

15 July 2009 [11-09]

PROPOSAL P1004 PRIMARY PRODUCTION & PROCESSING STANDARD FOR SEED SPROUTS (FIRST ASSESSMENT REPORT)

Executive Summary

Purpose

FSANZ has prepared this FIRST Assessment Report for public consultation. This Report is prepared in accordance with the principles of best practice regulation recommended by the Council of Australian Governments: identifying the problem that has prompted government action; the objectives of such action; and possible options for achieving the objectives. An overview of the industry sector, the proposed scope of the work, the food safety hazards and existing food safety measures (regulatory and non-regulatory) applying to the industry are provided.

To assist FSANZ undertake a comprehensive and informed impact analysis of the proposed options, affected parties are encouraged to provide comment and information on the issues raised in the report.

This Proposal is being assessed under the Major Procedure.

Introduction

A primary production and processing standard is an Australia-only standard and a set of obligations on primary producers and processors of food commodities. These standards are incorporated into Chapter 4 of the *Australia New Zealand Food Standards Code* (the Code) and along with other standards in the Code, they provide an approach to managing food safety and suitability in Australia that extends from production on the farm through to sale to the consumer.

To date, FSANZ has developed primary production and processing standards for the seafood and dairy sectors and is currently developing standards for poultry meat, eggs and red meat. The development of a primary production and processing standard for seed sprouts is now being considered following two outbreaks of food-borne illness in Australia attributed to the consumption of seed sprouts in 2005-2006.

Seed sprouts are a ready-to-eat sprouted form of seeds produced from a wide range of seeds including alfalfa, onion, radish, broccoli, sunflower, cress, snow peas, lentils, peas and mung beans.

Proposal P1004 will examine possible food safety measures that can be applied to primary production and processing of seed sprouts, covering the areas of seed production (pre-harvest and post-harvest activities) and sprout production.

A Standard Development Committee (SDC) consisting of representatives from the industry, retail, government regulators and consumers has been established by FSANZ to advise on this standard development Proposal.

The Problem

Seed sprouts contaminated by pathogenic micro-organisms present an unacceptable health risk to consumers. In recent years, outbreaks of food-borne illness have been associated with the consumption of seed sprouts both in Australia and overseas. In 2005 and 2006, two outbreaks of *Salmonella Oranienburg* in Australia were attributed to the consumption of alfalfa sprouts. The outbreaks affected the health of more than 130 people. Factors contributing to these adverse health events include:

- the inherent nature of the product (e.g. a ready-to-eat product in which the production process supports the growth of microbial pathogens if present)
- scientific uncertainty around the most effective pathogen mitigation steps
- a lack of through-chain risk mitigation measures (either regulatory or non-regulatory).

Objectives

The goal of government action is to minimise adverse health effects associated with the consumption of seed sprouts. The objectives of Proposal P1004 are to assess the need for and identify any appropriate through-chain control measures (regulatory and non-regulatory) that can be implemented nationally by industry to maximise the safety of seed sprouts.

Options

In order to decide the most effective and efficient approach for achieving the objectives, FSANZ must consider various risk management options. These options include the *Status Quo* (the situation if no action is taken) as a comparative measure against appropriate regulatory (government) and non-regulatory (industry) approaches. The options identified for Proposal P1004 are:

- Option 1 Self regulation. This approach requires food or primary production businesses to implement and enforce industry guidelines or codes of practice aimed at improving the safety of seed sprouts.
- Option 2 Status Quo. Currently there is a mixture of regulatory (State based and export requirements) and self-regulatory approaches developed for different pockets of the production chain for seed sprouts. A nationally consistent set of food safety measures across the production chain for seed sprouts is lacking.
- Option 3 Food Regulatory measures. This involves the development of food regulatory measures in the Code for sectors involved in the production of seed sprouts together with a regulatory impact analysis that demonstrates such measures are commensurate with risk and are cost effective.

Impact Analysis

The preferred option decided through the assessment of Proposal P1004 will be based on an analysis that considers:

- who is affected by the problem and the proposed solution
- scientific evaluation of the risks
- efficacy and practicality of risk mitigation measures (control measures) identified
- costs and benefits to affected parties of the interventions associated with each option.

In deciding the preferred option, an assessment of the costs and benefits of each of the identified options will be undertaken. This will include:

- looking at the scientific evidence that identifies the main hazards associated with seed sprouts and the factors along the supply chain that impact on the presence or level of that hazard
- assessing the factors that impact on the presence or level of these hazards in order to identify what control measures are needed and the extent to which potential hazards can or cannot be managed at steps in the chain to minimise public health risks.

FSANZ, with advice from the SDC and taking into consideration submissions made on this report, will undertake a detailed impact analysis of the costs and benefits to each affected party posed by each option. This assessment, together with the preferred option, will be detailed in the SECOND Assessment Report.

Conclusion

The comments and information provided during this consultation will be considered during the second assessment stage of this Proposal when a preferred option for implementing national through-chain food safety control measures for the seed sprout industry will be proposed.

Invitation for Submissions

FSANZ invites public comment on this Report based on the principles of best practice regulation for the purpose of preparing an amendment to the Code for approval by the FSANZ Board.

Written submissions are invited from interested individuals and organisations to assist FSANZ in further considering this Proposal. Submissions should, where possible, address the objectives of FSANZ as set out in section 18 of the *FSANZ Act*. Information providing details of potential costs and benefits of the proposed change to the Code from stakeholders is highly desirable. Claims made in submissions should be supported wherever possible by referencing or including relevant studies, research findings, trials, surveys etc. Technical information should be in sufficient detail to allow independent scientific assessment.

The processes of FSANZ are open to public scrutiny, and any submissions received will ordinarily be placed on the public register of FSANZ and made available for inspection. If you wish any information contained in a submission to remain confidential to FSANZ, you should clearly identify the sensitive information, separate it from your submission and provide justification for treating it as confidential commercial material. Section 114 of the FSANZ Act requires FSANZ to treat in-confidence, trade secrets relating to food and any other information relating to food, the commercial value of which would be, or could reasonably be expected to be, destroyed or diminished by disclosure.

Submissions must be made in writing and should clearly be marked with the word 'Submission' and quote the correct project number and name. While FSANZ accepts submissions in hard copy to our offices, it is more convenient and quicker to receive submissions electronically through the FSANZ website using the <u>Standards Development</u> tab and then through <u>Documents for Public Comment</u>. Alternatively, you may email your submission directly to the Standards Management Officer at <u>submissions@foodstandards.gov.au</u>. There is no need to send a hard copy of your submission if you have submitted it by email or the FSANZ website. FSANZ endeavours to formally acknowledge receipt of submissions within 3 business days.

DEADLINE FOR PUBLIC SUBMISSIONS: 6pm (Canberra time) 26 August 2009

SUBMISSIONS RECEIVED AFTER THIS DEADLINE WILL NOT BE CONSIDERED

Submissions received after this date will only be considered if agreement for an extension has been given prior to this closing date. Agreement to an extension of time will only be given if extraordinary circumstances warrant an extension to the submission period. Any agreed extension will be notified on the FSANZ website and will apply to all submitters.

Questions relating to making submissions can be directed to the Standards Management Officer at standards.gov.au.

If you are unable to submit your submission electronically, hard copy submissions may be sent to one of the following addresses:

Food Standards Australia New Zealand PO Box 7186 Canberra BC ACT 2610 AUSTRALIA Tel (02) 6271 2222 Food Standards Australia New Zealand PO Box 10559 The Terrace WELLINGTON 6036 NEW ZEALAND Tel (04) 473 9942

INTRODUCTION	3
1. PRIMARY PRODUCTION AND PROCESSING STANDARDS	3
2. SEED SPROUTS	4
2.1 Scope	4
2.2 The production chain	4
2.2.1 Seed production	4
2.2.2 Sprout production	5
THE PROBLEM	5
	6
3. PUBLIC HEALTH RISK	0 6
3.1 Oubleaks of 1000-bothe liness associated with seed sprouts	0 6
3.3 What controls are effective?	7
4 EXISTING REQUIREMENTS	7
4.1 Regulatory	7
4.1.1 Australia New Zealand Food Standards Code	7
4.1.1.1 Chapter 3 – Food Safety Standards	7
4.1.1.2 Chapter 1 – General Food Standards	8
4.1.2 State-based requirements	8
4.1.3 Export requirements	8
4.2 Industry measures	9
4.2.1 Seeu ploudeis	9 Q
4 2 3 Retailers	10
OBJECTIVES	.10
5 OBJECTIVE OF THE PROPOSAL	.10
5.1 Process for achieving the objective	. 10
5.2 Constraints	. 11
5.2.1 FSANZ Act	.11
5.2.2 Policy guidelines	.11
OPTIONS	.12
6 RISK MANAGEMENT OPTIONS	12
6.1 Option 1 – Self-regulation.	. 12
6.2 Option 2 – Status Quo	. 12
6.3 Option 3 – Food regulatory measures	. 12
IMPACT ANALYSIS	13
	10
7. CONSULTATION AND COMMUNICATION	.13
7.1 Consultation	.13
	13
0. AFFECTED PARTIES	. 14 1 <i>1</i>
8.1.1 Seed production	14
8.1.2 Sprout production	.14
8.1.3 Wholesalers and Retail	.14
8.2 Consumers	. 14
8.3 Government	. 15
8.4 World Trade Organization notification	.15
9. SCIENTIFIC EVALUATION OF THE RISK	.15
9.1 Scientific/Risk Assessment	.16
9.1.1 MICrobiological hazards	10
9.1.2 Factors that impact on the presence of level of microbiological hazalus	17
9.1.4 Factors that impact on the presence or level of chemical hazards	17
10. Risk mitigation (control) measures	.17

CONTENTS

 11. ASSESSMENT OF OPTIONS	19 19 19 19
CONCLUSION	20
12.CONCLUSIONATTACHMENT 1 - THE SEED SPROUT INDUSTRYATTACHMENT 2 - MICROBIOLOGICAL HAZARD EVALUATION OF SEED SPROUTS	20 21 28
ATTACHMENT 3 - CHEMICAL HAZARD EVALUATION OF SEED SPROUTS ATTACHMENT 4 - COST ESTIMATE OF THE 2005-06 AUSTRALIAN SEED SPROUT OUTBREAKS ATTACHMENT 5 - REGULATORY MEASURES APPLYING TO SPROUT PRODUCTION IN AUSTRALIA	40 43 46
ATTACHMENT 6 - SUMMARY OF INTERNATIONAL GUIDELINES/CODES OF PRACTICE ATTACHMENT 7 - SDC MEMBERSHIP REFERENCES	50 53 55

Introduction

1. Primary Production and Processing Standards

Since June 2002, Food Standards Australia New Zealand (FSANZ) has had responsibility for developing national food safety requirements that cover all parts of the food supply chain – an integrated paddock-to plate approach. To this effect, FSANZ has been developing primary production and processing standards for identified industry sectors for inclusion in the *Australia New Zealand Food Standards Code* (the Code).

A primary production and processing standard is a set of obligations on primary producers and processors of food commodities. These obligations include measures to control food safety hazards that could occur during the production and processing of agricultural produce. Such measures may include requirements for:

- control of inputs
- premises and equipment
- health and hygiene
- skills and knowledge
- storage and transportation
- traceability

Primary production and processing standards are incorporated into Chapter 4 of the Code and apply in Australia only. Along with other standards in the Code they provide an approach to managing food safety and suitability¹ in Australia that extends from production on the farm through to sale to the consumer. The process for developing such standards takes into account existing food safety requirements implemented by the sector, including any existing regulations (e.g. State legislation), industry codes of practice or guidelines and accredited food safety systems.

To date, FSANZ has developed primary production and processing standards for the seafood and dairy sectors and is currently assessing and developing standards for the poultry meat, and egg sectors. Work has also begun on meat and meat products. Concurrently, there have been preliminary scoping activities looking at the area of plants and plant products (e.g. fruit, vegetables, nuts, seed sprouts, fresh cuts) to consider how best to progress work on such a wide range of plant commodities. During this process the production of seed sprouts² has been identified as an area of public health concern (two outbreaks of food-borne illness in Australia were attributed to the consumption of seed sprouts in 2005-2006). Consequently FSANZ is now considering the development of a primary production and processing standard for seed sprouts.

A Standard Development Committee (SDC) consisting of representatives from the industry, retail, government regulators and consumers has been established by FSANZ to advise on this standard development Proposal.

¹ The term 'unsafe and unsuitable' covers hazards that could affect the health of consumers as well as levels of contaminants and residues which, while not unsafe, are in excess of the limits in the Code.

² Seed sprouts are sprouted seeds or beans (such as mung beans, alfalfa, mustard seed, onion, radish, soya bean etc) generally used and consumed as a salad vegetable.

2. Seed sprouts

2.1 Scope

Seed sprouts are a germinated form of seeds³ that are commonly consumed raw. There is a wide range of seeds that can be used for sprout production including (but not limited to) alfalfa, onion, radish, broccoli, sunflower, cress, snow peas, lentils, peas and mung beans. In Australia, bean sprouts, alfalfa sprouts (germinated lucerne seed) and snow pea sprouts are the main sprouts produced.

This Proposal will examine possible food safety measures that can be applied to primary production and processing of green sprouts (e.g. alfalfa sprouts, onion sprouts, radish sprouts etc.), bean sprouts (which are primarily produced from mung beans) and snow pea shoots.

2.2 The production chain

There are two aspects to the seed sprout industry – the production of seeds and the production of sprouts. In determining appropriate interventions across the supply chain, Proposal P1004 will look at both the seed production sector and the sprout production sector. The production stages to be considered in this process are outlined below. Further information on the industry and production processes is provided in Attachment 1.

2.2.1 Seed production

Seed production involves pre-harvest and post-harvest activities. These are presented in this report as on farm seed production and seed processing/grading.

In general, on farm seed production involves the following steps:

- field preparation/planting
- growth (including flowering and seed setting)
- seed harvest
- storage
- transport

At each of these steps, there are handling activities, equipment and inputs (e.g. irrigation water, fertilisers, agricultural chemicals) that need to be considered in determining possible food safety control measures.

Once seeds are transported to seed processing facilities there are a number of other steps involved:

- seed receipt/storage
- seed cleaning
- bagging
- storage
- transport

³ This Proposal is concerned with seeds from legumes, pulses, brassicas, bulb and root vegetables, and oilseeds such as sunflower seeds which, when sprouted, are used and consumed as salad vegetables. Sprouted forms of cereal grains (wheat, barley, oats etc.) which are used in the brewing industry or in juice manufacture (e.g. wheat grass) are excluded from the scope of this Proposal.

The handling activities undertaken, equipment used and premises/facilities for storage and processing also need to be considered in determining possible food safety control measures for seed production.

2.2.2 Sprout production

While there is a range of sprouted seed products, the steps generally involved in sprout production include:

- seed receipt/storage
- seed disinfection
- seed soaking
- germination/growth
- harvest
- washing/drying (depending on the variety and how it is grown)
- packaging
- chilling/storage
- transport

Given the range of seeds that can be sprouted, sprouting may occur in bins, tubs, punnets or beds, or involve a growth medium. Temperature and humidity conditions used during sprouting may also vary depending on the product. Possible food safety control measures that impact on the safety of sprouts need to have regard to all steps involved, from receipt of raw materials through to transportation of the final product to retail/wholesale.

The Problem

Seed sprouts contaminated by pathogenic micro-organisms present an unacceptable health risk to consumers. In recent years, outbreaks of food-borne illness have been associated with the consumption of seed sprouts both in Australia and overseas. Factors contributing to these adverse health events include:

- the inherent nature of the product (e.g. a ready-to-eat product in which the production process supports the growth of microbial pathogens if present)
- scientific uncertainty around the most effective pathogen mitigation steps
- a lack of through-chain risk mitigation measures (either regulatory or non-regulatory).

Since the most recent food-borne illness outbreaks in Australia in 2005-2006, seed sprouters have formed an industry association and developed industry guidelines to support the safer production of their products. However, the seed sprout industry consists of many small⁴ businesses and to date it has been difficult to achieve adequate coverage of the industry and comprehensive uptake of the guidelines. The industry association has sought government intervention and the development of regulatory measures (as appropriate) for the industry.

In regard to chemical hazards from seed sprouts, the limited data available do not indicate that these is a major concern for seed sprouts and, at this time, FSANZ does not consider there is a need for this issue to be further assessed (a Chemical Evaluation of Seed Sprouts is provided at Attachment 3).

⁴ The Australia Bureau of Statistics (ABS) defines a small business to be any business with less than 20 employees.

3. Public Health Risk⁵

3.1 Outbreaks of food-borne illness associated with seed sprouts

In the period 1988 to 2008, there have been over 40 reported outbreaks of food-borne illness worldwide attributed to consumption of contaminated seed sprouts. The most commonly reported aetiological agents in these outbreaks have been various serovars of *Salmonella* spp. and enterohaemorrhagic *Escherichia coli* (EHEC). Alfalfa and mung bean sprouts have been the most commonly reported seed sprouts implicated in these outbreaks of food-borne illness.

Most recently there have been two outbreaks of *S*. Oranienburg in Australia attributed to the consumption of alfalfa sprouts. From November 2005 to January 2006, there was an outbreak in Western Australia with 125 cases of salmonellosis reported, resulting in 11 hospitalisations. In May 2006, another outbreak of S. Oranienburg was reported in Victoria, with a total of 15 cases and two hospitalisations.

Outbreaks of food-borne illnesses are sporadic and unpredictable. An estimation of the cost of food-borne illness resulting from consumption of contaminated seed sprouts cannot be generated as an annual figure because of the sporadic and infrequent nature of such outbreaks. In this Proposal, the potential cost of adverse health consequences due to consumption of contaminated seed sprouts is estimated using the 2005-2006 outbreak data.

It has been estimated that for every one reported case of food-borne illness in the community there are 9 unreported cases⁶. Taking this underreporting into account, up to 1320 cases of salmonellosis may have been associated with the consumption of contaminated sprouts in Western Australia and Victoria during 2005-2006. When the productivity, welfare and medical costs are taken into consideration, this translates to an estimated social cost of \$AUD11.60 million⁷.

3.2 Factors contributing to risk

Epidemiological investigations suggest contaminated seed is the likely source of most, if not all, sprout-associated outbreaks.

Seeds and beans used for sprouting are raw agricultural products. During production in the field or during storage and conditioning, they may be exposed to pathogenic bacteria from a variety of sources such as contaminated soil, water, animal manure (grazing animals or applied as fertiliser), farming and processing equipment, rodents, insects, wild birds, and agricultural waste. There is, however, little specific data on how seeds used for sprouting become contaminated with pathogens during production or the relative contribution of potential sources of contamination.

If pathogenic bacteria are present on the seed or in the sprouting environment, the environmental conditions applied during the sprouting of seeds (moist conditions at temperatures of 20-30 °C) become ideal for their exponential growth. Therefore, even with very low initial numbers of pathogenic bacteria, there is the potential for pathogens to grow rapidly to high numbers during the sprouting process.

⁵ The information provided in this section is sourced from a Microbiological Evaluation of Seed Sprouts provided at Attachment 2.

⁶ Hall G. et al. (2005) Estimating food-borne gastroenteritis, Australia. Emerging Infectious Diseases 11(8): 1257-1264.

⁷ Refer to Attachment 4 for cost estimation.

As sprouts are a raw ready-to-eat product, there are no terminal processing steps (such as heat treatment) that can then be applied to eliminate any pathogenic micro-organisms that may be present. This means that the control of potential hazards must start with the seed itself and consider what measures can be implemented to reduce the likelihood of pathogens being present on seeds and during sprout production – a through-chain approach from seed production to seed sprouting.

3.3 What controls are effective?

Seed sprout safety has been assessed internationally by a number of countries and through forums such as the Codex Alimentarius Commission (Codex)⁸. Codex has developed a *Code of Hygienic Practice for Fresh Fruits and Vegetables* (CAC/RCP, 2003) which includes an annex specific to sprout production (ANNEX II Annex for Sprout Production). In general, these documents highlight the importance of implementing relevant Good Agricultural Practices for seed production and Good Manufacturing Practices for sprout production to minimise the presence of hazards. In addition there are three specific measures which have been identified for minimising risk:

- testing of seed lots for microbial pathogens
- microbiological decontamination of seed (seed sanitation) prior to use (by, for example, chemical treatment)
- pathogen testing of spent irrigation water.

While a number of scientific investigations (particularly in relation to seed sanitisation) have been undertaken around these control measures, and are described in Attachment 2, there is uncertainty and variability in terms of what is or should be achieved by them. Further examination of these measures will take place during the second assessment of the Proposal and in light of any additional information received during the public consultation.

4 Existing Requirements

4.1 Regulatory⁹

4.1.1 Australia New Zealand Food Standards Code

4.1.1.1 Chapter 3 – Food Safety Standards

Standards 3.2.2 - Food Safety Practices and General Requirements and 3.2.3 - Food Premises and Equipment set out specific requirements for food businesses, food handlers and the food premises and equipment with which they operate to ensure the safe production of food. The Chapter 3 Food Safety Standards apply in Australia only and apply to all food businesses involved in the handling of food intended for sale but specifically do not apply to primary food production¹⁰. Where food safety requirements are required for primary production activities they are developed as primary production and processing standards in Chapter 4.

⁸ The Codex Alimentarius Commission is the international body that develops food standards, guidelines and related texts such as codes of practice under the Joint FAO/WHO Food Standards Programme.

⁹A more detailed summary of existing regulatory measures is provided at Attachment 5.

¹⁰ Primary food production means the growing, cultivation, picking, harvesting, collection or catching of food and includes transportation or delivery, and the packing, treating (such as washing) or storing of food on the premises on which it was grown, cultivated, picked etc.

Seed producers are primary producers under the definition in the Code. In regard to sprout producers, while their growing and processing operations may involve a number of the food handling activities generally undertaken by food businesses (storing, cooling, and packaging), they also meet the definition of primary food production and Chapter 3 requirements have not been applied to them.

4.1.1.2 Chapter 1 – General Food Standards

The food standards in Chapter 1 apply to all food sold or traded at retail and wholesale level in Australia and New Zealand (except Standards 1.6.2 and 1.4.2 that apply to Australia only). These standards include labelling requirements; the maximum permitted levels for additives, processing aids, contaminants and natural toxicants; Maximum Residue Limits for agricultural and veterinary chemical residues; requirements for articles and materials in contact with food; and microbiological limits for food.

A microbiological limit has been set specifically for seed sprouts (described as cultured seeds and grains) in Standard 1.6.1 where *Salmonella* should not be detected in 25 g.

4.1.2 State-based requirements

As both seed and sprout producers are considered primary food producers under State and Territory Food legislation, any regulatory measures applying to them would need to have been developed under State or Territory primary production legislation (in lieu of any standards in Chapter 4 of the Code). To date, NSW¹¹ is the only jurisdiction to have developed requirements for sprout producers under the NSW Food Safety Scheme legislation. This essentially requires seed sprout businesses in NSW to:

- develop and implement an audited Food Safety Program (HACCP plan)
- verify their Food Safety Program through pathogen testing (spent irrigation water)
- have support programs, which may include a maintenance program, approved supplier program, cleaning and sanitation program, pest control program etc.

These requirements for sprout producers in NSW were introduced in 2005 and only apply to the production of sprouts, not the growing of seeds.

4.1.3 Export requirements

Seed producers¹² who export seeds are regulated by the *Export Control Act* 1982, certain provisions of the *Export Control (Prescribed Goods- General) Orders* 2005, and the *Export Control (Plants and Plant Products) Orders* 2005.

Exporters must meet both the requirements of relevant export legislation and any importing country requirements¹³ for the Australian Quarantine and Inspection Service (AQIS) to provide the necessary documentation to enable products to be exported.

¹¹ NSW Food Authority

¹² The term 'seed producers' is also covering businesses that purchase seed from producers and process it (clean it, export it etc- but does not cover sprout producers)

¹³ PHYTO is AQIS's plant and plant product export conditions database which contains information about the conditions to export plants and plant products, including fruit, vegetables, seeds, grains, cut flowers and timber from Australia.

Mung beans are prescribed goods for the purposes of the export legislation regardless of the intended end use of the beans. Mung beans for export must be prepared for export and presented for inspection at a registered establishment (that complies with the General Orders and meets the specific requirements for example, structural, operational and hygiene requirements of the Plants and Plant Products Export Control Orders). Exporters must notify the intention to export and the mung beans and transport arrangement must be inspected. The beans must be free of pests, live animals, animal carcasses and animal droppings and must meet any other phytosanitary requirements of the importing country. Export certification will be provided if requirements are met but this not intended to provide assurance of the suitability of the mung beans for consumption.

AQIS of the Department of Agriculture, Fisheries and Forestry is responsible for enforcing the export legislation.

4.2 Industry measures

4.2.1 Seed producers

Mung bean producers have formed an industry association (Australian Mungbean Association) that comprises all sectors of the mung bean industry. An industry Code of Hygienic Practice for Whole Mung Beans¹⁴ has been developed and is promoted by the Australian Mungbean Association as a minimum standard with which the industry should comply. The mung bean Code of Hygienic Practice covers:

- hygiene requirements on the farm and during transport to the mung bean grading establishment
- design and facilities of the mung bean processing establishment
- hygienic requirements for the mung bean processing establishment
- hygienic processing requirements in the mung bean processing establishment
- storage and transport of the end-product
- reference sampling of finished product.

Lucerne producers have also formed an industry association (Lucerne Australia) to represent all sectors of the lucerne industry. Lucerne seed is primarily grown as a non-food crop for pasture. However, as lucerne seeds have been used to produce alfalfa sprouts, and problems with contaminated lucerne seeds have been raised, microbiological testing (coliforms, *E. coli, Salmonella, Listeria monocytogenes*) of seed lots has been implemented by some lucerne seed producers and/or processors. Additionally, growers have been investigating on-farm measures they can implement to minimise contamination of lucerne seeds by microbial pathogens on-farm.

4.2.2 Sprout producers

The production of seed sprouts in Australia is a relatively small industry undertaken by small, often family owned businesses (there are approximately 30 sprout producers located throughout Australia). Historically, they have had no industry association or representation.

¹⁴ Code of Practice is available on the Australian Mungbean Association website at: <u>http://www.mungbean.org.au/foodsafetyandhygiene.html</u>

Following the *Salmonella* outbreak in Australia in 2005-2006 attributed to seed sprouts, sprout producers have formed an industry association¹⁵ and in consultation with State jurisdictions, have developed a set of industry guidelines to support the safer production of seed sprouts. Currently, this Association represents just over half of the industry.

The Guidelines prepared by the Australian New Zealand Sprout Producers Association categorise sprouts into four risk categories:

- Category A alfalfa
- Category B all others including sunflower
- Category C snow pea shoots/sprouts
- Category D sprouts/shoots grown using a growing medium

The guidelines essentially specify seed sanitisation, sampling and microbiological testing protocols for each category, with an overarching requirement for the business to implement a HACCP based food safety program. Uptake of these guidelines is voluntary. There are currently no certification mechanisms for demonstrating compliance.

4.2.3 Retailers

One large retailer has developed produce specifications for seed sprout products supplied to it. While these specifications cover a number of quality attributes, they also cover safety and generally specify microbiological limits (generally for *E. coli, Listeria monocytogenes* and *Salmonella*) and criteria for Use By Dates (e.g. not to exceed a certain number of days from date of packaging). Where sprout businesses supply product under the retailer's own label, they must be accredited and audited against food safety and quality management schemes such as Woolworths Quality Assurance (WQA), Safe Quality Food (SQF) 2000 and BRC (British Retail Consortium). Currently only one supermarket chain supplies seed sprout products (not alfalfa sprouts) under its own label.

Objectives

5 Objective of the Proposal

The overall objective of government action is to minimise adverse health effects associated with the consumption of seed sprouts.

5.1 **Process for achieving the objective**

This Proposal will assess the need for and identify any appropriate through-chain control measures (regulatory and non-regulatory) that can be implemented nationally by industry to maximise the safety of seed sprouts. Possible regulatory and or non-regulatory options are identified in Section 6.

FSANZ uses an internationally agreed risk analysis approach to inform its regulatory decision-making processes. The risk analysis approach¹⁶ includes an assessment of the main hazards associated with this product and the factors along the supply chain that impact on the presence or level of that hazard. This will inform what control measures are needed and the extent to which potential hazards can or cannot be managed at steps in the chain to minimise public health risks.

¹⁵ Australian New Zealand Sprouters Association

¹⁶ Information on the risk analysis framework in which FSANZ operates is available on the FSANZ website at: <u>www.foodstandards.gov.au/aboutfsanz/scientificcapabilities/riskanalysis.cfm</u>

Risk management considers how the hazards identified by the scientific assessment are currently being managed (outlined in Section 4), whether there are any gaps that need to be addressed and what interventions are needed to address them. This has involved undertaking an audit of existing control measures that have been developed for seed and sprout producers (domestic and international). FSANZ will now assess how best these control measures can be met, be integrated into a set of nationally consistent approaches, and implemented through the options identified below.

5.2 Constraints

5.2.1 FSANZ Act

Where regulatory interventions are required (e.g. by developing or varying a food standard), FSANZ is required by its legislation to meet three primary objectives which are set out in section 18 of the FSANZ Act. These are:

- the protection of public health and safety; and
- the provision of adequate information relating to food to enable consumers to make informed choices; and
- the prevention of misleading or deceptive conduct.

In developing and varying food regulatory measures, FSANZ must also have regard to:

- the need for standards to be based on risk analysis using the best available scientific evidence;
- the promotion of consistency between domestic and international food standards;
- the desirability of an efficient and internationally competitive food industry;
- the promotion of fair trading in food; and
- any written policy guidelines formulated by the Ministerial Council.

5.2.2 Policy guidelines

The Australia and New Zealand Food Regulation Ministerial Council (Ministerial Council) developed an *Overarching Policy Guideline on Primary Production and Processing Standards*. This policy guideline specifies a number of high order principles for primary production and processing standards outlining that they will:

- be outcomes-based
- have a consistent regulatory approach across the Standards
- be consistent with the approach outlined in Chapter 3 of the Code
- be consistent with Codex standards
- address food safety across the entire food chain where appropriate

- facilitate trade and comply with Australia's obligations under World Trade Organization (WTO) Agreements
- promote consumer confidence
- ensure the cost of the overall system should be commensurate with the assessed level of risk
- provide a regulatory framework that only applies to the extent justified by market failure.

Any regulatory measures developed should be commensurate with risk and not impose any unnecessary additional economic burden on the sprout industry.

Options

6. Risk management options

In order to decide the most effective and efficient approach for achieving the objectives stated in Section 5, FSANZ must consider various risk management options. These options include the *Status Quo* (the situation if no action is taken) as a comparative measure against appropriate regulatory (government) and non-regulatory (industry) approaches. The options identified for Proposal P1004 (covering both seed and sprout production) are outlined below.

6.1 Option 1 – Self-regulation

A self-regulatory approach requires food or primary production businesses to be able to implement and enforce (e.g. through certification schemes) industry guidelines or codes of practice aimed at improving the safety of seed sprouts.

The success of such an approach needs strong industry wide commitment and evidence that voluntary participation can work through, for example, the ability to apply sanctions or incentives (such as using a product logo which demonstrates compliance with a food safety scheme) to achieve maximum participation. Under this option industry would be responsible for enforcement and there would be no government applied food regulatory measures.

6.2 Option 2 – *Status Quo*

Option 2, the *Status Quo*, is largely characterised by the current requirements outlined in Section 4 which reflect a mixture of regulatory and self-regulatory approaches developed for different pockets of the production chain of seed sprouts. Under the *Status Quo* there is no nationally consistent set of food safety control measures for sprout production.

6.3 Option 3 – Food regulatory measures

Option 3 involves the development of food safety regulatory measures in the Code (which may be supported by self-regulatory measures). These measures would apply to sectors in the production chain (on-farm seed production, seed processing, and sprout production) where cost benefit analysis can demonstrate such measures are commensurate with risk and are cost effective. Such requirements would be subject to the impact analysis which will evaluate the costs and benefits accruing to all stakeholders.

Option 3 may result in a combination of regulatory and non-regulatory measures. For example, regulatory measures could be introduced to control specific activities that pose the greatest risk with voluntary or self-regulatory measures helping to support these controls.

Any regulatory measures developed would be included in a primary production and processing standard in Chapter 4. If warranted, additional measures could be included such as specific labelling requirements or microbiological limits in other Chapters of the Code.

The role of consumer education to maximise seed sprout safety will be explored within this option.

Impact analysis

The Assessment reports on this Proposal will provide information to comply with the Council of Australian Governments (COAG) requirements for regulatory impact analysis. FSANZ will continue to consult with the Australian Government's Office of Best Practice Regulation on meeting these requirements.

The preferred option decided through the assessment of Proposal P1004 will be based on an analysis that considers:

- who is affected by the problem and the proposed solution
- scientific evaluation of the risks
- efficacy and practicality of risk mitigation measures (control measures) identified
- costs and benefits to affected parties of the interventions associated with each option.

7. Consultation and communication

7.1 Consultation

The FSANZ process for the development of a standard involves a consultative and transparent process that reaches the industry concerned, State and Territory Government enforcement agencies, as well as consumers. A SDC is established for each primary production and processing standard with representatives from the industry sector, the relevant State and Territory government agencies and consumer organisations to provide ongoing advice to FSANZ throughout the standard development process. The SDC contributes a broad spectrum of knowledge and expertise covering industry, government, research and consumers (a list of SDC members for this standard development Proposal is provided at Attachment 7). In addition, targeted consultations have been undertaken with seed producers/processors and seed sprout producers through on-site visits to glean first hand perspectives and information from these parties. Additional targeted consultations will be undertaken throughout the standard development process as required.

This Report has been developed in consultation with the SDC and provides the first opportunity, in accordance with FSANZ statutory consultation processes, for stakeholders to comment on and supply information to FSANZ in regard to Proposal P1004.

7.2 Communication

As the assessment of Proposal P1004 proceeds, FSANZ will report its progress on its website at

http://www.foodstandards.gov.au/standardsdevelopment/proposals/proposalp1004primary43 61.cfm.

Organisations or individuals with an interest in this Proposal can seek to have their names listed as an interested party by emailing the Standards Management Officer at <u>standards.management@foodstandards.gov.au</u> their full contact details.

8. Affected parties

Parties that have been identified as being affected by this Proposal include: industry (including those involved in seed production, sprout production and retail of seed sprouts), consumers of seed sprouts, State and Territory Governments, and member nations of the World Trade Organization (WTO).

8.1 Industry

8.1.1 Seed production

Seed production includes the growing and cleaning/grading of seed. Therefore, both growers and seed processing establishments may potentially be affected by this Proposal. In considering impacts on these parties, FSANZ has consulted and will continue to consult (through the SDC and targeted consultation as required) with:

- associations representing grower and processor interests such as Lucerne Australia and the Australian Mungbean Association
- seed processors such as Booborowie Seed Pty Ltd and Keith seeds.

8.1.2 Sprout production

The seed sprout industry consists of mainly small businesses (around 30 businesses known to FSANZ located throughout Australia). An industry association has been established (Australian New Zealand Sprouters Association) and FSANZ has consulted and will continue to consult (through the SDC and targeted consultation as required) with the members and other sprout producers in assessing impacts on this sector.

8.1.3 Wholesalers and Retail

While seed sprouts may be distributed directly to retail outlets from seed sprout businesses, a large proportion is distributed via fresh food wholesale markets.

Some supermarkets have implemented requirements for seed sprouts, particularly for their own brand products. While this is an issue of market access and the impacts on the retail sector may not be assessed directly, any implications on the existing arrangements and requirements will be considered in the cost benefit analysis.

8.2 Consumers

People generally consume seed sprouts because of health and culinary factors (e.g. the use of bean sprouts in Asian dishes). There is also 'indirect' consumption of seed sprouts where they are incorporated in dishes as a garnish.

There is very limited Australian or international information on the extent of sprout consumption. Data from the 1995 National Nutrition Survey (Australia) indicates that, at that time, approximately 4% of respondents consumed seed sprouts.

Alfalfa sprouts were consumed most frequently whereas bean sprouts were consumed in the largest quantities. Since that time the range of seed sprout products has grown as has their availability at retail outlets and use by the food service sector.

To minimise adverse health effects associated with seed sprout consumption, this Proposal will include assessing the impacts on consumers of seed sprouts who may be considered at higher risk from food-borne illness, such as the very young, the elderly and the immunocompromised as well as the general population.

8.3 Government

Currently only NSW (NSW Food Authority) has developed and implemented specific regulatory requirements for seed sprout businesses. Any change from the status quo will mean State and Territory jurisdictions may need to consider implementation and enforcement costs associated with the specific application of possible food regulatory measures for the seed sprout sector developed as a result of this Proposal.

8.4 World Trade Organization notification

As members of the WTO, Australia and New Zealand are obligated to notify WTO member nations where proposed mandatory regulatory measures are inconsistent with any existing or imminent international standards and the proposed measure may have a significant effect on trade.

This issue will be fully considered during the further assessment of the Proposal and, if necessary, notification will be recommended to the agencies responsible in accordance with Australia's obligations under the WTO Technical Barriers to Trade (TBT) or Sanitary and Phytosanitary Measures (SPS) Agreements. This will enable other WTO member countries to comment on proposed changes to standards where they may have a significant impact upon them.

FSANZ invites comment and information in relation to the parties that may be affected by this Proposal.

9. Scientific evaluation of the risk

In deciding the preferred option to achieve the Objectives identified for this Proposal, an assessment of the possible measures to mitigate public health risks posed by consumption of seed sprouts will be undertaken. This will include:

- examining the scientific evidence that identifies the main hazards associated with seed sprouts and the factors along the supply chain that impact on the presence or level of that hazard
- assessing the factors that impact on the presence or level of these hazards in order to identify what control measures are needed and the extent to which potential hazards can or cannot be managed at steps in the chain to minimise public health risks.

9.1 Scientific/Risk Assessment

9.1.1 Microbiological hazards

A review of the scientific literature has been undertaken to identify and elaborate the public health risks associated with seed sprouts (Attachment 2) covering:

- outbreaks of food-borne illness associated with seed sprouts
- prevalence and levels of pathogens in seed sprouts
- the potential for pathogen contamination and/or proliferation during seed production and seed sprouting.

From reports of outbreaks of food-borne illness associated with seed sprouts, the main hazards identified have been *Salmonella* spp. and enterohaemorrhagic *E. coli* (EHEC). Surveys of sprouts have also demonstrated contamination by *L. monocytogenes*, coagulase-positive staphylococci, and *Bacillus cereus*.

Between 1988 and 2008, there have been over 40 reported outbreaks worldwide attributed to consumption of seed sprouts contaminated by pathogenic micro-organisms. The most commonly reported aetiological agents in these outbreaks have been various serovars of *Salmonella* spp. and EHEC. Alfalfa and mung bean sprouts have been the most commonly reported seed sprouts implicated in outbreaks of food-borne illness.

An outbreak due to *S*. Oranienburg occurred in Western Australia during November 2005 – January 2006 that was epidemiologically linked to consumption of alfalfa sprouts. This was later confirmed microbiologically, with *S*. *Oranienburg* being isolated from the implicated alfalfa sprouts. A total of 125 cases of salmonellosis were reported, resulting in 11 hospitalisations. In May 2006, another outbreak of *S*. *Oranienburg* was reported in Victoria, with a total of 15 cases attributed to consumption of alfalfa sprouts. In the latter outbreak, *S*. Oranienburg was isolated from the implicated alfalfa sprouts as well as from seed obtained from the sprouting facility. Molecular typing of the *S*. Oranienburg isolates from both the Victorian and Western Australian outbreaks showed indistinguishable patterns by pulsed field gel electrophoresis (Pers. Comm. Martyn Kirk, OzFoodNet, 23 April 2009). Trace back of seeds associated with these outbreaks found that seed originated from the same Australian state but from different seed suppliers.

Surveys conducted by various Australian State and Territory Governments between 2000 and 2008 have found occasional contamination of seed sprouts with pathogenic microorganisms. Potential microbiological pathogens detected include *Salmonella* spp., EHEC, *L. monocytogenes*, coagulase-positive staphylococci, and *B. cereus* (Attachment 2).

9.1.2 Factors that impact on the presence or level of microbiological hazards

Seeds grown for sprouting are raw agricultural products, and as such, may be exposed to microbiological pathogens from a variety of sources. These sources include soil, water, animal manure (grazing animals or manure applied as fertiliser), farming equipment, rodents, insects, wild birds and agricultural wastes (Attachment 2). Studies have found that, once attached to seeds, pathogens such as *Salmonella* and EHEC can survive for long periods of time under normal seed storage conditions. Contamination of seed with pathogens is considered to be sporadic and at low-levels, with concentrations of 1-100 MPN per kg of contaminated seed being reported in the literature.

Conditions during the sprouting of seeds (temperatures of 20-30°C, presence of water and availability of nutrients) permit the rapid growth of microorganisms if they are present.

To minimise and/or eliminate the potential for pathogen growth during sprouting, seeds are often sanitised prior to germination. There have been extensive investigations into the efficacy of sanitisation in reducing levels of pathogenic micro-organisms in contaminated seeds. Consensus amongst the scientific literature is that sanitising reduces, but does not necessarily eliminate pathogens from contaminated seed. Most seed sprouts are consumed raw and will therefore not receive any form of heat treatment prior to consumption.

9.1.3 Chemical hazards

Chemical hazards such as residues of agricultural and veterinary chemicals on seeds and beans, contaminants, processing aids, food additives and packaging material which are likely to be associated with seed sprouts, have been reviewed. The details of this review can be found in Attachment 3 to this report.

There are limited data currently available but these data do not indicate that chemical hazards are a major concern for seed sprouts.

9.1.4 Factors that impact on the presence or level of chemical hazards

Based on the available information, the current regulatory measures, including those in the Code, are considered adequate with respect to managing chemical hazards in seed sprouts. At this time and subject to any further data becoming available, FSANZ does not consider that there is a need for this issue to be further assessed under this Proposal.

FSANZ invites comment and information in relation to the scientific evaluation of the risks associated with seed sprouts.

10. Risk mitigation (control) measures

A number of guidelines and codes of practice have been developed internationally for seed sprout production including¹⁷:

- Codex Code of Hygienic Practice for Fresh Fruits and Vegetables Annex II Annex for Sprout Production
- Canadian Code of Practice for the Hygienic Production of Sprouted Seeds
- Reducing Microbial Food Safety Hazards for Sprouted Seeds Guidance for Industry (US FDA)
- Code of Practice for Food Safety in the Fresh Produce Supply Chain in Ireland (Chapter 4: Microbiological Safety of Sprouted Seed Production).

In Australia, specific food safety measures have been applied in NSW to seed sprout businesses under the NSW Food Safety Scheme legislation, and voluntary guidelines have been developed for the industry in South Australia by Primary Industries and Resources SA.

¹⁷ A summary of international guidelines/ codes of practice is provided at Attachment 6

A review of the control measures specified by these documents has been undertaken to identify what is considered best practice for sprout production.

The main control measures identified in these guidelines/codes of practice to prevent or eliminate microbial pathogen contamination in sprouted seeds are outlined in Table 1.

These control measures are generally supported by the scientific evidence and three specific control measures for sprout production have been identified:

- the testing of seed lots for microbial pathogens
- microbiological decontamination of seed (seed sanitisation) prior to use (by, for example, chemical treatment)
- pathogen testing of spent irrigation water.

The next stage of this standard development Proposal will look at any issues around the effectiveness, practicality and feasibility associated with these controls to determine how best they can be met and implemented.

FSANZ invites comment and information in relation to the efficacy and practicality of risk mitigation measures (control measures) identified.

Table 1: Summary of the main control measures identified within existing guidelines/codes of practice for sprout production.

Step:	Main Controls:
Seed	Good Agricultural Practices in particular:
production	Management of grazing animals on farm
	Management of fertilizers and other inputs
	Minimising damage to seed (damaged seed should not be used for sprouting)
	Segregation of seed for sprouting from seed for animal feed production
	Protection of seed during storage, packaging and transport from pests and environmental contamination.
Sprout	Good Hygienic Practices in particular
production	Minimise contamination from equipment and premises, sprout contact surfaces, sprout handlers etc.
	Use of potable water
	Have cleaning and sanitising and pest control programs etc.
	Approved seed supplier (evidence that the seed has been sourced under appropriated production practices and is not contaminated by pathogens)
	Microbiological testing of seeds to verify absence of pathogens
	Seed disinfection - using approved sanitiser at correct concentration and contact time
	Rinsing of final product (Use of chilled water/ use of sanitiser [2-4 ppm free available chlorine])
	Testing for pathogens recommended by sampling of irrigation water (also called spent water) collected within 48 hours from start of sprouting.
Storage and distribution	Temperature control (≤ 5°C)

11. Assessment of options

FSANZ, with advice from the SDC taking into consideration submissions made on this report, will undertake a detailed impact analysis of the costs and benefits to each affected party posed by each option. This assessment, together with the preferred option, will be detailed in the SECOND Assessment Report.

A preliminary assessment of the options is provided below.

11.1 Option 1 – Self–regulation

Self-regulation as a sole option across all sectors of the seed sprout industry is not considered viable. This is reflected by the fact that significant efforts invested by the Australian government through the Implementation Sub-Committee in the past three years have made very little progress in getting sprout producers to self-regulate. As described in the problem section, the Australian New Zealand Sprouters Association has met with difficulties in having its guidelines for safe production of sprouts taken up by the producers and has subsequently sought government intervention and the development of regulatory measures (as appropriate) for their industry.

Despite this, self-regulatory measures developed by the industry may be utilised to supplement the regulatory approach outlined under Option 3 and an analysis of the feasibility of such an approach will be undertaken during the preparation of the Second Assessment Report for this Proposal.

11.2 Option 2 – Status Quo

Status Quo refers to the situation if no action is taken.

Other than the government effort to facilitate industry self-regulation and the development of draft guidelines for safe production of seed sprouts by the Australian New Zealand Sprouters Association, little change in improving the safety of seed sprouts has occurred nationwide since 2006 (except in NSW where there is a food safety scheme for seed sprouts). This indicates that *status quo* is unlikely to be a viable option.

FSANZ assessed the economic impact of possible adverse health consequences of supplying unsafe seed sprouts to Australian consumers. The FSANZ evaluation estimated that community costs due to food-borne illness arising from consumption of pathogen contaminated seed sprouts could be in the range of \$AUD11.60 million based on the 2005-2006 outbreak data (see Attachment 4). This cost estimate did not taken into consideration of possible costs to industry and government such as loss of reputation, closure of business, loss of employment, fines and food recall costs.

11.3 Option 3 – Food Safety Regulations

In relation to Option 3, FSANZ is seeking input from stakeholders to provide cost and benefit information in light of any potential activities that may be required under a regulatory regime to achieve the desired food safety objectives.

As an example of information that may inform this assessment, the NSW Food Authority has recently undertaken a microbiological survey of seed sprouts produced in NSW (*Report on the Microbiological Quality of Sprouts*) as part of its evaluation of the NSW Plant Products Food Safety Scheme for seed sprouts.

In general, the results indicate that the microbiological quality of sprouts has improved¹⁸ since requirements were implemented by seed sprout businesses in that State (a baseline survey was undertaken in 2005), noting that the surveys cannot be compared statistically given the relatively small number of samples taken. Information on enforcement and compliance costs will also be useful.

In relation to seed production and processing, it is noted that current measures are largely industry driven (apart from export requirements) with improvements in production practices linked to market access and pricing incentives. As an example, lucerne seed producers have been looking at possible actions they could take that may reduce the likelihood of hazards being present. A control measure of not having livestock on lucerne paddocks for a full 12 months is estimated to add an additional cost of about \$AUD1.00-1.50 to a kilogram of seeds. However, there would be no guarantee that the seed would reach sprouting quality and that the additional cost incurred would be recouped (or that contamination further down the chain would be prevented).

FSANZ invites comment and information on the costs and benefits of the proposed risk management options from affected parties.

Conclusion

12. Conclusion

This First Assessment Report provides an opportunity for stakeholders to comment on and supply information to FSANZ in regard to Proposal P1004.

To assist FSANZ undertake a comprehensive and informed impact analysis of the proposed options, affected parties are encouraged to provide comment and information on the issues raised in the report. The comments and information provided during this consultation will be considered during the second assessment stage of the Proposal when a preferred option for implementing national through-chain control measures for the seed sprout industry will be proposed.

Attachments

- 1. The Seed Sprout Industry
- 2. Microbiological Hazard Evaluation of Seed Sprouts
- 3. Chemical Hazard Evaluation of Seed Sprouts
- 4. Estimated Costs of Australian Outbreaks
- 5. Regulatory Measures Applying to Seed Sprout Production in Australia
- 6. Summary of International Guidelines/Codes of Practice
- 7. SDC members

¹⁸ This determination is based on testing of hygiene indicators (*E. coli*).

The seed sprout industry

1. Seed Production

Seed production involves pre-harvest and post-harvest activities – from the growing of seed through to seed processing/grading and transport. This section discusses these activities under two main areas: on-farm production activities and seed processing.

1.1 On-farm seed production

There is a wide range of seeds that can be used for sprouting and thus a diverse range of agricultural practices may be associated with seed production. Crops involved may have annual or perennial production cycles and may not exclusively be grown for seed production. For example, beans, such as mung beans and soybeans, are annual crops and the seeds are harvested once per annum in autumn. Lucerne (alfalfa) on the other hand, is a perennial crop and is subject to grazing or repeated harvests for hay, with lucerne seeds harvested once per year.

The steps generally involved in seed growing are presented in a flow diagram below. As alfalfa sprouts and mung bean sprouts are two of the main seed sprouts produced in Australia, this section outlines the production systems used for lucerne seed (perennial crop) and mung bean (annual crop) production.



Figure 1: The steps involved in on-farm seed production, from planting to transport

1.1.1 Lucerne Production¹⁹

Lucerne is a perennial plant which has a life-span of 6-15 years depending on variety and crop management. It is grown predominantly as a fodder crop, either grazed directly, or made into silage or hay first. Lucerne pastures are drought-resistant perennials that produce green feed in all seasons.

More than 80% of lucerne seed production in Australia takes place in South Australia with the remaining occurring in NSW and Victoria (Hassall & Associates Pty Ltd, 2001; De Barro, 2005). Lucerne seed production within South Australia is centred in the region of Keith, Bordertown and Jamestown. In NSW, lucerne production takes place from Howlong through to Corowa.

The volume of lucerne seed produced in Australia varies from 4,000 to 7,000 tonnes per year. While most of this seed is exported as pasture seed, a proportion of lucerne seed is used to produce alfalfa sprouts for human consumption. Industry data indicates that an estimated 300 tonnes of lucerne seeds have been used for sprout production in Australia in 2006 and approximately 600 tonnes of exported lucerne seeds (~10% of all exported seeds) are used for sprout production in international market. With an estimated market price of \$AUD5.0 to \$AUD5.50 per kilogram of lucerne seeds, the value of lucerne seeds used for sprout production in Australia is approximately \$AUD1.5 million.

1.1.1.1 Paddock management

There are a number of management strategies that may be used by growers in the 12 months prior to harvest involving grazing and hay cutting. It is common practice to allow the grazing of animals (sheep and cattle) on a rotational basis in the period leading up to seed harvesting. For example, paddocks may be grazed up to October-December, which removes most of the vegetation from the paddock and encourages flowering of the plant. Alternatively, animals may be excluded from paddocks in July-August, and the crop cut for hay in October to early December. Therefore the amount of time that paddocks are kept free of grazing animals prior to seed harvesting could be approximately 120-150 days or 190-220 days, depending on crop management strategy.

For irrigated lucerne production, crops may be flood irrigated between one to six times per season, or three to ten times for spray irrigation, depending on local conditions. In Australia lucerne seed is harvested once a year only, in February, March and April.

1.1.1.2 Harvest

The lucerne plant flowers in January–February, at which time pollination occurs and the seed is set. Once set, the lucerne seeds develop within an enclosed seed pod, with each pod containing up to 12 or more seeds.

Harvesting of lucerne seeds can be undertaken by two methods:

- cutting and windrow curing followed by threshing with a combine harvester; or
- chemical desiccation followed by direct harvesting of the standing crop.

Windrowing involves cutting the crop just above the crown of the plant and laying the foliage in rows (windrows) on the ground. The plant material is allowed to dry for a number of days, until the moisture content of the foliage falls to approximately 12-18%.

¹⁹ Information on production practices supplied by Lucerne Australia

Once the plant material is suitably dry, it is picked up using a harvester which works close to the ground. Inevitably, the harvester will also pick up extraneous material from the ground including soil and other potential contaminants. The plant material is then threshed inside the harvester to separate the seed from the other material (e.g. stalks).

For direct harvesting, crops are sprayed with a chemical desiccant/defoliant, allowed to dry, then collected using a header and harvested.

Harvested seed is generally collected in mobile field bins, which are either stored on-site or transported to a seed processor for cleaning and packing.

1.1.2 Mungbean production

Mungbean is a specialised food crop which is cooked as whole beans, sprouted or processed into flour. It is predominantly produced in Australia for the export market.

The bulk of mung bean production in Australia is in central and southern Queensland and northern NSW. Production volumes vary from 30,000-50,000 tonnes annually depending on seasonal factors, varieties cultivated and farm management practices. The crop produced is graded as sprouting grade beans (which return the highest price per tonne), cooking grade beans or processing grade beans.

In 2006-07 it was estimated that the Australian mung bean industry produced 38 974 tonnes of mung beans of which 1,325 tonnes were used for sprouts. Most of Australian production falls into processing grade (medium quality) seed.

1.1.2.1 Crop management

Mungbean is a warm season annual pulse grown mostly in rotation with other crops such as cereals. Plants have a short growth duration (75-90 days) which means that it can fit easily into crop rotations. Sowing times vary depending on the location and variety grown.

Mungbean crops are managed with the aim of producing premium grade seed. Factors that need to be considered to maximise the yield include:

- choice of paddock (e.g. no soil variation, adequate soil moisture profile)
- seed variety
- time of planting
- planting rates
- pests and diseases
- seasonal variability

Mungbeans may be grown under dry land or irrigated crop production systems.

1.1.2.2 Harvest

Mungbeans have an indeterminate flowering habit. This means that they do not have a defined flowering period and consequently, can have flowers, green pods and black pods present on the plant at the same time. Harvest occurs when more than 90% of pods are mature and dry.

To minimise damage to seeds, they are harvested at seed moisture contents of 14-16%. A desiccant is often used before harvest to kill any green leaf and the few remaining green pods.

Mung bean plants grow erect with few branches carrying clusters of pods (containing 8-15 seeds) near the top of plants. Pods are mechanically harvested by combine harvesters.

Following harvest beans are trucked to a grading shed where they are cleaned, graded and bagged as soon as possible (seed processing).

1.2 Seed processing

Seed harvested from the field may contain extraneous material such as soil, weed seeds and other debris. This is removed during seed processing, whereby the seed received from the field is passed through a series of sieves (4-5 screens of different pore sizes) and then further cleaned via use of a gravity table, where seeds are separated by their weight. Once cleaned, seed is packed into 40 kg bags, or larger containers for the bulk seed market, and stored on-site prior to shipping. Seed purchased by sprouters is generally required in 25 kg bags.



The main steps that are generally involved are presented in Figure 2.

Figure 2: The steps involved in seed processing

1.2.1 Seed Quality - Alfalfa

The accepted market quality of sowing lucerne seed is²⁰:

- minimum rate of germination: 85%
- minimum normal seedling: 60%.

For sprouting purposes, sprout producers specify that seed should have a 4-day minimum germination rate of approximately 90% and a maximum hard/abnormal/dead seed count of approximately 10%. Germination rates are variable and seed suitable for sprouting cannot be assured until a seed germination test is performed.

²⁰ De Barro, J. (2006) Presentation from Lucerne Australia, Presented at the workshop of "Food Safety and Sprouts", held at the Tiffins on the Park, Adelaide, 20 July 2006.

Where there is a high hard seed count, the germination rate can be increased by scarifying the seeds during processing or by leaving seed in storage. Scarification is a process whereby the seed coat is broken or scratched. This makes it permeable to water and gases and thus aids germination. During seed processing this can be achieved by a mechanical process using spinning abrasive discs against which seed is dropped then collected.

Seeds used for production of seed sprouts are largely grown and processed in Australia.

2. Sprout production

Seed sprouts can generally be subdivided into three groups:

- green sprouts (e.g. alfalfa sprouts, onion sprouts, radish sprouts)
- bean sprouts/bean shoots (produced primarily from mung beans)
- shoots and grasses (e.g. snow pea shoots, wheat grass).

The scope of this proposal is green sprouts, bean sprouts and snow pea shoots.

2.1 Sprout producers

The seed sprout industry consists of approximately 30 small businesses located throughout Australia. Around two thirds of these businesses have been in operation for 16 years or longer with many being family owned and operated. While the majority of sprout businesses produce a range of products, a number of operations are single product producers (e.g. producing just bean sprouts or snow pea sprouts).

The sprouting industry has an annual turnover of approximately \$AUD30 million, employing approximately 300 people and producing more than twelve product lines. One of the largest sprout production businesses has a turnover of approximately \$AUD5.5 million per annum.

Mungbean sprouts are the predominant product (by volume) produced followed by snow pea sprouts and then alfalfa sprouts.

2.2 Sprout growing

Methods used for the germination and growth of seed sprouts, including water cycles used, vary depending on the type of sprout being produced and the size/sophistication of the sprouting operation. The basic process involves applying water to seeds and placing them in a warm humid environment for a period of 1-14 days. Sprouting may take place in temperature-controlled environments, which maintain air temperatures at 20-30°C, or at ambient temperatures. Once the sprout has reached the size required it is harvested, chilled and packaged ready for storage and distribution. The typical production steps undertaken are outlined in Figure 3 and discussed further below.



Figure 3: Typical production steps during production of seed sprouts

2.2.1 Seed disinfection

Prior to sprouting, seed should be washed to remove any dirt or debris. It is then recommended that seeds are treated with an antimicrobial agent, such as chlorine, prior to the sprouting process. Recommended seed disinfection regimes vary, however a treatment of 20,000 ppm calcium hypochlorite has been commonly suggested for alfalfa seed.

Following seed disinfection the seed is again rinsed to remove the antimicrobial agent used.

2.2.2 Pre-germination soak

Soaking is undertaken to improve germination. Seed is commonly soaked in potable water for 3 to 10 hours (depending on seed variety) at ambient temperature. Sufficient quantities of water need to be used during the soaking process as seed swells and may double its volume during this time.

2.2.3 Germination/Growth

The sprout growing process varies depending on the variety of seed being germinated and the sophistication of the sprouting operation. Examples of typical protocols for alfalfa sprouts, bean sprouts and snow pea shoots are outlined below.

2.2.3.1 Alfalfa sprouts

Alfalfa sprouts (including mixes) may be grown in large rotating drums/ tumblers (for larger operations) or in trays or punnets over a 3-6-day period. The sprouts are continually watered during this time and runoff water (spent irrigation water) removed. For businesses that have temperature controlled rooms, the temperature for growth is kept at 18-21°C and the irrigation water at 20-22 °C.

2.2.3.2 Bean sprouts and shoots

Bean shoots are generally grown in bins, buckets or on beds over a 5 to 6 day period to allow for shoot development. In large scale operations, bean sprouts are grown in temperature controlled rooms at 20-28 °C. Water is generally applied every 1-2 hours.

Shorter 'crunchy' style bean sprouts are grown under similar conditions to bean shoots but for only a 24-48 hour period.

2.2.3.4 Snow pea shoots

Snow pea shoots are generally grown in containers, using soil/compost mix or other medium. They are usually grown in green houses, particularly once the seed has germinated, to provide sufficient light and water requirements for growth. Snow peas are grown over an 8-12 day period, depending on the shoot and leaf development required.

2.2.4 Harvest, packaging and storage

Seed sprouts are generally harvested by hand once they have reached the desired size (some mechanical harvesting of bean shoots may occur in large scale operations). For green sprouts and bean sprouts/shoots, the whole product is collected. For snow pea shoots the product is cut away from the seed and root development. Some products may be grown in punnets and not require harvesting per se.

Green and bean sprouts are generally washed before packaging, often using cooled water to start chilling the product before storage. Water is drained away (spinning or shaking may be used to help dry off product) before product is hand packaged into plastic punnets/tubs or bags.

Packaged product is placed into cool rooms (<5°C) and stored and transported at refrigeration temperatures.

Microbiological hazard Evaluation of seed sprouts

1. Outbreaks of food-borne illness associated with seed sprouts

Between 1988 and 2008 there have been over 40 reported outbreaks worldwide attributed to consumption of contaminated seed sprouts (Appendix 1). The most commonly reported aetiological agents in these outbreaks have been various serovars of *Salmonella* spp. and enterohaemorrhagic *E. coli* (EHEC). *B. cereus* and *Yersinia enterocolitica* have also been responsible for outbreaks of food-borne illness associated with seed sprouts (Portnoy *et al.*, 1976; Cover and Aber, 1989). Alfalfa and mung bean sprouts have been the most commonly reported seed sprouts implicated in outbreaks of food-borne illness.

The majority of sprout-associated outbreaks have been reported in the United States, however, outbreaks have also occurred in Canada, Sweden, Finland, Denmark, United Kingdom, Japan and Australia. The largest reported outbreak occurred in Japan in 1996, with over 10,000 notified case and was attributed to consumption of radish sprouts contaminated with *E. coli* O157:H7 (Michino *et al.*, 1999; Watanabe *et al.*, 1999).

An outbreak due to *S*. Oranienburg occurred in Western Australia during November 2005-January 2006 that was epidemiologically linked to consumption of alfalfa sprouts. This was later confirmed microbiologically, with *S*. Oranienburg being isolated from the implicated alfalfa sprouts. A total of 125 cases of salmonellosis were reported, resulting in 11 hospitalisations.

In May 2006, another outbreak of *S. Oranienburg* was reported in Victoria, with a total of 15 cases attributed to consumption of alfalfa sprouts. In the outbreak, *S.* Oranienburg was isolated from the implicated alfalfa sprouts as well as from seed obtained from the sprouting facility. Molecular typing of the *S. Oranienburg* isolates from both the Victorian and Western Australian outbreaks showed indistinguishable patterns by pulsed field gel electrophoresis (Pers Comm. Martyn Kirk, OzFoodNet 23 April 2009). Trace back of seeds associated with these outbreaks found that the seed originated from the same Australian state but from different seed suppliers.

A number of contributing factors have been identified in reported sprout-associated outbreaks. In the US, a multi-state outbreak of *S. Mbandaka* associated with alfalfa sprouts occurred in Oregon, Washington, Idaho and California in 1999 with 89 confirmed cases (Gill *et al.*, 2003). The cases of salmonellosis were linked to two geographically separate sprout growers. A single lot of contaminated seed was identified which was used by five sprout growers during the outbreak period. Onsite investigations of the sprouting facilities associated with cases of salmonellosis identified that no form of seed sanitising was being employed prior to sprouting. No cases of illness were linked to the other three sprout growers that used the same lot of seed, all of whom employed seed sanitisation (20,000 ppm Ca(OCI)₂ or 500 ppm NaOCI).

There have, however, been reports of sprout-associated outbreaks where seed sanitising has been undertaken (Brooks *et al.*, 2001; Proctor *et al.*, 2001; Gill *et al.*, 2003). A multi-state outbreak of *E. coli* O157:NM associated with alfalfa sprouts occurred in Minnesota and Colorado in 2003 (Ferguson *et al.*, 2005). Trace-back investigations identified a common seed distributor who supplied seed (originally sourced from Australia) from the same lot to both implicated sprout growers.

During the on-site inspection of the Minnesota sprouting facility, a number of issues were identified that may have contributed to the outbreak including: use of lower hypochlorite concentration for seed disinfection than that recommended by FDA (15,000 ppm rather than the recommended 20,000 ppm for 15 min), inadequate agitation of disinfection solution, and weekly testing of spent irrigation water rather than by production lots as recommended. No deficiencies were identified at the sprouting facility implicated in the Colorado outbreak. *E. coli* O157 was not detected from samples taken at the sprouting facility, although a sample of the seed implicated in the outbreak was not available for testing.

Another multi-state alfalfa sprout-associated outbreak occurred in the US in 1999, in which there were 157 reported cases of *S. Munchen* (Proctor *et al.*, 2001). Outbreaks were epidemiologically, and microbiologically, linked to sprouting facilities that sanitised seed in 20,000 ppm calcium hypochlorite (Ca(OCI)₂) for 15 min prior to sprouting. These outbreaks illustrate that employing seed sanitising in isolation may not reliably prevent cases of foodborne illness from occurring.

2. Reported prevalence and levels of pathogens/microorganisms in seed sprouts

Microbiological surveys of seed sprouts, both domestically and internationally, have identified the presence of a variety of food-borne pathogens including *Salmonella* spp., EHEC, *B. cereus*, *Cryptosporidium* spp. and *Giardia* spp (Prokopowich and Blank, 1991; Beuchat, 1996; Robertson *et al.*, 2002; Kim *et al.*, 2004; Samadpour *et al.*, 2006).

In Australia, the Health Department of Western Australia undertook a microbiological survey of seed sprouts between January to March 2000 (Department of Health WA, 2002). In total, 261 sprouts (pre-packed and bulk) were sampled and included alfalfa, mung bean, bean sprouts, sunflower sprouts, snow pea shoots, onion sprouts [and "other sprouts"]. Samples were assessed as being acceptable/unacceptable based on whether they exceeded set criteria for total plate count (TPC), coliforms, *L. monocytogenes*, *Salmonella* spp., *E. coli*, *B. cereus* and coagulase-positive staphylococci. Of the pathogens tested, *L. monocytogenes* was detected in eight samples (at levels <5 cfu/g), and *Salmonella* spp. was detected in one sample (**Table**). *E. coli* was detected at >100 cfu/g in seven samples, however, none of these isolates were found to be toxigenic. Coagulase-positive staphylococci was detected at levels >100 cfu/g in two samples of sprouts.

Sprout type	n	<i>E. coli</i> ≥100 cfu/g (%)	Coagulase +ve Staph. ≥100 cfu/g (%)	<i>B. cereus</i> ≥100 cfu/g (%)	Salmonella (%)	L. monocytogenes (%)
Alfalfa	110	3 (2.7)	-	-	-	6 (5.5)
Bean sprout	42	-	-	1 (2.4)	-	-
Mung bean	20	-	-	-	-	1 (5.0)
Onion sprout	7	-	-	1 (14.3)	-	1 (14.3)
Snow pea	57	2 (3.5)	1 (1.8)	2 (3.5)	-	-
Sunflower	9	-	-	-	-	-
Other sprouts	13	2 (12.5)	1 (6.3)	3 (6.3)	1 (6.3)	-
Total	261	7 (2.7)	2 (0.8)	7 (0.4)	1 (0.4)	8 (3.1)

Table 1: Summary of results from WA survey of sprouts at retail (Department of Health WA, 2002)

A similar survey was conducted in the ACT during April to June 2001, where 62 samples of various seed sprouts were analysed for *E. coli*, coagulase-positive staphylococci, *B. cereus*, *Salmonella* spp. and *L. monocytogenes* (Millard and Rockliff, 2001). Again, samples were classified as satisfactory, marginal, unsatisfactory or potentially hazardous based on the level of organisms detected.

Salmonella spp. and L. monocytogenes was not detected in any of the samples. E. coli was detected in 11 samples (11.1%), with only one of these samples (mung bean sprouts) having over 100 cfu/g. Testing for pathogenic strains of E. coli was not undertaken. Coagulase-positive staphylococci was detect in one sample of snow pea shoots at levels deemed 'potentially hazardous', with $>10^4$ cfu/g, however, on further testing of a sample from the same manufacturer and retail outlet (although different lots), levels were considered satisfactory i.e. <100 cfu/g. For B. cereus, two samples were positive with ranges of 50-150 cfu/g reported.

Results from microbiological surveys of sprouts undertaken by the NSW Food Authority prior to, and following, the implementation of the NSW Plant Products Food Safety Scheme also demonstrate that sprouts are occasionally contaminated with pathogenic micro-organisms (NSWFA, 2008). In a limited sample of 30 seed sprout products in 2005, no *L. monocytogenes* or *Salmonella* spp. were detected, while *E. coli* was detected at levels <100 cfu/g in two samples. In 2006 (n=36), five samples of seed sprouts tested positive for *E. coli*, with two of these samples containing >100 cfu/g sample. Of most concern, a sample of broccoli sprouts tested positive for verotoxin-producing *E. coli* (VTEC) and was therefore rated as potentially hazardous. The survey was expanded in 2008 to a total of 122 samples of seed sprouts and included testing for *B. cereus*. Overall, no *L. monocytogenes*, *Salmonella* spp. or *E. coli* was detected. *B. cereus* was detected at levels of 100 – 1000 cfu/g in four samples and at 5500 cfu/g in one sample.

The National Enteric Pathogen Surveillance Scheme (NEPSS) collects data on isolates of *Salmonella* spp. and other pathogens submitted by primary diagnostic laboratories throughout Australia. Between 2000 and 2005 there were 13 *Salmonella* spp. isolates from seed sprouts submitted to laboratories (Table). While this data does not provide information on the prevalence of *Salmonella*-contaminated seed sprouts in Australia (only reports positive samples) it does give information on the range of *Salmonella* serovars associated with these products.

Year	Sample	Number of isolates	Serovar
2000	Sprouts	1	S. Zanzibar
2001	Lucerne seeds	1	S. Saintpaul
2002	-	-	
2003	Sprouts	8	S. Agona; S. Chester; S. Choleraesuis bv Kunzendorf Australia; S. Havana (2); S. Oranienburg; S. Orion; S. subsp I ser 16:1,v:-
	Mustard seed	1	S. subsp Illa ser 38:z53:-
2004	Sprouts	1	S. Oranienburg
2005	Bean sprouts	1	S. Infantis

Table 2: Salmonella isolates from seed sprouts, NEPSS data 2000 – 2005

As summarised by Harris *et al.* (2003), numerous international surveys have also detected pathogens in seed sprouts. It is difficult, however, to directly compare results between surveys due to differences in the number and type of samples analysed, the stage of production where samples were taken, and the methodologies used to isolate and/or enumerate the organisms.

In a microbiological survey of seed sprouts in Norway, Robertson *et al.* (2002) detected *Cryptosporidium* and *Giardia* from 9% and 2% samples respectively of mung bean sprouts (n=149), with reported level levels of 2-6 oocysts/100 g sprouts. *Cryptosporidium* and *Giardia* are highly infectious, with ingestion of one oocyst being considered sufficient to cause illness in humans (US FDA, 2008).

Protozoan parasites were also detected in six out of eight 100 g samples of unsprouted mung bean seeds – all of the positive samples contained *Cryptosporidium* (range 1-5 oocysts/100 g), with three samples also being positive for *Giardia* (1 cyst/100 g). *Cryptosporidium* was also detected in one sample of radish sprouts (n=6). No *Cryptosporidium* or *Giardia* was isolated from alfalfa sprouts, however the sample size was very low (n=6).

3. Potential pathogen contamination during seed production

While there is little specific data in the scientific literature on how seeds used for sprouting become contaminated with microbiological pathogens during on-farm production, and the relative contribution of potential sources of contamination, epidemiological investigations suggest contaminated seed is the likely source of most, if not all, sprout-associated outbreaks.

Grazing animals such as cattle and sheep are known reservoirs of *Salmonella* spp. and EHEC and infected animals may shed large numbers of these organisms in their faeces. A number of studies have shown that these pathogens can persist in animal faeces for significant periods of time. For example in Ireland, Bolton *et al.* (1999) found that when bovine faeces inoculated with *E. coli* O157:H7 (initial concentration of approximately 10⁸ CFU/g) was applied to grassland, numbers of organisms reduced by 4-5 log₁₀ within 50 days, however *E. coli* O157:H7 could still be recovered from surrounding soil for up to 99 days (the duration of the study period). In a similar study undertaken in New Zealand, Sinton *et al.* (2007) observed that the time required for a 1-log₁₀ (90%) decrease in numbers of *Salmonella* spp in bovine faeces during summer was 58 days. Survival of pathogens in the environment is extremely complex and is affected by many factors such as temperature, intensity of sunlight (UV), and moisture, hence inactivation of pathogens in animal faeces would vary significantly between geographic regions depending on environmental conditions.

Attachment of pathogens to seeds

Few studies have investigated the mechanisms by which seeds become associated with human pathogens such as *Salmonella* spp. and EHEC. Theoretically, seeds can become contaminated at any stage of production – from while they are being formed through to immediately prior to sprouting.

Pathogens may be able to enter seeds by a variety of routes such as the vascular system, pollen germ tube and the dorsal suture of the silique (seed pod) or hilum of the mature seed, (Mundt and Hinkle, 1976; Harman, 1983; Delaquis *et al.*, 1999; Thayer *et al.*, 2003). Cracks or openings in the seed coat increases the opportunity for bacterial attachment, and may enhance the potential for penetration into the seed (Charkowski *et al.*, 2001; Wade *et al.*, 2003; Fett, 2006b). The prevalence of seeds with cracks or other imperfections is highly variable, with rates of 3-85% being reported in alfalfa seeds (Wade *et al.*, 2003). Cooley *et al.* (2003) also demonstrated that pathogens can become associated with seed via direct contact with contaminated material such as chaff.

Cooley *et al.* (2003) found that following inoculation of the roots and shoots of thale cress (*Arabidopsis thaliana*) with *S.* Newport and *E. coli* O157:H7, pathogens were able to be recovered from the flowers and seeds of the mature plants. Both pathogens were found to persist longer on plants grown gnotobiotically (sterile agar) compared to those grown in sterile and non-sterile soil respectively. This suggests the role of competing microflora in reducing the colonisation and persistence of the pathogens tested. Pathogens were detected deep within the primary root system, but not in the vasculature (bacteria were not detected systemically throughout the plant).

The authors concluded that movement of pathogens to flowers and seed was therefore most likely via the plant exterior i.e. epiphytically. Results from other studies, however, have shown that *Salmonella* spp. and *E. coli* have the ability to enter plant tissue and move through the vascular system (Itoh *et al.*, 1998; Dong *et al.*, 2003).

Prevalence and levels of pathogens in seed

Very little data is available on the prevalence and levels of *Salmonella* and EHEC in seed destined for sprouting. This is, in some part, due to limitations in methodologies to isolate bacterial pathogens from contaminated seeds, where contamination is usually sporadic, at low levels and non-uniformly distributed within a sample (Inami and Moler, 1999; Wu *et al.*, 2001; Stewart *et al.*, 2001b). Reported levels of pathogens from naturally contaminated seeds range from 0.7 MPN/kg to 100 MPN/kg (NACMCF, 1999b; Fu *et al.*, 2008). The level of contamination in two lots of alfalfa seed associated with an outbreak of *S. Munchen* in the US in 1999 was reported to be 16.2 ± 1.9 MPN/kg (n=5) and 13.2 ± 3.5 MPN/kg (n=3) (Fu *et al.*, 2008).

Seed cleaning/processing

There are many opportunities for cross contamination to occur during seed processing. This may be via contact with contaminated material such as chaff (stalks etc) or with equipment that has residual contamination from previous contaminated lots (NACMCF, 1999a). Rodents, birds and other pests can harbour *Salmonella* and EHEC, and may therefore be a source of contamination if allowed access to the seed.

Seeds are sometimes scarified during processing to assist germination. Scarification involves intentionally damaging the seed coat. It has been suggested that this may provide additional sites for pathogens to attach to, and enter the seed, and potentially be protected from exposure to sanitising agents (Fett, 2006b). Results from studies investigating the efficacy of chemical treatments on reducing levels of pathogens from scarified and non-scarified seeds have been inconclusive. Holliday *et al.* (2001) found that there was a reduction in the efficacy of sanitising using scarified alfalfa seeds from one supplier compared to control (non-scarified) seeds, although this effect was not observed using seeds from a second supplier.

Survival of Salmonella and EHEC on contaminated seeds

Studies have demonstrated that once seeds are contaminated, *Salmonella* and EHEC can survive for long periods of time under normal seed storage conditions (Jaquette *et al.*, 1996; Taormina and Beuchat, 1999). In a study by Jaquette *et al.* (1996) using artificially contaminated alfalfa seeds, populations of *S. Stanley* were found to decrease by approximately 0.7-log₁₀ during storage at 8°C for 9 weeks. Storage of seed at 8°C for one week and then 21°C for 8 weeks resulted in reduction in of *S. Stanley* from initial levels of 339 cfu/g to 8 cfu/g (1.6- log₁₀ reduction). Taormina *et al.* (1999) investigated the survival of *E. coli* O157:H7 on artificially contaminated alfalfa seeds (initial concentration of approximately 10³ cfu/g), whereby the pathogen could be recovered by enrichment from 25 g samples of seed after storage for 38 weeks at either 25 or 37°C. When seed was stored at 5°C, populations of *E. coli* O157:H7 remained relatively stable during the study period of 54 weeks.

Seed sanitising

There have been extensive investigations into the efficacy of various chemical sanitising agents and other disinfection treatments in reducing levels of pathogenic micro-organisms in contaminated seeds. Consensus amongst the scientific literature is that sanitising reduces, but does not necessarily eliminate pathogens from contamination seed. Statistical analyses of published seed sanitisation studies to reduce levels of *Salmonella* and EHEC have revealed a high degree of variability in the results (Montville and Schaffner, 2004).

A summary of published studies into the efficacy of physical and chemical treatments for reducing levels of microbial pathogens from seeds is provided in Appendix 2. There are very few disinfection treatments that consistently achieve a substantial (i.e. > $5-\log_{10}$) reduction in pathogen numbers. Some disinfection treatments can have a negative effect on seed germination, which needs to be considered when determining the most appropriate method of treatment. In a review of published studies on reductions of *Salmonella* and *E. coli* O157:H7 levels in seeds treated with 20,000 ppm calcium hypochlorite (Ca(OCl)₂), Montville and Schaffner (2004) found that the most likely level of inactivation (mode) was 2.5 log₁₀, with a range of $1.0 - 6.5 \log_{10}$. Chemical sanitisers, such as chlorine, have reduced efficacy against naturally resistant pathogens such as oocysts of *Cryptosporidium* and *Giardia*, and bacterial spores (Venczel *et al.*, 1997).

The variability in reported efficacy of seed sanitisation may be due to a range of factors such as differences in: the initial pathogen load on contaminated seeds (either naturally or artificially contaminated), physiological status of the test microorganism (fresh laboratory cultures versus environmentally stressed micro-organisms), type and condition of the seed, treatment time and concentration of active compound, use of buffers, agitation of seeds during treatment and methods used to detect pathogens (e.g. direct plating on selective media versus enrichment in broth). While the same disinfection treatment may have been used in published studies, protocols for the application of the treatment and conditions for the growing of the sprouts often vary, which may affect the results observed.

As previously discussed, pathogens may be protected from chemical sanitising agents due to their location in cracks or other openings in the seed coat, or incorporation into biofilms (Fett, 2006b). Results from a study by Chrkowski *et al.* (2001) found that the efficacy of alfalfa seed sanitisation with Ca(OCl₂) varied significantly between different seed lots. When separated based on seed characteristics, it was observed that wrinkled alfalfa seeds had higher levels of total aerobic bacteria and were more difficult to sanitise compared with smooth seeds. When seeds were artificially inoculated with *S*. Newport and then treated with Ca(OCl)₂, no *Salmonella* was recovered from smooth seeds however > 10³ CFU/seed was recovered from wrinkled seeds. However, when sanitised seeds were sprouted, *Salmonella* was recovered from both smooth and wrinkled seed. This highlights the limitations of sanitising treatments in eliminating pathogens such as *Salmonella* from contaminated seed.

4. Potential pathogen contamination during sprout production

Germination/growth of sprouts

As previously discussed, seed has been identified as the likely source of contamination in many reported outbreaks of Food-borne illness associated with seed sprouts. Other possible sources of contamination during the sprouting process include water, pests, growing medium (e.g. soil).

Regardless of sprouting method, studies have demonstrated the growth of *Salmonella* and *E. coli* O157:H7 during the germinating process, with increases of 2–5 log_{10} within 48 hours being reported in the literature (Gandhi *et al.*, 2001; Stewart *et al.*, 2001a; Stewart *et al.*, 2001b; Charkowski *et al.*, 2002; Palmai and Buchanan, 2002; Howard and Hutcheson, 2003; Montville and Schaffner, 2005; Pao *et al.*, 2005; Liu and Schaffner, 2007; Fu *et al.*, 2008; Liao, 2008).

Methodologies used to investigate the growth of pathogens during the sprouting process vary widely, which may affect observed rates of growth. For example, studies have used naturally or artificially contaminated seed. For seeds that have been artificially contaminated prior to sprouting, there are differences in the methods used to inoculate the seed e.g. growth phase of the bacterial culture used, length of time and conditions in which inoculated seeds are dried before sprouting, and differences in the inoculum size. If the initial inoculum level in seeds is high (>10⁴ cfu/g), a reduced potential for growth may be observed as levels may quickly reach the maximum population density (Montville and Schaffner, 2005). The probability of naturally contaminated seeds having levels of contamination as high as this is also extremely low. Stewart *et al.* (2001a) found that the maximum level of *E. coli* O157:H7 reached in alfalfa sprouts grown from seeds with low (1.9 \log_{10} cfu/g) or high (3.9 \log_{10} cfu/g) inoculums was 5–6 \log_{10} cfu/g.

Methods used to sprout seeds in the laboratory often differ from that used commercially. For example in the laboratory small volumes of seeds may be sprouted in glass jars or "minidrums", with different methods and/or frequencies of irrigation utilised. Fu *et al.* (2008) demonstrated that irrigation frequency significantly affected the level of growth of *Salmonella* in alfalfa sprouts grown using a small-scale rotating drum. Decreasing the irrigation frequency from 20 minutes every 2 h to 20 min every 4 h resulted in a 2-log₁₀ increase in levels of *Salmonella*. Increasing the temperature during sprouting from 20°C-30°C also resulted in a 2-log₁₀ increase in *Salmonella* levels.

Pao *et al.* (2005) undertook a study to investigate the growth of *B. cereus* during the production of different sprout types. Results for sprouts grown in glass jars using naturally contaminated seed showed that levels of *B. cereus* increased by > 5 \log_{10} for radish and broccoli sprouts, however no significant growth was observed for alfalfa, lentil or mung bean sprouts. When seeds were sprouted using a "home-sprouting" drum with automatic watering, levels of *B. cereus* increased by 3 \log_{10} in radish, broccoli and mung bean sprouts, with no growth observed in alfalfa or lentil sprouts.

A number of studies have shown that *Salmonella* spp. and *E. coli* O157:H7 can become internalised in the tissue of seed sprouts during germination (Itoh *et al.*, 1998; Warriner *et al.*, 2003). In a study by Itoh *et al.* (1998) *E. coli* O157:H7 was found to be attached the inner tissue and stomata of cotyledons as well as the outer surface of radish sprouts. *E. coli* O157:H7 was isolated from sprouts after being surface-sterilised with mercuric chloride (HgCl₂), further suggesting internalisation. Warriner *et al.* (2003) also demonstrated the ability of *E. coli* and *Salmonella* spp. to become internalised in mung bean sprouts during germination. Treating the sprouts with 20,000 ppm sodium hypochlorite removed the majority of bacteria from the surface of hypocotyls, although viable organisms were recovered from the sprout tissue.

Microorganisms can also become incorporated into biofilms on the sprout surface (Fett, 2000; Warriner *et al.*, 2003; Fett and Cooke, 2003b). Biofilms are a complex structure of microorganisms adhered to a surface (usually inert) and encapsulated in self-produced extracellular material such as exopolysaccharides, lipids and proteins. This structure provides protection to microorganisms from antimicrobial agents such as chemical sanitisers (Costerton *et al.*, 1995).

When viewing alfalfa, broccoli, clover and sunflower sprouts by scanning electron microscopy, biofilms have been found to be most abundant on the cotyledon surface compared with hypocotyls and roots (Fett, 2000).

Several studies have shown that levels of bacterial pathogens in spent irrigation water during the germinating process is strongly correlated to levels found in the contaminated seed sprouts (Costerton *et al.*, 1995; Stewart *et al.*, 2001b; Howard and Hutcheson, 2003; Johnston *et al.*, 2005; Liu and Schaffner, 2007). In an analysis of published data, Montville and Schaffner (2005) found that the average concentration of pathogens was slightly higher in sprouts than in spent irrigation water (mean 0.5 log₁₀, range -0.75-2.25 log₁₀). Liu and Schaffner (2007) demonstrated that *S*. Stanley could be detected in irrigation water within 12 hours of germination of alfalfa sprouts.

Various treatments have been studied to reduce the levels of pathogens either during the sprouting process or post harvest. These treatments include the use of chemical agents, competitive exclusion (e.g. plant-associated pseudomonads), bacteriophages and irradiation (Rajkowski and Thayer, 2000; Fett, 2002c; Fett, 2006a; Kocharunchitt *et al.*, 2009). To date, however, most of these treatments have not been shown to result in consistent levels of pathogen reduction. This may be due to a number of factors such as protection of microbial pathogens in biofilms, or internalisation into the sprout tissue. One method that has been found to reduce levels of pathogens in sprouts is treatment by irradiation, with a minimum gamma radiation dose of 0.5 kGy shown to be effective in eliminating *Salmonella* from naturally contaminated alfalfa sprouts (Rajkowski and Thayer, 2000).

Sprout harvesting

Contamination of seed sprouts can also occur during harvesting. Possible sources of contamination include equipment, rinse waters and workers themselves. Gandhi *et al.* (2001) found that the transfer of *Salmonella* to non-contaminated sprouts via hands directly after harvesting contaminated sprouts (7.9 \log_{10} cfu/g) was approximately 5 \log_{10} cfu/g. While the level of *Salmonella* in the contaminated sprouts used in this study is considered higher than that observed under commercial conditions, it demonstrates the potential for pathogens to be transferred between batches of seed sprouts during harvesting.

Depending on the type of seed sprout being produced, they are often rinsed prior to packaging. While studies have demonstrated that there is limited/no reduction in pathogens from rinsing sprouts in water containing sanitisers, there is an opportunity for significant cross contamination if wash/rinse water becomes contaminated.

Retail/consumer

Once packaged, sprouts are generally stored under temperature control (<4°C) to limit the growth of microorganisms. Gandhi *et al.* (2001) found that levels of *S*. Stanley in contaminated alfalfa sprouts reduced slightly (0.09 \log_{10}) when stored at 4°C. In the survey of seed sprouts at retail undertaken by WA Department of Health in 2001, 78% (203/261) of samples were stored under refrigeration. Of the refrigerated samples, 75% (154 samples) were recorded to have temperatures above 10°C. Most seed sprouts are consumed raw and therefore will not receive any form of heat treatment prior to consumption (which would inactivate pathogens if present).

Reported sprout-associated outbreaks (adapted from Taormina et al., 1999)

Year	Pathogen	No. of culture-	Location	Type of sprout	Likely source of	Reference
		confirmed cases			contamination	
1988	S. Saintpaul	143	United Kingdom	Mung bean	Seed	(O'Mahony <i>et al</i> ., 1990)
1988	S. Saintpaul, S.	148	Sweden	Mung bean		
	Havana, S. Muenchen					
1988	S. Virchow	7	United Kingdom	Mung bean		(O'Mahony <i>et al.</i> , 1990)
1989	S. Gold-Coast	31	United Kingdom	Cress	Seed and/or sprouter	(Joce et al., 1990)
1990	S. Anatum	15	US (Washington)	Alfalfa		(CDC, 1990)
1992	S. enterica 4,5,12:b:-	272	Finland	Mung	ND	(Mattila <i>et al.</i> , 1994)
1994	S. Bovismorbificans	492	Finland,	Alfalfa	Seed	(Ponka et al., 1995; Puohiniemi
			Sweden			<i>et al.</i> , 1997)
1995	S. Stanley	242	Finland	Alfalfa	Seed	(Mahon <i>et al.</i> , 1997)
	-		6 US States			
1995-	S. Newport	>133	7 US States	Alfalfa	Seed	(van Beneden et al., 1999)
1996			Canada			
			Denmark			
1996	S. Stanley	30	US (Virginia)	Alfalfa	Seed	(CDC, 1996; Barret and Chaos,
	_					1996)
1996	<i>E. coli</i> O157:H7	>10,000	Japan	Radish	Seed	(Watanabe <i>et al.</i> , 1999)
1996	S. Montevideo and	~500	California, Nevada (USA)	Alfalfa	Seed and/or sprouter	(Taormina <i>et al.</i> , 1999;
	S. Maleagridis					NACMCF, 1999a; Mohle-
	_					Boetani <i>et al</i> ., 2001)
1997	S. Anatum and	109	Kansas, Missouri (USA)	Alfalfa	Seed	(Taormina <i>et al</i> ., 1999)
	S.Infantis					
1997	<i>E. coli</i> O157:H7	79	4 US States	Alfalfa	Seed	(Breuer <i>et al.</i> , 2001)
1997	S. Meleagridis	78	Canada	Alfalfa	Seed	(Sewell and Farber, 2001)
1997-	S. Senftenberg	60	US (California, Nevada)	Alfalfa	Seed and/or sprouting	(Mohle-Boetani et al., 2001)
1998					drum	
1998	S. Havana/S. Cubana	40	US (California)	Alfalfa	Seed	(CDC, 1998; NACMCF, 1999a)
1998	<i>E. coli</i> O157:NM	8	California, Nevada	Alfalfa, Clover	Seed and/or sprouter	(CDC 1998; Mohle-Boetani et
						al., 2001)
1999	S. Mbandaka	83	8 US states	Alfalfa	Seed	(CDC 1998; NACMCF, 1999a)
1999	S. Muenchen	157	10 US states	Alfalfa	Seed	(Proctor et al., 2001)
1999	S. paratyphi var Java	51	Canada	Alfalfa	Seed	(Stratton <i>et al.</i> , 2001)
1999	S. Saintpaul	36	US (California)	Clover	ND	(CDC 1998)
1999	Salmonella spp.	34	US (Michigan)	Alfalfa	ND	(CDC 1998)
1999	S. Typhimurium	120	Colorado (USA)	Alfalfa	Seed	(Winthrop <i>et al.</i> , 2003)

Year	Pathogen	No. of culture-	Location	Type of sprout	Likely source of	Reference
2000	S. Enteritidis phage type 4b	27	The Netherlands	Mung beans	Seed	(van Duynhoven <i>et al.</i> , 2002)
2000	S. Enteritidis	75	US	Mung beans	ND	(CDC, 2000)
2000	S. Enteritidis	8	Canada	Alfalfa	ND	(Harris et al., 2003)
2001	S. Enteritidis PT 913	84	Canada	Mung bean	Seed	(Honish and Nguyen, 2001)
2001	S. Kottbus	31	California	Alfalfa	Seed	(Mohle-Boetani et al., 2002)
2001	S. Enteritidis PT1	26	US (Hawaii)	Mung bean	Seed and/or sprouter	(CDC, 2002)
2002	S. Abony	13	Finland	Mung bean	ND	(Ministry of Agriculture and Forestry, 2003)
2003	S. Saintpaul	16	US	Alfalfa	ND	(CDC, 2003)
2003	<i>E. coli</i> O157:H7	7	US	Alfalfa	ND	(CDC 2003)
2003	E. coli O157:NM (H-)	13	US	Alfalfa	ND	(CDC 2003)
2003	S. Chester	26	US	Alfalfa	ND	(CDC 2003)
2004	E. coli O157:NM	2	US	Alfalfa	ND	(CDC 2003)
2004	S. Bovismorbificans	35	US	Alfalfa	ND	(CDC, 2004)
2005- 2006	S. Oranienburg	126	Australia (WA)	Alfalfa	Seed	(OzFoodNet, 2006)
2006	S. Oranienburg	15	Australia (Vic)	Alfalfa	Seed	(OzFoodNet, 2007)
2006	S. Braenderup	4	US	Mung bean	ND	(CDC, 2006)
2007	S. Weltevreden	45	Norway Denmark Sweden	Alfalfa	Seed	(Emberland <i>et al.</i> , 2007)

APPENDIX 2

Physical and chemical treatments for the inactivation of pathogens on inoculated sprouting seeds (Fett, 2006b)

Treatment	Conditions	Time	Seed Type	Bacterium	Logarithmic Reduction - (CFU/G)	Seed Germination	Ref
Acetic acid, vapour	242 μl/L air, 45°C	12 h	Mung bean	Salmonella	> 5, no survivors	No effect	(Delaquis <i>et al.</i> , 1999)
Acetic acid, vapour	242 μl/L air, 45°C	12 h	Mung bean	<i>E. coli</i> O157:H7	> 6, no survivors	No effect	(Delaquis <i>et al</i> ., 1999)
Acetic acid, vapour	242 μl/L air, 45°C	12 h	Mung bean	L. monocytogenes	4.0	No effect	(Delaquis <i>et al.</i> , 1999)
Acetic acid, vapour	300 mg/L air, 50°C	24 h	Alfalfa	Salmonella	0.8	No effect	(Weissinger <i>et al.</i> , 2001)
Acidic EO water	1,081 mV, 84 ppm chlorine	10 min	Alfalfa	Salmonella	1.5	No effect	(Kim <i>et al.</i> , 2003)
Acidic EO water	1150 mV, 50 ppm chlorine	64 min	Alfalfa	<i>E. coli</i> O157:H7	1.6	Significant reduction	(Sharma and Demirci, 2003b)
Acidic EO water	1,079 mV, 70 ppm chlorine	15 min	Alfalfa	Salmonella	2.0	No effect	(Stan and Daeschel, 2003)
Allyl isothiocyanate	50 μl/950-cc jar, 47°C	24 h	Alfalfa	<i>E. coli</i> O157:H7	>2.0, survivors present	Slight reduction	(Park <i>et al</i> ., 2000)
Ammonia, gas	300 mg/L	22 h	Alfalfa	Salmonella	2.0	No effect	(Himathongkham et al., 2001)
Ammonia, gas	300 mg/L	22 h	Mung bean	Salmonella	5.0	No effect	(Himathongkham <i>et al</i> ., 2001)
Ammonia, gas	300 mg/L	22 h	Alfalfa	<i>E. coli</i> O157:H7	3.0	No effect	(Himathongkham <i>et al</i> ., 2001)
Ammonia, gas	300 mg/L	22 h	Mung bean	<i>E. coli</i> O157:H7	6.0	No effect	(Himathongkham <i>et al</i> ., 2001)
Ca(OH)₂ (Calcium Hydroxide)	1%	10 min	Alfalfa	<i>E. coli</i> O157:H7.	3.2		(Holliday <i>et al</i> ., 2001)
Ca(OH)₂	1%	10 min	Alfalfa	Salmonella	2.8 - 3.8	No effect	(Weissinger and Beuchat, 2000; Holliday <i>et al</i> ., 2001)61,62
Ca(OCl) ₂ (Calcium Hypochlorite)	20,000 ppm	3 min	Alfalfa	<i>E. coli</i> O157:H7	> 2.3, survivors present	Reduced rate	(Taormina and Beuchat, 1999)
Ca(OCI) ₂	20,000 ppm	10 min	Alfalfa	Salmonella	2.0	Slight reduction	(Weissinger and Beuchat, 2000)
Ca(OCI) ₂	18,000 ppm	10 min	Alfalfa	Salmonella	3.9	No effect	(Fett, 2002a)
Ca(OCI) ₂	18,000 ppm	10 min	Alfalfa	<i>E. coli</i> O157:H7.	4.5	No effect	(Fett, 2002a)
Ca(OCI) ₂	16,000 ppm	10 min	Mung bean	Salmonella.	5.0	No effect	(Fett, 2002b)
Ca(OCI) ₂	16,000 ppm	10 min	Mung bean	E. coliO157:H7	3.9	No effect	(Fett, 2002b)
Chlorine dioxide, acidified	500 ppm	10 min	Alfalfa	E. coliO157:H7	>2.4, survivors present	Significant reduction	(Taormina and Beuchat, 1999)
Citrex [™]	20,000 ppm	10 min	Alfalfa	Salmonella	3.6	No effect	(Fett and Cooke, 2003a)
Citrex [™]	20,000 ppm	10 min	Alfalfa	<i>E. coli</i> O157:H7	3.4	No effect	(Fett and Cooke, 2003a)
Dry heat	50°C	60 min	Alfalfa	<i>E. coli</i> O157:H7	1.7	No effect	(Bari <i>et al</i> ., 2003)
Dry heat	70°C	3h	Alfalfa	Salmonella	3.0	Slight reduction	(Weissinger <i>et al.</i> , 2000)
Fit	According to label	15 min	Alfalfa	Salmonella	2	No effect	(Beuchat <i>et al.</i> , 2001)
Fit [™]	According to label	15 min	Alfalfa	<i>E. coli</i> O157:H7	>5.4	No effect	(Beuchat <i>et al.</i> , 2001)
H_20_2	8%	3 min	Alfalfa	<i>E. coli</i> O157:H7	>2.9, survivors present	No effect	(Taormina and Beuchat, 1999)

Treatment	Conditions	Time	Seed Type	Bacterium	Logarithmic Reduction - (CFU/G)	Seed Germination	Ref
H_2O_2	8%	10 min	Alfalfa	Salmonella	3.2	No effect	(Weissinger and Beuchat, 2000)
Hydrostatic pressure	300 mPa	15 min	Garden cress	Salmonella	5.8	Reduced rate	(Wuytack <i>et al</i> ., 2003)
Hydrostatic pressure	300 mPa	15 min	Garden cress	Shigella flexneri	4.5	Reduced rate	(Wuytack <i>et al</i> ., 2003)
Lactic acid	5%, 42°C	10 min	Alfalfa	E. coli 0157:H7	3.0	No effect	(Lang <i>et al</i> ., 2000)
Radiation, gamma	Various		Alfalfa	Salmonella	D-value of 0.97 kGy	Dosage dependent	(Thayer <i>et al</i> ., 2003)
Radiation, gamma	Various		Alfalfa	<i>E. coli</i> O157:H7	D-value of 0.60 kGy	Dosage dependent	(Thayer <i>et al</i> ., 2003)
Radiation, gamma	Various		Broccoli	Salmonella	D- value of 1.10 kGy	Dosage dependent	(Rajkowski <i>et al</i> ., 2003)
Radiation, gamma	Various		Broccoli	<i>E. coli</i> O157:H7	D- value 0f 1.11 kGy	Dosage dependent	(Rajkowski <i>et al</i> ., 2003)
Sodium chlorite, acidified	1,200 ppm, 55°C	3 min	Alfalfa	<i>E. coli</i> O157:H7	>1.9, survivors present	Slight reduction	(Taormina and Beuchat, 1999)
Sulphuric acid	2N	20 min	Alfalfa	<i>E. coli</i> O157:H7	5.0	No effect	(Pandrangi <i>et al</i> ., 2003)
Ozone, aqueous	21 ppm, w/sparging	64 min	Alfalfa	<i>E. coli</i> O157:H7	2.2	No effect	(Sharma <i>et al</i> ., 2002)
Ozone, aqueous	21.3 ppm, w/sparging	20 min	Alfalfa	L. monocytogenes	1.5	No effect	(Wade <i>et al</i> ., 2003)
Pulsed UV light	5.6 J/cm ² , 270 pulses	90 sec	Alfalfa	<i>E. coli</i> O157:H7	4.9	Significant reduction	(Sharma and Demirci, 2003a)
Dielectric heating, radio frequency	39 MHz, 1.6 kV/cm	26 sec	Alfalfa	Salmonella	1.7	No effect	(Nelson <i>et al</i> ., 2002)
Supercritical CO ₂	4000 psi, 50 C	60 min	Alfalfa	<i>E. coli,</i> generic	1.0	No effect	(Mazzoni <i>et al</i> ., 2001)
Water, hot	3-stage: 25 to 50 to 85°C	30 min, 9 sec, 9 sec	Alfalfa	<i>E. coli,</i> generic	>4, no survivors	No effect	(Enomoto <i>et al.</i> , 2002)
Water, hot	54°C	5 min	Alfalfa	Salmonella	2.5	No effect	(Jaquette <i>et al.</i> , 1996)
Water, hot	80°C	2 min	Mung bean	Salmonella	>6	No effect	(Weiss and Hammes, 2003)

Chemical hazard Evaluation of seed sprouts

Summary

There are legislative requirements²¹ that regulate the use and presence of chemical substances in food. These requirements ensure that public health and safety is protected and that chemical hazards in food are adequately managed.

These requirements include the provisions in the Code. There are certain Standards in the Code that are of relevance in relation to chemical hazards in seed sprouts and these are:

Standard 1.3.1 – Food Additives Standard 1.3.3 – Processing Aids Standard 1.4.1 – Contaminants and Natural Toxicants Standard 1.4.2 – Maximum Residue Limits Standard 1.4.3 – Articles and Materials in Contact with Food.

The limited data available (see below) do not indicate that chemical hazards are a major concern for seed sprouts. On this basis, the current regulatory measures including those in the Code are considered adequate with respect to managing chemical hazards in seed sprouts. Notwithstanding this, there are general hazard mitigation measures that could be considered to ensure that chemical hazards associated with seed sprout production are specifically managed.

Residues of agricultural and veterinary chemicals

Residues of agricultural and veterinary chemicals in food are managed by ensuring that only approved chemical products are used in food production, and that these products are used in accordance with approved conditions of use by food handlers and producers.

Chemical products may be used in the production of seed and then in the subsequent production of seed sprouts, including as sanitising agents. The use of products in these situations may result in residues of agricultural chemicals in seed sprouts. It would not be expected that veterinary chemical residues would be present in seed sprouts.

Standard 1.4.2 lists the maximum permissible limits for agricultural and veterinary chemical residues present in food. These limits apply to seeds and seed sprouts for human consumption.

The Maximum Residue Limits (MRLs) in Standard 1.4.2 are based on MRLs notified to FSANZ by the Australian Pesticides and Veterinary Medicines Authority (APVMA). The APVMA is a Commonwealth of Australia Government authority responsible for the assessment and registration of pesticides and veterinary medicines and for their regulation up to and including the point of retail sale. The APVMA administers the National Registration Scheme for Agricultural and Veterinary Chemicals (NRS) in partnership with the States and Territories and with the active involvement of other Australian government agencies. The MRLs in Standard 1.4.2 are based on the highest residues that may occur in food that has been legitimately treated with approved chemical products, including appropriate withholding periods.

²¹ These requirements are in State, Territory and Commonwealth of Australia legislation.

In addition to the MRLs in Standard 1.4.2, certain chemical product label restraints and precautions apply to the use of chemical products as seed treatments. These restraints and precautions require chemical products to be used appropriately on seeds and where relevant, that such seeds are not provided as food for human consumption.

There are very little data available on the presence of residues of agricultural and veterinary chemicals in seed sprouts (see below). Survey results available to FSANZ have not identified non-complying residues of agricultural and veterinary chemicals in seed sprouts.

While not a compliance survey, data from the Australian Total Diet Survey (ATDS) indicate that few residues of agricultural and veterinary chemicals have been detected in alfalfa sprouts²². This result may be considered representative for sprouts generally in that chemical products are likely to be used similarly to control similar pests and diseases.

FSANZ understands that nutrients are not used during the production of seed sprouts (e.g. fertiliser, growth nutrients).

Overall, the limited data currently available would suggest that residues of agricultural and veterinary chemicals in seed sprouts are not of concern. Notwithstanding this, it would be appropriate to ensure that food handlers and seed sprout producers only use approved chemical products in the production of seed sprouts and that these products are used in accordance with approved conditions of use.

Processing Aids in Food Production

Chemical products may be used as processing aids in the production of seed sprouts and the use of these products may result in residues of these processing aids in seed sprouts (e.g. chlorine). Chemical products used in the production of seed sprouts should be approved for the relevant purpose and used in accordance with approved conditions of use.

In addition, Standard 1.3.3 includes substances which may be used as processing aids and includes limits for these substances in seed sprouts or the inputs that may be used in seed sprout production (washing agents). Seed sprouts treated legitimately with processing aids should comply with this Standard.

It would be appropriate to ensure that food handlers and seed sprout producers only use approved processing aids in the production of seed sprouts and that these products are used in accordance with approved conditions of use.

Contaminants

There are very little data available on the presence of chemical contaminants in seed sprouts. These contaminants may include metals (cadmium, copper, zinc), non-metal contaminants (e.g. packaging monomers) and mycotoxins.

While not a compliance survey, data from the ATDS indicate that some metals have been detected in alfalfa sprouts²³. It would not be appropriate to extrapolate this to all seed sprouts because of the different types of seed sprouts.

The limited data currently available would not suggest that there are concerns with contaminants that may be present in seed sprouts.

²² 19th Australian Total Diet Survey - <u>http://www.foodstandards.gov.au/_srcfiles/tables%2023-26.pdf</u>

²³ 19th Australian Total Diet Survey - <u>http://www.foodstandards.gov.au/_srcfiles/tables%209-22.pdf</u>

Notwithstanding this, it would be appropriate to ensure that food handlers and seed sprout producers handle seed sprouts to minimise or prevent contamination.

Substances Added to Seed Sprouts

Food additives may only be used in seed sprouts in accordance with Standard 1.3.1. In addition, Standard 1.3.4 – Identity and Purity includes specifications for substances added to food. These Standards specify food additives that may be used by food type and include limits for specific food additives in foods. Depending on the degree of processing, seed sprouts may contain certain additives. FSANZ understands that food additives would be unlikely to be added to seed sprouts.

It would be appropriate to ensure that food handlers and seed sprout producers only use approved food additives in the production of seed sprouts.

Packaging

There are general provisions in food legislation²⁴ that require articles and materials in contact with food to be safe and suitable for that purpose. These requirements apply to seed sprouts.

The Code also includes requirements in Standard 1.4.3. The purpose of this Standard is to provide for articles and materials to be in contact with food, including packaging.

The Standard does not specify individual packaging materials for food contact or how they are produced or used but includes general requirements to ensure that such articles and materials do not cause harm.

While there is little information available on the articles and materials that may be used for seed sprouts, the general requirements are considered to be adequate at this stage. These requirements relate to ensuring:

- that equipment and inputs used in producing seed sprouts are suitable for their intended purpose to prevent contamination of seed sprouts
- that seed sprout handlers are aware of the need to prevent contamination of seed sprouts and how to prevent any contamination
- seed sprouts are protected from contamination including during storage and transport
- packaging used for seed sprouts is fit for its intended purpose and is therefore not likely to cause contamination.

Overall, seed sprout producers should ensure that procedures are instituted, including with seed sprout handlers, to prevent contamination of seed sprouts as far as is reasonably possible.

²⁴ State and Territory food legislation.

Cost Estimate of the 2005-06 Australian Seed Sprout outbreaks

Outbreaks of food-borne illnesses are sporadic and unpredictable. An estimation of the cost of food-borne illness resulting from consumption of contaminated seed sprouts cannot be generated as an annual figure because of the sporadic and infrequent nature of such outbreaks. The potential cost of adverse health consequences due to consumption of contaminated seed sprouts is estimated using the 2005-2006 outbreak data.

OzFoodNet ²⁵ reports that in Australia about 132 cases of food-borne salmonellosis have been identified to be associated with consumption of raw sprouts in Western Australia and Victoria during 2005-2006. According to an Australian study (Hall, et. al. 2005)²⁶, for very one reported case of food-borne illness in the community there are 9 unreported cases. Taking into account of underreporting there could be up to 1320 community cases of salmonellosis associated with consumption of contaminated sprouts in Western Australia and Victoria during 2005-2006.

Based on a US model to compute social cost of illness, and a Dutch study (Kemmeran et. al., 2006²⁷) that estimates disease burden of enteric pathogens, the cost of a general foodborne salmonellosis case has been estimated at approximately \$8,786 in 2009 prices. Cost per case in Table 1 is in the context of Australian estimates for Quality Adjusted Life Year (QALY) and Value of Statistical Life (VSL)²⁸. This estimate takes into account productivity, welfare and medical costs for a range of effects ranging from a mild gastro illness to extreme consequences like death. See Table 1 followed by Explanatory Notes for the breakdowns of this estimate.

Outcomes	Incidence	Total QALDs Lost per Illness	Health Loss per Case	Medical Costs per Case	Weighted Dollar Loss per Case
Gastroenteritis					
Mild	.857	5.58	\$2,466	\$0	\$2,113
Moderate	.154	10.65	\$4,707	\$73	\$736
Severe	.018	16.15	\$7,138	\$ 1,526	\$156
Reactive Arthritis					
Mild	.011	222	\$ 98,124	\$ 0	\$ 1079
Moderate	.002	222	\$ 98,124	\$110	\$ 196
Severe	.0002	222	\$ 98,124	\$ 4,063	\$ 20
Irritable Bowel					
Syndrome	.0002	Life Long	\$3,738,000	\$1,526	\$748
Death	001	9454	¢ 2 729 000		¢ 0 700
Death	.001	8404	φ <i>3,13</i> 8,000		\$ 3,738
Total Expected Loss	per Case				\$ 8,786

Table 1.	Social	cost of a	typical	food borno	Salmonalla illnoss	0000	
Table I.	Social	CUSL UL a	typical	1000-borne	Samonena miness	Lase	(JAUD)

²⁵ Kirk M., 2006. Outbreaks Associated with Raw Sprouts. Ozfoodnet presentation.

²⁶ Hall G. et al. (2005) Estimating food-borne gastroenteritis, Australia. Emerging Infectious Diseases 11(8): 1257-1264.

²⁷ Kemmeren, J.M. et al. (2006) RIVM: Priority setting of food-borne pathogens. Disease burden and costs of selected enteric pathogens: Report 330080001:55-58.

²⁸ Refer to Abelson, P. (2007) Office of Best Practice Regulation. Establishing a Monetary Value for Lives Saved: Issues and Controversies: WP 2008-02:21.

The product of the number of potential cases in the community and the cost per case provides an indicative social cost of approximately \$11.60 million of 2005/2006 outbreaks in current prices.

This cost estimation refers to the two outbreaks occurred in 2005-2006 that was a once off event. The cost estimation should not be treated as an annual cost of seed sprouts caused food-borne illnesses.

Explanatory Notes

Outcomes: A range of adverse health outcomes have been reported to be associated with human illness resulting from a food-borne salmonellosis. An occurrence could vary from a mild gastroenteritis illness (GE) to extreme consequences like death. Long term adverse health complications include Reactive Arthritis and Irritable Bowel Syndrome. These outcomes have been derived from the Dutch study (Kemmeren, et al. 2006).

Incidence: A Mild case of Gastroenteritis illness is classified as one that involves no visit to a general practitioner (GP), a moderate case involves a GP visit and a severe case would be one that requires hospitalisation. The breakdown of cases into Mild, Moderate and Severe cases of illness is based on Kemmeren et al. (2006) estimate of 35,000 community cases of *Salmonella*- associated gastroenteritis and sequelae illness. For example out of the 35,000 most likely community wide cases, 30,000 or approximately .857 or (approximately 86%) could experience mild symptoms.

Quality Adjusted Life Day (QALD): QALD refers to a day of life adjusted for its quality or its value. A day in perfect health is considered equal to 1.0 QALD. The estimated number of QALDs lost due to illness has also been derived from the Dutch study where a mild illness may only impact about 5 days whereas a severe illness could affect up to 16 days of an individual's life (Kemmerer, et al. 2006).

Health loss: Health loss is measuring what the community is willing to pay to avoid an adverse health outcome or consequence. It is obtained as a product of number of QALDs and value of QALD. The recommended Value of a Life Year (VLY) which may also be expressed as Quality Adjusted Life Year (QALY) in Australia is \$AUD151,000 (Abelson, 2007). Therefore the value of a Quality Adjusted Life Day (QALD) would be \$AUD151,000 divided by 365 or \$AUD414 in 2007 prices. Based on the Australian Taxation Office's (ATO's) Consumer Price Index (CPI) up till 2009 inflation adjusted value of QALD is \$AUD442 (increase of 6.8%). E.g. the health loss for a mild gastroenteritis illness affecting 5.58 days at the rate of \$AUD442 per day is \$AUD2,466. In case of Reactive Arthritis cases it is 222 days at the rate of \$AUD442 or \$AUD98,124.

Similarly for death, the health loss is estimated to equal to the Value of Statistical Life (VSL) at \$AUD3.5 million in 2007 prices (Abelson, 2007). In simple words it is assumed that the society is willing to pay approximately \$AUD3.74 Million in 2009 prices to avoid death for a healthy individual (after CPI inflation adjustment of 6.8%) Health loss is limited to loss of leisure, welfare and quality of life.

Medical costs: Medical costs include the health care and medical costs associated with the range of adverse health outcomes resulting from a food-borne salmonellosis illness. While a mild illness may not warrant any medical examination a moderate case could only involve a GP visit i.e. \$AUD60 in 2002 prices (Abelson, et al. 2006)²⁹. For a severe hospitalisation case of a gastroenteritis illness or Irritable Bowel Syndrome (IBD) the cost is estimated to be approximately \$AUD1,254 assuming average of hospital stay is 2 days. In the event of Reactive Arthritis, it is assumed one specialist visit at \$AUD90 for a moderate case and \$AUD3,339 for a severe case. Costs used are 2002 prices and derived from the annual cost of food-borne illness in Australia (Abelson, et al. 2006). As the above prices are of 2002 CPI inflation adjusted estimates for 2009 are \$AUD73 for a GP visit and/or \$AUD110 for a specialist visit, \$AUD1,526 for a hospitalisation or IBD case and \$AUD4,063 for a severe hospitalisation case (after ATO's CPI inflation adjustment of approximately 21.7% over 2002-09).

²⁹ Abelson, P. et al. (2006) Australian Government Department of Health and Ageing. The annual cost of food-borne illness in Australia.

Weighted dollar loss: is the sum of Health Loss and Medical costs proportioned to the incidence or case breakdown, e.g. in a moderate gastroenteritis illness outcome, the health loss was \$ 4,707. In addition there could be medical costs of a GP visit of \$AUD73. The sum of \$AUD4,780 apportioned to the incidence or likelihood of that event, i.e. 154 or 15.4% translates to \$AUD736 which has been placed in the weighted dollar loss column for a moderate gastroenteritis illness. In case of Death the VSL of \$AUD3,738,000 is then pro-rated to the incidence or likelihood of death at .1% (.001) to generate the weighted dollar loss for Death as \$AUD3,738.

Regulatory measures applying to sprout production in Australia

1. Australia New Zealand Food Standards Code

Chapter 3 – Food Safety Standards

Standards 3.2.2 – Food Safety Practices and General Requirements and 3.2.3 – Food Premises and Equipment set out specific requirements for food business, food handlers and the food premises and equipment with which they operate to ensure the safe production of food. Standard 3.2.2 specifies process control requirements to be satisfied at each step of the food handling process:

- receipt
- storage
- processing
- display
- packaging
- transportation
- disposal
- recall

In addition there are requirements for skills and knowledge, health and hygiene of food handlers and the cleaning, sanitising and maintenance or premises and equipment.

Standard 3.2.3 sets out requirements to ensure that food premises, fixtures, fittings, equipment and transport vehicles are designed and constructed to minimise opportunities for food contamination and are cleaned and sanitised where necessary.

The food safety standards apply to all food businesses in Australia. A food business is defined in the Code as follows:

food business means a business, enterprise or activity (<u>other than primary food production</u>) that involves:

- (a) the handling of food intended for sale; or
- (b) the sale of food;

regardless of whether the business, enterprise or activity concerned is of a commercial, charitable or community nature or whether it involves the handling or sale of food on one occasion only.

primary food production means the growing, cultivation, picking, harvesting, collection or catching of food, and includes the following:

- (a) the transportation or delivery of food on, from or between the premises on which it was grown, cultivated, picked, harvested, collected or caught;
- (b) the packing, treating (for example, washing) or storing of food on the premises on which it was grown, cultivated, picked, harvested, collected or caught; and
- (c) any other production activity that is regulated by or under an Act prescribed by the regulations for the purposes of this definition.

While the operation of a seed sprout business may involve a number of the food handling activities generally undertaken by food businesses, State and Territory jurisdictions (excepting NSW) have not been able to apply Chapter 3 requirements to them because, in accordance with these definitions, seed sprout businesses have been regarded as a primary food producer (a grower of sprouts).

Chapter 1 – General Food Standards

The food standards in Chapter 1 generally apply to all food sold or traded at retail and wholesale level in Australia and cover labelling requirements; the use of additives and processing aids; contaminants and natural toxicants; MRLs, articles and materials in contact with food, and microbiological limits for food. The only provision in Chapter 1 that is specific for seed sprouts is a microbiological limit in Standard 1.6.1.

Standard 1.6.1 – Microbiological Limits for Food specifies a microbiological limit for Salmonella in 'cultured seeds and grains' (alfalfa sprouts, bean sprouts etc.):

Food	Micro-organism	n	С	m	М
Cultured seeds and grains (bean sprouts, alfalfa etc)	Salmonella/25 g	5	0	0	

Where:

n means the minimum number of sample units which must be examined from a lot of food **c** means the maximum allowable number of sample units that can exceed m **m** means the acceptable microbiological level in a sample unit

M means the level, when exceeded in one or more samples, would cause the lot to be

M means the level, when exceeded in one or more samples, would cause the lot to be rejected.

Information on Chapter 1 requirements for covering the use of additives and processing aids; contaminants and natural toxicants; Maximum Residue Limits and articles and materials in contact with food, is discussed in Attachment 2.

2. State and Territory requirements

New South Wales food safety scheme- seed sprouts

The NSW Food Regulation 2004 was amended in September 2005 to include the Plant Products Food Safety Scheme, applying to specified high risk plant product industries including sprouting and processing of seed sprouts.

Businesses that produce, store or transport seed sprouts for supply to the retail and food service sectors must hold a licence with the New South Wales Food Authority stating the activities that they are authorised to undertake and specific controls relevant to the industry. Businesses producing or handling unsprouted seed, unsprouted beans or wheatgrass do not require a licence.

Businesses that receive seeds for sprouting and produce seed sprouts must comply with the NSW Food Act 2003, NSW Food Regulation 2004, the Australia New Zealand Food Standards Code and the NSW Plant Products Safety Manual³⁰. The manual outlines and explains the requirements of the Plant Products Food Safety Scheme.

³⁰ Plant Products Safety Manual NSW/FA/FI012/0711 version 1 issued 12/11/07 available on the website of the NSW Food Authority at <u>www.foodauthority.nsw.gov.au/industry/industry-sector-requirements/plant-products/</u>

Sprout producers must demonstrate compliance though implementing a food safety program, based on Codex HACCP or Standard 3.2.1, which is certified by the Authority and audited. Businesses that only transport, distribute or store seed sprouts do not require a food safety program and are inspected for compliance with the legislation and the manual.

As part of their food safety program, sprout producers must address the following:

- raw material receival and storage
- seed pre-screening for *Salmonella* (this may be certified by the seed supplier)
- raw material quality either by obtaining Authority approval to source seed from a supplier that can provide evidence that seed is produced under an audited HACCPbased food safety program or sanitising seed as specified in the manual;
- washing and sprouting
- testing of spent irrigation water for Salmonella
- post harvest washing
- sprout storage
- cleaning and sanitising of equipment and processing surfaces
- finished product testing for *E. coli*.

Sprout producers must also ensure they have documented procedures for notifying the Authority of tests that fail to meet the microbiological testing requirements in the manual and the microbiological and chemical standards in the Code. Laboratories testing these products are also required to notify failures to the Authority.

Specific requirements, detailed explanations and guidance for these activities are provided in the manual.

3. Export requirements

Schedule 3A of the *Export Control (Plant and Plant Products) Orders 2005* prescribes structural requirements and operational and hygiene requirements for establishments preparing mung beans aimed, primarily focussed on pest control, effective cleaning and personal hygiene. Clause 6 of this schedule specifies the following:

- A registered establishment in which mung beans are prepared or inspected for export:
 - must be equipped and operated in a manner which permits effective pest control and hygienic conditions to be maintained at the establishment
 - must have a defined program of hygiene and pest control.
- All machinery, equipment and surrounding floor area must be thoroughly cleaned of all waste material and debris at intervals not exceeding one week, or at such other times as an approved inspector considers necessary.
- Mung bean debris and waste must be removed from areas where mung beans are prepared each day and removed from the establishment each week.

- Any material likely to contaminate, infest or provide a source of infestation of mung beans must not be stored or handled in a building or area used for their preparation or storage or in any area likely to create a source of contamination.
- Toxic substances and other substances which may contaminate mung beans must not be stored in an area or a building where mung beans are handled or stored.
- Animals (including birds and rodents) must not be present in the establishment where preparation of mung beans takes place.
- A person who:
 - is suffering from a communicable disease; or
 - is a carrier of a communicable disease; or
 - may transmit pathogenic organisms to mung beans;
- must not enter any registered establishment used for the preparation of mung beans.
- All persons handling mung beans must maintain a high degree of personal cleanliness.
- Handwashing facilities and toilet facilities must be kept in a clean and sanitary condition at all times.

Additionally there are specific packaging requirements for mung beans (packaging materials must adequately protect the mung beans from contamination) as well inspection procedures for pests and contaminants (Schedule 6A).

Summary of international Guidelines/Codes of Practice

Codex Alimentarius

Codex has developed a Code of Hygienic Practice for Fresh Fruits and Vegetables which includes an Annex for Sprout Production. The Annex recommends control measures to occur in two areas: during seed production and during sprout production. During seed production, conditioning and storage, the application of Good Agricultural Practices (GAPs) and good Hygieninc Practices (GHPs) are aimed at preventing microbial pathogen contamination of seeds. During sprout production, good hygieninc practices are aimed at preventing the introduction of microbial pathogens and minimising their potential growth with a microbiological seed decontamination step included to reduce potential contaminants. A summary of the measures included in the annex is provided below.

Codex Code of Hygienic Practice for Fresh Fruits and Vegetables – ANNEX II Annex for Sprout Production							
Step in production chain		Control measures included (additional to those specified in the Code of Hygienic Practice for Fresh Fruits and Vegetables)					
Ρι	Primary production of seeds:						
•	Hygienic production of seeds	 Manure and biosolids: Wild or domestic animals should not be allowed to graze in the fields, Manure, biosolids and other natural fertilizers should only be used when they have undergone a pathogen reduction treatment. Agricultural chemicals: Only chemicals (e.g. pesticides, desiccants) which are acceptable for seeds intended for the production of sprouts for human consumption should be used. 					
•	Handling, storage and transport	 Segregation of seed intended for sprout production from seed intended for forage crops and clear labelling. Maintain sanitation in drying yards. 					
•	Analyses	 Lots of seeds should be tested for microbial pathogens (seed producers, distributors and sprout producers). If contamination found, seeds to be diverted or destroyed. 					
•	Recall Procedures	 Recall procedures in place to enable complete and rapid recall of implicated seed. Practices should minimise the quantity of seed identified as a single lot and avoid mixing of multiple lots. Records kept for each lot. Lot number, producer and country of origin should be indicated on each container. System in place to effectively identify lots, trace production sites and inputs. 					
E	stablishment for Sprout	Production:					
•	Design and layout of establishment	 Storage, seed rinsing, microbiological decontamination, germination and packaging area should be physically separated. 					
C	Control of Operation						
•	Water use	 Quality of water used dependent on stage of operation (clean water for initial washing staged, potable water in later production processes). 					
•	Initial rinse	 Seeds rinsed and thoroughly agitated in large volumes of clean water (maximise surface contact). Process should be repeated until rinse water remains clear. 					
•	Microbiological	 Recommended that seeds are treated prior to use. Seeds should be 					

decontamination	agitated in large volumes of antimicrobial agent to maximise surface contact. Duration of treatment/concentration of agent should be accurately recorded.		
Rinse after seed treatment	 As appropriate to eliminate any antimicrobial agent 		
Pre-germination soak	 Seeds should be soaked in cleaned water for the shortest possible time (to minimise microbial growth). After soaking seeds should be rinsed with potable water. 		
Germination	 Only potable water should be used Soils and other matrices should be treated to achieve a high degree of microbial reduction 		
Harvest	 Harvesting should be done with dedicated, cleaned and disinfected tools. 		
Final Rinse and cooling	 As appropriate, rinse with cool potable water Water should be changed to prevent cross-contamination Drain sprouts using appropriate equipment Steps to facilitate rapid cooling should be taken (if additional cooling time necessary) 		
Storage	 Sprouts should be kept under cold temperature (5°C to minimise microbial growth for the intended shelf life of the product (as appropriate) 		
 Microbiological and other specifications 	 Recommended that seed and sprouts or spent irrigation water be tested for the presence of pathogens. Each new lot of seeds received at the sprouting facility should be tested before entering production Producers should have in place sampling/testing plan to regularly monitor for pathogens at one or more stages after the start of germination (e.g. spent irrigation water, finished product). Recommended that every production lot is tested. 		
Microbiological cross- contamination	 Traffic patterns should prevent cross-contamination of sprouts 		
Incoming Material Require	ments		
Seed specifications	 Sprout producers should require evidence from seed producers that product was grown in accordance with measures outlined under primary production of seeds (assurance that chemical residues are within limits and certificates of analysis for microbial pathogens) 		
Control of incoming seeds	 Seed containers should be examined for physical damage and signs of contamination (particularly from pests). Seed lots analysed for the presence of microbial pathogens should not be used until results available. 		
Seed storage	 Seeds should be stored to prevent mould and bacterial growth and facilitate pest control Open containers should be stored such that they are protected from pests and other sources of contamination 		
Documentation and Recor	ds		
Documentation and Records	 Records should be maintained of the seed supplier, the lot number and country of origin to facilitate recall procedures. Records must include seed sources and lot numbers; water analysis results, production volumes, storage temperature monitoring, product distribution and consumer complaints. 		
Awareness and responsibi	lities		
 Awareness and responsibilities 	 Producer should have a written training program that is routinely reviewed and updated. Systems should be in place to ensure food handlers remain aware of all procedures necessary to maintain safety of product. 		

Weblinks for other international documents:

Canadian Code of Practice for the Hygienic Production of Sprouted Seeds http://www.inspection.gc.ca/english/fssa/frefra/safsal/sprointe.shtml

Reducing Microbial Food safety Hazards for Sprouted Seeds – guidance for Industry (US FDA) http://www.cfsan.fda.gov/~dms/sprougd1.html

http://www.cfsan.fda.gov/~dms/sprougd2.html

Code of Practice for food Safety in the Fresh Produce Supply Chain in Ireland (Chapter 4: Microbiological Safety of Sprouted seed Production) http://www.fsai.ie/assets/0/86/204/7332e0dd-fc90-45a0-a633-79c8066863ec.pdf

Attachment 7

SDC Membership

Name	Sector represented	Role
	Government	
Ms Catherine Bass	New South Wales	Manager - Program Evaluation, New South Wales Food Authority
Mr Bill Calder Mr Stan Goodchild – Proxy	Western Australia	Senior Project officer, Department of Health Western Australia
Mr Paul Dowsett	South Australia	Manager – Food Safety, Department of Primary Industries South Australia
Ms Katie Fullerton	Dept of Health and Ageing	Coordinating Epidemiologist, OzFoodNet
Ms Kira Goodall	Victoria	Policy Analyst - Agriculture and Forestry, Department Primary Industry. Victoria
Dr Olivia McQuestin	Tasmania	Senior Advisor, Environmental Health Unit, Tasmania Department of Health
Mr Phil Pond Mr Brian Witherspoon - Proxy	Queensland	General Manager - Strategy, Policy Development, Safe Food Production Queensland
Ms Usha Sriram-Prasad Ms Narelle Marro – Proxy	Dept of Agriculture, Fisheries and Forestry	Manager – Food Regulation and Safety, Department of Agriculture, Fisheries and Forestry
Ms Marion Castle	New Zealand	Programme Manager – Production and Processing, New Zealand Food Safety Authority
	Industry	
Mr Richard Bennett	Horticulture Australia Limited	Product Integrity Manager, Horticulture Australia Limited
Mr Andrew Boundy	Mungbean growers and processors	Executive Officer - Australian Mungbean Association
Mr Alan Davey	Rural Industry Research & Development Corporation	Senior Research Manager - New plant products, Asian Foods (Rural Industry Research & Development Corporation)
Ms Michele van der Sander	Sprout producers	Technical and Quality Assurance Manager - Parilla Fresh
Ms Patricia Donald	Sprout producers	Quality Manager – Healthy Sprout Company
Mr Stephen Donnelly	Rural Industry Development - seeds and pulses sales (nominated by the Grain Research & Development Corporation)	Director of Regal Seed and Grain, Managing Director of Blue Ribbon Seed and Pulse Exporters.
Ms Alison Gallagher	Woolworth Limited	Quality Manager - Fresh Foods
Mr Will Golsby Mr Tim Teague – Proxy	Sowing Seed industry	CEO of Australian Seed Federation
Dr Andreas Klieber	Coles Supermarkets	Technical Manager - Fresh Produce

Name	Sector represented	Role
Mr Andrew Phin Mrs Michele Phin – Proxy	Seed growers and processors	Managing Director of Booborowie Seed Pty Ltd
Mr James Rattray	Sprout producers	Director, Flowerdale Sprout Farm – Victoria
Mr Rob Sanders	Seed growers and processors	Director of Lucerne Australia
	Consumer	
Mr George Seymour- Dearness	Consumer	Legal professional
	FSANZ	
Mr Steve McCutcheon Ms Melanie Fisher - Alternative	Chair of the Standard Development Committee	Chief Executive Officer of FSANZ

References

Abelson, P., Forbes, M.P. and Hall, G. (2006) Australian Government Department of Health and Ageing. The annual cost of food-borne illness in Australia.

Abelson, P. (2007) Establishing a Monetary Value for Lives Saved: Issues and Controversies: WP 2008-02, Department of Finance and Deregulation, <u>http://www.finance.gov.au/obpr/docs/Working-paper-2-Peter-Abelson.pdf</u> (accessed 10 June 2009).

Bari, M.L., Nazuka, E., Sabina, Y., Todoriki, S. and Isshiki, K. (2003) Chemical and irradiation treatments for killing Escherichia coli O157:H7 on alfalfa, raddish, and mung bean seeds. *Food Protection* 66(5):767-7.

Barret, E. and Chaos, C. (1996) An outbreak of salmonellosis associated with eating alfalfa sprouts, Lexington, Virginia. Sproutnet. <u>http://www.sproutnet.com/Research/an_outbreak_of_salmonellosis.htm</u>. Accessed on 24 February 2009.

Beuchat, L.R. (1996) Pathogenic microorganisms associated with fresh produce. *Journal of Food Protection: 59* (2) 204-216 59(2):204-216.

Beuchat, L.R., Ward, T.E. and Pettigrew, C.A. (2001) Comparison of chlorine and a prototype produce wash product for effectiveness in killing Salmonella and Escherichia coli O157 : H7 on alfalfa seeds. *Journal of Food Protection* 64(2):152-158.

Bolton, D.J., Byrne, C.M., Sheridan, J.J., McDowell, D.A. and Blair, I.S. (1999) The survival characteristics of a non-toxigenic strain of Escherichia coli O157:H7. *J Appl Microbiol* 86(3):407-411.

Breuer, T., Benkel, D.H., Shapiro, R.L., Hall, W.N., Winnett, M.M., Linn, M.J., Neimann, J., Barrett, T.J., Dietrich, S., Downes, F.P., Toney, D.M., Pearson, J.L., Rolka, H., Slutsker, L. and Griffin, P.M. (2001) A multistate outbreak of Escherichia coli O157 : H7 infections linked to alfalfa sprouts grown from contaminated seeds. *Emerging Infectious Diseases* 7(6):977-982.

Brooks, J.T., Rowe, S.Y., Shillam, P., Heltzel, D.M., Hunter, S.B., Slutsker, L., Hoekstra, R.M. and Luby, S.P. (2001) Salmonella typhimurium infections transmitted by chlorine-pretreated clover sprout seeds. *American Journal of Epidemiology* 154(11):1020-1028.

CDC (1990) *Food-borne disease outbreak line listing.* Centers for Disease Control. <u>http://www.cdc.gov/food-borneoutbreaks/us_outb/fbo1990/fbofinal1990.pdf</u>. Accessed on 24 February 2009.

CDC (1996) *Listing of Food-borne Disease Outbreaks, United States, 1996.* Centres for Disease Control. <u>http://www.cdc.gov/food-borneoutbreaks/us_outb/fbo1996/fbofinal1996.pdf</u>. Accessed on 24 February 2009.

CDC (1998) *Listing of Food-borne Disease Outbreaks, United States, 1998.* Centres for Disease Control. <u>http://www.cdc.gov/food-borneoutbreaks/us_outb/fbo1998/fbofinal1998.pdf</u>. Accessed on 24 February 2009.

CDC (2000) *Listing of Food-borne Disease Outbreaks, United States, 2000.* Centres for Disease Control. <u>http://www.cdc.gov/food-borneoutbreaks/us_outb/fbo2000/fbofinal2000.pdf</u>. Accessed on 24 February 2009.

CDC (2002) Update on Salmonella serotype Enteritidis infections, outbreaks, and the importance for traceback. Centers for Disease Control. <u>http://www.cdc.gov/ncidod/dbmd/diseaseinfo/files/2001SECSTE.pdf</u>. Accessed on 24 February 2009.

CDC (2003) *Listing of Food-borne Disease Outbreaks, United States, 2003.* Centres for Disease Control. 24 February 2009.

CDC (2004) *Listing of Food-borne Disease Outbreaks, United States, 2004.* Centres for Disease Control. <u>http://www.cdc.gov/food-borneoutbreaks/us_outb/fbo2004/Outbreak_Linelist_Final_2004.pdf</u>. Accessed on 24 February 2009.

CDC (2006) *Listing of Food-borne Disease Outbreaks, United States, 2006.* Centres for Disease Control. <u>http://www.cdc.gov/food-borneoutbreaks/documents/2006_line_list/2006_line_list.pdf</u>. Accessed on 24 February 2009.

Charkowski, A.O., Barak, J.D., Sarreal, C.Z. and Mandrell, R.E. (2002) Differences in growth of Salmonella enterica and Escherichia coli O157 : H7 on alfalfa sprouts. *Applied and Environmental Microbiology* 68(6):3114-3120.

Charkowski, A.O., Sarreal, C.Z. and Mandrell, R.E. (2001) Wrinkled alfalfa seeds harbor more aerobic bacteria and are more difficult to sanitize than smooth seeds. *Journal of Food Protection* 64(9):1292-1298.

Cooley, M.B., Miller, W.G. and Mandrell, R.E. (2003) Colonization of Arabidopsis thaliana with Salmonella enterica and enterohemorrhagic Escherichia coli O157 : H7 and competition by Enterobacter asburiae. *Applied and Environmental Microbiology* 69(8):4915-4926.

Costerton, J.W., Lewandowski, Z., Caldwell, D.E., Korber, D.R. and Lappin-Scott, H.M. (1995) Microbial biofilms. *Annu.Rev Microbiol* 49:711-745.

Cover, T.L. and Aber, R.C. (1989) Yersinia enterocolitica. N Engl.J Med 321(1):16-24.

Delaquis, P.J., Sholberg, P.L. and Stanich, K. (1999) Disinfection of mung bean seed with gaseous acetic acid. *Journal of Food Protection* 62(8):953-957.

Department of Health WA. (2002) Microbiological safety and quality of sprouts in Western Australia. *Food Surveillance Newsletter* Summer:1-4.

Dong, Y.M., Iniguez, A.L., Ahmer, B.M.M. and Triplett, E.W. (2003) Kinetics and strain specificity of rhizosphere and endophytic colonization by enteric bacteria on seedlings of Medicago sativa and Medicago truncatula. *Applied and Environmental Microbiology* 69(3):1783-1790.

Emberland, K.E., Ethelberg, S., Kuusi, M., Vold, L., Jensvoll, L., Lindstedt, B.A., Nygard, K., Kjelso, C., Torpdahl, M., Sorensen, G., Jensen, T., Lukinmaa, S., Niskanen, T. and Kapperud, G. (2007) Outbreak of Salmonella Weltevreden infections in Norway, Denmark and Finland associated with alfalfa sprouts, July-October 2007. *Eurosurveillance* 12(11):

Enomoto, K., Takizawa, T., Ishikawa, N. and Suzuki, T. (2002) Hot-water treatments for disinfecting alfalfa seeds inoculated with Escherichia coli ATCC 25922. *Food Science and Technology Research* 8(3):247-251.

Ferguson, D.D., Scheftel, J., Cronquist, A., Smith, K., Woo-Ming, A., Anderson, E., Knutsen, J., De, A.K. and Gershman, K. (2005) Temporally distinct Escherichia coli O157 outbreaks associated with alfalfa sprouts linked to a common seed source - Colorado and Minnesota, 2003. *Epidemiology and Infection* 133(3):439-447.

Fett, W.F. (2000) Naturally occurring biofilms on alfalfa and other types of sprouts. *Journal of Food Protection* 63(5):625-632.

Fett, W.F. (2002a) Factors affecting the efficacy of chlorine against Esherichia coli O157 : H7 and Salmonella on alfalfa seed. *Food Microbiology* 19(2-3):135-149.

Fett, W.F. (2002b) Reduction of Escherichia coli O157:H7 and Salmonella spp. on laboratory-inoculated mung bean seed by chlorine treatmentt. *J Food Prot* 65(5):848-852.

Fett, W.F. (2002c) Reduction of the native microflora on alfalfa sprouts during propagation by addition of antimicrobial compounds to the irrigation water. *International Journal of Food Microbiology* 72(1-2):13-18.

Fett, W.F. (2006a) Inhibition of Salmonella enterica by plant-associated pseudomonads in vitro and on sprouting alfalfa seed. *J Food Prot* 69(4):719-728.

Fett, W.F. (2006b) Interventions to ensure the microbial safety of sprouts. In: Saper, G.M., Gorny, J.R., and Yousef, A.E. eds. *Microbiology of fruit and vegetables*. Chapter 8. CRC Press, Boca Raton, FL, US, pp. 187-210.

Fett, W.F. and Cooke, P.H. (2003a) Reduction of Escherichia coli O157 : H7 and Salmonella on laboratoryinoculated alfalfa seed with commercial citrus-related products. *Journal of Food Protection* 66(7):1158-1165.

Fett, W.F. and Cooke, P.H. (2003b) Scanning electron microscopy of native biofilms on mung bean sprouts. *Can.J Microbiol* 49(1):45-50.

Fu, T.J., Reineke, K.F., Chirtel, S. and Vanpelt, O.M. (2008) Factors influencing the growth of Salmonella during sprouting of naturally contaminated alfalfa seeds. *Journal of Food Protection* 71(5):888-896.

Gandhi, M., Golding, S., Yaron, S. and Matthews, K.R. (2001) Use of green fluorescent protein expressing Salmonella Stanley to investigate survival, spatial location, and control on alfalfa sprouts. *J Food Prot.* 64(12):1891-1898.

Gill, C.J., Keene, W.E., Mohle-Boetani, J.C., Farrar, J.A., Waller, P.L., Hahn, C.G. and Cieslak, P.R. (2003) Alfalfa seed decontamination in a Salmonella outbreak. *Emerging Infectious Diseases* 9(4):474-479.

Hall, G., Kirk, M.D., Becker, N., Gregory, J.E., Unicomb, L., Millard, G., Stafford, R., Lalor, K., and the OzFoodNet Working Group (2005) Estimating food-borne gastroenteritis, Australia. *Emerging Infectious Diseases* 11(8): 1257-1264.

Harman, G.E. (1983) mechanisms of seed infection and pathogenesis. *Phytopathology* 73(2):326-329.

Harris, L.J., Farber, J.N., Beuchat, L.R., Parish, M.E., Suslow, T.V., Garrett, E.H. and Busta, F.F. (2003) Outbreaks associated with fresh produce: Incidence, growth, and survival of pathogens in fresh and fresh-cut produce . *Comprehensive Reviews in Food Science and Food Safety* 2(s1):78-141.

Himathongkham, S., Nuanualsuwan, S., Riemann, H. and Cliver, D.O. (2001) Reduction of Escherichia coli O157 : H7 and Salmonella Typhimurium in artificially contaminated alfalfa seeds and mung beans by fumigation with ammonia. *Journal of Food Protection* 64(11):1817-1819.

Holliday, S.L., Scouten, A.J. and Beuchat, L.R. (2001) Efficacy of chemical treatments in eliminating Salmonella and Escherichia coli O157 : H7 on scarified and polished alfalfa seeds. *Journal of Food Protection* 64(10):1489-1495.

Honish, L. and Nguyen, Q. (2001) Outbreak of Salmonella enteritidis phage type 913 gastroenteritis associated with mung bean sprouts--Edmonton, 2001. *Can.Commun Dis Rep* 27(18):151-156.

Howard, M.B. and Hutcheson, S.W. (2003) Growth dynamics of Salmonella enterica strains on alfalfa sprouts and in waste seed irrigation water. *Applied and Environmental Microbiology* 69(1):548-553.

Inami, G.B. and Moler, S.E. (1999) Detection and isolation of Salmonella from naturally contaminated alfalfa seeds following an outbreak investigation. *Journal of Food Protection* 62(6):662-664.

Itoh, Y., Sugita-Konishi, Y., Kasuga, F., Iwaki, M., Hara-Kudo, Y., Saito, N., Noguchi, Y., Konuma, H. and Kumagai, S. (1998) Enterohemorrhagic Escherichia coli O157 : H7 present in radish sprouts. *Applied and Environmental Microbiology* 64(4):1532-1535.

Jaquette, C.B., Beuchat, L.R. and Mahon, B.E. (1996) Efficacy of chlorine and heat treatment in killing Salmonella stanley inoculated onto alfalfa seeds and growth and survival of the pathogen during sprouting and storage. *Appl Environ Microbiol* 62(7):2212-2215.

Joce, R., O'Sullivan, D.G., Strong, C., Rowe, B., Hall, M.L.M. and Threlfall, E.J. (1990) A national outbreak of *Salmonella* Gold-Coast. *Communicable Disease Report Review* 4:3-4.

Johnston, L.M., Elhanafi, D., Drake, M. and Jaykus, L.A. (2005) A simple method for the direct detection of Salmonella and Escherichia coli O157:H7 from raw alfalfa sprouts and spent irrigation water using PCR. *J Food Prot* 68(11):2256-2263.

Kemmeren, J.M., Mangen, M.-J.J., Duynhoven, Y.T.H.P. van, Havelaar, A.H., (2006) RIVM: Priority setting of food-borne pathogens - Disease burden and costs of selected enteric pathogens: http://www.rivm.nl/bibliotheek/rapporten/330080001.html (accessed 10 June 2009).

Kim, C., Hung, Y.C., Brackett, R.E. and Lin, C.S. (2003) Efficacy of electrolyzed oxidizing water in inactivating Salmonella on alfalfa seeds and sprouts. *Journal of Food Protection* 66(2):208-214.

Kim, H.J., Lee, D.S. and Paik, H.D. (2004) Characterization of Bacillus cereus isolates from raw soybean sprouts. *Journal of Food Protection* 67(5):1031-1035.

Kirk, M., (2006) Outbreaks Associated with Raw Sprouts. Ozfoodnet presentation, Presented at the workshop of "Food Safety and Sprouts", held at the Tiffins on the Park, Adelaide, 20 July 2006. Kocharunchitt, C., Ross, T. and McNeil, D.L. (2009) Use of bacteriophages as biocontrol agents to control Salmonella associated with seed sprouts. *Int J Food Microbiol* 128(3):453-459.

Lang, M.M., Ingham, B.H. and Ingham, S.C. (2000) Efficacy of novel organic acid and hypochlorite treatments for eliminating Escherichia coli O157:H7 from alfalfa seeds prior to sprouting. *International Journal of Food Microbiology* 58:73-82.

Liao, C.H. (2008) Growth of Salmonella on sprouting alfalfa seeds as affected by the inoculum size, native microbial load and Pseudomonas fluorescens 2-79. *Letters in Applied Microbiology* 46(2):232-236.

Liu, B. and Schaffner, D.W. (2007) Quantitative analysis of the growth of *Salmonella* Stanley during alfalfa sprouting and evaluation of *Enterobacter aerogenes* as its surrogate. *Journal of Food Protection* 70(2):136-322.

Mahon, B.E., Ponka, A., Hall, W.N., Komatsu, K., Dietrich, S.E., Siitonen, A., Cage, G., Hayes, P.S., Lambert-Fair, M.A., Bean, N.H., Griffin, P.M. and Slutsker, L. (1997) An international outbreak of Salmonella infections caused by alfalfa sprouts grown from contaminated seeds. *J Infect Dis* 175(4):876-882.

Mattila, L., Leirisalo-Repo, M., Koskimies, S., Granfors, K. and Siitonen, A. (1994) Reactive arthritis following an outbreak of Salmonella infection in Finland. *Br.J Rheumatol.* 33(12):1136-1141.

Mazzoni, A.M., Sharma, R.R., Demirci, A. and Ziegler, G.R. (2001) Supercritical carbon dioxide treatment to inactivate aerobic microorganisms on alfalfa seeds. *Journal of Food Safety* 21(4):215-223.

Michino, H., Araki, K., Minami, S., Takaya, S., Sakai, N., Miyazaki, M., Ono, A. and Yanagawa, H. (1999) Massive outbreak of Escherichia coli O157 : H7 infection in schoolchildren in Sakai City, Japan, associated with consumption of white radish sprouts. *American Journal of Epidemiology* 150(8):787-796.

Millard, G. and Rockliff, S. (2001) Microbiological quality of seed sprouts. ACT Health Protection Service.

Ministry of Agriculture and Forestry (2003) *Trends and sources of zoonotic agents in animals, feedingstuffs, food and man in Finland in 2002.* Department of Food and Health, Finland. <u>http://wwwb.mmm.fi/el/julk/kuvat/zoon/02/table12.0.pdf#search=%22s.%20abony%20and%20finland%20and%20 mung%20bean%20sprouts%22</u>. Accessed on 24 February 2009.

Mohle-Boetani, J., Werner, B., Polumbo, M., Farrer, J., Vugia, D., Komatsu, K., Tagg, K., Peterson, N., Painter, J., Van Dunn, S., Winthrop, K. and Beatty, M. (2002) Outbreak of Salmonella serotype Kottbus infections associated with eating alfalfa sprouts--Arizona, California, Colorado, and New Mexico, February-April 2001. *MMWR Morb Mortal Wkly Rep* 51(1):7-9.

Mohle-Boetani, J.C., Farrar, J.A., Werner, S.B., Minassian, D., Bryant, R., Abbott, S., Slutsker, L. and Vugia, D.J. (2001) Escherichia coli O157 and Salmonella infections associated with sprouts in California, 1996-1998. *Annals of Internal Medicine* 135(4):239-247.

Montville, R. and Schaffner, D. (2005) Monte Carlo simulation of pathogen behavior during the sprout production process. *Appl Environ Microbiol* 71(2):746-753.

Montville, R. and Schaffner, D.W. (2004) Analysis of published sprout seed sanitization studies shows treatments are highly variable. *Journal of Food Protection* 67(4):758-765.

Mundt, J.O. and Hinkle, N.F. (1976) Bacteria within ovules and seeds. Appl Environ Microbiol 32(5):694-698.

NACMCF. (1999a) Microbiological safety evaluations and recommendations on sprouted seeds. *International Journal of Food Microbiology* 52(3):123-153.

NACMCF (1999b) *Microbiological safety evaluations and recommendations on sprouted seeds.* http://www.cfsan.fda.gov/~mow/sprouts2.html. Accessed on 19 January 2009b.

Nelson, S.O., Lu, C.Y., Beuchat, L.R. and Harrison, M.A. (2002) Radio-frequency heating of alfalfa seed for reducing human pathogens. *Transactions of the Asae* 45(6):1937-1942.

O'Mahony, M., Cowden, J., Smyth, B., Lynch, D., Hall, M., Rowe, B., Teare, E.L., Tettmar, R.E., Rampling, A.M. and Coles, M. (1990) An outbreak of Salmonella saint-paul infection associated with beansprouts. *Epidemiol Infect* 104(2):229-235.

OzFoodNet. (2006) Burden and causes of food-borne disease in Australia: Annual report of the OzFoodNet network, 2005. *Commun Dis Intell* 30(3):278-300.

OzFoodNet. (2007) Monitoring the incidence and causes of diseases potentially transmitted by food in Australia: annual report of the Ozfoodnet Network, 2006. *Commun Dis Intell* 31(4):345-365.

Palmai, M. and Buchanan, R.L. (2002) Growth of Listeria monocytogenes during germination of alfalfa sprouts. *Food Microbiology* 19(2-3):195-200.

Pandrangi, S., Elweeell, M.W., Anantheswaran, R.C. and LaBorde, L.F. (2003) Efficacy of sulfuric acid scarification and disinfectant treatments in eliminating Escherichia coli O157 : H7 from alfalfa seeds prior to sprouting. *Journal of Food Science* 68(2):613-618.

Pao, S., Khalid, M.F. and Kalantari, A. (2005) Microbial profiles of on-line-procured sprouting seeds and potential hazards associated with enterotoxigenic Bacillus spp. in homegrown sprouts. *Journal of Food Protection* 68(8):1648-1653.

Park, C.M., Taormina, P.J. and Beuchat, L.R. (2000) Efficacy of allyl isothiocyanate in killing enterohemorrhagic Escherichia coli O157 : H7 on alfalfa seeds. *International Journal of Food Microbiology* 56(1):13-20.

Ponka, A., Andersson, Y., Siitonen, A., de, J.B., Jahkola, M., Haikala, O., Kuhmonen, A. and Pakkala, P. (1995) Salmonella in alfalfa sprouts. *Lancet* 345(8947):462-463.

Portnoy, B.L., Goepfert, J.M. and Harmon, S.M. (1976) An outbreak of Bacillus cereus food poisoning resulting from contaminated vegetable sprouts. *Am J Epidemiol* 103(6):589-594.

Proctor, M.E., Hamacher, M., Tortorello, M.L., Archer, J.R. and Davis, J.P. (2001) Multistate outbreak of Salmonella serovar Muenchen infections associated with alfalfa sprouts grown from seeds pretreated with calcium hypochlorite. *Journal of Clinical Microbiology* 39(10):3461-3465.

Prokopowich, D. and Blank, G. (1991) Microbiological evaluation of vegetable sprouts and seeds. *Journal of Food Protection:* 54 (7) 560-562 54(7):560-562.

Puohiniemi, R., Heiskanen, T. and Siitonen, A. (1997) Molecular epidemiology of two international sprout-borne Salmonella outbreaks. *Journal of Clinical Microbiology* 35(10):2487-2491.

Rajkowski, K.T., Boyd, G. and Thayer, D.W. (2003) Irradiation D-values for Escherichia coli O157:H7 and Salmonella sp. on inoculated broccoli seeds and effects of irradiation on broccoli sprout keeping quality and seed viability. *J Food Prot* 66(5):760-766.

Rajkowski, K.T. and Thayer, D.W. (2000) Reduction of Salmonella spp. and strains of Escherichia coli O157 : H7 by gamma radiation of inoculated sprouts. *Journal of Food Protection* 63(7):871-875.

RIRDC (2008) *Economic Analysis of the Australian Lucerne Seed Industry*. Rural Industries Resarch and Development Corporation. <u>http://www.rirdc.gov.au/reports/PSE/08-103.pdf</u>. Accessed on 7 January 9 A.D.

Robertson, L.J., Johannessen, G.S., Gjerde, B.K. and Loncarevic, S. (2002) Microbiological analysis of seed sprouts in Norway. *International Journal of Food Microbiology* 75(1-2):119-126.

Samadpour, M., Barbour, M.W., Nguyen, T., Cao, T.M., Buck, F., Depavia, G.A., Mazengia, E., Yang, P., Alfi, D., Lopes, M. and Stopforth, J.D. (2006) Incidence of enterohemorrhagic Escherichia coli, Escherichia coli O157, Salmonella, and Listeria monocytogenes in retail fresh ground beef, sprouts, and mushrooms. *Journal of Food Protection* 69(2):441-443.

Sewell, A.M. and Farber, J.M. (2001) Food-borne outbreaks in Canada linked to produce. *Journal of Food Protection* 64(11):1863-1877.

Sharma, R.R. and Demirci, A. (2003a) Inactivation of Escherichia coli O157 : H7 on inoculated alfalfa seeds with pulsed ultraviolet light and response surface modeling. *Journal of Food Science* 68(4):1448-1453.

Sharma, R.R. and Demirci, A. (2003b) Treatment of Escherichia coli O157 : H7 inoculated alfalfa seeds and sprouts with electrolyzed oxidizing water. *International Journal of Food Microbiology* 86(3):231-237.

Sharma, R.R., Demirci, A., Beuchat, L.R. and Fett, W.F. (2002) Inactivation of Escherichia coli O157 : H7 on inoculated alfalfa seeds with ozonated water and heat treatment. *Journal of Food Protection* 65(3):447-451.

Sinton, L.W., Braithwaite, R.R., Hall, C.H. and Mackenzie, M.L. (2007) Survival of Indicator and Pathogenic Bacteria in Bovine Feces on Pasture. *Applied and Environmental Microbiology* 73(24):7917-7925.

Stan, S.D. and Daeschel, M.A. (2003) Reduction of Salmonella enterica on alfalfa seeds with acidic electrolyzed oxidizing water and enhanced uptake of acidic electrolyzed oxidizing water into seeds by gas exchange. *J Food Prot* 66(11):2017-2022.

Stewart, D., Reineke, K., Ulaszek, J., Fu, T. and Tortorello, M. (2001a) Growth of Escherichia coli O157:H7 during sprouting of alfalfa seeds. *Lett Appl Microbiol* 33(2):95-99.

Stewart, D.S., Reineke, K.F., Ulaszek, J.M. and Tortorello, M.L. (2001b) Growth of Salmonella during sprouting of alfalfa seeds associated with salmonellosis outbreaks. *Journal of Food Protection* 64(5):618-622.

Stratton, J., Stefaniw, L., Grimsrud, K., Werker, D.H., Ellis, A., Ashton, E., Chui, L., Blewett, E., Ahmed, R., Clark, C., Rodgers, F., Trottier, L. and Jensen, B. (2001) Outbreak of Salmonella paratyphi B var java due to contaminated alfalfa sprouts in Alberta, British Columbia and Saskatchewan. *Can.Commun Dis Rep* 27(16):133-137.

Taormina, P.J. and Beuchat, L.R. (1999) Comparison of chemical treatments to eliminate enterohemorrhagic Escherichia coli O157:H7 on alfalfa seeds. *J Food Prot* 62(4):318-324.

Taormina, P.J., Beuchat, L.R. and Slutsker, L. (1999) Infections associated with eating seed sprouts: an international concern. *Emerg.Infect Dis* 5(5):626-634.

Thayer, D.W., Rajkowski, K.T., Boyd, G., Cooke, P.H. and Soroka, D.S. (2003) Inactivation of Escherichia coli O157 : H7 and Salmonella by gamma irradiation of alfalfa seed intended for production of food sprouts. *Journal of Food Protection* 66(2):175-181.

US FDA (2008) *Bad Bug Book*. US Food and Drug Administration. <u>http://vm.cfsan.fda.gov/~mow/intro.html</u>. Accessed on 25 February 8 A.D.

van Beneden, C.A., Keene, W.E., Strang, R.A., Werker, D.H., King, A.S., Mahon, B., Hedberg, K., Bell, A., Kelly, M.T., Balan, V.K., MacKenzie, W.R. and Fleming, D. (1999) Multinational outbreak of Salmonella enterica serotype Newport infections due to contaminated alfalfa sprouts. *Journal of the American Medical Association:* 281 (2) 158-162 281(2):158-162.

van Duynhoven, Y.T.H.P., Widdowson, M.A., de Jager, C.M., Fernandes, T., Neppelenbroek, S., van den Brandhof, W., Wannet, W.J.B., van Kooij, J.A., Rietveld, H.J.M. and van Pelt, W. (2002) Salmonella enterica serotype enteritidis phage type 4b outbreak associated with bean sprouts. *Emerging Infectious Diseases* 8(4):440-443.

Venczel, L.V., Arrowood, M., Hurd, M. and Sobsey, M.D. (1997) Inactivation of Cryptosporidium parvum oocysts and Clostridium perfringens spores by a mixed-oxidant disinfectant and by free chlorine. *Applied and Environmental Microbiology* 63(4):1598-1601.

Wade, W.N., Scouten, A.J., McWatters, K.H., Wick, R.L., Demirci, A., Fett, W.F. and Beuchat, L.R. (2003) Efficacy of ozone in killing Listeria monocytogenes on alfalfa seeds and sprouts and effects on sensory quality of sprouts. *Journal of Food Protection* 66(1):44-51.

Warriner, K., Spaniolas, S., Dickinson, M., Wright, C. and Waites, W.M. (2003) Internalization of bioluminescent Escherichia coli and Salmonella Montevideo in growing bean sprouts. *Journal of Applied Microbiology* 95(4):719-727.

Watanabe, Y., Ozasa, K., Mermin, J.H., Griffin, P.M., Masuda, K., Imashuku, S. and Sawada, T. (1999) Factory outbreak of Escherichia coli O157 : H7 infection in Japan. *Emerging Infectious Diseases* 5(3):424-428.

Weiss, A. and Hammes, W.P. (2003) Thermal seed treatment to improve the food safety status of sprouts. *Journal of Applied Botany-Angewandte Botanik* 77(5-6):152-155.

Weissinger, W.R. and Beuchat, L.R. (2000) Comparison of aqueous chemical treatments to eliminate Salmonella on alfalfa seeds. *Journal of Food Protection* 63(11):1475-1482.

Weissinger, W.R., Chantarapanont, W. and Beuchat, L.R. (2000) Survival and growth of Salmonella baildon in shredded lettuce and diced tomatoes, and effectiveness of chlorinated water as a sanitizer. *International Journal of Food Microbiology* 62(1-2):123-131.

Weissinger, W.R., McWatters, K.H. and Beuchat, L.R. (2001) Evaluation of volatile chemical treatments for lethality to Salmonella on alfalfa seeds and sprouts. *Journal of Food Protection* 64(4):442-450.

Winthrop, K.L., Palumbo, M.S., Farrar, J.A., Mohle-Boetani, J.C., Abbott, S., Beatty, M.E., Inami, G. and Werner, S.B. (2003) Alfalfa sprouts and Salmonella Kottbus infection: A multistate outbreak following inadequate seed disinfection with heat and chlorine. *Journal of Food Protection* 66(1):13-17.

Wu, F.M., Beuchat, L.R., Wells, J.G., Slutsker, L., Doyle, M.P. and Swaminathan, B. (2001) Factors influencing the detection and enumeration of Escherichia coli O157 : H7 on alfalfa seeds. *International Journal of Food Microbiology* 71(1):93-99.

Wuytack, E.Y., Diels, A.M.J., Meersseman, K. and Michiels, C.W. (2003) Decontamination of seeds for seed sprout production by high hydrostatic pressure. *Journal of Food Protection* 66(6):918-923.