RISK PROFILE:
SHIGA-LIKE TOXIN PRODUCING
ESCHERICHIA COLI IN UNCOOKED COMMINUTED
FERMENTED MEAT PRODUCTS

Prepared as part of a New Zealand Food Safety Authority
contract for scientific services

by

Dr Rob Lake
Dr Andrew Hudson
Peter Cressey

February 2003

A CROWN RESEARCH
INSTITUTE

RISK PROFILE:
SHIGA-LIKE TOXIN PRODUCING
ESCHERICHIA COLI IN UNCOOKED COMMINUTED
FERMENTED MEAT PRODUCTS

Professor Ian Shaw
Food Safety Programme Manager

Dr Rob Lake   Rosemary Whyte
Project Leader   Peer Reviewer

DISCLAIMER

This report or document ("the Report") is given by the Institute of Environmental Science and Research Limited ("ESR") solely for the benefit of the New Zealand Food Safety Authority ("NZFSA"), Public Health Services Providers and other Third Party Beneficiaries as defined in the Contract between ESR and the NZFSA, and is strictly subject to the conditions laid out in that Contract.

Neither ESR nor any of its employees makes any warranty, express or implied, or assumes any legal liability or responsibility for use of the Report or its contents by any other person or organisation.

CONTENTS

LIST OF TABLES

LIST OF FIGURES

1   INTRODUCTION

The purpose of a Risk Profile is to provide contextual and background information relevant to a food/hazard combination so that risk managers can make decisions and, if necessary, take further action. The place of a risk profile in the risk management process is described in "Food Administration in New Zealand: A Risk Management Framework for Food Safety" (Ministry of Health/Ministry of Agriculture and Forestry, 2000). Figure 1 outlines the risk management process.

Figure 1:   Risk Management Framework
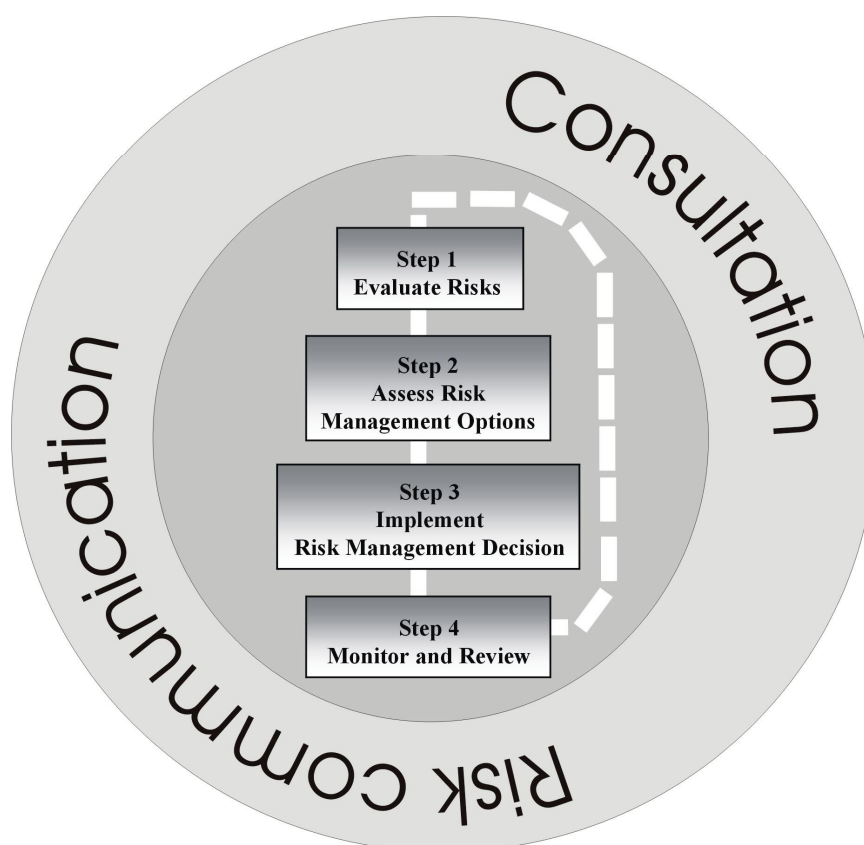
Figure reproduced from "Food Administration in New Zealand. A risk management framework for food safety" (Ministry of Health/Ministry of Agriculture and Forestry, 2000).

In more detail, the four step process is:

1. Risk evaluation

- identification of the food safety issue
- **establishment of a risk profile**
- ranking of the food safety issue for risk management
- establishment of risk assessment policy
- commissioning of a risk assessment
- consideration of the results of risk assessment

2. Risk management option assessment

- identification of available risk management options
- selection of preferred risk management option
- final risk management decision

3. Implementation of the risk management decision

4. Monitoring and review.

The Risk Profile informs the overall process, and provides an input into ranking the food safety issue for risk management. Risk Profiles include elements of a qualitative risk assessment. However, in most cases a full exposure estimate will not be possible, due to data gaps, particularly regarding the level of hazard in individual foods. Consequently the parts of a Risk Profile that relate to risk characterisation will usually rely on surveillance data.

The Risk Profiles also provide information relevant to risk management. Based on a Risk Profile, decisions are made regarding whether to conduct a quantitative risk assessment, or take action, in the form of gathering more data, or immediate risk management activity.

This Risk Profile concerns shiga-like toxin producing *Escherichia coli* (STEC) (shiga toxins are so named due to their similarity to those produced by some species of *Shigella*). The most well known of these is *E. coli* O157:H7 (and H-) but this profile also considers other serotypes. These organisms are important emerging pathogens, recognised for the first time in the United States in 1982. The first human case of illness caused by *E. coli* O157 in New Zealand occurred in 1993 (Baker *et al.*, 1999).

The food group to be addressed in this Risk Profile is uncooked comminuted fermented meat (UCFM) products. This type of food relies on pH and water activity reduction to control pathogens. There have been suggestions that some strains of STEC are more acid tolerant than *E. coli* bacteria in general (Tilden *et al.*, 1996) and so control of this bacterium is particularly important for UCFM.

The sections in this Risk Profile are organised as much as possible as they would be for a conventional qualitative risk assessment, as defined by Codex (1999).

Hazard identification, including:

- A description of the organism
- A description of the food group

Hazard characterisation, including:

- A description of the adverse health effects caused by the organism.
- Dose-response information for the organism in humans, where available.

Exposure assessment, including:

- Data on the occurrence of the hazard in the New Zealand food supply.
- Data on the consumption of the food group by New Zealanders.
- Qualitative estimate of exposure to the organism (if possible).
- Overseas data relevant to dietary exposure to the organism.

Risk characterisation

- Information on the number of cases of adverse health effects resulting from exposure to the organism with particular reference to the identified food (based on surveillance data)
- Qualitative estimate of risk, including categorisation of the level of risk associated with the organism in the food (categories are described in Appendix 1).

Risk management information

- A description of the food industry sector, and relevant food safety controls.
- Information about risk management options.

Conclusions and recommendations for further action

Note: Earlier versions of this document were produced as part of a project undertaken by ESR and jointly directed by the Ministry of Health and the Ministry of Agriculture and Forestry. Ministry responsibilities for food safety were combined into the New Zealand Food Safety Authority (NZFSA) in July 2002.

The Australia New Zealand Food Authority (ANZFA) became Food Standards Australia New Zealand (FSANZ), also in July 2002.

Information and reports published by the older organisations have been referenced to those names.

2   HAZARD IDENTIFICATION: THE ORGANISM

2.1   Shiga-like toxin producing *Escherichia coli* (STEC)

The following information is taken from data sheets prepared by ESR under a contract for the Ministry of Health. The data sheets are intended for use by regional public health units. Information for *E. coli* O157 is presented separately from other shiga-like toxin producing serotypes. The ability of the serotypes in the latter group to cause disease varies greatly.

2.1.1   Nomenclature

There are three acronyms that are in common use that pertain to this group of organisms. The two in most common use currently are VTEC (verocytotoxigenic *Escherichia coli*) and STEC (shiga toxigenic *Escherichia coli*). The acronym VTEC is derived from the fact that the toxin expressed causes a pathological effect on Vero cells in tissue culture, while the acronym STEC is derived from the fact that the toxins are shiga-like i.e. similar to those produced by *Shigella dysenteriae* (Chart, 2000).. The two acronyms have now become *de facto* synonyms. An alternative meaning to the acronym STEC is "Shiga-like toxin producing *E. coli*"; this is less commonly used although strictly more accurate. The term shiga-toxigenic *E. coli* is used in recent reviews (Baker *et al.*, 1999; Jaeger and Acheson, 2000) and by the International symposia and workshops on shiga toxin (verocytotoxin)-producing *Escherichia coli* infections.

The acronym EHEC, (Enterohaemorrhagic *Escherichia coli*). refers to those *E. coli* that have the same clinical, epidemiological and pathogenic features associated with the prototype EHEC organism *E. coli* O157:H7 (Karmali, 1989). Strictly EHEC are therefore a specific subset of the two groups of organisms described above as some STEC/VTEC have never been associated with human disease. However, EHEC is often used as a synonym of STEC and VTEC.

STEC will be the acronym used throughout this document.

Individual strains of STEC are denoted by their O and H serotypes; O= "ohne hauch" or the somatic antigen, H= "hauch" or the flagellar antigen. Non-motile isolates (normally recorded as NM) are considered here to be H-, i.e. without an H antigen. If the serotype cannot be determined it is described as NT: "non-typable". Some isolates of STEC react (i.e. agglutinate) with all sera; these are described as "rough".

Note that in the following text the term "D" is used. In microbiological terms "D" refers to a 90% (or decimal or 1 log cycle) reduction in the number of organisms.

2.2   *Escherichia coli* O157

2.2.1   The organism/toxin

E. coli O157 is a pathogenic variant of an organism that is generally regarded as innocuous. The H serotypes associated with this O type in cases of disease are H7 and H-.

2.2.2 Growth and Survival

Growth:

Temperature: Optimum 37°C, range 7-8 to 46°C. Doubling time approx. 0.4 hours at 37°C.

pH: Optimum 6-7, range 4.4 to 9.0. The limit at the low pH end depends on the acidulant used. Mineral acids such as HCl are less inhibitory than organic acids (e.g. acetic, lactic – as produced post mortem in meat) at the same pH. Growth was inhibited in the presence of 0.1% acetic acid (pH 5.1).

Atmosphere: Can grow in the presence or absence of oxygen. Growth can occur in vacuum-packed meat at $8-9^{o}C$, but not when the meat is packed under 100% CO_2. At $10^{o}C$ growth was not inhibited under 100% N_2 or 20% CO_2:80% N_2 but was under 100% CO_2. Growth on lettuce was not inhibited by the presence of 30% CO_2, or under 97% N_2 :3% O_2.

Water activity: Optimum growth is at a_w = 0.995 minimum a_w = 0.950

Survival:

Temperature: Survives well in chilled and frozen foods. For example little change was noted in numbers in hamburgers stored at -20°C for 9 months.

pH: Can survive in low pH (down to 3.6) environments. In fact the organism dies slowly under these conditions and persistence is proportionate to the degree of contamination. For example, numbers reduced by only 100 fold after 2 months storage at $4^{o}C$ on fermented sausage of pH 4.5. Prior exposure to acidic conditions can increase acid tolerance. Has been shown to survive stomach pH (1.5) for periods longer than that required to clear an average meal (three hours).

Experiments to determine the acid tolerance of strains of enterohaemorrhagic *E. coli* (EHEC) showed that a number of strains could survive (i.e. were able to be recovered at levels up to 100%) at a pH of 2.5 or 3.0 for a number of hours (Benjamin and Datta, 1995). These data were consistent with outbreaks of EHEC linked with the acidic foods apple cider and mayonnaise. There have been claims that pathogenic *E. coli* are significantly more acid tolerant than non-pathogenic strains, but this has not been clearly established (McClure and Hall, 2000). Significant interstrain variation with respect to acid tolerance is a common feature of both commensal and O157 strains of *E. coli* (Duncan *et al.*, 2000).

Atmosphere: An atmosphere of 100% CO_2 enhanced survival of uninjured cells at both 4 and $10^{o}C$. Survival on fermented meat was equivalent when packed under air or under vacuum.

Viable but Non-Culturable (VNC) Cells: Evidence indicates that low temperature is the primary signal for entry into the VNC state in water (Rigsbee *et al.*, 1997) although sunlight too has been shown to cause VNC cells to form (Pommepuy *et al.*, 1996). Entry into the VNC state is suspected in high salt foods (Makino *et al.*, 2000).

2.2.3   Inactivation (CCPs and Hurdles)

Temperature: Rapidly inactivated by heating at 71°C (recommended temperature for hamburger cooking in the USA, in the UK it is 70°C for 2 minutes). D time at 54.4°C = 40 minutes. D time at 60 = 0.5–0.75 minutes (4.95 minutes in minced beef). D time at 64.3°C = 0.16 minutes.

pH: Inactivation at pH 4.5 in fermented meat created by lactic acid production from glucose by starter cultures.

Water activity: Withstands desiccation well and has caused disease through carriage on venison jerky.

Preservatives: 8.5% NaCl inhibits growth at 37°C, growth retarded above 2.5%. The amount of salt required for inhibition reduces as other factors such as temperature and pH become sub-optimal. For example 5% salt inhibited *E. coli* O157 at 12°C.

Radiation: Sensitive to UV and γ irradiation. D (kGy) approx. 0.31 frozen, 0.24 refrigerated in ground beef. A 2-3 kGy dose is sufficient to decontaminate meat.

2.2.4   Sources

Human: Faecal-oral person-to-person transmission is often reported in family members of cases who contracted the disease from food or water.

Animal: Found in the guts of ruminant animals. Cattle are considered primary reservoirs but sheep and deer may also carry the organism. Carriage of the organism by cattle is generally considered to be low, but estimates of prevalence are rising with improved laboratory techniques. Calves are thought to shed the organism more often than adult cattle.

Food: Incriminated foods overseas have usually been contaminated by cattle manure. Foods involved in outbreaks have included hamburgers, other meat products, apple juice, salads, bean sprouts, raw milk, cheese, melons, lettuce and yoghurt. For one case in New Zealand, an indistinguishable isolate was obtained from both the infected person and raw milk present in the home, although the route of infection is uncertain (Anonymous, 2002).

Environment: Water contaminated from faecal sources has been the vehicle involved in a number of large outbreaks overseas. Such waters have included reticulated drinking water and swimming/paddling pool water. Two cases in New Zealand have been attributed to the consumption of contaminated water (neither was reticulated water). The organism has been shown to survive for 150 days in soil and 90 days in cattle faeces. It can also survive for at least 4 months in sediment in cattle drinking troughs.

Transmission Routes: In summary, any food or water source that has been contaminated by the faeces of a ruminant animal. Direct contact with carrier animals is also a recognised transmission route. Secondary transmission is also common. Poor personal hygiene can also result in infection; eight pop festival attendees became infected after the event, which was held in a muddy paddock on which cattle had recently been grazed. The relative importance of the various transmission routes is currently not well understood in New Zealand.

2.3 Non-O157 Shiga-like Toxin Producing *Escherichia coli* (STEC)

2.3.1 The organism/toxin

These organisms form a diverse group of *E. coli* that are capable of producing shiga-like toxin(s), as is *E. coli* O157:H7. However, they are of widely differing pathogenic potential, varying from those that can cause illnesses similar to that produced by *E. coli* O157:H7 to those that have never been associated with disease.

By definition all STEC must produce one of two groups of toxins (denoted Stx1 and Stx2 with subscripts to denote variants), but other factors are required for pathogenicity and it is the possession of these that seems to determine the virulence of any one serotype. Other factors known to be involved include the ability to adhere to intestinal cells (*eaeA* gene), and the ability to produce a haemolysin (*hlyA* gene).

New Zealand isolates of STEC that have caused disease have, to date, possessed virulence factors in addition to either Stx gene.

2.3.2 Growth and survival

The behaviour of these organisms is largely the same as for serotype O157.

2.3.3 Inactivation (CCPs and Hurdles)

The behaviour of these organisms is largely the same as for serotype O157.

2.3.4 Sources

Human: Some serotypes are reported to be restricted to people, e.g. O1, O55:H7 and H:10 and O48:H21 (Bettelheim, 2000)

Animal: Ruminant animals, notably bovines, seem to be a natural reservoir of many of the non-O157 STEC that cause disease in humans.

Food, environment, transmission routes: Little is known about the distribution of these organisms in food and the environment. However, it seems likely that the situation will be similar to that for serotype O157. Non-O157 STEC are likely to be much more common than serotype O157 in foods, but only a small proportion of the isolates will be pathogenic to humans.

Non-O157 STEC have been detected in beef, pork and lamb mince, and unpasteurised milk.

3   HAZARD IDENTIFICATION: THE FOOD

3.1   Relevant Characteristics of the Food: Uncooked Comminuted Fermented Meat (UCFM) Products

Uncooked comminuted fermented meats (UCFM) are products which contain minced, chopped or ground meat along with salt (2.5-3%), nitrite, glucose (0.4-0.7%), spices, seasonings, and ideally a bacterial inoculum as a starter culture chosen according to the temperatures used in the process and the level of lactic acid desired (Ricke and Keeton, 1997). Some of the older traditional processes may not involve the use of a starter culture.

The meat ingredients are primarily beef and pork, with lamb and mutton meats less commonly used. Lean poultry meat is sometimes used but is less desirable as the fat has a higher polyunsaturated content, which makes it more susceptible to oxidation.

The production process involves (i) reduction of particle size of the meat, (ii) combination of ingredients, (iii) mixing and further reduction of particle size to produce a "batter", (iv) vacuum stuffing into a semi-permeable casing (thereby keeping oxygen exposure low), (v) incubating (ripening, fermentation) at the temperature optimum of the starter culture, (vi) heating (of some products) to inactivate the inoculum and eliminate pathogens, (vii) drying (and sometimes smoking) (Ricke and Keeton, 1997). Fermentation typically occurs over a period of 1-3 days, while the drying period can range from a few days to several weeks, depending on the product (Ross and Shadbolt, 2001).

UCFM products include dry sausages (e.g. salami, cappicola, pepperoni) and semi-dry sausages (e.g. summer sausages or cervelat, medwurst, bologna) (Ricke and Keeton, 1997).

Fermented sausages rely on both a reduced pH (4.6-5.3) and a reduced water activity (a_w) of <0.95 for microbial stability (ICMSF, 1998). Alternatively either a pH less than 4.5 or a water activity of <0.91 may achieve the same result (Ross and Shadbolt, 2001). If the moisture reduction during drying is less than 15%, smoking and mild heat treatment may be used as additional steps to restrict microbial growth. Moisture losses are much higher (25-30%) for dry salami varieties (DeBauch and Savage, 1993).

Reduction in pH is primarily achieved by the production of lactic acid from added carbohydrates (usually glucose) by lactic acid bacteria belonging to the genera *Streptococcus*, *Lactococcus*, *Leuconostoc*, *Pediococcus*, *Bifidobacterium* and *Lactobacillus* (Ross and Shadbolt, 2001).   The bacteria are added to the mix as a starter culture; the earlier practice of adding a portion of the previous batch ("backslopping") has been discontinued (it is illegal in Australia).

Glucono-delta-lactone is sometimes added to the batter of UCFM to rapidly reduce the pH, and thus inhibit the growth of pathogens during the initial stages of fermentation. Nitrite is also added and is important in colour and taste development, but may only be effective against pathogen growth in the early stages of fermentation as it is rapidly consumed by microbial activity (Ross and Shadbolt, 2001).

3.2   Hurdles

The hurdle concept is now well accepted in food microbiology and is the technology upon which safe production of salami (other than pasteurised varieties) relies. Hurdle technology "advocates the deliberate combination of existing and novel preservation techniques in order to establish a series of preservative factors (hurdles) that any microorganisms present should not be able to overcome" (Leistner and Gorris, 1995). It is an essential concept for the preservation of foods using "mild" techniques as it allows for the concerted and sometimes synergistic effects of a number of preservative factors that may be required at lower levels than if only one preservative factor, or hurdle, was used. In many cases none of the individual hurdles would be able to provide for a safe product on their own. Salami is an excellent example of a food that is preserved by the application of multiple hurdle technology where the hurdles used are the presence of nitrite, a low redox potential, a preferred competitive flora (inoculum), salt, low pH (from lactic acid), a low water activity and in some cases the presence of antibacterial compounds resulting from smoking. Some salamis may then receive a thermal treatment which is sufficient to pasteurise the food, but these heat treated products are not included in this Risk Profile.

Each producer of raw fermented sausages will have a recipe and process unique to the establishment, which is regarded as intellectual property and is commercially important to the manufacturer. In Europe considerable differences exist in the styles of the products and this may be reflected in both the recipe formulation and the nature and outcome of each processing step. For example, some products may be quick fermented while others take many days or even weeks.

It was demonstrated in 1992 that *E. coli* O157:H7 can survive but not grow during fermentation, drying and storage (for 2 months) of salami (Glass *et al.*, 1992). Given that the processing of UCFM products is complex, some more discussion of the process and controls follows. This is broken into three sections 1) the batter 2) production processes and 3) storage.

3.2.1   Batter

The treatment of the batter prior to processing has been shown to have some effect on the survival of *E. coli* O157:H7 (Faith *et al.*, 1998a). Various treatments of the batter ((i) refrigerated, (ii) frozen and thawed or tempered, (iii) frozen and thawed) were investigated and shown to influence the final numbers of *E. coli* O157:H7. Treatments (ii) and (iii) resulted in higher D reductions (2.1 and 1.6 $\log_{10}$ cfu/g respectively) compared to treatment (i) (1.1 $\log_{10}$ cfu/g) when assessed immediately after drying. The relativity persisted throughout storage at 21 and 4°C. There seemed to be some advantageous effect of the freezing component of the pre-treatments, possibly by the production of injured cells, which were more susceptible to the treatments that followed.

3.2.2   Production

Table 1 shows data from a number of studies of initial and final concentrations of *E. coli* O157:H7 resulting from salami production. The table shows that fermentation and drying alone do not result in large reductions in the number of *E. coli* O157:H7 with most processes resulting in an approximate 2D reduction in numbers.

In experiments where various parameters in salami manufacture (e.g. NaCl and pH) have been adjusted, a rapid fall in pH to a suitably low level in the presence of a mild heat treatment has been shown to be pivotal in the destruction of *E. coli* O157:H7. Addition of preservatives that inhibit the action of the starter cultures impedes acid production and results in reduced inactivation of *E. coli* O157:H7 (Chikthimmah *et al.*, 2001). Experiments with pepperoni showed that the conventional recipe and process resulted in a 0.84 D reduction in numbers, while by increasing the NaCl and sodium nitrite concentrations and using a lower final pH (4.4-4.6) this could be increased to a 4.79 D reduction (Riordan *et al.*, 1998).

In simulated meat and fermented meat systems Uyttendaele *et al.* (2001) provided data to suggest that survival could be longer under more extreme conditions (e.g. survival at pH 4.5 > pH 5.4 >7.0 at 7 and –18°C). It was suggested that this is because multiple challenges to the organism result in a general stress response, and that the use of hurdle technology would actually result in increased survival. Genes involved in stress response may also make the organism more virulent. However, these observations were made using a model system, which was essentially pH-adjusted artificial gravy. Hence the number of stress factors involved would be less than in an UCFM product.

Some investigation into the survival of *E. coli* O111 in Hungarian salami has also been carried out (Pidcock *et al.*, 2002). Here salami batter was inoculated with the pathogen and various starter organisms followed by fermentation at 25°C for 7d, conditions not typical for this product (where fermentation is for 72 h). With a commercial starter culture only around a 0.5 D reduction in *E. coli* O111 had occurred after 72 h, while the addition of other cultures could improve on this only by the order of 0.5-1.0 D. While these workers did produce salamis using the usual process they did not test them for reductions in pathogen numbers.

Table 1: Effect of UCFM product fermentation and drying on numbers of *Escherichia coli* O157:H7

Fermentation conditions	Drying conditions	Concentration of *Escherichia coli* O157:H7 ($\log_{10}$ cfu/g)			Overall D reduction	Reference
		Initial	After fermentation	After drying		
24°C, 90% RH to pH ≤ 4.8 (60–64 h)	13°C, 65% RH to M/Pr ratio ≤1.9:1 (approx. 21 days)	7.5–7.7	6.0 – 6.8	5.4–6.6	0.9–2.1	Faith *et al.*, 1998a
36°C, 90% RH to pH<4.8 (14-16h)	13°C, 65% RH to M/Pr ratio ≤ 1.6:1 (12-16 days)	7.5	7.0 (15% fat) 6.9 (20% fat) 6.4 (30% fat)	6.5 6.2 5.8	1.0 1.3 1.7	Faith *et al.*, 1998b
26°C, 85% RH to pH ≤ 5.0 (14-18 h)	13°C, 65% RH to M/Pr ratio ≤ 1.6:1 (15-21 days)	6.9	6.8	5.7 (21 days drying)	1.2	Hinkens *et al.*, 1996
36°C, 90% RH to pH ≤ 4.8 (16 to 20 h)	13°C, 65% RH to M/Pr ratio ≤1.6:1 (18 days)	7.8	6.8	5.9	1.9	Faith *et al.*, 1997
24°C, 90-95% RH, 3 days	22°C, 80-85% RH until 40% moisture achieved	7.7	NR	+ starter culture 5.7 - starter culture 7.4	2.0 0.3	Calicioglu *et al.*, 2001
29 rising to 41°C, 80% RH to pH 4.6 or 5.0	49 rising to 66°C (sausage internal temp 54°C), 60% RH	8.0 7.8	6.6 (to pH 4.6) 7.5 (to pH 5.0)	<1.0 after heating. 4.6 after heating 2.8 after heating and holding for 30 min <1.0 after heating and holding for 60 min	>7.0 3.2 5.0 >6.8	Calicioglu *et al.*, 1997
21 rising to 38°C, 75% RH to pH 4.8	15°C 64-70% RH until a_w <0.8 (approx. 7 days)	6.7	6.3	5.9	0.8	Riordan *et al.*, 1998
15.6 rising to 35.6°C, to pH 4.8 (13-14 h)	12.8°C, 70% RH until M:Pr ratio <1.9:1 (18-21 days)	4.7	4.4	3.7	1.0	Glass *et al.*, 1992

RH = Relative humidity, M/Pr = moisture to protein ratio, NR = Not Reported

3.2.3 Storage

Data for the effect of storage conditions on the survival of *E. coli* O157:H7 in UCFM products are shown in Table 2. Storage at room temperature under air is an effective way to decrease numbers of *E. coli* O157:H7 in correctly manufactured salami. However, such storage may result in undesirable product characteristics and control of microbiological hazards other than *E. coli* O157:H7 also has to be considered. Slightly less effective but technologically acceptable storage conditions may need to be used to increase inactivation. Heat shocking of cells was also found to increase inactivation during storage. It should be noted that both papers by Faith *et al.* (1997, 1998a) and Clavero and Beuchat (1996) focused on sliced (as opposed to whole) salami only.

The effect of temperature on the survival of the organism at a fixed low pH has also been shown in a model system by Uyttendaele *et al.* (2001).

Table 2: **Data for reductions in *Escherichia coli* O157:H7 numbers during storage of UCFM products**

Conditions	Temperature (^{o}C)	Storage time (days)	Concentration of *E. coli* O157:H7 ($\log_{10}$ cfu/g)		D reduction	Reference
			Initial	**Final**		
Air	4	90	5.4-6.6	<1-3.0	2.4->6.6	Faith *et al.,* 1998a
	21			<1	>5.4->6.6	
Vacuum	4	90	5.4-6.6	1.4-3.6	1.8-5.2	Faith *et al.,* 1998a
	21			<1	>5.4->6.6	
Air						Faith *et al.,* 1998b
15% fat	4	28	6.5	6.0	0.5	
20% fat	4	28	6.2	5.8	0.4	
35% fat	4	28	5.8	5.5	0.3	
15% fat	21	14	6.5	2.0	4.5	
20% fat	21	14	6.2	1.4	4.8	
35% fat	21	14	5.8	1.3	4.5	
Vacuum						Faith *et al.,* 1998b
15% fat	4	28	6.5	5.8	0.7	
20% fat	4	28	6.2	5.8	0.4	
35% fat	4	28	5.8	5.6	0.2	
15% fat	21	14	6.5	3.0	3.5	
20% fat	21	14	6.2	2.0	4.2	
35% fat	21	14	5.8	1.2	4.6	
Air	-20	90	5.2	3.6	1.6	Faith *et al.,* 1997
	4	90	5.9	3.7	2.2	
	21	28	5.5	<1.0	>4.2	
Vacuum	4	90	6.0	4.3	1.7	Faith *et al.,* 1997
	21	90	5.6	<1	>4.6	
CO$_2$	-20	90	5.1	3.9	1.2	Faith *et al.,* 1997
	4	90	5.8	4.1	1.7	
Vacuum						Calicioglu *et al.,* 2001
+ starter	21	28	5.7	<1	>4.7	
- starter		28	7.4	5.4	2.3	
+ starter	4	28	5.7	3.7	2.0	Calicioglu *et al.,* 2001
- starter		28	7.4	7.2	0.2	

Conditions	Temperature (°C)	Storage time (days)	Concentration of *E. coli* O157:H7 (log$_{10}$ cfu/g)		D reduction	Reference
pH 5.0, heat to 54°C	25	7	4.6	4.1	0.6	Calicioglu *et al.,* 1997
	4	7	4.6	4.7	0.0	
pH 5.0, heat to 54°C, hold 30 min	25	7	2.8	<1.0	>1.8	Calicioglu *et al.,* 2001
	4	7	2.8	2.0	0.8	
pH 4.8, vacuum packed	4	56	3.7	2.7	1.0	Glass *et al.,* 1992
pH 4.8, a$_w$ 0.95	5	32 (UH)	4.9	2.4	2.5	Clavero and Beuchat, 1996
	5	32 (HS)	4.5	1.1	3.4	
	20	16 (UH)	4.9	1.9	3.0	
	20	4 (HS)	4.5	1.9	2.6	
pH 4.63, a$_w$ 0.90	5	32 (UH)	4.9	2.2	2.7	Clavero and Beuchat, 1996
	5	32 (HS)	4.5	1.2	3.3	
	20	16 (UH)	4.9	1.9	3.0	
	20	4 (HS)	4.5	2.1	2.4	

HS= heat stressed, UH=unheated

Some modeling work has been carried out in Australia, which is aimed at predicting the effect of the steps in salami production on any STEC present. The model is currently being validated, and is some way off from being available (Paul Vanderlinde, personal communication). One such attempt at modeling has already appeared in the literature (Pond *et al.*, 2001). These authors used three different models, but some aspects of the process were not included, some simplifications made and the need for more data points identified. Nevertheless the models seem to be reasonably useful.

As an alternative method not involving cooking, the application of irradiation to meat prior to the production of pepperoni to give a >5 log$_{10}$ reduction in *E. coli* O157:H7 produced a food with acceptable organoleptic qualities, unlike heat-treated salami where texture and colour were adversely affected (Johnson *et al.*, 2000).

Experiments to determine the survival of *E. coli* O157:H7 inoculated onto salami (pepperoni) slices on top of frozen pizza showed, unsurprisingly, that the amount of inactivation was dependent on the time and temperature of cooking (Faith *et al.*, 1998b). Baking of the pizza at 246°C for 15 minutes or at 191°C for 20 minutes was necessary to reduce the numbers from around 5 log$_{10}$ cfu/g to non-detectable by enrichment.

3.3 The Food Supply in New Zealand

According to data from Statistics New Zealand, as of February 2001 there were 75 business enterprises engaged in "Bacon, Ham and Smallgood Manufacturing" (New Zealand Standard Industrial Category C211300). These represented 85 geographic locations (i.e. some enterprises operated at more than one location). However, only a proportion of these businesses will be involved in the production of UCFM. The actual number of businesses involved in producing UCFM is probably approximately ten, including six major manufacturers (Francis Clement, Pork Industry Board, personal communication).

The New Zealand Pork Industry Board (NZPIB) has provided an estimate of total UCFM annual production in New Zealand at 343,367 kg.

3.3.1 Imported food

Data from Statistics New Zealand show that meat preparations (with a non-poultry base) (Code 1601.00.00.29) comprised 170 tonnes for the year to September 2001. The majority came from Australia (145 tonnes) and Canada (20 tonnes). This import category includes salami as well as frankfurters, liver sausages, saveloys, bologna, paté, meat pastes etc., so the amount of imported UCFM products is likely to be much less than 170 tonnes. However, it does appear that Australia is the most common source of imported food of this type.

4 HAZARD CHARACTERISATION: ADVERSE HEALTH EFFECTS

Infection with STEC results in the organism invading the gut and then producing one or more toxins. Toxins are not produced in foods, but only after infection.

This can cause a wide range of outcomes. Some cases will be asymptomatic, others will experience diarrhoea, and a proportion will go on to suffer more serious outcomes including haemorrhagic colitis (HC), haemolytic uraemic syndrome (HUS), thrombotic thrombocytopaenic purpura (TTP) and death (Desmarchelier and Grau, 1997).

4.1 Symptoms

Incubation: 3 to 9 days (mean 4 days) following ingestion of the bacteria.

Diarrhoea Symptoms: Diarrhoea is accompanied by severe abdominal cramps. Vomiting may occur (30-60% of cases) but fever is infrequent (less than 30% of cases) (Dundas and Todd, 2000).

Condition: More serious consequences of infection include:

> *Haemorrhagic Colitis (HC):* Bloody diarrhoea, inflammation of the large bowel, severe abdominal pain, vomiting, no fever.

> *Haemolytic Uraemic Syndrome (HUS):* HUS follows HC and is normally associated with children. The condition is characterised by renal failure and the consequences of that including seizures, coma, death.

> *Thrombotic Thrombocytopaenic purpura (TTP):* A version of HUS most often experienced by the elderly. Involves loss of platelets, skin coloration, fever and nervous system disorder (seizures and strokes) in addition to HUS signs and symptoms. There is no prior episode of diarrhoea. Illness lasts from 2-9 days.

Treatment: Dialysis, maintenance of fluid balance and treatment of hypertension in cases of HUS.

Long Term Effects: HUS: kidney problems, hypertension, neurological deficits.

4.2 Serotypes Causing Disease

4.2.1 Non-O157 serotypes

Over 200 non-O157 STEC serotypes have been isolated from humans and are clearly recognized as human pathogens. The World Health Organisation has identified the most important non-O157 STEC serogroups, from an epidemiologic perspective, as O26, O103, O111 and O145 (WHO, 1998). An updated list of STEC, with literature references, is maintained by Dr K. Bettelheim (National *E. coli* Reference Laboratory, Melbourne,

Australia) and can be found on the World Wide Web at: http://www.microbionet.com.au/frames/feature/vtec/brief01.html

Approximately 90% of notified STEC infections in New Zealand are caused by *E. coli* O157 (H7 or H-). Other serotypes causing infections have included O113:H21, O26:H-, O91:H21, O145:H-, ONT:H18, ONT:H6, and O128:H-.

Analysis of patient stool samples submitted to the Dunedin Hospital laboratory during 1996 identified two serotypes, O26:H11 and O128:H2, that were toxigenic and typable (Brooks *et al.*, 1997).

Further details of serotypes found in New Zealand are given in Section 6.1.3.

4.2.2   Overview of international situation

It has long been held that serotype O157 is the predominant cause of STEC related disease in the USA. However, some recent data indicate that there may be a re-thinking of this position. In a recent review of the impact of foodborne disease in the USA, Mead *et al.* (1999) estimated that illness attributable to non-O157 STEC was approximately 50% of that caused by *E. coli* O157:H7. If these estimates are correct then approximately 33% of STEC-related illness is caused by non-O157 serotypes in the USA, and this represents a major shift in the way this group of organisms is regarded.

A study from Canada (Rowe *et al.*, 1993) reported that of 30 isolates from HUS patients 26 were *E. coli* O157:H7 and four belonged to other serotypes (two of the isolates could not produce verotoxin and so may have not caused the disease, although expression of toxin can be lost on subculture). An earlier study in Alberta (Pai *et al*., 1988) of faecal samples submitted at hospitals for bacteriological examination found 130 patients infected with *E. coli* O157:H7, 29 with non-O157 STEC and seven with both.

Bitzan *et al.* (1991) demonstrated that 20 of 22 HUS patients in Germany had been infected with type O157, one with O26 and one with O55. This represents an approximate 10% of the cases being caused by non-O157 serotypes.

An Italian study into HUS cases (Luzzi *et al.*, 1995) revealed a somewhat higher proportion of non-O157 cases, with 45 cases having antibodies to O157, 12 to O111, 6 to O26 and 2 to O103 (30.8% non-O157), although the significance of antibodies to STEC remains controversial.   In Britain a similar proportion (28.3%) of non-O157 STEC has been recorded in children with HUS (Kleanthous *et al.*, 1990), although an earlier study had shown a smaller proportion, 21% (Scotland *et al.*, 1988).

In Belgium, only 18% of STEC strains were reported to belong to serotype O157:H7 (Pierard, 1992), and a French study reported isolating only O103:H2 from the faeces of six of 69 HUS patients, i.e. no other STEC were isolated (Mariani-Kurkdjian *et al.*, 1993). A more recent French study focused on children with HUS found that 86% of these cases had evidence of STEC infection. Of the HUS cases, 75% showed evidence of infection from *E. coli* O157, but other serotypes identified included O103, O126 and O26 by microbiological testing and, in addition, O9, O103 and O145 by serum antibody testing (Decludt *et al.,* 2000).

Caprioli *et al.* (1997) observed that during 1996 there was a sudden increase in the proportion of non-O157 isolations in Europe. In HUS cases from 1996 up to the time of publication 11% were caused by O103 and 33% by O26 compared to 1.5% and 6.6% respectively in previous years. This trend was describes as "worrisome" because of the lack of implementation of reliable methods for detecting these infections.

The pattern of transmission of sporadic STEC infection in continental Europe may be atypical because of the lack of an epidemiological link between STEC infection and beef products (Pierard *et al.*, 1999).

Tamura *et al.* (1996) reported on investigations of diarrhoeal specimens tested from Asian countries. Only 20.3% of the isolates typed were of serotype O157. The other serotypes identified were similar to those identified in other countries.

Australia has been known to be unusual in respect to STEC types isolated, as type O157 represents a low proportion of the isolates (Goldwater and Bettelheim, 1995), with type O111:H- being more common.

4.3   Dose Response

The survival of *E. coli* (one non-pathogenic strain and one enterohaemorrhagic strain) during passage through the stomachs of young and elderly people has been investigated using mathematical modeling, and a fermentor that mimicked the human gastric pH (Takumi *et al.*, 2000).   On average 20-80% of the ingested *E. coli* were estimated to arrive at the small intestine without inactivation by low pH.   This was attributed to the temporary increase in gastric pH after consumption of food, as well as acid tolerance of *E. coli*. To illustrate this last point, the *E. coli* O157:H7 isolate tested showed no decline in numbers after incubation for two hours at pH 2.5, and 26% of the cells survived when the pH was 2.0.

This acid tolerance and gastric survival will presumably be reflected in the dose response relationship.

4.3.1   Dose-response for *Escherichia coli* O157:H7

Haas *et al.* (2000) developed a dose-response relationship for *E. coli* O157:H7 based on a prior animal (rabbit) relationship. This model was validated by reference to two well documented human outbreaks; one involving water-borne organisms and the other involving venison jerky. The model gave a dose for infection of 50% of the exposed population of 5.9×10^5 organisms and a risk for consumption of 100 organisms of 2.6×10^{-4}.

Based on a retrospective analysis of foods involved in outbreaks, the capability of person-to-person transmission, and the ability of the pathogen to tolerate acidic conditions, which enables survival in the acidic environment of the stomach, Doyle *et al.* (1997) estimated the infectious dose of *E. coli* O157:H7 to be less than a few hundred cells. A similar estimate of infectious dose has been proposed by CAST (1994). However, the concept of an infectious dose has now been replaced by estimates of the probability of infection from exposure to differing numbers of cells.

Recently an estimate of the dose response for *E. coli* O157:H7 using a beta-Poisson model gives a value of 1.9 x 10^5 cells as the median dose (50% exposed become symptomatic), with a probability of 0.06 (6 x 10^{-2}) of infection when exposed to 100 cells (Powell *et al.*, 2001).

4.3.2   Dose-response for non-O157:H7 STECs

Haas *et al.* (1999) developed dose-response relationships for *E. coli* O111 and O55 using human volunteers. The relationship gave a dose for infection of 50% of the exposed population of 2.6 x 10^6 organisms and a risk for consumption of 100 organisms of 3.5 x 10^{-4}.

5   EXPOSURE ASSESSMENT

5.1   The Hazard in the New Zealand Food Supply: STEC in Red Meat and UCFM

5.1.1   STEC in meat animals: O157

Only one paper attempting to measure the prevalence of *E. coli* O157:H7 (specifically) in New Zealand livestock has been published. Buncic and Avery (1997) sampled the faeces of 371 cows originating from 55 farms on arrival at a single slaughterhouse in the Waikato area. Two (0.5%) of these samples were positive for the presence of *E. coli* O157:H7. A further 160 cattle from the farm of one of the positive animals tested negative for *E. coli* O157:H7.

5.1.2   STEC in meat animals: other serotypes

There are no reports of isolations of non-O157 STEC from the faeces of New Zealand farmed meat animals.

5.1.3   STEC in meat: O157

The New Zealand Ministry of Agriculture and Forestry has been monitoring meat products for the presence of *E. coli* O157 since 1998 (see Section 7.1.1.2). Baseline surveys of bovine (2400) and ovine (500) carcasses from meat processing plants did not detect any *E. coli* O157:H7. Records from the National Microbiological Database (NMD) indicate that to January 2003, from 144,023 cartons of bulk beef only two confirmed positives and one screen positive (unable to be confirmed or otherwise) for *E. coli* O157 were detected (0.002%; 95% confidence limits of 0.0004% to 0.018%) (Dr Roger Cook, NZFSA, personal communication). While these samples were all taken from export meat works their ultimate destination may have been either export or domestic consumption. Approximately 61% of total domestic beef supply originates from export meat works (Roger Cook, NZFSA, personal communication). The remaining 39% of domestic beef supply comes from local abattoirs and is not subjected to the *E. coli* O157 testing programme. The prevalence of *E. coli* O157:H7 in meat produced by local abattoirs is therefore not known.

No co-ordinated *E. coli* O157 testing programme is currently in place for ovine or porcine meat produced in New Zealand (Roger Cook, NZFSA, personal communication).

5.1.4   STEC in meat: other serotypes

In addition to the isolates from human cases, the ESR Enteric Reference Laboratory has been asked to serotype isolates from meat samples. The isolates derive from meat produced in New Zealand, but the details have not been made available for commercial reasons. Nevertheless, the serotypes have been reported (Bennett and Bettelheim, 2002). Seven isolates from New Zealand beef were typed, and found to belong to four different serotypes. From sheep meat 40 isolates were identified as including 18 different serotypes (although many of these were non-typable or rough). Seven of the meat isolate serotypes (one from beef and six from sheep meat) have also been associated with human disease. Three of the isolates (O5:H-, O91:H-, O104:H-) have been associated with cases of HUS, while the others (O6:H-, O104:H7, O128:H2, O163:H19) have been associated with diarrhoea only.

A study of the isolation of STEC using a specific agar examined 15 retail meat products (raw minced beef or pork, vacuum packed sliced meat and salami) (Hudson *et al.*, 2000). Four of the five minced beef or pork samples yielded presumptive STEC colonies, of which all but one were serotype O163:H19, which has not been involved in HUS cases. The remaining isolate was non-typable.

Brooks *et al.* (2001) examined beef (91 samples), mutton and lamb (37 samples), pork (35 samples), chicken (36 samples), mutton/beef mince (10 samples), and sausage mixtures (9 samples) obtained from Dunedin supermarkets and butcheries. STEC were isolated from 12% of beef, 17% of lamb, and 4% of pork retail raw meat samples (chicken samples tested were negative). Serotypes obtained were, from beef: O128:H2, ONT:H21, O144:H2, O27:H21, ONT:H-, O8:H-, O15:H27, O81:H26, from lamb: O91:H-, O171:H2, ONT:H4, O128:H-, O81:H26, O5:H-, from pork: O156:H-, and from beef and lamb mince O15:H27.

All isolates were tested for the presence of factors associated with virulence i.e. Stx1, Stx2, Ehly (i.e. *hlyA*) and *eaeA*. All were positive for Stx1 and/or Stx2, and five (ONT:H21, O128:H2, O144:H2, O81:H26, O5:H-) were positive for Ehly. None were positive for the *eaeA* factor.

Serotypes O5:H-, O128:H-, O128:H2, and O91:H-, have been reported to be involved in diarrhoea and HUS cases, but not outbreaks, overseas. Serotype O128:H- has caused a case of STEC infection in New Zealand.

Work carried out by ESR for the Ministry of Health during 2000-2001 tested 97 beef, 65 chicken, 66 lamb and 73 pork samples of minced or cubed meat for all STEC. Only five lamb samples (7.6%) were positive. The isolates obtained were; ONT:H-, ONT:H8, O123:H51, O rough:H-, O75:H8, and O128:H2 (ONT = non-typable from O antigen, O rough = agglutinates with all sera). The prevalences found in this study were low, but the method used was focused on obtaining isolates that could be typed. Other methods such as PCR detection might give higher prevalences but need not necessarily have yielded any isolates for serotyping.

5.1.5   The Hazard in the New Zealand Food Supply: STEC in UCFM Products

No data for STEC in UCFM in the New Zealand domestic food supply have been located.

5.1.6   Conclusion

The main conclusion that can be drawn from these data is that while STEC can be isolated from raw meats in New Zealand, only two of the serotypes (O128:H-, O128:H2) isolated from meat correspond to serotypes that have been associated with illness in humans living in New Zealand. Serotype O128:H- has been associated with a notified case of STEC infection, while serotype O128:H2 was from a patient suffering from diarrhoea. Despite these serotypes having been isolated from New Zealand beef and lamb (see Section 5.1.4), there is no evidence to associate meat consumption with transmission of infection for these cases.

Internationally the most common serotypes causing outbreaks or clusters of serious infections (i.e. HUS, TTP) are; O26:H11, O111:H-, O113:H21 and O157. Many STEC serotypes are

infrequently associated with disease or have yet to be associated with disease. By far the majority of isolates from New Zealand meat fall into these two categories.

Isolation rates of *E. coli* O157 from raw meats from export meat processing plants in New Zealand appear to be particularly low. These meat processing plants represent the majority (61%) of the domestic meat supply.

5.2 Food Consumption: Uncooked Comminuted Fermented Meat Products

Analysis of 24 hour dietary recall records from the 1997 National Nutrition Survey (NNS; Russell *et al.*, 1999) only identified one descriptor (salami) which could be used to describe an UCFM product. It should be noted that not all products described as 'salami' will be UCFM, as some cooked and unfermented products are commonly referred to as salami.

Only 73 NNS respondents (1.6%) reported consuming salami in the previous 24 hour period, mainly as a component of sandwiches, filled rolls or bagels. Salami will be a common component of pizza, a more commonly reported food in the 1997 NNS, however, the thermal processing which salami will receive during pizza manufacture means it is not relevant to the current risk profile.

The mean serving size of uncooked salami consumed by the 73 respondents was 33.4 g, while the median serving size was 15.5 g, with a range from 2 to 276 g. The mean daily consumption of uncooked salami by the whole population would be approximately 0.5 g/day.

The New Zealand Pork Industry Board (NZPIB) has provided an estimate of total UCFM annual production in New Zealand at 343,367 kg. Based on a total population of 3,737,490 (2001 Census) this equates to 0.25 g/person/day. The difference between this estimate of consumption and that in the previous paragraph may be due to a range of factors. The higher figure (0.5 g/person/day) may include some imported product and/or product which has been cooked during the production process.

5.3 Qualitative Estimate of Exposure

5.3.1 Number of servings and serving sizes

UCFM is a minor component of the New Zealand diet. Salami (uncooked) makes up less than two percent of food consumption, on the basis of servings and only 0.5% of food consumption on a weight basis. It should be reiterated that these figures represent upper bound estimates of UCFM consumption, as not all salami will be UCFM.

Median serving size for uncooked salami is 15.5 g, while the 75, 95 and 99th percentile serving sizes are 30, 114 and 275 g respectively.

5.3.2   Frequency of contamination

Given that no data are available for New Zealand, overseas data are all that can be used to evaluate this. Those data would suggest that contamination of UCFM products is extremely rare.

5.3.3   Predicted contamination level at retail

Given that the prevalence is rare, then the level of contamination on these comminuted products that are contaminated is likely to be low.

5.3.4   Growth rate during storage and most likely storage time

Even if the UCFM products were contaminated during manufacture or distribution, the pH and water activity of UCFM products should be such that growth should not occur. The main factor that is relevant is how long *E. coli* O157 would survive on the products. If errors during manufacture were so great that a sufficiently low pH was not achieved then this may not apply.

5.3.5   Heat treatment

Not relevant to UCFM products. Some salami-like products may be heat treated, but they are not considered here, as they are essentially a cooked meat product. A proportion of UCFM products will be consumed after cooking (e.g. on pizza) but these were not able to be separated in this consumption analysis.

5.3.6   Exposure summary

From overseas data the exposure of the population to *E. coli* O157 and other STEC serotypes from UCFM consumption is likely to be very low. The food is a small component of the New Zealand diet.

5.4 Overseas Context

5.4.1 STEC in UCFM Products: O157

Information summarising data for the prevalence of *E. coli* O157 in UCFM products is given in Table 3.

Table 3 **Prevalence of *Escherichia coli* O157 in UCFM products from overseas surveys**

Country	Products tested	Number tested	% positive	Reference
Argentina	Dry sausage	30	3.3	Chinen *et al.*, 2001
England, Wales and Northern Ireland	Dried and fermented meat and meat products	2,981	0	Little *et al.*, 1998
Holland	Cooked or fermented RTE meats	328	0.3	Heuvelink *et al.*, 1999
USA	Dry and semi dry fermented sausages	3,445	0	Levine *et al.*, 2001

In general, reported contamination rates for UCFM are low.

6 RISK CHARACTERISATION

The public health significance of infection with STEC derives from the high proportion of cases which have serious consequences, following gastrointestinal disease. Infection with STEC can affect any age group but most often causes disease in infants (< 4 years) and the elderly (>65 years).

These consequences, HC, HUS, and TTP, are described in section 4.1. Children under five years are most susceptible to HUS whereas the elderly are more likely to develop TTP (Baker *et al.*, 1999).

6.1 Adverse Health Effects in New Zealand

6.1.1 Incidence and outbreaks

The first New Zealand case of infection with STEC was detected in 1993, and the illness was made a notifiable disease in June 1996. The number of cases of infection with STEC in New Zealand increased steadily throughout the late 1990s. The rates are shown in Table 4. The trend over the period 1993-2001 is also shown in Figure 2.

Table 4:   Rates of infection with STEC in New Zealand 1998 – 2001

Year	Rate per 100,000 (number of cases)	Reference
1998	1.3 (48)	Baker *et al.*, 1999
1999	1.8 (64)	Kieft *et al.*, 2000
2000	1.9 (68)	Lopez *et al.*, 2001
2001	2.0 (76)	Sneyd *et al.*, 2002

Figure 2:   STEC notifications by year, 1993 –2001

Notified cases of VTEC / STEC infection by year, 1993 - 2001
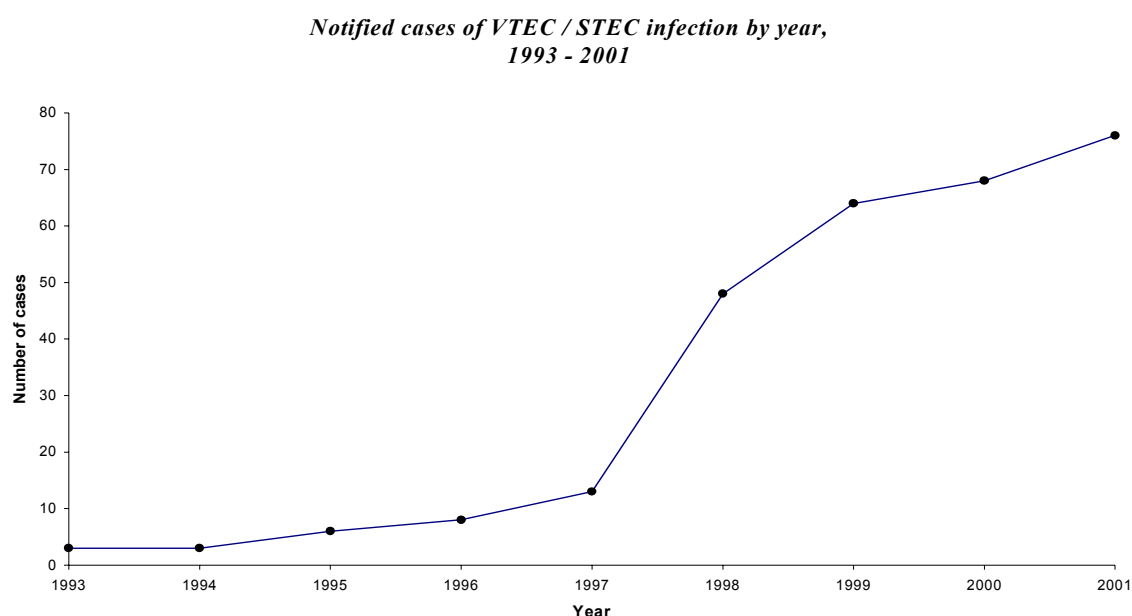
An analysis of data up to 1998 showed that rates in children are much higher than for adults. The highest rates of STEC infection in 1998 were in children aged less than one year (14.6 per 100,000) and those aged 1-4 years (10.2 per 100,000). Rates for males and females (1.1 and 1.5 per 100,000 respectively) were similar, as were rates in Europeans and Pacific Islanders (1.4 and 1.2 per 100,000). Rates were lower in Maori (0.4 per 100,000) (Baker *et al.*, 1999).

Small numbers of outbreaks, involving relatively low numbers of cases, have been reported to the national surveillance system each year since 1998, with the highest number being in 1998 (8 outbreaks, 20 cases). These events are probably better described as clusters. In 2001 there were 4 outbreaks reported, involving 10 cases (Sneyd *et al.*, 2002).

Note that these rates are for <u>all</u> STEC and serotype O157 (H7 and H-) accounts for around 90% of the notified cases. In 2001 the Enteric Reference Laboratory at ESR received a total of 75 isolates, of which 73 were O157 (Sneyd *et al.*, 2002).

Based on studies in Canada, it is possible that in New Zealand 10-12 unreported cases of STEC infection occur for each reported case (Baker *et al.*, 1999). These cases are likely to have milder symptoms than reported cases.

6.1.2   Clinical consequences of STEC infection

The clinical consequences of STEC infection of cases in New Zealand are summarised in Table 5.

Table 5:   Summary of clinical consequences of STEC infection in New Zealand

Period	Hospitalised*	HC*	HUS*	TTP*	Fatalities
Oct 1993-Dec 1998[1]	24/58 (41.4%)	21/59 (35.6%)	18/59 (30.5%)	1/59 (1.7%)	2/79 (2.5%)
1999[2]	20/60 (33%)	NR	2/64 (3.1%)	NR	0
2000[3]	11/65 (16.9%)	NR	3/68 (4.4%)	NR	0
2001[4]	16/74 (21.6%)	NR	6/76 (7.9%)	NR	0

1 Baker *et al.*, 1999
2 Kieft *et al.*, 2000
3 Lopez *et al.*, 2001
4 Sneyd *et al.*, 2002

*Percentages are determined on the basis of cases for which information was available
NR Not reported

6.1.3   Serotypes causing disease

Approximately 90% of notified STEC infections in New Zealand are caused by *E. coli* O157. Other serotypes causing infections have included O113:H21, O26:H-, O91:H21, O145:H-, ONT:H18, ONT:H6, and O128:H-. Some serotypes causing infection in New Zealand are ONT:H- i.e. have not been able to be classified There have been two deaths attributed to

STEC (both prior to 1999), one to serotype O157 and the other to O113:H21 (Carolyn Nicol, ESR Enteric Reference Laboratory, personal communication).

Stool specimens (484) from children suffering from diarrhoea submitted to laboratories at Dunedin Public Hospital were examined in a study in 1996 (Brooks *et al.,* 1997). Sixteen cultures were identified as *E. coli* cytotoxic to Vero cells, but only serotypes O26:H11, which is capable of causing HUS outbreaks, and O128:H2, were toxigenic and typable. Retrospective analysis of five of these STEC showed that the O26:H11 isolate was positive for the Stx1, *hlyA* and *eaeA* factors, while the others (O128:H2, OR:H2, OR:H-) were positive for Stx1 and Stx2 but not the other factors (Brooks *et al.*, 2001).

The serotypes O91:H-, O128:H2 and O128:H- have recently been isolated from New Zealand retail meat samples (Brooks *et al.*, 2001). The latter serotype has also been isolated from a notified human case (see above). Serotype O128:H2 is another that has been isolated from both meat and a person suffering from diarrhoea, although this was not a notified case of STEC infection.

6.1.4   Case control studies and risk factors

There have been no New Zealand case control studies to identify risk factors. The overview of 79 cases of STEC in New Zealand reported that in 1998 there were four household clusters including 9 cases, of which four were classified as caused by secondary transmission. Over the six year period 1993 to 1998 six cases reported living on a farm or visiting a farm regularly. Consumption of unpasteurised milk was reported by eight cases (Baker *et al.*, 1999).

An analysis of risk factors associated with STEC infection for cases from June – December 1999 was given in the Annual Surveillance Summary (Kieft *et al.*, 2000). A high (>50%) proportion of cases reported consumption of beef, poultry, processed meats, and raw fruit and vegetables, but not consumption of pink/undercooked meat. Animal contact was another common factor. A further collation of risk factor information was reported for 2001 (Sneyd *et al.*, 2002). Again there were high (>50%) proportions of cases who reported consumption of dairy products, beef, poultry, processed meats, raw fruit/vegetables, contact with animals and contact with raw meat/offal. However, both analyses cautioned that these are common factors in New Zealanders lives and the proportions may simply reflect that fact, and the number of cases was too low to draw meaningful conclusions.

There have been a few episodes where an indistinguishable STEC bacterial strain has been isolated from both a human case and a potential source in New Zealand. Contaminated untreated drinking water (one spring and one roof supply) was linked to two episodes of infection, affecting a total of three people in 1999, and one case has been attributed to contact with a calf (Anonymous, 2000). For one case in New Zealand, an indistinguishable isolate was obtained from both the infected person and raw milk present in the home, although the route of infection is uncertain (Anonymous, 2002).

To date, there are no surveillance data to link STEC transmission with meat products, including UCFM, in New Zealand.

6.2 Adverse Health Effects Overseas

6.2.1 Incidence

Incidence rates for a selection of countries/states are given in Table 6. Note that there is a mix of reporting of STEC cases in general on some occasions, and all STEC infections in others. New Zealand's incidence has been included for comparison and is on a par with other countries.

Table 6: New Zealand and international rates of reported infections with STEC

Country	Year	Incidence (per 100,000)	Reference
New Zealand	**2001**	**2.0**	**Sneyd _et al._, 2002**
Australia	2001 (Jan-Mar)	0.4	Communicable Diseases Australia[1]
Canada	1996	4.1	Williams _et al._, 2000
England and Wales	1990-1998	<2	Pennington, 2000
Finland	1996-1997	0.16	Keskimakii and Siitonen, 1997
Scotland	1990-1998	2-10 (approx.)	Pennington, 2000
USA	1998	2.8	http://www.cdc.gov/mmwr/preview/ mmwrhtml/mm5013a1.htm
USA	1999	2.1	http://www.cdc.gov/mmwr/preview/ mmwrhtml/mm5013a1.htm
USA	2000	2.1	http://www.cdc.gov/mmwr/preview/ mmwrhtml/mm5013a1.htm

[1] http://www.health.gov.au/pubhlth/cdi/cdi2502/pdf/cdi2502o.pdf

The proportion of STEC infected cases hospitalised in the United States has been estimated as 29.5%, with 0.8% of cases resulting in death (Mead _et al._, 1999). Although New Zealand's hospitalisation and fatality rates to the end of 1998 were higher than this, more recent data show rates closer to overseas values (see Table 5).

HUS has been estimated to occur in approximately 4% of cases (Mead _et al._, 1999). HUS is the most common cause of acute renal failure in children. Mortality is approximately 5% and approximately 10% of survivors are left with severe sequelae (Park _et al._, 1999).

For the US it has been estimated that for each confirmed case of infection with STEC that is reported, 13-27 cases of _E. coli_ O157 infection occur in the community (Mead _et al._, 1999). These cases have mild symptoms. The total number of cases of infection with non-O157 STEC has been assumed to be 50% of the rate for O157:H7 (Mead _et al._, 1999).

6.2.2 Contributions to outbreaks and incidents

The proportion of outbreaks caused by _E. coli_ O157 overseas is summarised in Table 7. Only a small proportion of outbreaks are attributable to STEC.

Table 7: **Proportions of outbreaks and incidents caused by *Escherichia coli* O157 in overseas countries**

Country	Year	Proportion of outbreaks (%)	Reference
Canada	1982	0.2	Todd, 1992
Canada	1983	0.2	Todd, 1992
Canada	1984	0.1	Todd, 1992
England and Wales	1992-1994	1	Djuretic *et al.*, 1996
England and Wales	1995	1	Evans *et al.*, 1998
England and Wales	1996	1.4	Evans *et al.*, 1998
Sweden	1992-1997	<1	Lindqvist *et al.*, 2000

The Food Safety and Inspection Service (FSIS) of the USDA risk assessment for *E. coli* O157:H7 in ground beef summarised information from 154 *E. coli* O157:H7 outbreaks during the period 1982-1997 (FSIS, 1998). Ground beef was identified as the likely vehicle for infection in 25% of outbreaks, while whole cuts were identified with only 2% of outbreaks and salami with less than one percent.

6.2.3 <u>Specific incidents and case control studies linking STEC infection with UCFM</u>

Meat and meat products are often associated with STEC outbreaks and incidents overseas, however, a much smaller number of incidents have been linked to consumption of UCFM products. These are summarised for *E. coli* O157 and other STEC in Table 8.

Table 8: **Specific incidents of disease reported for STEC associated with UCFM products**

Location	Serotype	No. affected	No. deaths	Source	Reference
Australia	O111:H-	23 HUS (one death), 30 bloody diarrhoea, 3 adults with TTP, 105 other GI symptoms	1	Mettwurst	CDC, 1995b
Canada	O157:H7	39	NS	Genoa Salami	Williams *et al.*, 2000
Canada	O157:H7	5 HUS; 150 ill	0	Hungarian style sausage	Health Canada, 2000
Germany	O157:H- (sorbitol +ve)	28 children with HUS, estimated 300-600 other persons	3	Mortadella and teewurst[*]	Ammon *et al.*, 1999
USA	O157:H7	23 (including 6 hospitalised and 2 HUS)	0	Dry-cured salami	CDC, 1995a

NS=Not Stated. [*] Mortadella is a cooked product, but teewurst is substantially fermented and dried. The latter product was considered to be the likely vehicle in this outbreak.

A dry-cured salami was the source of infection for 20 laboratory confirmed cases of STEC caused diarrhoea reported in Washington in late 1994 (CDC, 1995a). These cases were subsequently linked with three more in California. A case control study involving 16 cases linked the cases with salami, and salami samples taken from the grocery chain involved were found to contain isolates indistinguishable by Pulsed Field Gel Electrophoresis and Restriction Fragment Length Polymorphism analysis from isolates from the cases.

A subsequent study investigated several hypotheses for the occurrence of the bacteria in this product by conducting evaluations of the production facility (Tilden *et al.*, 1996). The hypotheses were:

- Organisms present on raw meat ingredients survived a substandard fermentation and drying process;
- Organisms survived a fermentation and drying process that met existing industry and regulatory standards; or,
- Contamination occurred after fermentation and drying, either as a result of the slicing process or as a result of subsequent handling.

The investigation found that the methods used to produce the salami were typical of those used in the plant throughout 1994, and were typical of industry-wide methods used to produce Italian style salami. Record keeping was good, as the plant participated in the USDA FSIS Total Quality Control Program, and these records indicated that the methods complied with existing regulations and recommended good manufacturing practices. The recovery of *E. coli* O157:H7 from intact packages indicated that contamination did not occur at retail level. Extensive environmental and product testing failed to find *E. coli* O157:H7 and it was concluded that the most likely hypothesis was that a limited amount of contaminated meat was introduced into the plant.

The 1998 Canadian outbreak was investigated by a case-control study that identified Genoa salami from amongst a range of foods (Williams *et al.*, 2000). Although *E. coli* O157:H7 was not detected in any of the original food samples taken from retailers or the homes of cases, it was detected in samples taken from a single plant. The phage typing and pulsed field gel electrophoresis patterns of these isolates were indistinguishable from those for the isolates obtained from cases. An investigation of the plant by the Canadian Food Inspection Agency revealed significant problems with the manufacturing process: the use of natural fermentation (i.e. instead of a starter culture), poor record keeping, no microbiological tests of incoming raw ingredients or final products, no records of lot specific pH or degree-hours measurements, faulty pH measurement methods and no written procedures for how to manage products with abnormal results.

In this investigation it was considered possible that some cases, who ate deli sliced products but who did not recall eating Genoa salami, were in fact infected by cross contamination via the slicer.

A further outbreak in November 1999 in Western Canada involved 150 people who became ill and 5 developed HUS. The outbreak was traced to a type of raw, fermented, Hungarian-style sausage. In the investigation that followed this outbreak it became clear that the interventions recommended by Health Canada to improve the safety of UCFM products were

not being followed in some establishments, and regulatory action was instituted (Health Canada, 2000).

The outbreak in South Australia in December 1994 – January 1995 was linked to uncooked semi-dry fermented sausage product, mettwurst, from a single manufacturer by a preliminary investigation of cases (CDC, 1995b). Of ten sausage samples taken from the homes of patients, eight (all from the same manufacturer) were positive for the Shiga toxin producing genes (detected using polymerase chain reaction) and *E. coli* O111:H- was isolated from four of these samples. During the inquest into the death of the 4 year old child with HUS it was reported that the manufacturing company had been implicated in prior foodborne outbreaks due to *Salmonella* spp. (Desmarchelier, 1997). The product was manufactured from a combination of frozen mutton, pork back fat and fresh beef and mutton, any of which may have been a source of contamination. A starter culture was not used in the fermentation, no monitoring of pH or water activity was evident, and no pasteurisation process was employed.

The outbreak in Germany began in late 1995 and was identified from an increase in the number of children suffering from HUS that were reporting to pediatric haemodialysis centres (Ammon *et al.*, 1999). On the basis that 5-10% of people with EHEC infections develop HUS, it was estimated that 300-600 people may have been infected. Two case-control studies were conducted to determine the cause; the first gathered information on a variety of potential risk factors, while the second gathered detailed information about consumption of meat products. Of the sliced sausage products eaten by cases, only mortadella and teewurst were more likely to have been eaten by cases than controls. Mortadella was considered less likely to be the source of the infection as it is heated during production; however, post heating contamination could not be ruled out. No investigation of the manufacturer was reported.

6.2.4   Risk assessments and other activity overseas

No risk assessments that specifically address UCFM have been located. The USDA FSIS Risk Assessment for STEC only considered ground beef.

6.2.5   Secondary transmission

Secondary transmission of STEC infection is a significant cause of cases. In a large beefburger-associated outbreak in the USA 11% of the identified cases were secondary. A study in Wales between 1994 and 1996 indicated that 11% of cases were secondary, while the household transmission rate was estimated at 7% (summarised in Parry and Palmer, 2000).

6.3   Qualitative Estimate of Risk

Data on the prevalence of STEC in beef, sheep and pork meat in New Zealand are limited and the existing reports have found only non-O157 serotypes. Many of these isolates lack the full complement of virulence factors, although some have been associated with cases of serious disease (HUS) overseas (Brooks *et al.*, 2001; Bennett and Bettelheim, 2002). Two serotypes isolated from meat have also been isolated from ill people in New Zealand, but these is no information to indicate a source for these infections.

The prevalence of the dominant disease causing serotype, O157, appears to be extremely low (0.002%) in New Zealand raw meat samples from exporting plants, compared with up to 2% reported for surveys overseas (see data summarised in the "STEC in red meat" Risk Profile). This may be due to effective control in meat processing facilities, as the data from a limited study for the presence of the organism in faeces from cows indicate a prevalence of 0.5%.

Only small amounts of beef and sheep meat (approximately 5% of the total supply) are imported, principally from Australia. Approximately 30% of the pigmeat supply in New Zealand is imported from Australia and Canada.

Little information on transmission is available from the analysis of cases in New Zealand between 1993 and 1998 (Baker *et al.*, 1999) or subsequent surveillance summaries. Information on potential sources of infection is available for a few cases of STEC infection, but meat has not been implicated as a transmission vehicle.

It was concluded by the Risk Profile on STEC in red meat that there is currently little information to suggest that transmission of STEC via red meat is occurring in New Zealand. There are similarly no data to link UCFM with transmission of STEC in New Zealand. The acquisition of data on the prevalence and numbers of STEC in meat and meat products at retail would assist in reinforcing these conclusions.

6.4 Risk Categorisation

The rationale for categorisation of food/hazard combinations is presented in Appendix 1.

The proportion of severe outcomes (hospitalisation, long term sequelae, and death) resulting from STEC infection in New Zealand is approximately 10% (Lake *et al.*, 2000) placing this infection in the highest severity category.

For the purposes of estimating the numbers of cases of foodborne disease in New Zealand (Lake *et al.*, 2000) it was assumed that 20% of STEC infections were due to foodborne transmission. The total rate of STEC infection (including unreported cases) attributable to food contamination in New Zealand was thus estimated to be of the order of 1.4 per 100,000 of population.

However, there is currently no evidence linking red meat consumption in general or UCFM consumption in particular to cases of STEC infection in New Zealand. The prevalence of the dominant pathogenic serotype (O157) in red meat in New Zealand is low by international standards.

Thus the rate of STEC infection due to transmission in UCFM will be considerably less than 1 per 100,000 of population. This places STEC in UCFM in the lowest incidence category.

6.5   Summary

Food/hazard combination	Severity	Incidence	Trade importance	Other considerations
STEC in uncooked comminuted fermented meats	1 (>5% serious outcomes)	4 (<1 per 100,000)		

7 RISK MANAGEMENT INFORMATION

7.1 Relevant Food Controls: New Zealand

7.1.1 Meat processing

The United States (New Zealand's largest beef market) requires that HACCP plans are in place in processing plants, and countries in the European Union also require a partial application of HACCP principles.

Currently New Zealand meat processing plants are registered under the Meat Act 1981. The Meat Regulations 1969, Game Regulations 1975, and subsidiary Industry Standards and Technical Directives apply.

This legal situation is changing with the introduction of the Animal Products Act.

7.1.1.1 The Animal Products Act

The Animal Products Act 1999 reforms the New Zealand law that regulates the production and processing of animal material and animal products to:

- manage associated risks; and
- facilitate overseas market access.

The Animal Products Act requires all animal products traded and used to be "fit for intended purpose". This means they must meet New Zealand animal product standards. The New Zealand animal product standards are contained in Part 1 of the Animal Product Regulations 2000.

The Animal Products Act (except for Part 2) and the transitional Act commenced on 1 November 1999. Part 2 of the Animal Products Act commenced on 20 November 2000. Part 2 provides the requirements for risk management programmes.

The risk management system potentially applies anywhere in the value chain from production, through processing to the market. The risk management system comprises the following main types of controls:

- risk management programmes;
- regulated control schemes; and
- controls relating to the export of animal material and animal products.

All animal product primary processing businesses, except those exempt under the Act or under the Animal Products (Exemptions and Inclusions) Order 2000, must have a risk management programme. The transition time to the Animal Products Act (previously to 1 November 2002), has been extended during a phase-in period from July 2003 to July 2006. Red meat processors, export seafood processors and packing houses will have until July 2003 to have their programmes registered, and other processors will be phased in over the following three years.

A risk management programme is a documented programme to identify and manage biological, chemical and physical hazards. The programme is to be based on the principles of Hazard Analysis and Critical Control Point (HACCP): identifying the hazards, the systems of control, and demonstrating that the controls are effective. Risk management programmes are to be designed by individual businesses for the animal materials used, the processes performed and the product range produced.

7.1.1.2 Monitoring compliance with standards

All US listed beef and sheep slaughter premises and packinghouses in New Zealand participate in a mandatory microbiological monitoring programme. The results are collated by the National Microbiological Database (NMD), which is operated by the NZFSA. The rationale for the scheme was to demonstrate the equivalence of New Zealand's food safety controls to those of other countries, in particular the "US Pathogen Reduction; Hazard Analysis and Critical Control Point (HACCP) Systems" rule. Procedures are defined for sampling and analysis for aerobic plate count, *E. coli* and *Salmonella*. Samples are collected from boxed manufacturing beef at the end of the production system, immediately prior to freezing. (Source:
http://www.maf.govt.nz/animalproducts/publications/manualsguides/nmd/index.htm).

In 1998 MAF assessed the needs of New Zealand beef exporters with respect to the food safety and due diligence requirements of the USDA Food Safety and Inspection Service with respect to *E. coli* O157. It was concluded that the New Zealand meat hygiene programme along with a national testing regime for *E. coli* O157 as part of the NMD satisfied the US requirements for a "validated pathogen reduction intervention on beef carcasses". Such a testing programme was then developed by the New Zealand Meat Industry Standards Council, and industry results are provided to the NMD. Provision of results to the NMD is not mandatory, but is highly recommended. While this testing regime is not mandatory, contributions are currently received from 100% of export processing plants (Roger Cook, NZFSA, personal communication).

7.1.2   Raw comminuted meat/salami processing

Food Standards Australia New Zealand (FSANZ) is in the process of developing a Standard for the production of UCFM products (see below). However, the proposed Standard will only apply to Australia. New Zealand considers this issue to be outside the scope of the Joint Food Standards System and is developing its own requirements.

In the late 1990s the New Zealand Pork Industry Board led the development of an industry wide standard, the Pork Quality Improvement Process (PQIP), which applied the HACCP approach to the New Zealand pork production process. This programme included requirements for UCFM of a pH of less than 4.6 and water activity less than 0.9. The programme also stated that "For fermented products acidification must not allow cross-contamination between batches. Back slopping is forbidden." An appendix to the PQIP programme dealt with *E. coli* O157:H7 in fermented sausages (Buncic, 1995). It described the effects of the various antimicrobial factors in such products, and stated that "In non-pasteurised, fermented sausages *E. coli* O157:H7 is not expected to grow, but can survive the

production process and subsequent storage. Therefore, *E. coli* O157:H7 free uncooked sausages can be obtained only if raw material is assured to be *E. coli* O157:H7 free."

In 1999 a survey of the manufacturing practices of some of the larger salami processors in New Zealand was conducted by ESR on behalf of the Ministry of Health with the support of the Pork Industry Board (Hasell, 2000). Five processors, representing a large proportion of the salami and UCFM production in New Zealand were interviewed or visited. None of the companies were testing raw meat, or batter for *E. coli* as identified in the FSANZ Code, and none had verified their process in terms of *E. coli* reduction. Overall, in 1999, the move towards HACCP-based food safety programmes amongst UCFM manufacturers in New Zealand was described as "only just beginning".

The report also considered the requirements and advisory guidelines developed by FSANZ for Australia as to their appropriateness for New Zealand. At that time FSANZ (then ANZFA) were proposing that UCFM production processes should achieve a 3D (1000 fold) reduction in *E. coli*. The comment was made that if the FSANZ code was followed correctly then the product must pose minimal risk. However, it was crucial to know which CCPs were required to obtain the 3D reduction, and the wide variety of products means that each manufacturing plant would need its own discrete HACCP programme to achieve the 3D reduction. An alternative to exhaustive testing of raw meat for the presence of *E. coli* was suggested: that companies producing salami source their raw meat from suppliers with HACCP-based food safety programmes in place. This was considered likely to result in a more enduring control on the quality of the raw material than testing alone, and mitigated against the adoption of the FSANZ code (Hasell, 2000).

The report concluded that the UCFM industry in New Zealand should be supported in its initiative to develop generic food safety programmes (in collaboration with MAF), and that once these were available the Ministry of Health should consider making such programmes compulsory. It was also recommended that the Ministry of Health provide industry with guidance as to the outcomes and performance outcomes that need to be achieved by a food safety programme.

7.1.3 Current status of HACCP based food safety programmes in the New Zealand UCFM industry

The PQIP programme contains a number of modules for different sectors of the pork industry. The module for Pork Processors (PQIP07) has been approved as a Code of Practice by the Ministry of Health (now New Zealand Food Safety Authority) for all processed pork products, except for UCFM.

The UCFM section of PQIP07 has been largely finalised in terms of water activity and other controls. The HACCP plan described in the UCFM section has been implemented by the major New Zealand UCFM manufacturer, Mainland. The main outstanding issue for the UCFM section is process validation. Industry and regulators are working towards the establishment of appropriate and acceptable process controls and microbiological criteria suitable for the control of pathogens in UCFM.

Issues being discussed are:

- Appropriate indicator organisms for challenge testing;
- How validation of processes will be performed;
- The value of computer modeling of processes to assess pathogen control; and,
- The standard which will have to be achieved by the process.

Towards this, a challenge protocol is being developed by Dr. Tecklok Wong (formerly with Envirolink, now with ESR) Envirolink together with Verkerks, another UCFM manufacturer. The earlier generic *E. coli* protocol has been updated to use non-pathogenic acid tolerant *E. coli*. Trials using this protocol are underway at Mainland (Rob Scott, Mainland, personal communication).

7.2   Relevant Food Controls: Overseas

The outbreaks that have occurred in the US, Canada, and Australia have prompted a variety of regulatory and industry responses for controls on UCFM production.

7.2.1   United States

In 1993 beef producers (National Cattlemen's Beef Association) in the United States established a Blue Ribbon Task Force to address the *E. coli* O157:H7 problem. One of their reports addressed UCFM products (Nickelson *et al.*, 1996). Using preliminary information from this report and other data, in 1995 the USDA issued a Directive to fermented meat processors which offered the following options for the control of *E. coli* O157:H7 (Bacus, 1997).

- Use a heat processing step (e.g. 63°C for 4 minutes)
- Include a validated 5-D inactivation treatment
- Conduct a "hold and test" programme for finished product
- Propose other processes to assure at least a 5-D inactivation
- Initiate a HACCP system that includes raw batter testing and a validated 2-D inactivation

All of the above options were also required to address the control of *Salmonella*, *Trichinella*, and *Staphylococcus aureus*.

In February 2001 a proposed rule for the federal meat and poultry inspection regulations which covered ready-to-eat meats and partially heat-treated meat and poultry products was published in the federal register (see: http://www.fsis.usda.gov/oppde/rdad/frpubs/97-013p.htm). Draft compliance guidelines were issued at the same time, and discussed the following options, in relation to fermented products:

See: http://www.fsis.usda.gov/oppde/rdad/frpubs/97-013p/rteguide.pdf

1. Heating processes to achieve a 6.5-$\log_{10}$ reduction of *Salmonella* and 5-$\log_{10}$ reduction of *E. coli* O157:H7 (formerly Option 1)

2. Processes to achieve a 5-$\log_{10}$ reduction or equivalent reduction of *E. coli* O157:H7 only (does not apply to *Salmonella*)

 a) Validated processes for a 5-$\log_{10}$ reduction of *E. coli* O157:H7 (formerly Option 2) – adapted from Table 6 of the Blue Ribbon Task Force report
 b) No *E. coli* O157:H7 detected in raw product and a validated 2D, 2-log reduction, process (formerly Option 5)

3. Fermentation processes that achieve at least a 5-$\log_{10}$ reduction of *E. coli* O157:H7 in specific products

The basis for the 5 $\log_{10}$ reduction of *E. coli* is derived from a hypothetical, worst case raw product of 10^4 CFU/g *E. coli* O157:H7 on red meat. The worst case raw product was deduced by the US National Advisory Committee for Microbiological Criteria for Foods (NACMCF) using data from the risk assessment of *E. coli* O157:H7 in ground beef performed by the USDA Food Safety and Inspection Service Office of Public Health and Science.

The option of a 2 $\log_{10}$ reduction is presumably predicated on sampling of raw ingredients to demonstrate the absence of *E. coli* O157:H7 and provides a safety margin over this.

7.2.2 Canada

In December 1996 and again in September 1998 the Canadian Food Inspection Agency notified UCFM manufacturers of the five interventions developed by the United States agencies and recommended that establishments adopt one of these. However, following the second outbreak of *E. coli* O157:H7 linked with UCFM, Health Canada decided to institute regulatory action. Interim guidelines using the five options were issued in February 2000, along with a challenge protocol to evaluate the process in terms of its ability to control *E. coli* O157:H7 (Health Canada, 2000). Following appropriate consultation with industry and consumer groups the guidelines were intended to be developed into a regulation.

It is also worth noting that a survey in Toronto in 1994-1995 found that non-heat treated salami and sausage products made on-site at small deli-type establishments were more likely to be left constantly unrefrigerated, and have a higher pH and water activity than products made at larger commercial plants (Lee and Styliadis, 1996). This suggests that smaller processors of UCFM may need special attention for the introduction of controls.

7.2.3 Australia

In 1995, as an emergency response to the Australian outbreak from mettwurst, FSANZ (then ANZFA) amended Standard C1 60A of the National Food Standards Code to incorporate additional requirements for UCFM products. In addition, an Advisory Guide for making UCFM products was published in 1996.

The additional requirements in the Advisory Guide were (Bacus, 1997):

- That the finished product must be free of *E. coli* in 0.1g of the food (specified method);
- That the number of *E. coli* in raw meat ingredients and finished product must be "monitored and recorded"; and,
- That processing must allow for a reduction in the numbers of *E. coli* that may have been present by "99.9% or 1000 fold".
- Alternatively the product must be cooked at 65°C for at least 10 minutes.

The use of starter cultures was also mandatory, and the transport and storage of raw meat was required to occur below 5°C. Additional requirements were stipulated for the control of coagulase positive staphylococci and *Salmonella*.

The basis for the choice of a 3D or (1000 fold) reduction appears to be the limit specified in the Advisory Guide that raw ingredients should be less than 100 CFU/g generic *E. coli*. Thus, a 1000-fold reduction, together with the relatively low prevalence and concentration of EHEC on meats used in UCFM manufacture, is deemed adequate to protect public health (Ross and Shadbolt, 2001).

In March 2002, FSANZ (then ANZFA) initiated a proposal to review the Food Standards Code with regard to UCFM production. This will apply in Australia only, as New Zealand considers this issue to be outside of the Joint Standards System. The review took note of a report commissioned by Meat and Livestock Australia (Ross and Shadbolt, 2001) which concluded that:

- Many UCFM processes currently used, either in Australia or overseas, can not comply with the 3D kill requirement; and,
- Given the inability of most processes currently used in Australia to satisfy the requirement of Clause 60A, that the utility of this prescriptive regulation be reconsidered.

In December 2002, FSANZ released the Draft Assessment Report of Proposal P251 "Review of processing requirements for uncooked fermented meat (UCFM) products". In this the requirement for, *inter alia*, a 3D reduction was rejected because:

- "The key performance criterion in this option, i.e. the production process must reduce the number of *Escherichia coli* organisms by 99.9% or greater, could not be implemented and enforced effectively. Limitations in the predictive model used for assessing industry compliance with the performance criterion, prevent a comprehensive evaluation of all processing protocols.
- The performance criterion in this option is not food safety outcome based as it correlates neither with the initial *E. coli* load in the ingoing meat ingredients; nor the end product *E. coli* specification; and
- The performance criterion is unnecessarily prescriptive."

The recommended option is:

"Production of UCFM must:
(a) Implement a HACCP based food safety program in accordance with Standard 3.2.1 of the Code that has been verified and audited to ensure that the production process effectively reduces the number of *Escherichia coli* organisms in the UCFM product to a level specified in 1.6.1 of the code and satisfies the following requirements.
(1) Fermentation of a comminuted meat product, which will not be cooked, must be initiated through the use of a starter culture.
(2) A previously fermented or fermenting meat product must be treated so as to eliminate all the microbial pathogens and toxins prior to use as an ingredient in a comminuted fermented meat product, which will not itself be cooked.
(3) The maximum number of *Escherichia coli* organism in the ingoing raw meat ingredients should not exceed 20 MPN per gram unless an equivalent food safety outcome can be demonstrated with existing processing protocol(s); and the number of *Escherichia coli* organisms in a comminuted fermented meat product which will not be cooked must be monitored and recorded for the-
      (i)   ingoing raw meat ingredients; and
      (ii)   product after fermentation and any subsequent process.
(4) The pH of fermenting comminuted meat products which will not be cooked, measured in accordance with Method 1 in the schedule of Standard 1.6.2 of the *Food Standards Code*, and the temperature and time of the fermentation and maturation/drying steps must be monitored and recorded.
(5) Measurements recorded under subclasses (3) and (4) must be kept either for 1 year after the minimum durable shelf life of the product, or 2 years whichever is greater.
(6) Meat for a comminuted fermented meat product, which will not be cooked, must, if stored by the manufacturer, be stored at $5^{o}C$ or lower prior to fermentation.
<div align="center">OR</div>
(b) Implement a measure that achieves an equivalent food safety outcome as that achieved through the implementation of HACCP based food safety programs, and this measure is specified in the *Food Standards Code*.
<div align="center">OR</div>
(c) Conduct microbiological end product testing of each production lot (a lot corresponds to products with the same diameter manufactured under the same conditions in a single day) and hold the lots pending satisfactory compliance of the results with the *Escherichia coli* limit specified for Standard 1.6.1 of the Code. If the product has a pH of 5.3 or lower and a water activity of 0.90 or lower, the sampling prescribed in Standard 1.6.1 applies. Otherwise 30 samples of 25g each from the finished products must be taken for microbiological testing and none of the samples should exceed the level specified in column 5 of the Schedule in Standard 1.6.1 of the Code."

For reference the end-product testing specified in part (c) needs to conform to the following:

      Product pH <5.3 and a_w <0.90 n=5, c=1, m=3.6, M=9.2 MPN *E. coli* /g.
      Other UCFM n=30, c=0, m=3.6

The recommended option "…is recommended as the preferred regulatory option in order to minimise the potential risk posed to consumers by EHEC in UCFM products. The risk management measures encapsulated …. are comprehensive and have sufficient flexibility to ensure effective implementation. The proposed HACCP based food safety programs and the equivalent measures are preventative and are an outcome-based approach to managing the risk of EHEC contamination in UCFM products. The "test and hold" measure provides an alternative means of enforcement to cater for the situations where a HACCP based food safety program cannot be verified or audited".

Essentially the preferred option gives a microbiological goal that must be attained, with the means of achieving it being left to the manufacturer. This is in contrast to the Standard that may be replaced which did not give a microbiological goal. Comments on this proposal are due early in 2003 before the proposal proceeds to final assessment, at which point no further public consultation is performed.

7.3 Feasibility of Controls

There are significant practical difficulties with reaching a 5D reduction in STEC numbers in UCFM products. Incze (1998) concluded that the only practical way to achieve a 5D kill is to apply some form of heating process as commonly used methods only achieved a 1-2D kill. Altering other components, such as the salt concentration, was considered to result in unacceptable organoleptic qualities of the product (as does thermal processing). Traditionally fermented dry salamis with long fermentation and drying times were considered to be of "adequate safety".

The heating of UCFM products does not necessarily have to be such that it cooks the product, and a number of low temperature (*circa* 50°C) protocols have been described by Bacus (1997). Typically holding times at these temperatures are of the order of 60 minutes.

As already mentioned the report prepared for Meat and Livestock Australia (Ross and Shadbolt, 2001) concluded that most UCFM processes in current use in Australia do not reliably achieve a 3D kill of *E. coli*. A range of experiments were proposed in the report to examine the extent of inactivation of *E. coli* caused by several parameters. The effects of temperature were regarded as well understood, but the effects of other factors (pH, organic acid levels, water activity, redox potential) were regarded as data gaps.

The report suggested that the maturation temperature could be raised by a few degrees to increase control of *E. coli*. This was considered unlikely to greatly affect the aesthetic qualities of the final product, but would accelerate inactivation rates. The faster drying from ripening at higher temperatures, and the attendant risk of case hardening, could be offset by higher relative humidity in the ripening rooms.

The data summarised in Tables 1 and 2 in Section 3.2 suggest that pathogen reductions during fermentation and drying are relatively modest. However, a storage period at approximately 21°C following fermentation and maturation could achieve D kills of 3 $\log_{10}$ or greater.

The efficacy of a 3D inactivation of generic *E. coli* is influenced greatly by the proportion of generic *E. coli* that are pathogenic STEC. The smaller that this ratio is (i.e. less STEC per

generic *E. coli*), then the greater the assurance that a 3D inactivation (along with the associated requirements) will result in safe product.

Some data from the West Indies showed that of 92 isolates from dairy cow faecal samples 36.6% (range 20.0 to 60.0%) were identified as being able to produce verocytotoxin (Adesiyun *et al.*, 1997). However, the ratio in foods derived from these animals may be different as the same study showed that the ratio in bulk milk was 18.5% (range 0-35.3%). Contrasting data come from Australia where only 0.8% of *E. coli* colonies isolated from bovine faecal samples testing positive for the presence of the *stx* gene by PCR were identified as STEC (Midgley *et al.*, 1999). Most faecal samples (64.6%) were not found to contain STEC. Estimates from data for *E. coli* enriched from meat in the UK showed that 20% of samples yielded *E. coli* colonies which were >2% STEC, 30% yielded 0.5-2% STEC and the remainder <0.5% STEC (Willshaw *et al.*, 1993).

Further to this, not all STEC are of equal pathogenic potential, and some STEC serotypes which are have never been associated with human disease. The proportion of STEC isolates which have the genetic components necessary to be pathogenic seems to be low. For example, of 39 STEC isolates from the carcasses of (previously) healthy cattle sampled in France, none contained the full complement of pathogenicity factors (a toxin gene (*stx*1 and/or *stx*2) as well as the *eaeA* and *hlyA* genes) (Rogerie *et al.*, 2001). Three of 14 isolates tested in Australia had this genotype (Midgley *et al.*, 1999). Of 37 STEC isolates from British beef samples, none contained the *eaeA* gene (Willshaw *et al.*, 1993).

It is difficult to extrapolate data from overseas as farming systems in New Zealand are unique. It is therefore not possible to speculate on the likely ratio of STEC to generic *E. coli* in meat in New Zealand. It also seems unlikely that this ratio will be fixed as variables such as animal stress and differences in processing are likely to influence it. We do know, that of STEC isolates identified from New Zealand retail meats to date, none have been of the more important STEC serotypes. These factors are likely to contribute a safety margin to any given requirement to attain a certain "D kill".

Despite expressions of doubt as to the ability of UCFM manufacturing processes to achieve a 3D reduction in *E. coli* numbers (Ross and Shadbolt, 2001) the FSANZ Draft Assessment Report Proposal P251 December 2002 included data that showed a marked improvement in the number of assessed manufacturing protocols that did achieve such a reduction. The data showed that 91% of South Australian UCFM producers were using processes that achieved a 3D or greater reduction in *E. coli*. This appeared to be a result of collaboration between regulators and producers, guided by application of a predictive model of *E. coli* inactivation developed by Meat and Livestock Australia.

7.4   Economic Costs

An analysis of the incidence and costs of foodborne disease in New Zealand estimated that STEC cost $507,000 in direct and indirect costs (Lake *et al.*, 2000; Scott *et al.*, 2000). This was based on an estimated total of 248 reported and unreported cases, of which 20% were assumed to be caused by foodborne transmission. This low estimation of the proportion foodborne was based on the lack of data showing a link between food and transmission in New Zealand, as well as population attributable risk determinations in overseas studies. The

dollar estimate represented 0.9% of the total foodborne illness cost. This analysis represented all potential food vehicles.

In the United States the estimated annual foodborne illness cost of *E. coli* O157:H7 and *E. coli* non-O157 STEC has been estimated as US$1 billion (figures for 1998 updated for 2000). These costs derive from estimated annual cases of approximately 94,000, with approximately 2,800 hospitalisations and 78 deaths. This is from a total foodborne illness cost of US$6.9 billion which also includes diseases caused by *Campylobacter* species, non-typhoidal *Salmonella* and *Listeria monocytogenes* (Crutchfield and Roberts, 2000). These figures are high in comparison with New Zealand as they include productivity losses due to chronic illness caused by STEC infection, which were not included in the New Zealand estimate. The estimate also assumed that 80% of cases were caused by foodborne transmission (Buzby *et al.*, 1996), which is unlikely to be appropriate for New Zealand given current information. The percentage of cases caused by foodborne transmission in the United States has more recently been estimated as 85% (Mead *et al.*, 1999).

8   CONCLUSIONS

8.1   Description of Risks to New Zealand Consumers

8.1.1   Risks associated with UCFM

The current rate of STEC infection in New Zealand is similar to overseas countries at approximately 2 notified cases per 100,000 population. All cases have been sporadic or in clusters involving small numbers of cases; no widespread outbreaks affecting large numbers of people have yet been detected in New Zealand. Information on transmission routes is meager, but as was concluded in the Risk Profile on STEC in red meat, there is little evidence to suggest that red meat currently represents a vehicle for transmission of pathogenic STEC in New Zealand.

Nevertheless *E. coli* O157 has been found in the faeces of cows in New Zealand, as well as a small number of raw meat samples from export processing plants. Non-O157 STEC serotypes have been detected in samples of meat from retail sources. Consequently there is potential for the presence of STEC in raw meat in New Zealand, which then may be used as an ingredient in the production of UCFM. The amount of UCFM imported into New Zealand each year is unclear, but the majority will originate from Australia, where production will be subject to the standard created by FSANZ.

However, the consumption of UCFM is low in comparison with other red meat types, both in terms of servings (1.6% of all red meat servings) and weight (0.5% of all red meat consumed). The relatively low frequency of consumption of UCFM and the smaller serving sizes than for most other meat and meat products may mitigate the relative risk from this meat type.

STEC infections have a relatively high proportion of serious outcomes compared to other bacterial infections, and certain population groups in New Zealand (children up to 4 years) have higher rates of infection.

8.1.2   Risks associated with other foods

In the United States ground beef/hamburger is the food vehicle most frequently implicated in outbreaks of *E. coli* O157:H7 infection, while the limited information from Europe suggests that meat consumption is not associated with sporadic cases. Other food vehicles implicated in outbreaks have been contaminated foods not cooked prior to consumption (lettuce, salads, coleslaw) or unpasteurised foods (milk, apple juice). Contact with animals, and consumption of contaminated drinking water or other water sources have also been identified as transmission pathways.

8.1.3   Quantitative risk assessment

The main barrier to a comprehensive risk assessment is the limited data on the prevalence of contamination by STEC of New Zealand meats at the retail level (or at other points in the production chain), and the absence of quantitative levels of contamination. Further, there is no information from human surveillance studies to link meat or meat products with cases so far detected in New Zealand, and therefore no means to validate a QRA model. Current

methodology will need to be improved to provide the same sensitivity for broad screen STEC detection techniques as are available for specific *E. coli* O157 methods.

The relative importance of foodborne transmission of STEC in New Zealand is unclear from the information gathered on cases to date. Consequently a quantitative risk assessment for STEC in red meat and meat products would be premature, and could not be conducted with currently available data.

8.2   Commentary on Risk Management Options

Given the serious consequences of STEC infection it is essential that efforts continue to prevent foodborne transmission in red meat products generally. The high proportion of meat production that is exported means that mandatory HACCP based programmes will exist in most New Zealand meat processing plants and this will act to protect the portion of the domestic meat supply derived from the same source. This approach matches efforts in the United States to control STEC in red meat.

The prevalence of *E. coli* O157, and indeed *E. coli*, in meat from plants which deliver only to the domestic market (approximately 40% of the total supply) is unknown and represents a significant data gap. Information of this type would be of value as a baseline, as these suppliers develop Risk Management Programmes as required under the Animal Products Act.

Options for control of STEC specifically in UCFM include the following:

1. A large reduction in bacterial levels (5D) to cover all scenarios of bacterial contamination (US requirement);
2. Lower reductions in bacterial levels (2D-3D) providing a safety margin to back up low levels of generic *E. coli* contamination (or the absence of STEC), based on sampling and testing of raw ingredients (earlier FSANZ proposal);
3. As above with raw ingredients obtained from suppliers with a HACCP programme allied with microbiological testing programmes; and,
4. HACCP based food safety programmes that are validated to achieve a defined low level of *E. coli* in the finished product, as well as defined limits for *E. coli* in ingoing ingredients (later FSANZ proposal option (a));
5. An alternative measure that achieves an equivalent food safety outcome (later FSANZ proposal option (b)); and,
6. Microbiological testing of all end product before release (later FSANZ proposal option (c)).

Option 1 appears to be very difficult to achieve without some form of heat treatment, which may adversely affect the characteristics of the product

Due to the wide variety of UCFM products and processes used to make them, generic Critical Control Points are unlikely to be achievable. Therefore Options 2 - 5 would require process verification in the form of challenge testing using an agreed protocol and indicator bacterium, as well as accepted and validated sampling and testing procedures.

While a 1-2D reduction is all that might be expected during fermentation and drying/maturation, the scope for further reductions during storage are manifested. A greater

reduction (perhaps up to 3D) should be able to be achieved by introducing only minor process changes.

Data from the New Zealand National Microbiological Database Programme for 1997 to 2000 indicates that 8.0% of boneless beef samples intended for export (i.e. from plants with a HACCP based Risk Management Programme) were positive for generic *E. coli*. The mean $\log_{10}$ CFU/g was 1.11, with the 99th percentile being $\log_{10}$ 3.0, and the maximum was $\log_{10}$ 4.31 (Roger Cook, New Zealand Food Safety Authority, personal communication). The very low levels of *E. coli* O157 in such meat products have been reported in Section 5.1.3.

At least two overseas outbreaks of STEC infection involving UCFM involved processes that did not use starter cultures. Their use would seem to be crucial to reduce risks associated with UCFM production.

Any risk management options chosen are likely to be effectively implemented via the New Zealand Pork Processors Association and the PQIP programme, which will cover the majority of UCFM production in New Zealand. There may be a few "traditional" or "boutique" manufacturers outside this coverage, and a targeted intervention may be needed to achieve complete industry implementation of food safety measures.

8.3   Data gaps

The data gaps identified in this report are:

- Prevalence of contamination by STEC of New Zealand meats at the retail level (or at other points in the production chain);
- Quantitative levels of contamination by STEC of New Zealand meats at the retail level (or at other points in the production chain);
- Methods to detect non O157 STEC with the same sensitivity as *E. coli* O157;
- Relative importance of transmission pathways for STEC in New Zealand; and,
- The prevalence of *E. coli* O157 and *E. coli* in meat from processing plants which deliver only to the domestic market.

9   REFERENCES

Adesiyun AA, Webb LA, Romain H, Kaminjolo JS. (1997) Prevalence and characteristics of strains of *Escherichia coli* isolated from milk and feces of dairy farms in Trinidad. Journal of Food Protection; 60, 1174-1181.

Ammon A, Petersen LR, Karch H. (1999) A large outbreak of hemolytic uremic syndrome caused by unusual sorbitol-fermenting strain of *Escherichia coli* O157. Journal of Infectious Diseases; 179: 1274-1277.

Anonymous. (2000) Verocytotoxin-producing *Escherichia coli* (VTEC/STEC). Lablink; 7(1):, 8.

Anonymous. (2002) Verocytotoxin-producing *Escherichia coli* (VTEC/STEC). Lablink; 9(1): 8.

Bacus J. (1997) Processing procedures to control *Salmonella* and *E. coli* in fermented sausage products. Food Australia; 49: 543-545.

Baker M, Eyles R, Bennett J, Nicol C. (1999) Emergence of verotoxigenic *Escherichia coli* (VTEC) in New Zealand. New Zealand Public Health Report; 6: 9-12.

Benjamin MM, Datta AR. (1995) Acid tolerance of enterohemorrhagic *Escherichia coli*. Applied and Environmental Microbiology; 61: 1669-1672.

Bennett J, Bettelheim KA. (2002) Serotypes of non-O157 verocytotoxigenic *Escherichia coli* isolated from meat in New Zealand. Comparative Immunology, Microbiology and Infectious Disease; 25: 77-84.

Bettelheim KA. (2000) Role of non-O157 VTEC. Journal of Applied Microbiology Symposium Supplement; 88: 38S-50S.

Bitzan M, Moebius E, Ludwig K, Müller-Wiefel DE, Karch H. (1991) High incidence of serum antibodies to *Escherichia coli* O157 lipopolysaccharide in children with hemolytic-uremic syndrome. Journal of Pediatrics; 199: 380-385.

Brooks HJL, Bettelheim KA, Todd B, Holdaway MD. (1997) Non-0157 Verocytotoxin producing *Escherichia coli*: aetiological agents of diarrhoea in children in Dunedin, New Zealand. Comparative Immunology, Microbiology and Infectious Diseases; 20(2):163-70.

Brooks HJL, Mollison BD, Bettelheim KA, Matejka K, Paterson KA, Ward VK. (2001) Occurrence and virulence factors of non-O157 shiga toxin-producing *Escherichia coli* in retail meat in Dunedin, New Zealand. Letters in Applied Microbiology; 32: 118-122.

Buncic S. (1995) Assessment of fate of *Escherichia coli* O157:H7 in fermented sausages produced by processes described by New Zealand Pork Industry Board. Report prepared by the Meat Industry Research Institute of New Zealand. Appendix D to Standard for Food Safety New Zealand Bacon Curing Industry Part 1. September 1998.

Buncic S, Avery SM. (1997) *Escherichia coli* O157:H7 in healthy dairy cows. New Zealand Veterinary Journal; 45: 45-48.

Buzby JC, Roberts T, Lin C-TJ, MacDonald JM. (1996) Bacterial foodborne disease: Medical costs and productivity losses. Agricultural Economic Report Number 741. Washington DC: United States Department of Agriculture Economic Research Service.

Calicioglu M, Faith NG, Buege DR, Luchansky JB. (1997) Viability of *Escherichia coli* O157:H7 in fermented semidry low-temperature-cooked beef summer sausage. Journal of Food Protection; 60: 1158-1162.

Calicioglu M, Faith NG, Buege DR, Luchansky JB. (2001) Validation of a manufacturing process for fermented semidry Turkish Soudjouk to control *Escherichia coli* O157:H7. Journal of Food Protection; 64: 1156-1161.

Caprioli A, Tozzi AE, Rizzoni G, Karch H. (1997) Non-O157 shiga toxin-producing *Escherichia coli* infections in Europe. Emerging Infectious Diseases; 3: 578-579.

CAST (Council for Agricultural Science and Technology). (1994) Foodborne pathogens: risks and consequences. Task Force Report No. 122.

CDC. (1995a) *Escherichia coli* O157:H7 outbreak linked to commercially distributed dry-cured salami- Washington and California, 1994. Morbidity and Mortality Weekly Reports; 44: 157-160.

CDC. (1995b) Community outbreak of hemolytic uremic syndrome attributable to *Escherichia coli* O111:NM-South Australia, 1995. Morbidity and Mortality Weekly Reports; 44: 550-551, 557-558.

Chart H. (2000) VTEC enteropathogenicity. Journal of Applied Microbiology Symposium Supplement; 88: 12S-23S.

Chikthimmah N, Anantheswaran RC, Roberts RF, Mills EW, Knabel SJ. (2001) Influence of sodium chloride on growth of lactic acid bacteria and subsequent destruction of *Escherichia coli* O157:H7 during processing of Lebanon Bologna. Journal of Food Protection; 64: 1145-1150.

Chinen I, Tanaro JD, Miliwebsky E, Lound LH, Chimelli G, Ledri S, Baschkier A, Scarpin M, Manfredi E, Rivas M. (2001) Isolation and characterization of *Escherichia coli* O157:H7 from retail meats in Argentina. Journal of Food Protection; 64: 1346-1351.

Clavero MRS, Beuchat LR. (1996) Survival of *Escherichia coli* O157:H7 in broth and processes salami as influenced by pH, water activity, and temperature and suitability of media for its recovery. Journal of Food Protection; 62: 2735-2740.

Codex. (1999) Draft principles and guidelines for the conduct of microbiological risk assessment. Report of the thirty first session of the Codex Committee on Food Hygiene. ALINORM 99/13A. Rome: Codex Alimentarius Commission.

Crutchfield SR, Roberts T. (2000) Food safety efforts accelerate in the 1990s. Food Review; 23 (3): 44-49.

DeBauch ED, Savage GP. (1993) Salami: brewing a sausage. The Food Technologist; Autumn 1993: 31-39.

Decludt B, Bouvet P, Mariani-Kurkdjian P, Grimont F, Grimont PAD, Hubert B, Loirat C and the Société de Néphrologie Pédiatrique. (2000) Haemolytic uraemic syndrome and shiga toxin-producing *Escherichia coli* infection in children in France. Epidemiology and Infection; 124: 215-220.

Desmarchelier PM. (1997) Enterohaemorrhagic *Escherichia coli* – the Australian perspective. Journal of Food Protection; 60: 1447-1450.

Desmarchelier PM, Grau FH. (1997) *Escherichia coli*. In: Foodborne microorganisms of public health significance. Fifth Edition. Sydney: Australian Institute of Food Science and Technology Inc.

Djuretic T, Wall PG, Ryan MJ, Evans HS, Adak GK, Cowden JM. (1996) General outbreaks of infectious intestinal disease in England and Wales 1992 to 1994. CDR Review; 4: R57-R63.

Doyle MP, Zhao T, Meng J, Zhao S. (1997) *Escherichia coli* O157:H7. In: Doyle MP, Beuchat LR, Montville TJ. (eds). Food Microbiology Fundamentals and Frontiers. Washington, DC: American Society for Microbiology Press. pp 171-191.

Duncan SH, Booth IR, Flint HJ, Stewart CS. (2000) The potential for the control of *Escherichia coli* O157 in farm animals. Journal of Applied Microbiology Symposium Supplement; 88: 157S-165S.

Dundas S, Todd WTA. (2000) Clinical presentation, complications and treatment of infection with verocytotoxin-producing *Escherichia coli*. Challenges for the clinician. Journal of Applied Microbiology Symposium Supplement; 88: 24S-30S.

Evans HS, Madden P, Douglas C, Adak GK, O'Brien SJ, Djuretic T, Wall PG, Stanwell-Smith R. (1998) General outbreaks of infectious intestinal disease in England and Wales: 1995 to 1996. Communicable Disease and Public Health; 1: 165-171.

Faith NG, Parniere N, Larson T, Lorang T, Luchansky JB. (1997) Viability of *Escherichia coli* O157:H7 in pepperoni during the manufacture of sticks and the subsequent storage of slices at 21, 4 and –20°C under air, vacuum and CO_2. International Journal of Food Microbiology; 37: 47-54.

Faith NG, Parniere N, Larson T, Lorang T, Kaspar CW, Luchansky JB. (1998a) Viability of *Escherichia coli* O157:H7 in salami following conditioning of batter, fermentation and drying of sticks, and storage of slices. Journal of Food Protection; 61: 377-382.

Faith NG, Wierzba RK, Ihnot AM, Roering AM, Lorang TD, Kaspar CW, Luchansky JB. (1998b) Survival of *Escherichia coli* O157:H7 in full- and reduced-fat pepperoni after

manufacture of sticks, storage of slices at 4°C or 21°C under air and vacuum, and baking of slices of frozen pizza at 135, 191 and 246°C. Journal of Food Protection; 61: 383-389.

FSIS (Food Safety and Inspection Service, US Department of Agriculture). (1998) Preliminary Pathways and Data for a Risk Assessment of *E. coli* O157:H7 in Beef. http://www.fsis.usda.gov/ophs/ecolrisk/prelim.htm

Glass KA, Loeffelholz JM, Ford JP, Doyle MP. (1992) Fate of *Escherichia coli* O157:H7 as affected by pH or sodium chloride and in fermented, dry sausage. Applied and Environmental Microbiology; 58: 2513-2516.

Goldwater PN, Bettelheim KA. (1995) The role of enterohaemorrhagic *Escherichia coli* serotypes other than O157:H7 as causes of disease in Australia. Communicable Diseases Intelligence; 19: 2-4.

Haas CN, Rose JB, Gerba CP. (1999) Quantitative microbial risk assessment. New York: John Wiley.

Haas CN, Thayyar-Madabusi A, Rose JB, Gerba CP. (2000) Development of a dose-response relationship for *Escherichia coli* O157:H7. International Journal of Food Microbiology; 56: 153-159.

Hasell S. (2000) Hazard management guidelines for fermented meat products. ESR Client Report FW0092. Christchurch: ESR.

Health Canada. (2000) Interim guidelines for the control of Verotoxinogenic *Escherichia coli* including E. coli O157:H7 in ready to eat fermented sausages containing beef or a beef product as an ingredient. Available from: http://www.hc-sc.gc.ca/food-aliment/mh-dm/mhe-dme/rfao-aoca/e_guideline_for_fermented_sausages.html

Heuvelink AE, Zwartkruis-Nahuis JTM, Beumer RR, De Boer E. (1999) Occurrence and survival of verocytotoxigen-producing *Escherichia coli* O157:H7 in meats obtained from retail outlets in the Netherlands. Journal of Food Protection; 62: 1115-1122.

Hinkens JC, Faith NG, Lorang TD, Bailey P, Buege D, Kaspar CW, Luchansky JB. (1996) Validation of pepperoni processes for control of *Escherichia coli* O157:H7. Journal of Food Protection; 59: 1260-1266.

Hudson JA, Nicol C, Capill J, Bennett J. (2000) Isolation of shiga toxin-producing *Escherichia coli* (STEC) from foods using EHEC agar. Letters in Applied Microbiology; 30: 109-113.

ICMSF. (1998) Micro-organisms in food. 6 Microbial ecology of food commodities. 1 Meat and meat products. VI Raw and comminuted meats. International Commission on Microbiological Specifications for Foods (ICMSF). London: Blackie Academic and Professional.pp39-42.

Incze K. (1998) Dry fermented sausages. Meat Science; 49: S169-S177.

Jaeger JL, Acheson DW. (2000) Shiga toxin-producing *Escherichia coli*. Current Infectious Disease Reports; 2: 61-67.

Johnson SC, Sebranaek JG, Olson DG, Wiegland BR. (2000) Irradiation in contrast to thermal processing of pepperoni for control of pathogens: Effects on quality indicators. Journal of Food Science; 65: 1260-1265.

Karmali MA. (1989) Infection by verocytotoxin-producing *Escherichia coli*. Clinical Microbiological Reviews; 2: 15-38.

Keskimaskii M, Siitonen A. (1997) From Finland: Surveillance of STEC infections. www.iss.it/scientifica/pubblica/notiziario/v10n10s1/v10n10s1.htm.

Kieft C, Perks M, Baker M, Galloway Y, Sutherland H. (2000) Annual surveillance summary 1999. Client Report FW0059. Kenepuru: ESR.

Kleanthous H, Smith HR, Scotland SM, Gross RJ, Rowe B, Taylor CM, Milford DV. (1990) Haemolytic uraemic syndrome in the British Isles, 1985-8: association with verocytotoxin producing *Escherichia coli*. Part 2: microbiological aspects. Archives of Diseases in Childhood; 65: 722-727.

Lake RJ, Baker MG, Garrett N, Scott WG, Scott HM. (2000) Estimated number of cases of foodborne infectious disease in New Zealand. New Zealand Medical Journal; 113: 278-281.

Lee MB, Styliadis S. (1996) A survey of pH and water activity levels in processed salamis and sausages in metro Toronto. Journal of Food Protection; 59: 1007-1011.

Leistner F, Gorris LGM. (1995) Food preservation by hurdle technology. Trends in Food Science and Technology; 6: 41-46.

Levine P, Rose B, Green, S, Ransom, G, Hill W. (2001) Pathogen testing of ready-to-eat meat and poultry products collected at federally inspected establishments in the United States, 1990 to 1999. Journal of Food Protection; 64: 1188-1193.

Lindqvist R, Andersson Y, de Jong B, Norberg P. (2000) A summary of reported foodborne disease incidents in Sweden, 1992 to 1997. Journal of Food Protection; 63: 1315-1320.

Little CL, Monsey HA, Nichols GL, DeLouvois J. (1998) The microbiology of ready-to-eat dried and fermented meat and meat products. International Journal of Environmental Health Research; 8: 277-284.

Lopez L, Baker M, Perks M. (2001) Annual surveillance summary 2000. Client Report FW0156. Kenepuru: ESR.

Luzzi I, Tozzi AE, Rizzoni G, Niccolini A, Benedetti I, Minelli, F, Caprioli A. (1995) Detection of serum antibodies to the lipopolysaccharide of *Escherichia coli* O103 in patients with hemolytic uremic syndrome. Journal of Infectious Diseases; 171: 524-515.

Makino S-I, Kii T, Asakura H, Shirahata T, Ikeda T, Takeshi K, Itoh, K. (2000) Does enterohaemorrhagic *Escherichia coli* O157:H7 enter the viable but nonculturable state in salted salmon roe? Applied and Environmental Microbiology; 66: 5536-5539.

Mariani-Kurkdjian P, Denamur E, Milon A, Picard B, Cave H, Lambert-Zechovsky N, Loirat C, Goullet P, Sansonetti PJ, Elion J. (1993) Identification of a clone of *Escherichia coli* O103:H2 as a potential agent of hemolytic-uremic syndrome in France. Journal of Clinical Microbiology; 31: 296-301.

McClure PJ, Hall S. (2000) Survival of *Escherichia coli* in foods. Journal of Applied Microbiology Symposium Supplement; 88: 61S-70S.

Mead PS, Slutsker L, Dietz V, McCaig LF, Breese JS, Shapiro C, Griffin PM, Tauxe RV. (1999) Food-related illness and death in the United States. Emerging Infectious Diseases; 5: 607-625.

Midgley J, Fegan N, Desmarchelier P. (1999) Dynamics of shiga toxin-producing *Escherichia coli* (STEC) in feedlot cattle. Letters in Applied Microbiology; 29, 85-89.

Ministry of Health/Ministry of Agriculture and Forestry. (2000) Food Administration in New Zealand: A Risk Management Framework for Food Safety. Wellington: Joint Ministry of Health and Ministry of Agriculture and Forestry Food Harmonisation Project.

Nickelson R II, Luchansky J, Kaspar C, Johnson E. (1996) Dry fermented sausage and *E. coli* O157:H7. Blue Ribbon Task Force, National Cattlemen's Beef Association, Chicago, Research Report No. 11-316.

Pai CH, Ahmed N, Lior H, Johnson WM, Sims HV, Woods DE. (1988) Epidemiology of sporadic diarrhoea due to verocytotoxin-producing *Escherichia coli*: a two year prospective study. Journal of Infectious Diseases; 157: 1054-1057.

Park S, Worobo RW, Durst RA. (1999) *Escherichia coli* O157:H7 as an emerging foodborne pathogen: A literature review. Critical Reviews in Food Science and Nutrition; 39: 481-502.

Parry SM, Palmer SR. (2000) The public health significance of VTEC O157. Journal of Applied Microbiology Symposium Supplement; 88: 1S-9S.

Pennington TH. (2000) VTEC: lessons learned from British outbreaks. Journal of Applied Microbiology Symposium Supplement; 88: 90S-98S.

Pidcock K, Heard GM, Henriksson A. (2002) Application of nontraditional meat starter cultures in production of Hungarian salami. International Journal of Food Microbiology; 76:75-81.

Pierard D. (1992) Infections with verotoxin-producing *Escherichia coli*. Acta Clinica Belgica; 47: 387-396.

Pierard D, Crowcroft N, De Brock S, Potters D, Crabbe G, Van Loock F, Lauwers S. (1999) A case-control study of sporadic infection with O157 and non-O157 verocytotoxin-producing *Escherichia coli*. Epidemiology and Infection; 122: 359-365.

Pond, TJ, Wood DS, Mumin IM, Barbut S, Griffiths MW. (2001) Modeling the survival of *Escherichia coli* O157:H7 in uncooked, semidry, fermented sausage. Journal of Food Protection; 64:759-766.

Powell MR, Ebel E, Schlosser W, Walderhaug M, Kause J. (2001) Dose-response envelope for *Escherichia coli* O157:H7. Quantitative Microbiology; 2: 141-163.

Pommepuy M, Butin M, Derrien A, Gourmelon RR, Cormier M. (1996) Retention of enteropathogenicity by viable but nonculturable *Escherichia coli* exposed to seawater and sunlight. Applied and Environmental Microbiology; 62: 4621-4626.

Ricke SC, Keeton JT. (1997) Fermented meat, poultry and fish products. In: Food Microbiology: Fundamentals and Frontiers (eds. MP Doyle, LR Beuchat, Montville TJ). ASM Press: Washington DC: 610-628.

Rigsbee W, Simpson LM, Oliver JD. (1997) Detection of the viable but nonculturable state in *Escherichia coli* O157:H7 . Journal of Food Safety; 16: 255-262.

Riordan DCR, Duffy G, Sheridan JJ, Eblen BS Whiting RC, Blair IS, McDowell DA. (1998) Survival of *Escherichia coli* O157:H7 during the manufacture of pepperoni. Journal of Food Protection; 61:146-151.

Rogerie F, Marecat A, Gambade S, Dupond F, Beaubois P, Lange M. (2001) Characterization of shiga toxin producing *E. coli* and O157 serotype *E. coli* isolated in France from healthy domestic cattle. International Journal of Food Microbiology; 63: 217-223.

Ross T, Shadbolt CT. (2001) Predicting *E. coli* inactivation in uncooked comminuted fermented meat products. Prepared for Meat and Livestock Australia by the Centre for Food Safety and Quality, School of Agricultural Science, University of Tasmania. North Sydney: Meat and Livestock Australia.

Rowe PC, Orbine E, Lior H, Wells GA, McLaine PN. (1993) A prospective study of exposure to verotoxin-producing *Escherichia coli* among Canadian children with hemolytic uremic syndrome. Epidemiology and Infection; 110: 1-7.

Russell DG, Parnell WR, Wilson NC *et al.* (1999) NZ Food: NZ People. Key results of the 1997 national Nutrition Survey. Wellington: Ministry of Health.

Scotland SM, Rowe B, Smith HR, Willshaw GA, Gross RJ. (1988) Verocytotoxin-producing strains of *Escherichia coli* from children with haemolytic ureamic syndrome and their detection by specific DNA probes. Journal of Medical Microbiology; 25: 237-243.

Scott WG, Scott HM, Lake RJ, Baker MG. (2000) Economic cost to New Zealand of foodborne infectious disease. New Zealand Medical Journal; 113: 281-284.

Sneyd E, Lopez L, Eglinton M, McDowell R, Margolin T. (2002) Annual surveillance summary 2001. Client Report FW0156. Kenepuru: ESR.

Takumi K, de Jonge R, Havelaar A. (2000) Modelling inactivation of *Escherichia coli* by low stomach pH: application to passage through the stomach of young and elderly people. Journal of Applied Microbiology; 89: 935-943.

Tamura K, Sakazaki R, Murase M, Kosako Y. (1996) Serotyping and categorisation of *Escherichia coli* strains isolated between 1958 and 1992 from diarrhoeal diseases in Asia. Journal of Medical Microbiology; 45: 353-358.

Tilden J Jr, Young W, McNamara A-M, Custer C, Boesel B, Lambert-Fair MA, Majkowski J, Vugia D, Werner SB, Hollingsworth J, Morris JG Jr. (1996) A new route of transmission for *Escherichia coli*: infection from dry fermented salami. American Journal of Public Health; 86: 1142-1145.

Todd EC. (1992) Foodborne disease in Canada- a 10 year summary from 1975 to 1984. Journal of Food Protection; 55: 123-132.

Uyttendaele M, Taverniers I, Debevere J. (2001) Effect of stress induced by suboptimal growth factors on survival of *Escherichia coli* O157:H7. International Journal of Food Microbiology; 66: 31-37.

WHO. (1998) Zoonotic Non-O157 Shiga Toxin-Producing *Escherichia Coli* (STEC). Report of a WHO Scientific Working Group Meeting. Berlin, Germany, 23-26 June 1998. Available from: http://www.who.int/emc-documents/zoonoses/whocsraph988c.html

Williams RC, Isaacs S, Decou ML, Richardson EA, Buffett MC, Slinger RW, Brodsky MH, Ciebin BW, Hockin J. (2000) Illness associated with *Escherichia coli* O157:H7 in Genoa salami. Canadian Medical Association Journal; 162: 1409-1413.

Willshaw GA, Smith HR, Roberts D, Thirlwell T, Cheasty T, Rowe B. (1993) Examination of raw beef products for the presence of verocytotoxin producing *Escherichia coli*, particularly those of serogroup O157. Journal of Applied Microbiology; 75: 420-426.

APPENDIX 1: CATEGORIES FOR RISK PROFILES

The assignment of a category for a food/hazard combination uses two criteria: incidence and severity.

1. Incidence

The incidence is an estimate of the proportion of the foodborne disease rate due to an individual hazard, that is transmitted by a single food or food group.

The overall rate of foodborne disease caused by individual hazards can be derived from information in the published estimate of foodborne disease (Lake *et al*., 2000). This estimate has been updated to reflect more recent notifications rates for the 12 months to June 2001, but still using 1996 census figures (3,681,546 population). Rates include estimates for unreported cases who do not present to a GP.

Disease/organism	Food rate (/100,000 population) Calculated for 12 months to June 2001	Food rate (/100,000 population) Calculated for 12 months to December 1998
Campylobacteriosis	1320	2047
Listeriosis	0.4	0.4
VTEC/STEC	1.9	1.4
Salmonellosis	176	230
Yersiniosis	38	62
Shigellosis	7	7
NLV*	478	478
Toxins*	414	414
Typhoid*	0.3	0.3
Hepatitis A*	0.4	0.4

 * not recalculated.

These are **total** foodborne rates, so it is probably safe to assume that in most cases the rates associated with a particular food are likely to be an order of magnitude lower. For instance, a category of ">1000" would only be assigned if it was decided that all campylobacteriosis was due to a single food/food type.

The following categories are proposed for the rates attributable to a single hazard/food (or food group) combination:

Category	Rate range	Comments/examples
1	>100	Significant contributor to foodborne campylobacteriosis Major contributor to foodborne NLV
2	10-100	Major contributor to foodborne salmonellosis Significant contributor to foodborne NLV
3	1-10	Major contributor to foodborne yersiniosis, shigellosis
4	<1	Major contributor to foodborne listeriosis

A further category, of "no evidence for foodborne disease in New Zealand" is desirable, but it was considered more appropriate to make this separate from the others. Also separate is another category, of "no information to determine level of foodborne disease in New Zealand".

The estimation of the proportion of the total foodborne disease rate contributed by a single food or food group will require information from a variety of sources including:

- exposure estimates
- results from epidemiological studies (case control risk factors)
- overseas estimates

For illnesses where the rate is <1 per 100,000 the ability to assign a proportion is unlikely to be sensible. For such illnesses it may be more useful to consider a Risk Profile across the range of all high risk foods, rather than individual foods or food groups.

2. Severity

Severity is related to the probability of severe outcomes from infection with the hazard.

The outcomes of infectious intestinal disease are defined in the estimate of the incidence (Lake *et al.*, 2000) as:
- death
- hospitalised and long term illness (GBS, reactive arthritis, HUS)
- hospitalised and recover
- visit a GP but not hospitalised
- do not visit a GP

The first three categories of cases were classed as severe outcomes. Some hospitalisations will result from dehydration etc. caused by gastrointestinal disease. However, for infections with *Listeria* and STEC hospitalisation will result from more severe illness, even if recovery is achieved.

The proportion of severe outcomes resulting from infection with the hazards can be estimated from the proportion of cases hospitalised and recover, hospitalised and long term illness, and deaths (Lake *et al*., 2000).

Disease/organism	Percentage of outcomes involving death or long term illness from foodborne cases
Campylobacteriosis	0.3
Listeriosis	60.0
VTEC/STEC	10.4
Salmonellosis	1.0
Yersiniosis	0.4
Shigellosis	2.7
NLV	Assumed to be <0.5%
Hepatitis A	15.4
Typhoid	83.3
Toxins	Assumed to be <0.5%

Categories for the probability of severe outcomes are suggested as follows:

Severity Category	Percentage of cases that experience severe outcomes	Examples
1	>5%	listeriosis, STEC, hepatitis A, typhoid
2	0.5 – 5%	salmonellosis, shigellosis
3	<0.5%	campylobacteriosis, yersiniosis, NLV, toxins

There are a number of hazards for which the incidence of foodborne disease is uncertain. These have been assigned to the above severity categories as follows:

Severity category 1:

Bacteria

Clostridium botulinum

Protozoa

Toxoplasma

Severity category 3:

Bacteria

Aeromonas/Plesiomonas
Arcobacter
E. coli (pathogenic, other than STEC)
Pseudomonas
Streptococcus
Vibrio parahaemolyticus

Viruses

Others (e.g. rotavirus)

Protozoa

Giardia
Cryptosporidium
Cyclospora
Others (e.g. *Entamoeba*)

Proposed Category Matrix

Incidence	>100	10-100	1-10	<1
Severity 1				
Severity 2				
Severity 3				

Alternatives:

No evidence for foodborne disease in New Zealand

No information to determine level of foodborne disease in New Zealand