The order of delivery was initially included as a covariate. However, this did not yield any additional information, so it was not included in the final analyses. An overall analysis of variance was also completed with time of slaughter as a factor.

# **Results and discussions**

The consumer families cooperated very conscientiously and a high proportion (89%) of all possible replies were received (slaughter 1, 90%; slaughter 2, 91%; slaughter 3, 87%). Replies on the cooks' forms were summarized as percentages, and a taster score for each family was obtained by averaging scores for COOK'S FORM

Package Code

#### CSIRO MEAT RESEARCH LABORATORY

#### Steak Consumer Acceptance Study

Please examine the meat when you unwrap it and answer the questions below when you have cooked the meat.

- 1. How frequently would you normally serve steak?
  Once a month \_\_\_\_\_ Twice a month \_\_\_\_\_ Once a week \_\_\_\_\_
  More frequently \_\_\_\_\_.
- In your opinion was the raw steak too fat \_\_\_\_\_, about right \_\_\_\_\_, too lean \_\_\_\_\_.
- 3. Did you thaw the steak before cooking? YES \_\_\_\_\_ NO \_\_\_\_\_ If YES, how? \_\_\_\_\_

4. Did you use a tenderiser on the meat? YES \_\_\_\_\_ NO \_\_\_\_\_

- 5. How did you cook the steak? Grilled \_\_\_\_, Fried \_\_\_\_, BBQ'd \_\_\_\_, other \_\_\_\_\_. If other, how? \_\_\_\_\_.
- Was the steak "Well done" (no pink meat) \_\_\_\_, Medium (some pink meat) \_\_\_\_, Rare (some raw meat) \_\_\_\_.
- Would you be satisfied with steak like this if you bought it from your normal butcher? YES \_\_\_\_\_, NO \_\_\_\_\_.
- 8. Any further comments (both favourable and unfavourable comments are useful and greatly appreciated)

Fig. 2. Score sheet supplied to the person cooking the steaks.

TASTER'S FORM

Package Code

# CSIRO MEAT RESEARCH LABORATORY

# Steak Consumer Acceptance Study

Please fill in this form as soon as you have finished eating the steak and before you discuss your opinions with other members of the family. Place a cross in the appropriate space for each question.

- In your opinion was the meat? Too Fat \_\_\_\_, about right \_\_\_\_, too lean \_\_\_\_.
- How did the meat taste? Poor \_\_\_\_, fair \_\_\_\_, good \_\_\_\_, excellent \_\_\_\_.
- Was the meat? Very tender \_\_\_\_, tender \_\_\_\_, fairly tender \_\_\_\_, fairly tough \_\_\_\_, tough \_\_\_\_, very tough \_\_\_\_\_.
- Was the meat? Very juicy \_\_\_\_, juicy \_\_\_\_, fairly juicy \_\_\_\_, fairly dry \_\_\_\_, dry \_\_\_\_, very dry \_\_\_\_.
- 5. What best describes your reaction to this steak?



 Additional comments (both favourable and unfavourable comments are useful and greatly appreciated)

Signed	Age	if	under	18		Date	
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Fig. 3. Score sheet supplied to each taster in the family.

all members of the family over 12 years of age.

Most of the cooks (79%) thawed the steaks before cooking. Frying (57%) and grilling (39%) were the most frequently used cooking techniques and well done (62%) and medium (32%) were the most common degrees of cooking.

Tasters judged tenderstretched steaks significantly (P < 0.001) more tender and juicy than the steaks from conventionally hung sides, and also awarded them higher scores for taste and overall acceptability. Differences between sample scores for the three different cuts used were non-significant. The histograms in Fig. 4 show the scores for each attribute as means for each slaughter and an overall mean for the whole experiment. Mean scores for all attributes for slaughter 3 were significantly different (P < 0.001) from those for the first two slaughters. As can be seen from Fig. 4, the greatest effect of tenderstretching was obtained in slaughter 3, where the chiller temperature was lowest, namely 0°C, and where greater toughening of conventional samples owing to cold-shortening would be expected to occur.

There was a high degree of correlation among the tasters' scores for all attributes. All correlation coefficients were greater than 0.73, thus indicating a measure of consistency of response with respect to the different sensory properties. There is no logical explanation for tenderstretching to improve the scores for taste. It is probable that consumers subconsciously include tenderness in their assessment of this attribute, although the question was intended to measure flavour qualities. The improvement in juiciness detected by consumers has been noted in previous laboratory taste panel experiments. In answer to question 7 (Fig. 1), 75% of



Fig. 4. Mean taster scores for each slaughter (S1, S2, S3) and for the whole experiment (All) for 'conventional' and 'tenderstretched' steaks.

cooks receiving tenderstretched samples said they were satisfied with the steaks, but only 55% of those receiving conventional samples said they were satisfied. This difference is statistically significant (P < 0.01) and shows that the improvement in meat tenderness produced by using the tenderstretched hanging technique is apparent in a real life situation. An acceptance of 55% is low for good quality steak, and probably reflects a prejudice against frozen meat by consumers. Unfortunately it would be impracticable to conduct this type of experiment with fresh meat.

Since the two samples were not compared side by side on a plate at the same meal, the scores assigned by tasters could only have been arrived at by comparison with their notional standards of meat quality. These are purely subjective standards developed subconsciously from past experience and used by consumers to assess any meat they eat.

The greater satisfaction of tasters with steak from tenderstretch sides is surely an indication that a more extensive adoption of the tenderstretch hanging technique would guarantee the retail butcher a larger proportion of happy customers.

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# Improved media for enumeration of fungi from foods\*

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One problem inherent in counting moulds from foods is the spreading nature of fungal growth. Fungi from the order Mucorales are especially troublesome, as their effuse, freely sporing growth habit inhibits colony formation by less vigorous moulds and may cause widespread laboratory contamination. A second inherent problem is that standard mould-counting media are of very high water activity and unsuited to the detection and enumeration of the xerophilic fungi encountered in dried foodstuffs. This paper outlines the development of two new media devised to overcome these problems.

In the past decade several new media have been developed specifically for counting moulds in foods (Overcast and Weakley 1969; Mossel *et al.* 1970; Jarvis 1973). The most effective of these is rose bengal chlortetracycline agar (RBC; Jarvis 1973), which contains 50  $\mu$ gml<sup>-1</sup> of rose bengal to limit spreading mould growth, and 10  $\mu$ gml<sup>-1</sup> of chlortetracyline to inhibit bacterial growth.

In surveying the fungi present in some Australian foods (King *et al.* 1979), we found that even RBC was inadequate for the control of the spreading growth characteristic of such genera as *Mucor*, *Rhizopus*, *Syncephalastrum* and *Rhizoctonia*. In the examination of fresh foodstuffs, particularly fruits and vegetables and products made from them, the presence of a few spores of any of these fungi makes inaccurate the enumeration of other moulds present, and their isolation difficult if not impossible (Fig. 1).

An improved modification of RBC was therefore sought. Much of the developmental work was carried out by Dr A. D. King (U.S. Department of Agriculture, Albany, California) as a guest worker at this laboratory in 1977–78. King tested 31 antifungal agents (dyes, heavy metals, phenols, commercial fungicides and

\*This paper was presented at the Second Australian Food Microbiology Conference, Sydney, July 1979. Table 1. Screening of antifungal compounds for selective inhibition of *Rhizopus stolonifer*<sup>A</sup>

	Minimum inhibitory concentration $(\mu g/ml)$ for:			
Compound	R. stolonifer	Aspergillus and Penicillium spp. <sup>B</sup>		
Benlate	50	<2.5		
Dichloran	2.5	12.5		
Difolatan	2	< 0.1		
Thiram 80	1.25	5		
Phenol	500	500		
Pentachlorophenol	2	5		
Pentachloronitro-				
benzene	<5	50		
Sodium benzoate	1000	>1000		
Sodium propionate	>1000	>1000		
Potassium sorbate	>1000	>1000		
Salicylic acid	500	500		
Gentian violet	5	10		
Malachite green	12.5	25		
Rose bengal	250	500		
Cadmium chloride	100	25		
Cupric sulfate	100	500		

<sup>A</sup>Adapted from King et al. (1979).

<sup>B</sup>Aspergillus flavus, Penicillium expansum and P. islandicum.

preservatives) against *Rhizopus stolonifer*, *Penicillium expansum*, *P. islandicum* and *Aspergillus flavus* in the search for a compound that would control the spreading growth of





Fig. 1. RBC medium overgrown by colonies of *Rhizopus stolonifer* (left); DRBC medium (right). Colonies of *R. stolonifer* are shown by arrow heads.

*R. stolonifer*, while allowing near normal growth of more important spoilage fungi such as *Aspergillus* and *Penicillium* species.

Representative results are shown in Table 1. Of the 31 compounds tested, 27 showed little or no selectivity, and two allowed *R. stolonifer* to grow well while inhibiting the three other test organisms. Of the two compounds which inhibited *Rhizopus*, pentachloronitrobenzene was considered unsuitable because of reports of carcinogenicity (Fairchild 1977). The remaining compound was dichloran (2,6dichloro-4-nitroaniline), with a minimum inhibitory concentration for *Rhizopus* five times less than for the other test fungi (2.5 and 12.5  $\mu$ gml<sup>-1</sup> respectively).

Trials with concentrations of dichloran between 0 and 5  $\mu$ gml<sup>-1</sup> showed a level of 2  $\mu$ gml<sup>-1</sup> to be adequate to inhibit the spreading growth of *R. stolonifer* without affecting its germination. However, dichloran alone proved inadequate for controlling some other species of *Rhizopus* and *Mucor*, even at increased concentrations.

Consequently, combinations of dichloran and rose bengal were tested. The combination of these two compounds which gave highest counts and most satisfactory control of mucoraceous fungi was  $2 \mu \text{gml}^{-1}$ dichloran and  $25 \mu \text{gml}^{-1}$  rose bengal. This modification of Jarvis's RBC medium was designated DRBC (King *et al.* 1979).

The efficacy of DRBC as a fungal

enumeration medium was compared with RBC and acidified potato dextrose agar (APDA), two media commonly used for this purpose (Table 2). Mould counts were higher on DRBC than on the other two media. As noted in Table 2, the three media were of widely differing pH which undoubtedly influenced the counts obtained. Comparisons were made in the manner indicated because RBC and APDA are standard formulations, while King *et al.* (1979) showed that pH 5.6 is more inhibitory than pH 7.2 to mucoraceous fungi.

To determine whether DRBC would allow adequate colony development of food spoilage fungi, plates were spot inoculated with fungi representing 14 genera, including

Table 2.	Comparison	of mould	counts	from	foodsA
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Medium <sup>B</sup>	pН	Moulds/g <sup>C</sup>	Yeasts/g <sup>D</sup>
RBC	7.2	94	50
DRBC	5.6	102	45
APDA	3.5	59	68

<sup>A</sup>Adapted from King et al. (1979).

<sup>B</sup>RBC, rose bengal chlortetracycline agar; DRBC,

Dichloran rose bengal chlortetracycline agar; APDA, acidified potato dextrose agar.

<sup>C</sup>Average of six foods (black pepper, white pepper, sage, spice powder, wheat and minced meat).

<sup>D</sup>Average of four foods (raisins, dried currants and grapes (two samples).

11 different species of *Aspergillus* and 18 species of *Penicillium*. Some representative results are shown in Table 3. Colony diameters of fungi such as the *Aspergillus* and *Penicillium* species were only slightly smaller on DRBC than on RBC and APDA. However, data in the lower section of Table 3 show how well DRBC controls the colony diameters of a number of mucoraceous fungi. These promising results were confirmed when food samples were examined.

Thus DRBC (Table 4) was chosen as the most satisfactory medium providing good selective inhibition of mucoraceous fungi, while permitting growth of all other species examined.

In common with most other media currently recommended for enumerating fungi in foods, DRBC has a high water activity (0.999  $a_w$ ). While such media are satisfactory for enumerating and isolating yeasts and moulds from fresh foods such as fruit, vegetables, dairy products, and meat, they are inadequate for sampling the fungal flora of dried and semi-dried foods like cereals, nuts, condiments, dried fruits, confectionery, and dried meat and fish products.

Media of reduced  $a_w$  have been in use for many years for the isolation of osmophilic yeasts. As early as 1929 (Ingram 1958), Lockhead and Heron used media containing large proportions of honey for isolating yeasts from honey. Later workers have used media based on glucose and sucrose for cultivation of osmophilic yeasts (Ingram 1959; Scarr 1959) but reduced  $a_w$  media for moulds in foodstuffs have traditionally been based on

Organism	RBC	DRB	C APDA
Aspergillus (11 species)	18	13	17
Penicillium (18 species)	16	12	19
Paecilomyces variotii	30	16	36
Fusarium sp.	53	18	35
Cladosporium sp.	21	12	16
Wallemia sebi	5	4	0
Rhizopus arrhizus	>70	16	>70
R. stolonifer	63	11	>70
Mucor circinelloides	>70	19	>70
M. racemosus	42	16	>70
Syncephalastrum racemosum	>70	10	>70

\*Adapted from King et al. (1979).

sodium chloride (Christensen 1946; Mislivec and Bruce 1977).

In 1946, Christensen published his formulation for Malt Salt Agar (MSA), containing 7.5% wt/wt salt and 2% malt extract. He reported this medium to be particularly effective for enumerating such xerophilic species as *Aspergillus candidus* and the *A. glaucus* group. He also reported the pH to be 5.0, but depending on the source of the malt extract, this can be as low as about pH 4.0.

Other reduced water activity media based on glucose and sucrose have been developed more recently (Pitt and Christian 1968; Udagawa and Tsuruta 1973) but MSA is still the most widely used and recommended medium for enumerating xerophilic fungi.

MSA has a number of weak points as an enumeration medium. Because of the lowered  $a_{m}$  (0.95), species of the Aspergillus glaucus group spread rapidly, overgrowing some of the more slowly developing species, and heavy sporulation can cause problems with secondary colonies, as plates are incubated for up to a week. Because of the high concentrations of malt extract (2%) and salt (7.5%), the pH is quite low and the agar tends to break down, giving a soft, granular medium that is difficult to use for spreadplating, because glass spreaders sink into the medium. Depending on the type of malt extract used, the medium can vary from translucent to moderately opaque, and some malt extracts produce a brownish precipitate that can interfere with counting.

In view of these problems, and the difficulties associated with media containing high concentrations of sugars (viscosity, crystallization), an alternative was sought. Pitt and Hocking (1977) showed that glycerol is a suitable solute for the cultivation of a

Table 4.	Dichloran rose bengal chlortetracycline agar
(DRBC)A	

· ·	
Glucose	1.0%
Peptone	0.5%
KH <sub>2</sub> PO <sub>4</sub>	0.1%
$MgSO_4 \cdot 7H_2O$	0.05%
Agar	1.5%
Rose bengal	25 μg ml <sup>-1</sup>
Dichloran	2 μg ml <sup>-1</sup>
+ Chlortetracycline	10 μg ml <sup>-1</sup>
pH 5.6	

<sup>^</sup>King et al. (1979)

Table 5. Dichloran-glycerol agar (DG18)A

18% wt/wt
1.0%
0.5%
0.1%
0.05%
1.5%
$2 \mu \text{g ml}^{-1}$
$10 \mu g  ml^{-1}$
-

AHocking and Pitt (1980).

range of xerophilic fungi. It is easy to handle in high concentrations as it does not crystallize and is less viscous than sugar solutions, and glycerol-based agars set firmly and are very clear and transparent.

However, even media formulated with glycerol do not resolve the problem of controlling the spreading growth of the *Aspergillus glaucus* group at lowered  $a_w$ . Having successfully controlled spreading growth in a high  $a_w$  medium with dichloran, it was incorporated and tested in a low  $a_w$  medium.

Using this anti-spreading property of dichloran, a new medium, DG18 (Hocking and Pitt 1980), has been developed for enumeration of moulds from low moisture foods (Table 5). DG18 contains 18% wt/wt glycerol (giving an  $a_w$  of 0.955), and 2  $\mu$ gml<sup>-1</sup> dichloran. Chlortetracycline is incorporated to inhibit those bacteria which will grow at  $a_w$  0.95 and pH 5.6, *Bacillus* species being the most troublesome.

The question may be asked: is a special low  $a_w$  medium really necessary?

The need for a special medium for enumerating the fungal flora of low moisture

Table 6. Comparison of counts obtained on DRBC and  $\mathsf{DG18A}$ 

Total mould count per gram			
DRBC	DG18		
2.7 x10 <sup>5</sup>	4.1 x 10 <sup>6</sup>		
$1.4 \ge 10^4$	$1.0 \ge 10^{7}$		
$8.2 \ge 10^4$	1.6 x 10 <sup>8</sup>		
0	8.5 x 10 <sup>4</sup>		
7.3 x 10 <sup>5</sup>	$2.7 \ge 10^{6}$		
$1.3 \ge 10^4$	4.5 x 10 <sup>7</sup>		
$2.0 \ge 10^5$	$2.3 \ge 10^{6}$		
	Total mould DRBC 2.7 x10 <sup>5</sup> 1.4 x 10 <sup>4</sup> 8.2 x 10 <sup>4</sup> 0 7.3 x 10 <sup>5</sup> 1.3 x 10 <sup>4</sup> 2.0 x 10 <sup>5</sup>		

\*DRBC, Dichloran rose bengal chlortetracycline agar; DG18, Dichloran-glycerol agar. foods becomes obvious after examination of the values in Table 6. While DRBC is an excellent medium for fungal enumeration in fresh foods, it is significantly less effective than DG18 in counting moulds from low moisture foods. In some instances, there is a thousand-fold difference in counts.

These differences are mainly due to three groups of fungi: the Aspergillus restrictus group, the Â. glaucus group and the xerophile Wallemia sebi. The two commonly occurring species in the first group are A. restrictus, which is capable of weak growth on high  $a_m$ media, and A. penicilloides, an extreme xerophile which will not grow at all under these conditions. In the A. glaucus group, species with small ascospores such as A. chevalieri will grow slowly on DRBC, but species with large ascospores such as A. ruber are generally more xerophilic and grow only weakly, if at all, on media of high  $a_w$ . Wallemia sebi is a very common xerophilic fungus found on cereals and cereal products and salted meat and fish products. However, it is not often detected as it rarely grows on high  $a_w$  media, although pinpoint colonies may appear after about 5 days' incubation. Other less common xerophiles such as Scopulariopsis halophilica, and some unidentified fungi from salt fish, also contribute to differences in counts on DG18 and DRBC.

Mould counts on DG18 compare favourably with those on MSA as shown in Table 7. Counts are significantly (P < 0.05) higher on DG18 than on MSA.

An unexpected advantage of DG18 is that members of the *Aspergillus glaucus* group form more diverse colony types on this medium than on MSA, enabling more ready recognition of the various species. This is due

Table 7. Comparison of counts obtained on DG18 and MSAA

Commodity	Total mould count per gram			
	DG18	MSA	MSA/ DG18%	
Cayenne pepper	4.9 x 10 <sup>6</sup>	4.3 x 10 <sup>6</sup>	87	
Semolina	$2.0 \ge 10^{7}$	1.5 x 10 <sup>7</sup>	75	
Fish flour	$6.2 \ge 10^3$	$5.0 \ge 10^3$	80	
Dried fish	$5.4 \ge 10^{7}$	5.1 x 10 <sup>7</sup>	94	
Dried chillies	5.6 x 10 <sup>6</sup>	6.7 x 10 <sup>6</sup>	119	
Black pepper	2.6 x 10 <sup>5</sup>	$1.9 \ge 10^{5}$	73	
Paprika	1.7 x 10 <sup>6</sup>	1.5 x 10 <sup>6</sup>	88	
Mixed spice	$1.3 \ge 10^4$	$3.3 \ge 10^3$	25	

<sup>A</sup>DG18, Dichloran-glycerol agar; MSA, malt salt agar.



to the development of characteristic coloured hyphae and yellow cleistothecia on DG18, while on MSA mostly grey-green conidia are produced.

Restriction of the colony sizes of the Aspergillus glaucus group by dichloran makes DG18 plates easier to count. In addition, species in the A. restrictus group are often overgrown on MSA; this happens less on DG18. Other Aspergillus species including A. flavus, A. niger, A. ochraceus and most Penicillium species will grow well on DG18. In commodities which have no xcrophiles, mould counts obtained with DRBC and DG18 are comparable.

DG18 is not a perfect medium: it will not support the growth of such fastidious xerophiles as *Xeromyces bisporus* and *Chrysosporium fastidium* (both important in the spoilage of prunes) but neither will MSA. Despite this shortcoming, DG18 is recommended as a replacement for MSA for counting xerophilic fungi in low moisture and intermediate moisture foods.

Although dichloran is a relatively inexpensive chemical, it can be difficult to obtain in the small quantities needed for producing these media. This laboratory will supply dichloran free of charge to Australian laboratories on application to the author. It is hoped that both DRBC and DG18 will be available commercially in the near future.

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# Microbial ecology and interactions in chilled meat\*

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Aerobically, the selective pressures that determine the mix of microflora growing on meat are temperature, water activity, pH and inhibitors such as nitrite or carbon dioxide. Competition for sources of energy does not seem to be important. However, amino acids can be degraded earlier in meat that has a low glucose content. On vacuum-packed meat, the high carbon dioxide and low oxygen tensions exert additional selective pressures resulting in a changed flora. Anaerobically, undissociated lactic acid becomes another agent restricting growth, and lactobacilli may appear to inhibit some strains by a mechanism as yet unknown.

A great variety of microorganisms — derived from the hide, or fleece, or intestinal tract can contaminate meat. However, out of this large variety, only a very restricted number are able to achieve any significant growth on chilled meat.

# Meat stored aerobically

# Temperature

Temperature is one of the prime selective factors determining which organisms will grow and how rapidly they grow. When the temperature of storage is below 7 °C, mesophiles cannot grow, but psychrotrophs can. Under aerobic, moist conditions, the fastest growing psychrotrophs on fresh meat at temperatures near 0 °C are non-pigmented pseudomonads (Table 1) (Scott 1937; Gill and Newtown 1977). Most, if not all, of the strains referred to by Scott as achromobacters would now be classified as non-pigmented pseudomonads (Brown and Weidemann 1958).

As the temperature of growth is raised from near  $0^{\circ}$ C, the rate of growth of different strains increases, but not by exactly the same ratio.

In Table 1 the growth rates, for an increase in growth temperature from 2° to 10°C, increase from 2.6- to 3.5-fold. Those strains whose growth rate increases fastest with rise in temperature will increase their

\*A paper presented at the Second Australian Food Microbiology Conference, Sydney, July 1979. representation most in the final flora at higher temperatures. In spite of this, from the data in Table 1, non-pigmented pseudomonads will still be dominant at 10°C as they are still the fastest growing. Ingram and Dainty (1971) quote two examples where, when the storage temperature was raised from  $1^{\circ}-4^{\circ}C$  to  $10^{\circ}-15^{\circ}C$ , there

Table 1. Effect of temperature on the rates of microbial growth on meat incubated aerobically

Grov	vth rate (h/	gen.)	Growth rat	e ratio
10°C	2°C	−1°C	10°/2°C	10°/-1°C
	Pseudo	monas (non	pigmented)	
2.5	7.1	10.7	2.8	4.3
2.54	7.8	11.6	3.1	4.6
2.8	7.6		2.7	
	Psei	domonas (pij	gmented)	
3.0	8.2		2.7	
3.95	13.8	22	3.5	5.6
		Acinetobac	ter	
5.2	15.6		3.0	
		Enterobac	ter	
3.5	11.1		3.2	
		M. thermosph	actum	
3.4	12.0	-	3.5	
		Candida	2	
5.9	15.2	22	2.6	3.7
		Mycotoru	la	
7.1	22.8	44	3.2	6.2

From the data of Scott 1937 and of Gill and Newton 1977.

were more *Enterobacteriaceae* at the higher temperature, although pseudomonads still predominated.

# Water activity

Water activity  $(a_w)$  is a second, selective, externally applied factor that can change the relative growth rates of microorganisms on meat and therefore the composition of the flora. The accompanying Figure, derived from the data of Scott (1936), shows the growth in pure culture of three psychrotrophs on beef muscle at  $-1^{\circ}$ C and two  $a_w$  values. Strain 7 (presumably a non-pigmented pseudomonad) grows more rapidly than either the pigmented pseudomonad (strain 1) or the yeast *Geotrichoides sp.* (Y9). As the  $a_w$  of the meat is reduced, the pigmented pseudomonad fails to grow at  $a_m$  0.98, the non-pigmented pseudomonad at 0.96, and the yeast at 0.90. However, even a moderate reduction in  $a_w$  to 0.97 will change the flora. In this example, pigmented pseudomonads will not grow, and the yeast Geotrichoides will be found in greater proportion. For instance, it can be seen from the Figure that when the bacterial count reaches  $10^8 \text{ cm}^{-2}$  at  $a_w 0.993$ , the yeast count is about  $10^5$  cm<sup>-2</sup>, but at  $a_w$ 0.97 the yeast is about  $10^6$  cm<sup>-2</sup>.

Lactobacilli are able to grow at a lower  $a_w$ (about 0.94) than pseudomonads (Wodzinski and Frazier 1961*a,b*; Lanigan 1963). A reduction in the  $a_w$  of meat will therefore inhibit the growth of pseudomonads before that of lactobacilli. Smith and Palumbo (1973) found it necessary to add 3% salt to beef during ageing in the production of



Effect of water activity on microbial growth on beef muscle stored aerobically at --1°C (from Scott 1936). a, pigmented pseudomonad strain 1; **A**, non-pigmented 'Achromobacter' (pseudomonad) strain 7; •, Geotrichoides sp. strain Y9.

Lebanon bologna to restrict pseudomonad growth and select for an increased lactic flora. Australian smallgoods have an  $a_m$  of 0.96–0.98 (Brownlie 1969). Again, such an  $a_w$ would select against pseudomonads in favour of lactics. As M. thermosphactum can also grow at an  $a_w$  down to 0.94, this organism could also be expected to grow on lightly salted smallgoods. Shay et al. (1978) sometimes found *M. thermosphactum* to be a significant part of the flora of smallgoods. However, because nitrite in the undissociated form inhibits *M. thermosphactum* much more readily than it inhibits the lactics (Brownlie 1966), it is not surprising that lactics are more common on meats containing both salt and nitrite.

# pH

The pH of fresh meat can range from about 5.4 to over 6.0. Perhaps somewhat surprisingly the range of pH of fresh meat has only a restricted role in exerting a selective pressure on the aerobic flora. For instance, Barnes and Impey (1968) found that, in heart infusion broth at 1°C, the generation times of both pigmented and nonpigmented pseudomonads were unaffected over a pH range 5.8 to 7.4. The generation time of acinetobacter (Grp C) was reduced by less than 10%. On the other hand, strains of acinetobacter Grp B (i.e. Moraxella sp., see Barnes and Melton 1971) were unable to grow at pH 5.8, and the growth rate of Altermonas putrefaciens was more than halved by a decrease in pH from 6.2 to 5.8. On chicken breast muscle (pH 5.7-5.9) acinetobacter (Grp C) was unable to grow even though it grew in heart infusion broth at pH 5.8, and though it grew on chicken leg muscle (pH 6.4–6.7). This suggests that acinetobacter is sensitive to something other than simply pH in relatively low pH meat.

Barnes and Impey also observed that pigmented pseudomonads had a longer lag time than the non-pigmented pseudomonads on chicken muscle, particularly on the breast muscle, which had a lower pH. The shorter lag time of the non-pigmented pseudomonads was the major reason that these organisms were more numerous than the pigmented pseudomonads at spoilage, even though the latter were initially present in greater numbers. From the Figure it can be seen that on beef at -1 °C and  $a_w$  0.993, the pigmented pseudomonad (strain 1) also has a longer lag time than the non-pigmented strain 7. In Scott's data (1936), the same applies to a second strain of each group.

Gill and Newton (1977) used meat juice adjusted to pH 5.5-7.0 to study the aerobic growth of a number of psychrotrophs isolated from meat. In agreement with Barnes and Impey (1968), they found no effect of pH on the growth rate of either pigmented or nonpigmented pseudomonads, and a reduction in the growth rate of acinetobacter at pH 5.7 and below. The growth rates of *Enterobacter sp.* and *M. thermosphactum* were not affected by pH over the range pH 5.5-7.0. Brownlie (1969) observed only a 30% reduction in the growth rate of M. thermosphactum, measured at 25°C in APT broth (All Purpose Medium with Tween 80), when the pH was reduced from pH 7.0 to 5.5.

# Soluble components

Ingram and Dainty (1971) stated in their review that, from the evidence, 'it is difficult to draw any conclusions about the changes in concentrations of soluble constituents to be expected in spoiling meat'. Since then, New Zealand workers have shed considerable light on the interaction between organisms growing on meat and the soluble constituents of the meat.

Using aqueous extracts of meat, Gill (1976), and Gill and Newton (1977) showed that under aerobic conditions, pseudomonads, enterobacter and *M. thermosphactum* all preferentially used glucose. When glucose was exhausted, these organisms were then able to grow at an unchanged rate on a secondary substrate. For pseudomonads, the secondary substrate was lactate and amino acids simultaneously; for enterobacter, it was glucose-6-phosphate; and, for *M. thermosphactum*, it was glutamate. Glucose appeared to exert catabolite repression, ensuring its use before that of other energy-yielding substrates.

When meat on which these organisms were grown was examined for constituents of low molecular weight, the New Zealand workers found that pseudomonads, enterobacter or M. thermosphactum each exhausted glucose at the surface of the meat. A concentration gradient of glucose extended into the meat for up to c. 15 mm. However, because the organisms were able to grow at the same rate on their secondary substrate, active growth was maintained. When pseudomonads used amino acids and lactate, there was a rise in surface pH and in the concentration of ammonia. When the growth of pseudomonads ceased, lactate and all 16 amino acids examined were still present at the surface, although six amino acids had decreased in concentration and five had increased. That is, growth was not limited by the availability of growth substrates. Similarly, when *M. thermosphactum* ceased growing, glutamate was still abundant at the surface.

For enterobacter, the secondary substrate (glucose-6-phosphate) was depleted at the surface. However, this depletion occurred only when enterobacter approached its maximum cell density. Gill and Newton (1977) thought that the limited availability of glucose-6-phosphate was unlikely to have an appreciable effect on the growth of enterobacter. However, later work by Newton and Gill (1978b) showed that glucose could not be detected in muscles whose pH was above 6.4 and often was absent in meat with a pH of 6.0. The reduced glucose concentration in high pH meat should, it seems, mean an earlier exhaustion of glucose-6-phosphate, unless there was a compensating increase in the concentration of glucose-6-phosphate with increase in meat pH. Enterobacter would then have to grow on lactate and amino acids - substrates giving a slower growth rate. Acinetobacter did not use glucose for growth on meat but was able to use both amino acids and lactate, neither of which were exhausted at the surface by growth (Gill and Newton 1977).

Pseudomonads bring about spoilage odours when they attack the amino acids (Gill 1976). As pseudomonads preferentially use glucose, meat with a low concentration of glucose would be expected to be spoilt earlier, with a lower number of pseudomonads. On high pH meat (pH 6.3) with no glucose present, Newton and Gill (1978b) showed that spoilage occurred when the pseudomonad count was only about 10<sup>6</sup> cm<sup>-2</sup>. Reducing the pH of such meat to 5.6 with L-lactate still allowed early spoilage. However, the addition of glucose to the high pH meat delayed spoilage until the pseudomonad count was above 10<sup>8</sup> cm<sup>-2</sup>. With meat of pH 5.85 in which the natural glucose content was  $112 \,\mu g \, g^{-1}$  wet weight, a pseudomonad count of over 108 cm<sup>-2</sup> was required to cause spoilage. Thus they were able to explain the observation that high pH meat spoils aerobically more readily than low pH meat even though the growth rate of

pseudomonads is little affected by the difference in pH.

Although many of the organisms growing on meat are using the same substrates to provide energy, it is unlikely that competition for these substrates could be a major factor in one part of the flora inhibiting another, or in determining the predominant flora. The substrates are present in excess. The one exception is perhaps enterobacter, particularly on high pH meat. Gill and Newton (1977) found no evidence for an effect on the aerobic growth rate when organisms were inoculated onto meat in similar numbers. However, when there were very large differences in count between different organisms, competition was observed. When pseudomonads were present at their maximum count, they reduced both the growth rate and final population of enterobacter and M. thermosphactum. When enterobacter cells were at their final population on meat they reduced the growth rate of M. thermosphactum but not of the pseudomonads. Similarly, maximum numbers of *M. thermosphactum* reduced the growth rate of enterobacter but not of the pseudomonads. Gill and Newton (1977) suggested that this result, at high cell populations, might be because of competition for oxygen - pseudomonads probably being more efficient in utilizing oxygen at low tensions.

# Vacuum-packed meat

When meat is vacuum-packed, the contaminating flora are exposed to an atmosphere containing 20–25% carbon dioxide and less than 1% oxygen. Both the high carbon dioxide and low oxygen tension depress the growth of pseudomonads (Scott 1938; Shaw and Nicol 1969; Clark and Burki 1972), and the predominant flora becomes one of facultative anaerobes — lactic acid bacteria, *M. thermosphactum* and *Enterobacteriaceae* (Ingram and Dainty 1971; Newton *et al.* 1977; Gill and Newton 1978; Grau 1978).

# Temperature

The temperature of storage of vacuumpacked meat can be expected to exert a selective influence on the composition of the developing flora. Newton and Gill (1978a) found that lactobacillus grew faster on meat anaerobically than either enterobacter or *M. thermosphactum*, over the temperature range  $2^{\circ}$  to  $15^{\circ}$ C. The growth rate advantage for lactobacillus was greater at the lower temperatures. At  $2^{\circ}$ C, *M. thermosphactum* grew *c.* 1.7 times faster than enterobacter. At  $5^{\circ}$ C, both these organisms grew at almost the same rate, while at  $10^{\circ}$ C, and above, enterobacter grew faster than *M. thermosphactum*.

Other things being equal, the final flora on vacuum-packed meat stored at c. 2°C should therefore be composed predominantly of lactic acid bacteria, with smaller numbers of *M. thermosphactum* and still fewer *Enterobacteriaceae.* At higher temperatures, 10°C and above, the relative proportions of the last two groups of organisms should be reversed. These simple expectations are not always fulfilled. Other interactions must be involved in controlling growth.

# M. thermosphactum

If we consider *M. thermosphactum*, we find that according to Pierson *et al.* (1970) and Roth and Clark (1972) this organism will not grow on vacuum-packed meat, while a number of other workers (e.g. Sutherland *et al.* 1975; Seideman *et al.* 1976; Patterson and Gibbs 1977) found that it did.

Roth and Clark (1975), in seeking an explanation of their earlier failure to observe growth of M. thermosphactum, examined the interactions between homofermentative lactobacilli and M. thermosphactum. They found that, whereas in pure culture M. thermosphactum grew to about  $3 \times 10^7$  cm<sup>-2</sup> on vacuum-packed meat, it grew to only about 10<sup>5</sup> cm<sup>-2</sup> in the presence of lactobacilli. They suggested that the lactobacilli produced an inhibitory substance (most likely not lactic acid) that restricted the growth of M. thermosphactum, and that competition for a substrate was an unlikely explanation. Aerobically there was no inhibition of M. thermosphactum by lactobacilli.

Newton and Gill (1978a) further examined competition between a strain of lactobacillus and *M. thermosphactum*, and included a consideration of the substrates used for energy production. In agreement with Roth and Clark, they observed that lactobacillus, when it reached about  $10^7 \text{ cm}^{-2}$ , inhibited anaerobic growth of *M. thermosphactum*. While glucose and arginine were the sources of energy anaerobically for the lactobacillus growing on meat, only glucose was used by *M. thermosphactum*. Glucose is relatively quickly exhausted from the surface of the meat and the organisms growing on the meat have then to depend on the glucose diffusing from underlying tissues. Under such conditions lactobacillus would have an advantage in that it can use arginine. However, M. thermosphactum should still be able to grow and compete with the lactobacillus provided its affinity for glucose is greater. Newton and Gill therefore examined the dominance pattern for these two organisms when growing under glucose limitation in continuous culture. M. thermosphactum displaced lactobacillus from the continuous culture, implying that M. thermosphactum had a greater affinity for glucose. They therefore concluded, in agreement with Roth and Clark's suggestion, that lactobacillus must be producing a substance inhibitory for *M. thermosphactum* on meat. Attempts to demonstrate the production of this inhibitory material in both synthetic medium and in aqueous extracts of meat were unsuccessful.

While the results of Roth and Clark (1975) and of Gill and Newton (1978) appear to provide an explanation for the occasional failure to observe *M. thermosphactum* growing on vacuum-packed meat, recent observations suggest that this is not the full explanation.

The first consideration is that Shaw and Nicol (1969) noted that in pure culture, i.e. without the presence of lactobacilli, M. thermosphactum did not give sustained growth in the absence of oxygen on meat, the count increasing from  $10^3$  to only  $10^5$  cm<sup>-2</sup>. However, Newton and Gill (1978a) indicated that the maximum count attained anaerobically on meat depended on the concentration of fermentable substrate and that the only low molecular weight energy substrate in meat used by M. thermosphactum is glucose. As the pH of meat increases, the concentration of glucose decreases. On high pH meat therefore the limited glucose might be expected to give only slight, i.e. not sustained, growth of *M. thermosphactum*. The limited growth found by Shaw and Nicol might then be due simply to their having used high pH meat. We (Campbell et al. 1979) therefore reexamined the ability of M. thermosphactum to grow aerobically and anaerobically on meat with pH values ranging from pH 5.4 to 6.4. Anaerobically, M. thermosphactum did not grow when the pH was 5.7 or lower. It did grow when the pH was 6.0 and higher. When meat was vacuumpacked, the amount of growth depended on both the pH of the meat and the

permeability of the packaging film to oxygen. When film of very low permeability was used, *M. thermosphactum* did not grow when the pH of the meat was 5.8 or lower. It did grow when the pH was 6.0 and above. With film of intermediate permeability to oxygen, only limited growth occurred on low pH meat. Aerobically, growth occurred over the entire pH range studied - 5.4 to 6.4.

The failure of *M. thermosphactum* to grow anaerobically on meat of pH 5.7 and lower was surprising as Brownlie (1969) had observed anaerobic growth in APT broth down to about pH 5.0. However, the pH of meat is directly related to its content of lactic acid (Newbold and Scopes 1967). We have subsequently examined the effect of pH and of the lactic acid concentrations on the growth of this organism. Aerobically, it grew down to at least pH 5.5 and in the presence of 150 mM L-lactate. Anaerobically, growth was more sensitive. At 25°C, the concentration of lactic acid found in meat of pH 5.7–5.8 was sufficient to stop growth. The growth rate was halved by about 0.5 mм undissociated lactic acid - the concentration occurring in meat of about pH 6.1. Two strains of lactic acid bacteria examined were able to grow at pH 5.4 in the presence of 150 mм lactate.

# Enterobacteriaceae

Enterobacteriaceae (principally Enterobacter *spp.*) often reach a count of over  $10^6$  cm<sup>-2</sup> and can comprise 10-40% of the total flora on vacuum-packed fresh meat stored at 0-2°C (Patterson and Sutherland 1973; Seideman et al. 1976; Beebe et al. 1976; Patterson and Gibbs 1977, 1978; Newton et al. 1977; Grau 1978). They frequently outnumber M. thermosphactum. Patterson and Gibbs (1978) found that *Enterobacteriaceae* were more common on vacuum-packed lamb at 7°C than at 1-2°C. This agrees with the data of Newton and Gill (1978a) who found that as the temperature increased the growth rate of enterobacter increased more rapidly than the growth rate of either lactobacillus or M. thermosphactum. There is a suggestion in the results of Patterson and Gibbs (1977) that *Enterobacteriaceae* grow better on high pH meat, and Enterobacter liquefaciens was shown to have a shorter generation time on vacuumpacked, high pH meat (Gill and Newton 1979).

The preferred energy substrates for enterobacter growing anaerobically on meat were glucose and glucose-6-phosphate (Newton and Gill 1978a) and perhaps serine (Gill and Newton 1979). When glucose was exhausted, other amino acids were degraded and spoilage odours were produced. On high pH meat, the reduced content of glucose allowed enterobacter to attack amino acids earlier.

Lactobacilli at  $10^7$  cm<sup>-2</sup> also inhibited the anaerobic growth of enterobacter on meat (Newton and Gill 1978*a*). Again, the continuous culture technique was used with limiting glucose to show that enterobacter had a greater affinity for glucose than did lactobacillus. Inhibition on meat by lactobacillus therefore could not be caused by competition for glucose. In the same way as they explained the competition between lactobacillus and *M. thermosphactum*, they suggested that lactobacillus produced an inhibitor also active against enterobacter.

To explain the increased occurrence of enterobacter on high pH meat, Newton and Gill (1979) suggested that both high pH and the absence of glucose in the high pH meat were necessary to allow enterobacter to compete successfully with lactobacilli. However, just as occurs for *M. thermosphactum*, it may be that on high pH meat the lower concentration of lactate enables *Enterobacteriaceae* to grow better. We are currently examining this possibility.

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# International Refrigeration Conference in New Zealand

Following the commemoration in Australia of the centenary of the first successful commercial shipment of frozen meat in 1880 (see CSIRO Food Res. Q. 40(3/4)), it is interesting to note that the analogous event in New Zealand was a shipment of frozen meat and butter on s.s. Dunedin which sailed from Port Chalmers, Otago, on 15 February 1882 and arrived in London on 14 May. The centenary of this historic shipment will be recognized by the holding of a conference of the International Institute of Refrigeration (IIR) in New Zealand, the first to be held there. The theme of the conference is Refrigeration of Perishable Products for Distant Markets, the dates are 26–29 January, 1982, and the venue is the University of Waikato, Hamilton.

The conference will be a joint meeting of the IIR Commissions: C2, Food Science and technology; D2, Refrigerated land transport; and D3, Refrigerated sea transport. The keynote speaker will be Professor Gustav Lorentzen of Norway.

Papers are invited from the following

fields: Influence of chilling and freezing on product quality; Factors influencing food quality during storage and transport; Trends in cold store construction and operation; Microprocessors for control of refrigeration systems; Developments in container technology; Port facilities for refrigerated containers; Refrigerated ship design; Energy consumption in the cold chain; Test procedures for evaluating refrigerated transport; and Refrigeration systems for fishing vessels.

Abstracts of papers (maximum 200 words) must be received by the organizers by 1 June 1981, and full manuscripts (maximum of eight pages) by 30 September 1981.

Abstracts should be sent to, and further information obtained from:

I.I.R. Conference Organizer,

Meat Industry Research Institute of New Zealand Inc., P.O. Box 617,

Hamilton, New Zealand.

# The Australian defence force food specifications

By P. W. Board

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The Australian Defence Force Food Specifications is the title which was given in October 1974 to what until then had been called the Commonwealth Food Specifications. In 1945 the Commonwealth Food Specifications Committee was established by the Controller, Defence Foodstuffs, under the National Security Act. The terms of reference were, 'to prepare and continually revise specifications for foodstuffs for the Defence Forces'.

This activity has continued to the present time and in fact the Committee had its 109th meeting at the Royal Australian Army Ordnance Centre, Bandiana in August 1980. The change in name in 1974 simply emphasized the primary purpose of the Specifications, i.e. to serve as standards for the procurement of food for the Services. It must be emphasized, however, that the ADFF Specifications are also used in purchasing food for other organizations and they are used unofficially as manuals on food processing by many food manufacturers in Australia and overseas; the Specifications are sent to about 40 countries.

# **ADFFS Committee**

The ADFFS Committee has features that are unusual. For instance, it has representatives of the manufacturers, purchasers and the regulatory authorities sitting together to define not only the safety aspects of the various food products but also quality aspects. Despite the obvious risks of conflict of interests between these parties the ADFFS Committee is an efficient, productive and happy body which has made progress in technically difficult areas, e.g. microbiological standards for foods and retort processes for canned foods, where other committees have found progress difficult.

The ADFFS Committee comprises a Chairman and Secretariat supplied by the Department of Primary Industry, Canberra. The contracting authority for the Services,

the Department of Administrative Services, is represented, as are the Army, RAN and RAAF. The manufacturers are represented by the Council of Australian Food Technology Associations (CAFTA) and there is a representative of the Australian Government Analytical Laboratories (AGAL). Other members of the Committee include representatives of the Armed Forces Food Science Establishment, Scottsdale, CSIRO Division of Food Research and of the Horticultural Crops Division, Dairy Division, Food Services Branch and Bureau of Animal Health of the Department of Primary Industry.

In recent years the Committee has met for 2 days twice a year to develop new specifications and to revise existing specifications and the methods of testing for compliance with the Specifications. Much of the work of the Committee is done outside the meetings by specialist members of the Committee and draft specifications are examined and appropriately modified, rejected or accepted at the meetings. Sometimes specialist subcommittees are formed to draft specifications, e.g. a Microbiological Subcommittee recently prepared microbiological standards for most of the foods covered by the ADFF Specifications. The drafting of less extensive specifications is usually done by one or other of the specialist officers of the Committee.

In Part A of the ADFF Specifications about 180 food items are covered, ranging from biscuits, bread and cake to dairy, fruit, meat, and vegetable products, beverages, confectionery and grocery items. Part B contains about 15 specifications for food packaging systems such as bags, drums, flexible packages, tinplate containers and aluminium tubes. Part C gives details of the methods to be used for determining whether the food products and packages comply with the appropriate Specification. Clearly, the ADFF Specifications contain a vast amount





of realistic information on fresh and processed foods and packaging and it is little wonder that they are used as guidelines by many food processors supplying the non-Service market.

# Format of specifications

With few exceptions, the Specifications have a common format. The first paragraphs have a side heading 'General' and usually say that the product 'shall be prepared, packed and stored under the supervision of the Department of Primary Industry and shall comply with the appropriate Exports Regulations, excepting any parts thereof which are inconsistent' with the particular specification.

Not all products covered by the Specifications are manufactured under the supervision of the Department of Primary Industry or included in the Exports Regulations. In these instances, the first paragraphs say, in part, that the products 'shall be prepared and packed in accordance with the best commercial practice and, where considered necessary, be under the supervision of the purchasing authority or its agent'. These are most important requirements as they emphasize the need for supervision of the product at all times during the critical stages of manufacture and storage prior to receival by the Services. Such supervision is without doubt more efficient than end-product examination in ensuring that the product is of the nature demanded.

The specifications for most processed products require that the manufacturer gives a warranty, the period of which is specified in the contract. This, of course, is essential for products intended for survival and combat rations but it also indicates that the Specifications are designed to give a stable, safe product.

The product is described in the next part of the Specifications and then the 'Detailed Requirements' are given. This section deals with the separate quality factors or aspects of processing which must be defined to give a product which complies with the requirements of the Services. Every effort is made to avoid telling the processor how to manufacture the product unless particular operations are considered to be essential to give the kind of product required. The Specifications also say that the product shall be examined by the methods set out in Part C of the ADFF Specifications and these examinations are usually done by AGAL.

Towards the end of each specification there are details of the packaging system and the branding to be used and there is a list of 'Related Documents'. These include the relevant State Food Regulations and the specifications for items, usually packaging, used in the manufacture of the product being specified. Fig. 1 is a copy of ADFFS 2-4-1 for margarine, a typical example of an ADFF specification.

#### Sampling plan

An important recent development in the ADFFS Committee has been the preparation of a sampling plan to be used by the inspecting authority when taking samples of the finished product to determine whether it complies with the specification. As sampling procedures are not defined for the State or Exports Regulations, it must be assumed that every unit or item in the batch must meet the Regulation. A similar situation existed with the ADFF Specifications and it is only now that a sampling plan has been developed to the stage where it is undergoing practical trials with products being purchased to ADFF Specifications.

Any sampling plan should be based on probability theory, of course, but often other factors, which may be subjective in character, influence the final form of the sampling procedure. For instance, the extent and efficiency of inspection of the manufacturing operations by the purchasing authority may influence the stringency of inspection of the end product. The sampling procedure must be such that it can be carried out by existing staff and must not overload available laboratory facilities. The potential hazard involved in accepting a defective unit also influences the extent of sampling; stringent sampling is appropriate if the hazard is large but it is probably wasteful if the hazard is small. Less stringent sampling is often appropriate when a manufacturer has a consistent record of producing to specification and there is no reason to suspect that his procedures have changed. Finally, there are advantages in having a sampling plan which allows retesting if the initial sample is found to be defective and which permits recovery of sound product from batches containing defective units. These points were taken into consideration in developing the sampling plan for the ADFF Specifications; the essential features of the

# AUSTRALIAN DEFENCE FORCE FOOD SPECIFICATION

**ADFFS 2-4-1** 

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# MARGARINE

# GENERAL

- (a) Margarine shall be prepared, packed and stored under the supervision of the Department of Primary Industry. The product shall comply with the Exports (Dairy Produce) Regulations and with the relevant regulations governing food production of the State of manufacture excepting any parts thereof which are inconsistent with this Specification.
  - (b) The premises in which Margarine is prepared, packed and stored shall be approved by, and be under the supervision of the Department of Primary Industry and shall comply with the Exports (Dairy Produce) Regulations.
- 2. Margarine shall be subject to warranty as provided in the contract.
- 3. Samples shall be provided and delivered as specified by the purchasing authority.

## DEFINITION

4. Margarine shall be the solid, or semi-solid, emulsion, of the types specified below, prepared from fats, oils, sodium chloride and water; and with or without the addition of non-fat milk solids, approved colourings, flavourings, emulsifiers, anti-oxidants and vitamin concentrates.

#### TYPES

- 5. Margarine shall be of the following types, as specified in the contract:
  - (a) TYPE I-MARGARINE, PASTRY. Shall be a mixture of refined and deodorised (and/or hydrogenated) vegetable fats and oils, prepared with suitable texture for the manufacture of pastry.
  - (b) TYPE II-MARGARINE, CAKE. As in (a) above, but prepared with suitable texture for the manufacture of cakes.
  - (c) TYPE III-MARGARINE, TABLE. Shall be a mixture of refined and deodorised (and/or hydrogenated) vegetable fats and oils and shall have the sensory qualities expected in a table margarine.
  - (d) TYPE IV-MARGARINE, TABLE (POLYUNSATURATED). As in (c) above, but in which the total fatty acids present contain not less than 40% of cismethylene interrupted polyunsaturated fatty acids and not more than 20% saturated fatty acids.
  - (e) TYPE V-MARGARINE, TABLE, HIGH MELTING. Shall be a mixture of refined and deodorised (and/or hydrogenated) animal and/or vegetable fats and oils.

# DETAILED REQUIREMENTS

6. Margarine shall comply with the following:

- (a) Fat:
  - (i) Not less than 80%;
  - (ii) Shall have a Kirschner value not greater than the figure obtained by using the formula: Polenske value of fat + 0.5

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- (b) Moisture-not more than 16%;
- (c) Sodium Chloride-not more than 4%;
- (d) Free fatty acids-not more than 0.15%, expressed as oleic acid;
- (e) Coliforms-5 samples shall be examined; coliforms shall be absent in 0.1 g in each sample.
- 7. Margarine, Types I, II, III and IV, shall also comply with the following:
  - (a) A melting point between 35°C and 37°C.
  - (b) Vitamin A-Not less than 8 g/g.

Fig. 1. ADFFS 2-4-1, 'MARGARINE', is reproduced here to typify the specifications to be found in 'Australian Defence Force Food Specification'.

#### ADFFS 2-4-1

- (c) Vitamin D-not less than 0.05 g/g.
- 8. Margarine, Type V, shall also comply with the following:
  - (a) Melting point-not less than 37.5°C and not more than 40°C.
  - (b) Vitamin A-not less than 8 g/g.
  - (c) Vitamin D-not less than 0.05 ug/g.

9. All margarine types shall be free from impurities and contamination. They shall be sweet, clean and free from rancidity or foreign odour, and shall have a pleasing flavour and uniform finish.

10. Margarine Types III, IV and V shall score not less than 92 points when graded in accordance with the Exports (Dairy Produce) Regulations.

11. Analyses and Examinations shall be made in accordance with the Standard Methods set out in Part C of the Australian Defence Force Food Specifications.

# PACKAGING

12. Margarine shall be packed as specified in the contract.

13. Unless otherwise specified in the contract, tinplate containers and collapsible aluminium tubes shall comply with the detailed requirements of the relevant Australian Defence Force Food Specification(s) current at the date of placing the order(s).

# LACQUERING

14. Unless otherwise specified in the contract, all cans and collapsible tubes be lacquered as follows:

- (a) Internally, with an approved stoving lacquer.
- (b) Externally, with a lacquer conforming to the relevant Australian Defence Food Specification(s) current at the date of placing the order(s).

# BRANDING

15. Unless otherwise specified in the contract, all containers shall be branded to comply with the relevant Australian Defence Force Food Specification(s) current at the date of placing the order(s). However, the following conditions shall always apply:

- (a) The description of contents shall always include the Type: and,
- (b) Margarine, Type V shall always bear the words:

'FOR HOT CLIMATES' as part of the description of contents.

# **RELATED DOCUMENTS**

16. Any reference in this Specification to items or documents listed in the following schedule should relate to the latest edition of these documents:

Items	Documents
Branding of Foodstuffs for the Australian Army	ADFFS 15-2-1
Containers, Fibreboard, Laminated, Solid, for Foodstuffs	ADFFS 15-4-1
Lacquer, External	ADFFS 15-6-1
Tinplate for Cans for Food Products	ADFFS 15-7-3
Cans, Tinplate, Cylindrical, for Hermetic Sealing, for Foodstuffs	ADFFS 15-7-4
Collapsible Aluminium Tubes	ADFFS 15-9-1
Methods of Analysis and Examination	ADFFS Part C

Relevant State Regulations Exports Regulations

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Fig. 1. continued.



Fig. 2. Flow diagram for the sampling plan for the ADFF Specifications.

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plan are shown in Fig. 2.

Stage 1 of the sampling plan would often be done by the field staff of the inspecting or purchasing authority and it is designed to determine whether the product is essentially as ordered and whether it is free of obvious defects. Stage 2 sampling is done when the product is to be examined by AGAL or another approved laboratory. Again, the samples are taken by the field staff and sent to the laboratory under conditions which ensure that they arrive in their as-sampled state. The batch may be accepted after either Stage 1 or Stage 2 sampling depending on the nature of the product and perhaps on the past performance of the manufacturer.

Should the samples fail the Stage 1 or Stage 2 examinations the batch may be subjected to Stage 3 sampling which involves examination and analysis of larger numbers of units than the earlier Stages. Again, the batch may be accepted or rejected on the basis of the results of the Stage 3 examination. However, if the batch is rejected it is possible, with the approval of the purchasing authority, for the manufacturer to carry out a recovery operation and

Relationship of sample size to the probability of accepting a batch of product containing units having defective attributes

Defective units in batch (%)	Probability of acceptance for sample sizes of			
	200	20	10	5
10	→0.0	0.12	0.35	0.59
5	->0.0	0.36	0.60	0.77
1	0.13	0.82	0.90	0.95
0.5	0.37	0.90	0.95	0.98
0.1	0.82	0.98	→1.0	→1.0
0.01	0.98	→1.0	→1.0	→1.0
0.001	→1.0	→1.0	⇒1.0	⇒1.0

resubmit the batch to the purchasing authority for a new round of sampling and assessment.

There appears to be a popular misconception that a sampling plan based on statistical or probability theory always gives, in some magical way, a true picture of the properties of the consignment under test. It must be emphasized that a statistically based sampling plan does no more than allow the samples to be taken in the most efficient way in terms of obtaining an accurate measurement of the quality of the batch, and it allows the efficiency of the sampling to be calculated. The data in the Table are instructive in that they show the risk a purchaser takes in accepting batches having varying levels of faulty and randomly distributed units under different levels of sampling; in all instances it is assumed that the number of units in the consignment is large. Perhaps the most important conclusion to be drawn from the data in this Table is that any practical level of sampling gives poor protection against the purchasing authority accepting a batch containing faulty units. This fact emphasizes the importance of having effective and continuous inspection of the product during manufacture.

#### Conclusion

The ADFF Specifications have proved their usefulness over many years in defining the safety and quality of foods supplied to the Australian Defence Forces. In addition, the Specifications are widely used as sources of detailed information on food processing and packaging both in Australia and overseas.

The composition of the ADFFS Committee and its methods of operation allow rapid responses to be made to new demands by the Defence Forces and to new developments in food processing and packaging technology.

# News from the Division

# New appointment

On 23 December 1980, Dr J. H. B. Christian, Chief, CSIRO Division of Food Research, announced that the CSIRO Executive had appointed Mr L. L. Muller as Assistant Chief of the Division and Officer-in-Charge of the Dairy Research Laboratory.

Mr Muller joined the, then, Division of Dairy Research, CSIRO, in 1958 after previous experience in the Dairy Research Laboratory of the, then, Queensland Department of Agriculture and Stock and the Downs Cooperative Dairy Association, Toowoomba, Queensland.

Since joining CSIRO, Mr Muller's research activities have been mainly in the development of processes for the manufacture of dairy products. He and his colleagues developed techniques for the



Mr L. L. Muller

manufacture of edible grade hydrochloric acid casein which have been widely used in Australia and overseas, and for the manufacture of co-precipitates of casein and whey proteins. When the, then, Australian Dairy Produce Board entered into joint ventures in South-East Asia to manufacture recombined sweetened condensed milk, Mr Muller led the research team which developed the process, worked with Australian man ifacturers to establish processing conditions to make the specialized milk powders required, and assisted in the transfer of the technology to the commercial plants.

In recent years Mr Muller and his team turned their attention to problems of whey utilization, with a particular interest in applications of membrane technology and in the functional properties of whey protein concentrates.

Mr Muller has been very active in the Australian Society of Dairy Technology, having been President of both the Queensland and Victorian Divisions, Federal Secretary, 1951–1967, and Federal President 1969/70; he is now a Life Member.

His other notable activities, which provide liaison with industry and participation in industry organizations, include chairmanship of the Dairying Standards Board, Standards Association of Australia, and of the Working Party of the Australian Dairy Products Standards Organization. He represents CSIRO on the Gilbert Chandler Institute of Dairy Technology Advisory Committee, the Dairying Research Committee, and the Australian National Committee of the International Dairy Federation.

Mr Muller's appointment is timely, as several aspects of the Dairy Research Laboratory's program are moving towards the stage of commercial application. His wide experience and knowledge of the dairy industry in Australia and overseas, and the breadth of his understanding in dairy science will assist him to provide the leadership needed to bridge the gap successfully between research and development.



Dr. A. Sharp speaking at the symposium held at FRL on the occasion of the opening of a new Container Test Facility.

# New test facility for containers

A symposium organized by the Container Disinfestation Working Party of the Entomology Committee of the Standing Committee on Agriculture was held at FRL in October 1980 to demonstrate to the shipping industry the technique of incontainer disinfestation with carbon dioxide generated from dry ice.

The symposium was made the occasion for the opening of a new Container Test Facility of the Division of Food Research. The facility provides a controlled environment suitable not only for the thermal testing of containers, but also for reproducing the temperatures experienced during land and sea transport, i.e. the range  $0^{\circ}-50^{\circ}$ C. The facility was financed partly from a contribution from the Australian Chamber of Shipping and is available also for use by industry.

Speakers at the symposium included Dr Jonathan Banks of the CSIRO Division of Entomology and Dr Alister Sharp of FRL's Applied Food Science Group which has developed the technique of in-container disinfestation. Dr Banks described how insect pests present in grain and other durable foodstuffs can be killed by exposure to a high level of carbon dioxide maintained for 10 days. This requires a relatively gas-tight container, but a high level of gas-tightness is also necessary for effective in-container fumigation using conventional fumigants such as methyl bromide and phosphine. Dr Sharp explained the use of dry ice in an insulating box to provide a continuing addition of carbon dioxide gas to replace gas lost by air leakage. The technique will permit the use of containers having a somewhat lower level of gas tightness. Some containers with plywood floors in current use are sufficiently gas-tight to be used without modification. Thus it is possible to achieve an adequate level of gas-tightness in normal commercial practice, and shipping companies would find it advisable to consider specifying gas-tightness when ordering new containers.

The test procedure used to measure the gas-tightness of a container was demonstrated at the symposium. Air from a compressor or gas cylinder is forced into the container to produce a desired over-pressure which is then allowed to decay. The time taken for the pressure to fall from one value, say 200 Pa (0.8" W.G.) to half that value is the measure of gas-tightness. The test can be performed with relatively simple equipment, but may be further simplified by the use of the automatic pressure-decay timer, 'Contestor', developed by CSIRO.

The symposium was attended by approximately 50 people including grain exporters and representatives of the shipping industry. The talk and demonstration were repeated in the evening for the benefit of delegates attending the plenary meeting of ISO TC 104, the ISO Committee concerned with freight containers. Representatives from China, France, Germany, Japan, South Africa, Sweden, the United Kingdom and the United States were present.

#### Experimental slaughter facility at MRL

On Thursday 24 July a new experimental slaughtering facility at the Meat Research Laboratory, Cannon Hill, Queensland, was opened by the Minister for Science and the Environment, the Hon. David Thomson M.C., M.P.

Following the opening of the new facility a number of demonstrations of the current research programs at MRL were inspected by a party of invited guests from the meat and allied industries, the CSIRO Executive, and the CSIRO Advisory Council, including the Chairman Sir Victor Burley and the Chairman of the Australian Science and Technology Council Sir Geoffrey Badger.

The new facility consists of animal-holding yards, a slaughtering and dressing area, a

follow-on processing area, besides auxiliary rooms including chillers and freezers. The new facility was erected at a cost of \$800 000 contributed by CSIRO, the Australian Meat Research Committee, and a levy on the meat-processing industry. It will now be possible for MRL to carry out meatprocessing research under conditions very similar to a commercial abattoir.

# Dairy industry research

Two developments have increased the Dairy Research Laboratory's close relationships with industry. The Australian Dairy Corporation has contracted with the Australian Industrial Research and Development Incentives Board to organize a project aimed at increasing productivity in the cheese industry. DRL has subcontracted to undertake research on increasing the yield during manufacture of Cheddar cheese by using ultrafiltration in a process designed to increase retention of whey proteins in the cheese. Three new staff have been appointed for the term of the project.

A further appointee has commenced work on a project studying the problems of age gelation in UHT milk. Six dairy companies interested in this problem have joined forces to fund the work on a contract basis.

Both of these activities involve regular reports of progress to technical committees established for the purpose. These meetings increase the avenues for liaison with the industry.

#### Fuel from food wastes

Two workshop days were held at the Letona Cooperative Cannery, Leeton, N.S.W., in November 1980, to demonstrate the anaerobic digestion process for converting food processing wastes into methane gas. These were attended by about 60 representatives from food-processing companies, consulting engineers and the N.S.W. State Pollution Control Commission. At these workshops, Dr Alan Lane described the operation of the 25-m<sup>3</sup> pilot-scale digester which has now operated continuously and successfully for two years.

## Awards

Dr J. R. Vickery, former Chief of the Division, received a double honour at the Joint AIFST/NZIFST Convention held in Sydney in June 1980. His services to food science were recognized by the granting to



him of Honorary Fellowships by both the AIFST and the IFST of the U.K. The former was presented to Dr Vickery by the AIFST President, Mr R. E. Leverington, the latter by Dr George Elton, Chief Scientific Adviser (Food) to Britain's Ministry of Agriculture, Fisheries & Food.

# Committees

The Chief of the Division, Dr J. H. B. Christian, is now Chairman of the International Commission for Microbiological Specifications of Foods (ICMSF). He has also accepted a second 5-year term of the WHO Expert Advisory Panel on Microbiological Aspects of Food Hygiene.

# Visiting workers

Visiting workers in the Division of Food Research during 1980 were Dr Yoshinori Ueda, from Osaka Prefecture University, Japan, (FRL, 10 months); Dr A. J. Møller from the Royal Veterinary and Agricultural University, Copenhagen, Denmark (MRL, 12 months); Dr T. Nakayama, from Mie University, Japan (MRL, 12 months); Dr Lim Chin Lam, from the University of Science, Penang, Malaysia (FRL, 3 months); and Mrs Sing Ching Chen Tongdee from the Thailand Institute of Scientific and Technological Research, Bangkok (FRL, 6 months).

# **FICA** meeting

On Wednesday 15 December 1980 at the invitation of the Chief of the Division of Food Research, the Food Industry Council of Australia (FICA) held a meeting at the Food Research Laboratory. After the meeting the members of FICA were entertained at lunch and were then conducted on a tour of the Laboratory.



Some members of the Food Industry Council of Australia during a visit to the Food Research Laboratory; from right: Dr J. H. B. Christian, Chief of the Division of Food Research, Mr C. W. Love, Deputy Chairman and Mr J. R. Urquhart, Chairman of FICA.

