



Minimum Growth Temperatures of Foodborne Pathogens and Recommended Chiller Temperatures

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Scientific Interpretative Summary

This SIS is prepared by MPI risk assessors to provide context to the following report for MPI risk managers and external readers.

Standardisation of parameters for pathogen control in food: Minimum growth temperatures of foodborne pathogens and recommended chiller temperatures

ESR FW1104

This report reviews minimum growth data for the foodborne pathogens *Campylobacter jejuni*, *Campylobacter coli*, *Staphylococcus aureus*, *Salmonella*, *STECs*, *Clostridium perfringens*, *Listeria monocytogenes* and *Yersinia enterocolitica*. The report notes that there are relatively few studies that have been undertaken to specifically identify the lowest growth temperature for a pathogen, so data needs to be abstracted from other growth studies. Within this limitation, minimum growth temperatures have been arrived at. These data have allowed the limits cited by ICMSF in 1996 to be refined.

The implications of the minimum growth temperatures on chilled food storage and recommendations by competent authorities are discussed. This report provides a basis for reviewing the relevancy of these recommendations. This report would also be useful for industry to ensure storage temperatures for potentially contaminated foods are adequate to prevent pathogen growth and toxin production where this potential exist.

Bacillus cereus has not been evaluated in this report. Data for this bacterium, which has cold-tolerant strains, should also be assessed. This information would contribute to assisting industry arrive at appropriate 'use-by' date marking should cold-tolerant strains of *B.cereus* be of concern for a ready-to-eat food.

MINIMUM GROWTH TEMPERATURES OF FOODBORNE PATHOGENS AND RECOMMENDED CHILLER TEMPERATURES

Prepared for MAF Food Safety Authority under project MFS/07/07 – Standardisation
of parameters for pathogen control in foods, as part of overall contract for scientific
services

by

Dr. J. A. Hudson

January 2011

Client Report
FW1104

MINIMUM GROWTH TEMPERATURES OF FOODBORNE PATHOGENS AND RECOMMENDED CHILLER TEMPERATURES

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1 SUMMARY

This report is one of a series of four commissioned by MAF Food Safety to address an identified lack of conformity with respect to temperature advice given by regulators.

This report presents a review of the scientific literature to determine the minimum growth temperatures of foodborne pathogens for which temperature is used as a control. Temperatures recommended for chilled holding of foods are discussed in light of the findings of this review to determine if they are appropriate. The results are also compared with a similar review undertaken over 45 years ago (Michener and Elliott 1964).

Growth data for *Listeria monocytogenes*, *Yersinia enterocolitica*, *Salmonella*, shiga toxin producing *Escherichia coli* (STEC), *Staphylococcus aureus*, *Campylobacter* and *Clostridium perfringens* were extracted and analysed. A summary of the minimum growth temperatures for the pathogens considered on food is shown in Table 1.

Table 1. Minimum Recorded Growth Temperatures of Pathogens on Foods

Pathogen	Minimum growth temperature (°C)
<i>Campylobacter jejuni</i>	25 ¹
<i>Staphylococcus aureus</i>	6.7 ¹
<i>Salmonella</i>	6.7
STEC	6
<i>Clostridium perfringens</i>	12.8
<i>Listeria monocytogenes</i>	-1.5
<i>Yersinia enterocolitica</i>	-1.5

¹ A caution regarding these data is given under Tables 3 and 4.

It was concluded that, of the species considered, *Listeria monocytogenes* and *Yersinia enterocolitica* are those which can grow at the lowest temperatures. Both species have been recorded as growing at 0°C or less in several peer-reviewed papers. It is clear, therefore, that no storage temperature other than one which would freeze food is sufficiently low to prevent the growth of these two species, although the rate of growth is slowed at low temperatures.

Salmonella, STEC and *S. aureus* have similar low temperature growth characteristics and the growth of these organisms would be prevented by sufficiently low temperatures. In the case of *S. aureus* the temperature limit for growth is not the same as for toxin production. With some exceptions in the literature, the minimum growth temperature for these three organisms is approximately 6-7°C, but toxin production by *S. aureus* has not been shown below 10°C.

The growth minima for the remaining organisms considered, *Campylobacter* and *Clostridium perfringens*, are not of relevance as the temperatures are high enough that growth would only occur under considerable temperature abuse.

Chilled food storage temperatures recommended by competent authorities vary quite widely; from 4°C (Canada) to “below 8°C” (UK). At 4°C only *L. monocytogenes* and *Y. enterocolitica* could grow, but at 7.9°C (which is “below 8°C”) all of the pathogens other than *Campylobacter* and *Clostridium perfringens* could grow. From the data there may be

little advantage in using a 4°C limit in preference to one of 5°C as predominantly the same range of pathogens or toxin production is controlled, although the growth of *L. monocytogenes* and *Y. enterocolitica* will be slower at the lower temperature. The use of 4°C, as opposed to 5°C, would allow a greater safety margin and prevent the growth of *S. aureus* which was reported to grow at 5°C in one paper. However, this would have to be weighed against greater energy requirements.

The reports of some pathogens growing beneath their accepted temperature minimum is of interest, but may not necessarily have practical implications. Even if growth were to occur in some circumstances at temperatures a degree or so lower than expected the lag phase would be long and the rate of subsequent growth slow. The effects would therefore only be relevant for products stored over extended periods. If the risk associated with such growth needs to be assessed then more data would be required to reveal the factors (strain, extrinsic conditions etc.) associated with it.

2 INTRODUCTION

MAF Food Safety has identified that there is a lack of conformity with respect to temperature advice given by regulators and food industry organisations to the consumer and food industry. This applies across thermal treatment times and temperatures, chilled storage temperatures and hot holding temperatures. This report is one of four; the other three being a discussion document considering the factors affecting thermal death of pathogens (“Background Document on Factors Influencing the Heat Inactivation of Bacteria in Foods”), a consideration of hot holding temperatures (“Maximum Growth Temperatures of Foodborne Pathogens and Recommended Hot Holding Temperatures”), and an analysis of time/temperature combinations for the treatment of various meat and seafood products (“Analysis of D- and z-values of Selected Foodborne Pathogens in Meat and Seafood”).

This report presents a review of the scientific literature along with analysis to establish the lower growth temperatures of foodborne pathogens of relevance to food safety in foods stored at chiller temperatures. Temperatures recommended for chilled storage of foods are discussed in light of the findings of this review to determine whether they are appropriate for control of the hazards considered.

Comparisons are drawn to a similar project establishing minimum growth temperatures published almost 50 years ago (Michener and Elliott, 1964), and some preliminary recommendations are also made.

3 METHODS

The pathogens considered were;

- *Campylobacter jejuni* and *coli*
- *Staphylococcus aureus*
- *Salmonella*
- *Escherichia coli* O157 and others (The five European Union specified Group A strains)
- *Clostridium perfringens*
- *Listeria monocytogenes*
- *Yersinia enterocolitica*

A raft of chiller temperatures are described in the various pieces of NZ and international legislation and guidance. This report identifies the growth minima for pathogens of concern in refrigerated foods and identifies temperatures that can effectively control these hazards.

Infotrieve (<http://www4.infotrieve.com/search/databases/newsearch.asp>) was used to source publications which could be used to identify variability or consensus in the minimum growth temperatures reported. The data used were from papers which described growth in foods held at temperatures below the optimum for the organism. Temperatures at which growth was and was not recorded were used where possible, but in several papers only one of these values was given.

Data from papers describing attempts to measure growth at considerably higher and lower temperatures were excluded from assessment of the minimum growth temperature as they do not assist in defining the growth boundary. Also excluded are data for foods or media in which the pathogens cannot grow.

For the purposes of this report, growth is defined as a sustained period of increasing cell concentration shown in the data presented, irrespective of the amount of growth. A few exceptions have been noted where growth appeared to have commenced but the experiment was ended early so removing the possibility of a sustained period of growth.

4 RESULTS

4.1 Summary of data

The minima extracted from the ICMSF (1996) are given in Table 2 and provide initial estimates for these parameters. These can be taken as the “conventional wisdom” However, this book is 14 years old and so additional data are now available.

Table 2. Growth minima of the specified pathogens according to ICMSF (1996)

Pathogen	Minimum growth temperature (°C)
<i>Campylobacter jejuni</i>	32
<i>Staphylococcus aureus</i>	7
<i>Salmonella</i>	5.2 ¹
<i>Escherichia coli</i>	7-8
<i>Clostridium perfringens</i>	12
<i>Listeria monocytogenes</i>	-0.4
<i>Yersinia enterocolitica</i>	-1.3

¹ Most serotypes fail to grow at <7°C

The following data give growth temperatures close to the minimum growth temperature of the organism; there are many other papers describing experiments at higher and lower temperatures but these do not add to assessment of the minimum growth temperature. Excluded are data for foods in which the pathogens cannot grow.

Factors affecting the boundaries at which growth ceases to occur include isolate to isolate variation within the same species, the quality and quantity of the competing microbiota, and the physicochemical parameters of the food. It might be possible, therefore, to have different recommended refrigeration temperatures for different kinds of foods, but this is not the approach that has generally been taken.

There are some considerations that need to be taken into account when considering the data in the following tables.

Firstly, the papers cited generally only use a few temperatures and so this can lead to quite large gaps between temperatures at which growth did or did not occur, for example, data for *Campylobacter* showing no growth at 25°C and growth at 37°C. These are the temperatures at which measurements were made and it is not possible to

predict from these data what might have happened at particular temperatures between 25°C and 37°C.

Secondly, incubation periods differ between studies. Therefore a given isolate may be reported to grow and not to grow on the same food if the incubation period was longer in the first instance than the second.

4.2 Summary of data for each pathogen

Note that the data given below are for reported growth in food, and so the temperature ranges shown may differ from those in Table 1 which consider all data (i.e. in bacteriological broth too).

4.2.1 *Campylobacter*

The information in Table 3 reflects the meagre data available for *Campylobacter*. For this organism it is clear that growth will not occur at temperatures <25°C. The data are consistent and it can be concluded that the actual minimum growth temperature lies between 25 and 37°C. This observation makes the behaviour of *Campylobacter* under refrigeration as largely irrelevant with respect to recommendations concerning chiller temperatures. In fact, with *Campylobacter*, colder temperatures are likely to promote the survival of the organism (Moore and Madden 2001).

Table 3. Data defining the minimum temperature of growth for *Campylobacter* spp.

Food	Temperature not allowing growth (°C)	Temperature allowing growth (°C)	Incubation period (d)	Reference
Chicken breast skins	4	25	7	Lee <i>et al.</i> 1998 ¹
Irradiated chicken skin	25	37	2	Solow <i>et al.</i> 2003
Irradiated pork skin	25	37	2	
Sterile chicken mince	25	37	8	Chynoweth <i>et al.</i> 1998
Chicken nuggets	22	37	8	
Sterile minced chicken	23	37	18	Blankenship and Craven 1982
Sterile high pH meat	ND	37	4	Gill and Harris 1982
Sterile normal	25	ND	40	

pH meat				
Chicken skin	25	ND	2	Chantarapanont <i>et al.</i> 2003

¹ The data in this paper need to be treated with caution. The paper itself claims growth at 4°C whereas inspection of the graph presented show significant oscillations in count of over 2 log₁₀ /cm² from one sampling point to another representing, in all likelihood, experimental error rather than growth. Therefore this datapoint is not included in the table above. The data representing growth at 25°C are more convincing, but no error bars are shown on the graphs. Given the weight of the other literature this apparent rise in numbers may have more to do with recovery of injured cells or the breaking up of filamentous cells rather than real growth.

4.2.2 *Staphylococcus aureus*

The data in Table 4 show that the minimum growth temperature recorded was at 5°C on bacon (Farrell and Upton 1978). Growth was recorded at a minimum temperature of 6.7°C in chicken à la king, although there are some caveats on these data, see the footnote to the table. Growth at 7.8°C did occur in that food. However, in other instances, the organism failed to grow at temperatures higher than these, presumably because of physico-chemical properties of the food. It should be noted that these data are for growth and not toxin production.

There is little information on the production of toxin (where available data for toxin production are shown in Table 2), but the data available suggest that a temperature of more than 10°C is required for this to occur. This is not an absolute as one paper reports the absence of toxin when incubation was at 18°C (Yang *et al.* 2001). It is possible for staphylococci to grow to high concentrations in the absence of toxin formation. For example, toxigenic *S. aureus* reached 4.2 x 10⁸ cfu/g in defrosting pies yet no toxin was detected (Scheusner and Harmon 1973). The factors that determine toxin formation are complex and beyond the scope of this review. It has been assumed for the purposes of this review that, if growth is possible, then a food safety risk exists but this also needs to be in the context of the minimum temperature for toxin production.

Table 4. Data defining the minimum temperature of growth for *Staphylococcus aureus*

Food	Temperature not allowing growth (°C)	Temperature allowing growth (°C)	Incubation period (d)	Reference
Bacon	ND	5	35	Farrell and Upton 1978
Chicken à la king	5.5	6.7 ¹	5	Angelotti <i>et al.</i> 1961
Custard	6.7	7.8	5	
Ham salad	10	35	5	
Bologna	5	8	28	Nielsen and Zeuthen 1984
Ham	ND	10 [*]	up to 112	Genigeorgis <i>et al.</i> 1969
Cooked minced beef	ND	10 [*]	NS	Tatini 1973

Ham	ND	10 [*]	NS	
Bologna	ND	10 [*]	NS	
Cooked ham	6.5	10	33	Castillejo-Rodríguez <i>et al.</i> 2002
Cooked turkey	6.5	10	33	
Cooked chicken	10	13.5	33	
Minced beef	6	12	15	Kennedy <i>et al.</i> 2000
Roast beef	4.4	12.8	35	Hintlian and Hotchkiss 1987
Povi Masima	5 [#]	20 [*]	98	Wong <i>et al.</i> , 2004
Bacon	5	15	42	Dempster and Kelly 1973
Minced raw turkey	10	15	5	Yang <i>et al.</i> 1988
Minced beef	12.5	ND	14	Goepfert and Kim 1975
Minced pork	12.8	15.6	<2	Ingham <i>et al.</i> 2007
Minced beef	15.6	18.4	<2	
Minced turkey	12.8	15.6	<2	
Steamed egg	5	18 [#]	1.5	Yang <i>et al.</i> 2001
Scrambled egg	5	18 [#]	1.5	
Vanilla pudding	ND	19 [*]	2-4	Scheusner and Harmon 1973

* Toxin detected [#] No toxin detected NS Not stated ND Not done

¹ The graph indicates that the lag time may have been just resolved by the end of the experiment, although the time point representing growth could have been a result of variability in counting. Growth at 7.8°C did occur.

4.2.3 *Salmonella*

One paper reported growth of *Salmonella* at a temperature of 6.7°C in chicken à la king (Table 5). In the same study growth did not occur in ready-to-eat custard and ham salad even when the temperature was 10°C, but this may be due to other physico-chemical properties of the foods. Also, the incubation period was only 5 days in this study. At a temperature approximately 1°C higher (i.e. at 8°C) there are several reports of growth on a variety of foods, and many papers have reported growth at 10°C. The data are generally consistent with the accepted norm of a 7°C minimum growth temperature for most serotypes of this organism.

The data presented are for a range of different *Salmonella* serovars.

Table 5. Data defining the minimum temperature of growth for *Salmonella*

Food	Temperature not allowing growth (°C)	Temperature allowing growth (°C)	Incubation period (d)	Reference
Skim milk	ND	8 ¹	4	Mattick <i>et al.</i>

				2003
Pasteurised milk	ND	12	120	Kasrazadeh and
Hispanic soft cheese	6	8	70	Genigeorgis 1994
Chicken à la king	5.5	6.7	5	Angelotti <i>et al.</i> 1961
Custard	10	35	5	
Ham salad	10	35	5	
High pH beef	ND	8	6	Gill and DeLacy 1991
Silverside	ND	8	NS	Shaw and Nicol 1969
Blended meat	8.2	10	NS	Smith 1985
Beef	7-8	10	NS	Mackey <i>et al.</i> 1980
Fresh chicken	3	10	12	Nychas and
Fresh fish	3	10	17	Tassou 1996
Pork chops	4	10	35	Michaelsen <i>et al.</i> 2006
Reconstituted skim milk	5	10	5	Julseth and Deibel 1964
Minced pork	4	10	16	Alford and Palumbo 1969
Beef steaks	ND	10	9	Luiten <i>et al.</i> 1982
Minced beef	ND	10	7	Nissen <i>et al.</i> 2000
Minced pork	ND	10	<2	Ingham <i>et al.</i> 2007
Minced beef	ND	10	<2	
Minced turkey	ND	10	<2	
Minced beef	ND	10	>4	Mackey and Kerridge 1988
Roast beef	4.4	12.8	35	Hintlian and Hotchkiss 1987
Bologna	5	12	28	Nielsen and Zeuthen 1984
Minced beef	7	12.5	7	Goepfert and Kim 1975
Bacon	5	16	32	Farrell and Upton 1978
Steamed egg	5	18	1.5	Yang <i>et al.</i> 2001
Scrambled egg	5	18	1.5	
Brie	8	20	35	Little and Knøchel 1994
Sweet cream whipped salted butter	4.4	21	21 ²	Holliday <i>et al.</i> 2003
Crabmeat	11	22	28	Matches and Liston 1968

English sole	6	8	14
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¹ as determined by filament formation, a viable count was not performed. NS Not stated.

² Some growth followed by death

4.2.4 Shiga Toxigenic *Escherichia coli* (STEC)

Of note from the data in Table 6 is one paper reporting growth of *E. coli* O157:H7 in sterile minced beef at 6°C as this is below the accepted norm for this organism (7-8°C as listed by the ICMSF, 1996). Growth was recorded in other foods at 6.5°C (milk) and 7°C (minced beef). In contrast another study reported the absence of growth on minced beef at 15°C, although the experiment was only continued for 4 days. There are many reports of growth in a variety of foods at 8-10°C.

Salmonella, STEC and *S. aureus* therefore have similar low temperature growth characteristics and the growth of these organisms would be prevented by sufficiently low temperatures.

The data in the table are predominantly for *Escherichia coli* O157:H7 but some data for other serotypes are included in two of the papers (Kauppi *et al.* 1996; Palumbo *et al.* 1995).

Table 6. Data defining the minimum temperature of growth for STEC

Food	Temperature not allowing growth (°C)	Temperature allowing growth (°C)	Incubation period (d)	Reference
Minced beef (sterile)	5	6	23	Tamplin <i>et al.</i> 2005
Milk	5.5	6.5	42	Kauppi <i>et al.</i> 1996
Minced beef	4 (?)	7	14	Barkocy-Gallagher <i>et al.</i> 2002
Milk	5	8	21	Wang <i>et al.</i> 1997
UHT milk	ND	8	12	Palumbo <i>et al.</i> 1997
Pasteurised milk	ND	8	18	
Sterile minced beef	ND	8	15	
Minced beef	15	ND	4	Kasrazadeh and Genigeorgis 1995
Milk	ND	12	120	
Hispanic style cheese	8	10	60	
Minced beef	ND	10	8	Nissen <i>et al.</i> 2000
Minced pork	ND	10	<2	Ingham <i>et al.</i>
Minced beef	ND	10	<2	2007
Whey cheeses	2	12	30	Govaris <i>et al.</i>

				2001
Minced beef	6	12	15	Kennedy <i>et al.</i> 2000
Minced beef	ND	12	9	Walls and Scott 1996
Raw milk	7	ND	5	Farrag <i>et al.</i> 1992
Steamed egg	5	18	1.5	Yang and Chou 2000
Scrambled egg	5	18	1.5	
Sweet cream whipped salted butter	4.4	21 ¹	21	Holliday <i>et al.</i> 2003

? Indicates that there was some evidence of growth, but the increase was very small and within the margins of errors that might be experienced during counting.

¹ Some growth followed by death.

4.2.5 *Clostridium perfringens*

Most of the limited data located for *C. perfringens* were obtained from papers describing experiments with dynamic cooling conditions, and there is relatively little information for growth under static conditions. However, the minimum growth temperature recorded was 12.8°C on roast beef (Table 7) indicating that this organism need not be considered as relevant with respect to refrigeration temperatures. In another meat product no growth was recorded at a temperature of 12.5°C, but the incubation was relatively short at 14 days, and in two other foods no growth was measured at 12°C with longer incubations.

Since the minimum growth temperature observed is in excess of 12°C there is little prospect of this organism growing in chilled food unless it is subjected to quite marked temperature abuse.

Table 7. Data defining the minimum temperature of growth for *Clostridium perfringens*

Food	Temperature not allowing growth (°C)	Temperature allowing growth (°C)	Incubation period (d)	Reference
Roast beef	4.4	12.8	35	Hintlian and Hotchkiss 1987
Cooked cured pork	12	13.5	21	Juneja <i>et al.</i> 2006
Vacuum-packaged cooked beef	12	15	40	Juneja <i>et al.</i> 1994
Minced beef	12.5	ND	14	Goepfert and Kim 1975
Pea soup	10	15	56	de Jong <i>et al.</i> 2004
Cooked sous-	4	15	28	Juneja and

vide turkey mince				Marmer 1996
Cooked turkey	4	15	28	Juneja <i>et al.</i> 1996
Cooked beef	ND	17	NS	Huang 2002

NS Not stated.

4.2.6 *Listeria monocytogenes*

The data in Table 8 reflect the cold-tolerant nature of this organism. Growth was recorded at 0 and -1.5°C in several reports which means that, for foods where there are no other inhibiting factors, refrigeration retards but does not prevent growth of the organism. In broth culture growth has been measured at -2°C (Bajard *et al.* 1996). The organism should therefore be regarded as being able to grow, albeit slowly, at refrigeration temperatures in any food supporting its growth, and therefore need not be considered in setting a recommended chiller temperature. For foods which support the growth of *L. monocytogenes*, refrigeration alone cannot be used as the primary means of its control.

Table 8. Data defining the minimum temperature of growth for *Listeria monocytogenes*

Food	Temperature not allowing growth (°C)	Temperature allowing growth (°C)	Incubation period (d)	Reference
Sliced roast beef	ND	-1.5	96	Hudson <i>et al.</i> 1994
Smoked blue cod	ND	-1.5	40	Bell <i>et al.</i> 1995
Spadefish	1 ¹	ND	21	Harrison <i>et al.</i> 1991
Weakfish	1	ND	21	
Shrimp	1	ND	21	
Cooked meats	ND	0	35	Duffy <i>et al.</i> 1994
Pasteurised chicken breast	ND	0	NS	Nyati 2000
Pasteurised beef sirloin	ND	0	NS	
Cooked crawfish tail meat	ND	0	20	Dorsa <i>et al.</i> 1993
Fresh coconut cut	ND	2	9	Sinigaglia <i>et al.</i> 2006
Cooked chicken nuggets	ND	3	18	Marshall <i>et al.</i> 1991
Fresh endive	ND	3	10	Carlin <i>et al.</i> 1996
Vanilla cream	ND	3	38	Panagou and Nychas 2008
Bratwurst	ND	3	42	Glass <i>et al.</i> 2002

Queso fresco	ND	3	21	Mendoza-Yepes <i>et al.</i> 1999
Milk	ND	4	4	Donnelly and Briggs 1986
Bologna	ND	4	60	Yoon <i>et al.</i> 2009
Frankfurters	ND	4	40	
Turkey frankfurters	ND	4	14	Islam <i>et al.</i> 2002a
Chicken luncheon meat	ND	4	14	Islam <i>et al.</i> 2002b
Hard boiled egg	ND	4	21	Claire <i>et al.</i> 2004
Egg salad	ND	4	21	Hwang and Marmer 2007
Pasta salad	ND	4	32	
Light butter	ND	4	32	Lanciotti <i>et al.</i> 1992
Minced beef	4	ND	14	Johnson <i>et al.</i> 1988
Raw milk	4	ND	7	Gaya <i>et al.</i> 1991
Chocolate milk	ND	4	21	Rosso <i>et al.</i> 1996
Whole milk	ND	4	21	
Skimmed milk	ND	4	21	
Cream	ND	4	21	
Minced beef	ND	4	NS	
Smoked salmon	ND	4	21	
Sweet cream whipped salted butter	ND	4.4	21	Holliday <i>et al.</i> 2003
Pasteurised milk	ND	6	16	Bearns and Girard 1958
Chicken breast meat	1	6	10	Hart <i>et al.</i> 1991
Steamed egg	ND	5	1.5 ²	Yang and Chou 2000
Scrambled egg	ND	5	1.5 ²	
Whey cheeses	ND	5	38	Papageorgiou <i>et al.</i> 1996
Cooked beef	ND	5	33	Hudson and Mott 1993
Cooked mussels	ND	5	25	Hudson and Avery 1994
Cooked MAP shrimps	0	5	120	Dalgaard and Jørgensen 2000
RTE salmon	ND	5	14	Burnett <i>et al.</i> 2005
RTE turkey	ND	5	14	
RTE ham	ND	5	14	
Soy milk	ND	5	30	Ferguson and Shelef 1990
Minced beef	0	6	15	Kennedy <i>et al.</i> 2000
Ricotta cheese	ND	6-8	63	Davies <i>et al.</i>

				1997
Soy milk	ND	8	31	Tipparaju <i>et al.</i> 2004
Asparagus	4	8	22	Rodríguez <i>et al.</i> 2000
Minced beef	0	10	28	Duffy <i>et al.</i> 2000
Minced beef	4	10	14	Nissen <i>et al.</i> 2000

¹ Samples stored “on ice”

² The graph indicates that the lag time may have been just resolved by the end of the experiment, although this could have been a result of variability in counting.

4.2.7 *Yersinia enterocolitica*

The data (Table 9) reflect the cold-tolerant nature of this organism. Growth was recorded at 0 and -1.5°C in several reports which means that for most foods refrigeration only retards growth and does not prevent it. The organism should therefore be regarded as being able to grow, albeit slowly, at refrigeration temperatures in any food supporting its growth, and therefore need not be considered in setting a recommended chiller temperature.

Table 9. Data defining the minimum temperature of growth for *Yersinia enterocolitica*

Food	Temperature not allowing growth (°C)	Temperature allowing growth (°C)	Incubation period (d)	Reference
Sliced roast beef	ND	-1.5	96	Hudson <i>et al.</i> 1994
Smoked blue cod	ND	-1.5	40	Bell <i>et al.</i> 1995
Raw beef	ND	0	14	Hanna <i>et al.</i> 1977
Raw minced beef	ND	0*	13	Bhaduri and Phillips 2008
Bologna	ND	2	30	Nielsen and Zeuthen 1984
Milk	ND	3	20	Stern <i>et al.</i> 1980
Raw milk	ND	4	5	Farrag <i>et al.</i> 1992
Pasteurised milk	ND	4	10	Hellmann and Heinrich 1985
UHT Milk	ND	4	15	
Fresh pork	4	ND	3	
Minced beef	ND	4	14	Nissen <i>et al.</i> 2000
Spring water	ND	4	448	Karapinar and Gönül 1991

Light butter	ND	4	32	Lanciotti <i>et al.</i> 1992
Cooked chicken meat (sterile)	ND	4	NS	Wei <i>et al.</i> 2001
Brie	ND	4	35	Little and Knøchel 1994
Cooked beef	ND	5	21	Hudson and Mott 1993
Cooked mussels	ND	5	38	Hudson and Avery 1994
Bologna	ND	5	23	Nielsen and Zeuthen 1984

* data for *Yersinia pseudotuberculosis*

4.3 Summary of data for all pathogens of interest

A similar exercise to this was undertaken 45 years ago, albeit with a shorter list of pathogens and fewer data (Michener and Elliott 1964). The conclusions reached for *Salmonella*, *C. perfringens* and *S. aureus* (the only pathogens in common to both studies) have not changed significantly over that period, and many issues concerning methodology and interpretation of the data endure. They concluded that the minimum growth temperature for *S. aureus* and *Salmonella* was 6.7°C. *C. perfringens* was not discussed, presumably, as only three data points were listed.

ICMSF (Table 2) provide a lower growth temperature for *Salmonella* of 5.2°C, with the caveat that it is <7°C for most serovars. However no specific confirmatory data are provided. They state that some of these low temperature observations have not been confirmed and that “Additionally, because the duration of incubation will be very long at low temperatures, it is important that storage temperatures are monitored continuously to substantiate claims of growth at <5°C”.

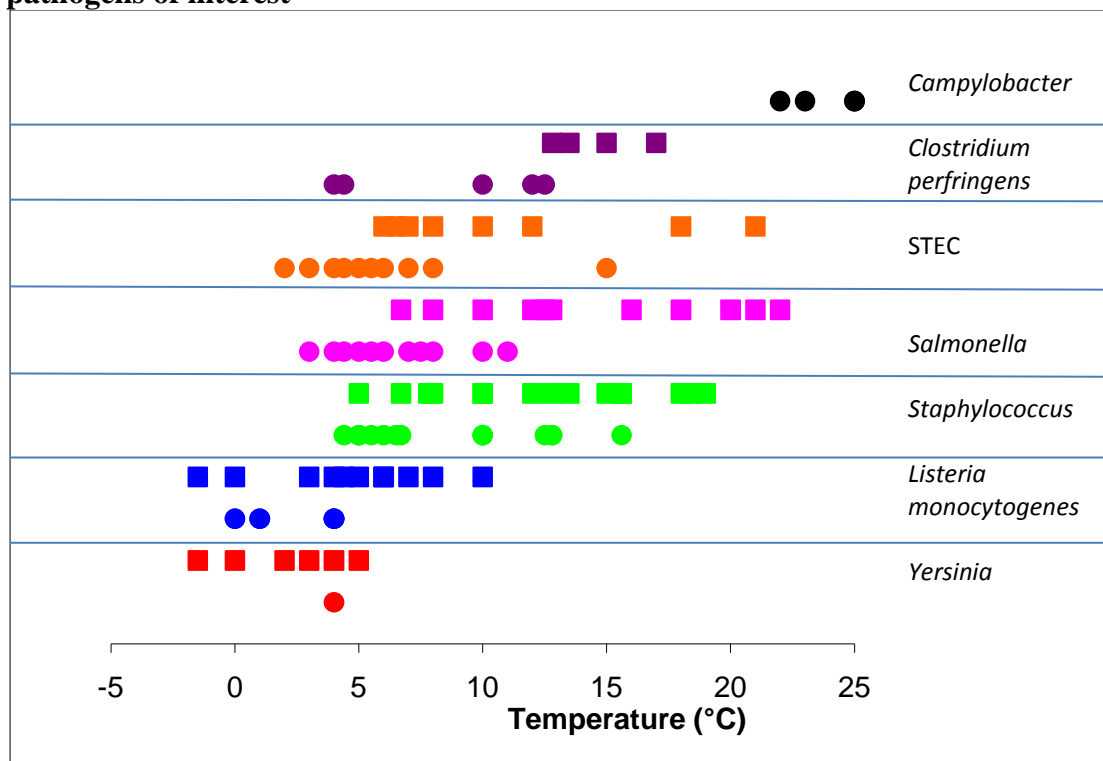
For *E. coli*, which was one of their faecal indicators, growth was reported at very low temperatures, down to -2°C in broth and 3-5°C in milk. However, other data they present show no growth at temperatures from 2°C (in medium) to 7.2°C (in liquid egg). The data shown in the table for STEC above were mostly for *E. coli* O157:H7 and it is possible that there is heterogeneity among the other serotypes. However, there is little in the literature to suggest that *E. coli* has anything other than a 6-7°C minimum growth temperature.

The data obtained in the present study were plotted to show the ranges of temperatures found in the literature (Figures 1 and 2).

Consideration needs to be given to the lower temperature end of the datapoints representing growth and the upper temperature end of the datapoints representing no growth. Particular consideration must be paid to the overlap between the two as this represents the area of uncertainty. This uncertainty is a result, *inter alia*, of strain variation, physicochemical parameters of different foods and the length over which the experiments were conducted. It is particularly pronounced with STEC where the overlap is from 5 to 15°C. An appropriate approach may be to consider the lowest

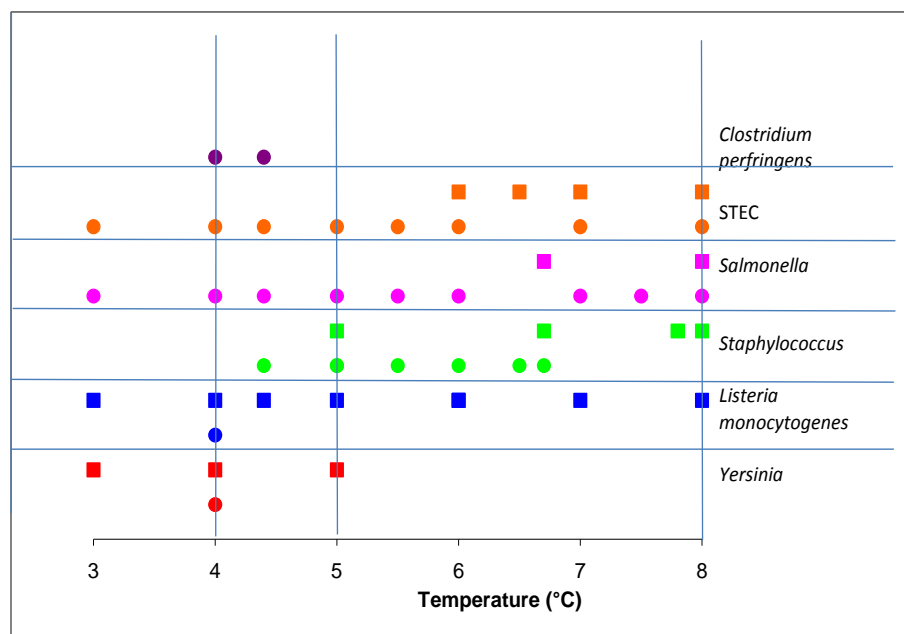
temperature at which growth was observed or the highest temperature at which it was not, depending on the available data.

Figure 1: Graphical representation of the minimum growth temperature of pathogens of interest



Circles represent instances where growth did not occur, squares where growth did occur.

Figure 2: Figure 1 blown up in the region 0-8°C and showing recommended chiller temperatures at 4, 5 and 8°C.



Circles represent instances where growth did not occur, squares where growth did occur

For *Campylobacter* there is no overlap, but neither are there data that can define the minimum growth temperature in food with any greater accuracy than $31 \pm 6^\circ\text{C}$. For *Listeria* and *Yersinia* some temperatures recorded for conditions not allowing growth are higher than others at which growth occurred. For both of these species there are very few data for conditions, other than freezing, where no growth was recorded. Also for both, it must be assumed that growth can occur on foods which allow it under normal commercial refrigeration. None of these pathogens therefore have characteristics which make them of relevance to the setting of chiller temperature recommendations.

For *Staphylococcus* the temperatures at which growth did not occur are a subset of those at which it did. Possibly growth of this species at low temperatures is very sensitive to the conditions it experiences in the food; i.e. there is considerable variation depending on food type.

Both *Salmonella* and STEC datasets also show a degree of overlap which appears to be less than that for *Staphylococcus*. For *Salmonella* the maximum temperature not permitting growth was 11°C and the minimum allowing growth on food 6.7°C . For STEC the equivalent values were 8°C and 6°C , respectively.

Only for *Clostridium perfringens* was there a clear and quite precise division; the maximum temperature at which growth was found not to occur was 12.5°C , while the

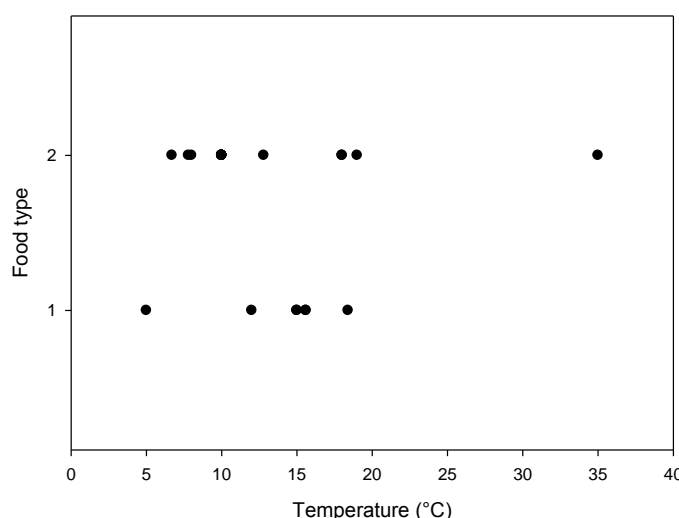
minimum allowing growth was 12.8°C. No overlap occurred, but this could be a result of the comparatively small dataset.

Commentary should be given on some of the papers reporting growth at temperatures regarded as being lower than that those regarded as “conventional wisdom” (ICMSF 1996). For *Staphylococcus*, growth is reported on bacon at 5°C. In this case there was an initial rise in the concentration of the pathogen, followed by a reduction, but then a longer, significant and sustained increase. An increase of almost 3 log₁₀ cfu/g occurred over a period of approximately four weeks. No information on storage conditions is given.

The lowest growth temperature for *Salmonella*, 6.7°C, was recorded in chicken à la king (Angelotti *et al.* 1961). These data are more equivocal than for *Staphylococcus* but the shape of the curve is consistent with a bacterial growth curve and there was a lag phase of 3 days followed by an almost 1 log₁₀ cfu/g increase in concentration over the following two days. This study would have been more useful if the incubation period had been longer. The data showing growth of STEC in sterile raw ground beef at 6°C are not shown in the original paper (Tamplin *et al.* 2005) but a growth rate of 0.003 ln/h and no lag phase (the growth curve was reported to be non-sigmoidal) were reported. Ten isolates were tested and, of these, nine were reported to grow at 6°C.

Examination of the overall data set tends to show that whether the food is uncooked or cooked does not seem to affect the minimum growth temperature. The most likely case where such a difference might exist is with *Staphylococcus* where the minimum growth temperature may be higher in raw foods than cooked. A plot of the data (Figure 3) shows that there is no difference, although a single data point (for bacon) extends the range of data for uncooked foods to 5°C. However, if that datapoint was removed then there is a suggestion that *Staphylococcus* needs a temperature of 12°C or above in order to grow on raw meats.

Figure 3: Comparison of low temperatures permitting the growth of *Staphylococcus* on cooked and uncooked foods.



Legend

- 1: Temperatures allowing growth of *Staphylococcus* in uncooked foods
- 2: Temperatures allowing growth of *Staphylococcus* in cooked foods

4.4 Advice from Overseas Competent Authorities

Table 10 shows some chilled storage temperature recommendations used by competent authorities overseas. Of note is the higher temperature recommended by competent authorities in the UK compared to Australia, Canada and the USA.

Table 10. Summary of Chiller Temperatures Recommended by Overseas Competent Authorities

Country	Temperature	Link
Australia	5°C or colder	www.foodstandards.gov.au/_srcfiles/FSTemp_control_Edition_for_printing.pdf
Canada	4°C or less	www.inspection.gc.ca/English/fssa/concen/tipcon/storage.shtml
England, Wales and Northern Ireland	Below 8°C (required)	www.food.gov.uk/multimedia/pdfs/tempcontrolguideuk.pdf
USA	Air temperature: 3.3°C food temperature: 5°C 5°C	www.idph.state.il.us/about/fdd/fdd_fs_foodservice.htm http://www.fda.gov/Food/FoodSafety/RetailFoodProtection/FoodCode/FoodCode2009/ucm186451.htm

5 DISCUSSION

This analysis was made difficult because of the wide variety of approaches taken to generate the data. A more precise definition of minimum growth temperatures would have been produced if data were available for growth experiments carried out near the minimum with small increments of temperature. In most of the literature reviewed the purpose of the publication was not to identify the lower temperature boundary and so many data sets provided data with quite a large temperature difference between the minimum allowing growth and the maximum at which growth did not occur.

The results are largely consistent with the “conventional wisdom” with a few reports of growth occurring at temperatures beneath those regarded as the norm. Chilled storage will not prevent the growth of some of these pathogens in some foods, while other pathogens will not grow except under gross temperature abuse to the extent that overt spoilage is likely. STEC, *Staphylococcus* (with one report of growth at 5°C) and *Salmonella* are the three pathogens considered here that have growth minima at temperatures in the 6-7°C range, and so could potentially grow under conditions of mild temperature abuse.

L. monocytogenes and *Y. enterocolitica* can grow at temperatures below 0°C and so only freezing would prevent their growth. If prevention of growth is the goal of chilled storage then it cannot be achieved in these two species by chilled temperatures alone.

Recommended chilled food storage temperatures vary quite widely; from 4°C (Canada) to “below 8°C” (UK). At 4°C only *L. monocytogenes* and *Y. enterocolitica* could grow, but at 7.9°C (which is “below 8°C”) all of the pathogens other than *Campylobacter* and *Clostridium perfringens* could grow. From the data there is no real advantage in using a 4°C limit in preference to one of 5°C as the same range of pathogens is controlled. An exception might be *S. aureus* where growth has been reported at 5°C. However, toxin is not produced at temperatures less than 10°C. The use of 4°C as opposed to 5°C does allow a greater safety margin but also would require a greater energy input. Use of the lower temperature will also increase the shelf life of these foods.

The generation of systematic data to obtain a better estimate of the minimum growth temperature would need to be carefully planned. It would need a representative number of isolates to be tested in a number of foods. A comprehensive screening of isolates in bacteriological media at a range of low temperatures could be used as an initial screen as an isolate unable to grow under ideal conditions could be regarded as unlikely to grow when extrinsic conditions, such as those encountered in food, are harsher.

Another factor is the capacity for *Staphylococcus* to produce toxin at low temperatures. A comprehensive review of the factors affecting growth/toxin production at low temperatures has not been undertaken. However, the lower temperature limit for toxin production found was 10°C suggesting that this may not be important in properly chilled foods.

6 PRELIMINARY RECOMMENDATIONS

A summary of the minimum growth temperatures for the pathogens considered on food is shown in Table 11.

Table 11. Minimum Recorded Growth Temperatures of Pathogens on Foods

Pathogen	Minimum growth temperature (°C)
<i>Campylobacter jejuni</i>	25 ¹
<i>Staphylococcus aureus</i>	6.7 ¹
<i>Salmonella</i>	6.7
STEC	6
<i>Clostridium perfringens</i>	12.8
<i>Listeria monocytogenes</i>	-1.5
<i>Yersinia enterocolitica</i>	-1.5

¹ A caution regarding these data is given under Tables 3 and 4.

Because of their growth characteristics the data for *Yersinia* and *L. monocytogenes* are not useful for determining a recommended chiller temperature. These organisms have been reported to grow at temperatures $\leq 0^{\circ}\text{C}$ in several papers. Because of this food would be frozen to prevent growth. If prevention of growth is the goal of chilled storage then it cannot be achieved in these two species by chiller temperatures alone.

Similarly the data for *Campylobacter* and *C. perfringens* are uninformative because of the high minimum growth temperatures of these bacteria. Foods would need to be significantly temperature abused for the conditions at which these organisms could grow would arise.

Guidelines therefore need to consider data for *Salmonella*, *S. aureus* and STEC. These have similar growth temperature minima at around 6-7°C. There is one report at temperature lower than this, i.e. 5°C for *S. aureus* in bacon, but the minimum temperature for toxin production is 10°C. Other work showing potential growth of *Salmonella* in broths at low temperatures does not seem to have been replicated in food.

International recommendations for chiller temperatures (4-5°C) are therefore adequate in respect to the fact that they prevent the growth (or toxin production) of STEC, *Salmonella* and *S. aureus*. There may be a need for a safety factor, in which case the use of 4 rather than 5°C provides a greater margin. Additionally it will slow the growth of *L. monocytogenes* and *Y. enterocolitica*, as well as assisting with maintaining product quality. There is a cost, however, in the increased power consumption needed to operate refrigeration units at the lower temperature.

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