

Survey of plant products not within the NSW Plant Products Food Safety Scheme



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EXECUTIVE SUMMARY

In 2005, the Plant Products Food Safety Scheme was introduced into legislation. The Scheme required businesses (with the exception of retail businesses) handling certain plant products to be licensed and operate under a food safety program. The products covered by the Scheme include fresh-cut fruits and vegetables, vegetables (or fruits) in oil, unpasteurised juices and seed sprouts. These products were included in the scheme based on a report by Food Science Australia (2000) which identified them as high risk.

Since then other plant-based product groups have increased their presence in the market such as soy products, fermented vegetables, vegetable-based dips & sauces, mixed salads, and fresh cut vegetables excluded in the plant products scheme (for example fresh herbs).

The Authority conducted a desktop (literature) review of these products (*Background Information – Plant Products not regulated under the NSW Plant Products Safety Scheme*) which found no specific hazards that would justify the products being classed as high risk and therefore included in the Food Safety Scheme.

As little information was available on the current microbiological status of these products, a survey was carried out to assess the microbiological quality and/or chemical properties of those products to confirm the findings of the desktop review.

The microbiological testing illustrated that the plants products tested were, in general, of a good or acceptable microbiological standard. The survey supported the finding of the CSIRO scoping study and literature review that the products are generally of lower risk when compared to those covered by the Scheme.

Results for tofu (100 samples), mixed salads (101 samples) and vegetable-based dips & sauces (100 samples) were assessed against the Authority's *microbiological guide for ready to eat food*. The microbiological testing illustrated that the products were, in general, of a good or acceptable microbiological standard, with 96.7% of samples tested falling into these categories.

A total of 10 samples (3.3%) were classified unsatisfactory due to an elevated level of *B. cereus,* CPS and/or *E. coli*.

- Three tofu samples were found to be unsatisfactory due to elevated level of coagulase positive staphylococci, *B. cereus*, and *B. cereus* & *E. coli*. Another sample was also unsatisfactory due to very low level of *Salmonella* (only positive by PCR and not by cultural method). *Bacillus cereus* is known to be present on soybeans and its heat resistant spores can germinate in the products. *E. coli* and *S. aureus* can be present in tofu due to its high protein content and the amount of handling subsequent to the heating process. However, correct refrigeration will inhibit the growth of these organisms.
- Three mixed salad samples were found to be unsatisfactory due to presence of *B. cereus*. This is not unexpected due to the presence of raw vegetables. *B. cereus* is present in soil and may contaminate vegetables used in the salad. The pH limit for growth of *B. cereus* is 5.0 under ideal conditions (ICMSF, 1996). One product had a pH of 4.4, thus it is expected that no growth would occur. The other two products had a pH of 5.0 and 5.2. However, the refrigerated storage of these products also means that growth is unlikely as reductions in storage temperature below the optimum will increase the minimum pH value for growth.
- Three (3%) vegetable-based dips and sauces tested in this survey were found to be unsatisfactory due to elevated level of *B. cereus* and *E. coli*. The unsatisfactory samples were hommus topped with paprika, hommus, and beetroot dip (containing tahini and chickpeas). These products have high water activity, high nutrient content and relatively neutral pH, which make for a favourable microbial media. However, the growth may be inhibited by presence of acids and refrigeration.



Thus, this survey highlighted the importance of keeping these products under refrigeration to ensure safety is maintained.

Bunched fresh herbs (103 samples) were also tested and were acceptable when compared to UK guidelines and EU regulation. Chopped fresh herbs, other than in mixed salads, were not found in the market place and were not included in the survey. However, there does appear to be a market for chopped parsley for use in kebab preparation observed in NSW. Further work on food service products, such as chopped herbs, is likely to result as a consequence of an investigation into a foodborne illness outbreak, which occurred in early 2014.



1. INTRODUCTION

In 2000, the NSW Food Authority (then SafeFood NSW) commissioned Food Science Australia to conduct a scoping study to determine the relative food safety risk of various plant products for human consumption, produced and/or marketed in NSW. The scoping study was conducted to assess the need to introduce a Food Safety Scheme for plant products.

The scoping study looked at twelve groups of plant products, including cereals and their products, vegetables and fruits and their products, spices, and plant based beverages. This scoping study identified five products as high risk based: fresh-cut vegetables consumed raw; fresh-cut fruits; vegetables (or fruits) in oil; unpasteurised juices; and seed sprouts (Table 1). In 2005, the Plant Products Food Safety Scheme was introduced into legislation. The Scheme required businesses (with the exception of retail businesses) handling those five product groups to be licensed and operate under a food safety program.

Table 1. Microbiological hazards associated with plant products regulated by the Plant Products Food Safety Scheme

Plant product	High risk ranking	Medium risk ranking
Fresh cut vegetables – may be consumed raw	Pathogenic <i>E. coli Salmonella</i> spp. <i>L. monocytogenes</i>	
Fresh cut vegetables – chilled, modified atmosphere packaging MAP) or extended shelf life	L. monocytogenes C. botulinum	
Fresh cut fruit	Pathogenic <i>E. coli</i> Salmonella spp. L. monocytogenes	Cryptosporidium parvum Enteric viruses
Fruit juice / drink (unpasteurised)	Pathogenic <i>E. coli Salmonella</i> spp.	
Vegetables in oil	C. botulinum	
Seed sprouts	Pathogenic <i>E. coli</i> Salmonella spp.	B. cereus L. monocytogenes

Adapted from Scoping study on the risk of plant products (FSA, 2000)

Since then the variety and volume of plant-based products have increased in at least some of the market categories. Chilled dips experienced 9 to 10% volume growth¹ in 2009 and 2010. Prepared salads had compound annual growth² in value of 5.8% from 2008 to 2013. Kimchi supply has grown as a consequence of Korean migration which has been running³ at about 5000 people per annum over the last 5 years. Most Korean immigrants settle in NSW and their influence is obvious in Sydney's inner west.

¹ Retail World Annual Reports

² Euromonitor International Analysis

³ Australian Government Department of Immigration and Citizenship



These products were included in the Food Science Australia scoping study and found to have low to medium microbiological risk. With the increase in variety and volume of these products, a review was required to assess if the risk rating assigned in the scoping study was still appropriate. To support the review, a survey of these products was undertaken to provide information on the intrinsic properties (pH, water activity and organic acid profile) of the products and to give an indication of their microbiological status.

Table 2. Plant product groups considered

Group	Example of products	Risk rating according to the scoping study (FSA, 2000)
Soy products	Tofu & fermented soy products (eg tempeh)	Medium risk for <i>B. cereus</i>
	Soy milk & milk products (eg soy yoghurt & soy cheese)	Low risk for <i>Salmonella</i> spp. and <i>Y. enterocolitica</i>
Fermented vegetables	Kimchi	
Vegetable based dips & sauces	Sesame-based dips (eg tahini, hommus & baba ghanoush) Salsa-style dips	For tahini & hommus: Medium risk for <i>Salmonella</i> spp. and <i>B. cereus</i>
	Pesto-style dips (eg pesto, tapenades)	For guacamole & olive tapenade: Medium risk for pathogenic E. coli, acid tolerant Salmonella spp., psychrotrophic B. cereus and Cl. botulinum
Salads (excluding fresh cut vegetables)	eg potato salad, rice salad, coleslaw, and other mixed salads	Salad with mayonnaise based dressing: Medium risk for pathogenic <i>E. coli</i> and <i>Salmonella</i> and low risk for <i>L. monocytogenes</i>
Fresh cut vegetables excluded in the FSS	Fresh herbs	Medium for Pathogenic <i>E. coli</i> , <i>Salmonella</i> spp., <i>Shigella</i> spp.
	Snow pea sprouts	Low for <i>B. cereus, L. monocytogenes,</i> Enteric parasites and viruses

Further information on these products can be found in the *Background Information – Plant Products not regulated under the NSW Plant Products Food Safety Scheme* document.



2. MATERIALS AND METHODS

2.1 Method for laboratory analysis

From March 2012 to May 2013, 404 samples were collected from retail outlets across Sydney. Samples included tofu, mixed salads, vegetable-based dips & sauces, and fresh herbs, all grown or manufactured in Australia. All samples, except fresh herbs, were pre-packaged.

Kimchi samples were purchased for method verification studies. However, they were not included in the final survey because all kimchi tested inhibited microbiological growth and the microbiological methods could not be verified.

Samples were analysed by the Food Microbiology and General Chemistry Laboratories of the NSW Forensic and Analytical Science Services (NSW FASS) at Lidcombe. Samples were kept in the original packaging, under temperature control (less than 5°C) at all times and were tested within the use-by-date. All samples were analysed within 24 hours of receipt at the laboratories using NATA accredited methods as detailed in Table 3.

Table 3. Tests conducted and their corresponding method

Tests	Method
Bacillus cereus enumeration	AS 1766.2.6
Coagulase positive Staphylococcus (CPS) enumeration	AS 1766.2.4
Listeria monocytogenes detection	PCR-based method for screening and AS 1766.16.1
Listeria monocytogenes enumeration	AS 5013.24.2
Salmonella detection	PCR-based method for screening and AS 1766.2.5
Standard Plate Count (SPC)	AS 1766.2.1
Thermotolerant coliforms & <i>E. coli</i> enumeration	AS 1766.2.3
рН	In house method F50.39 based on AOAC 981.12 & AS 2300.1.6 method
Water activity (a _w)	In house method F50.40 based on CSIRO published paper and AOAC 978.18 method
Organic acid profile	In house method F26



Due to the nature of the products, different food categories underwent different tests, as outlined in Table 4.

Table 4. Samples included in the survey and tests conducted

Food category	Tests
Tofu	B. cereus, CPS, L. monocytogenes detection, Salmonella, E. coli, pH, a _w
Mixed salads	<i>B. cereus</i> , CPS, <i>L. monocytogenes</i> enumeration, <i>Salmonella</i> , <i>E. coli</i> , pH ⁴
Vegetable-based dips & sauces	B. cereus, CPS, L. monocytogenes enumeration, Salmonella, E. coli, pH, a _w , organic acid profile
Fresh herbs	B. cereus, CPS, L. monocytogenes enumeration, Salmonella, E. coli

Before the survey commenced, method verification testing was undertaken for each food category to assess the ability to recover the microorganisms from the products. A selection of products from the different categories was inoculated with organisms (as listed in Table 4) and recovery levels were recorded.

For kimchi, it was planned that samples were to be tested for standard plate count, *B. cereus, CPS, L. monocytogenes* enumeration, *Salmonella, and E. coli*. However, during the verification testing, it was found that the recovery levels for pathogens from five different kimchi samples (manufactured by different manufacturers) were low, in particular for *B. cereus*. This means that the products were inhibitory to those organisms. This finding was similar to other studies where the growth of pathogenic or spoilage microorganisms were suppressed in such products, due to the following factors (Kim et al., 2008; Lee, 1997):

- The fermented kimchi has a pH of 4.1 to 4.5 and salt concentration of 3 to 5%. The salt concentration will suppress the growth of undesirable bacteria without affecting that of the lactic acid bacteria responsible for the fermentation,
- Some of the kimchi ingredients such as garlic, green onion, red pepper, and ginger are known to contain sulphur containing compounds that have antimicrobial activity against pathogens, and
- Lactic acid bacteria are known to produce a variety of antimicrobial agents such as organic
 acids, diacetyl, and hydrogen peroxide. Some also produce bacteriocins, which inhibit a
 variety of foodborne pathogens such as *B. cereus*.

This information would suggest that kimchi is inhibitory towards pathogens, resulting in the growth of the organisms being retarded or the inhibitory substances destroying or damaging the organisms. As such it was decided that kimchi would not be included in the survey.

⁴ Where the components of the salad were naturally separated within the package during storage, these were gently mixed (not stomached) to combine the salad ingredients prior to testing the pH. Because the combined ingredients in a mixed salad might have different pH values, three separate pH readings were taken and reported to provide a representative pH value. The pH value stated in this report is the average value of the three readings.



2.2 Method for data analysis

All food products tested in this survey, except for fresh herbs, fall into a large category of ready-to-eat (RTE) foods for which there are no microbiological standards in the Australian New Zealand Food Standards Code (the Code). The NSW Food Authority's *Microbiological quality guide for ready-to-eat foods*, as shown in Table 5. , was used to assess the microbiological results for all products except herbs. The results from the herb samples where compared to previous studies identified in the background information document.

Table 5. Guidelines for determining the microbiological quality of ready-to-eat foods (NSW Food Authority, 2009)

		Microbiological result (CFU/g)								
Test	Good	Acceptable	Unsatisfactory	Potentially hazardous						
Indicators										
Enterobacteriaceae ⁵	<10 ²	$10^2 \text{ to } < 10^4$	≥10 ⁴	N/A						
E. coli	<3	3 to <10 ²	≥10 ²	N/A						
Pathogens										
B. cereus	<10 ²	$10^2 \text{ to } < 10^3$	10 ³ to <10 ⁴	≥10 ⁴						
Coagulase positive staphylococci	<10 ²	10 ² to <10 ³	10 ³ to <10 ⁴	≥10⁴						
Salmonella	Not detected in 25g	-	-	Detected in 25g						
Listeria monocytoger	nes									
Food Group 1 ⁶	Not detected in 25g	-	-	Detected in 25g						
Food Group 2 ⁶	Not detected in 25g	Detected but <10 ²	-	≥10 ²						

All results were entered into a Microsoft[™] Excel spread sheet. Where enumeration was conducted, microbiological counts were transformed into log cfu/g. Frequency distributions were produced using the Histogram functionality of the data analysis tools of Microsoft[™] Excel.

 Food Group 1 applies to RTE food that will support the growth of L. monocytogenes and has been stored prepared for greater than one day (eg tofu)

⁵ In the absence of a guideline for thermotolerant coliform, these were compared with the guideline for *Enterobacteriaceae* as both groups of bacteria are similar (the bacteria detected by the coliform test are members of several genera within the family *Enterobacteriaceae*).

⁶ L. monocytogenes:

⁻ Food Group 2 applies to RTE food that will not support the growth of *L. monocytogenes* and has been stored prepared for greater than one day (eg mixed salads and vegetable-based dips & sauces)



3. RESULTS

3.1 Microbiological results - excluding fresh herbs

A summary of the microbiological results for the samples is shown in Table 6. When compared with the NSW Food Authority's *Microbiological quality guide for ready-to-eat foods*, 96.7% of samples were classified good or acceptable.

A total of 10 samples (3.3%) were classified unsatisfactory due to an elevated level of *B. cereus,* CPS and/or *E. coli*.

One sample of tofu produced a presumptive positive result for *Salmonella* by PCR, but the result could not be confirmed using cultural methods. Three further samples of the same product (different batches) were tested and *Salmonella* was not detected in any of them. This implies that *Salmonella* might be present in the sample at a very low level, likely less than one organism in 25 gram of product.

Where samples were found to be unsatisfactory, follow up action was undertaken by the Authority in accordance with the level of risk posed. Follow up action included notifying the manufacturer of the issue, to take appropriate action.

Table 6. Assessment of results for products using the microbiological criteria for ready-to-eat foods (NSW Food Authority, 2009)

Food category	No. of samples	Microbiological quality (%)							
		Good	Acceptable	Unsatisfactory	Potentially hazardous				
Tofu	100	86 (86%)	10 (10%)	4 (4%)	-				
Mixed salads	101	70 (69%)	28 (28%)	3 (3%)	-				
Vegetable-based dips & sauces	100	90 (90%)	7 (7%)	3 (3%)	-				

Frequency distributions for thermotolerant coliforms and *E. coli* for tofu, mixed salads and vegetable-based dips & sauces are presented in Figures 1, 2 and 3.



Figure 1. Frequency distribution for thermotolerant coliforms and $\it E.~coli$ in tofu samples

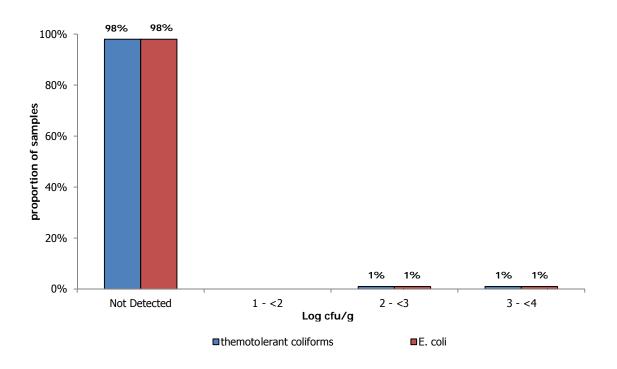


Figure 2. Frequency distribution for thermotolerant coliforms and $\it E.~coli$ in mixed salads samples

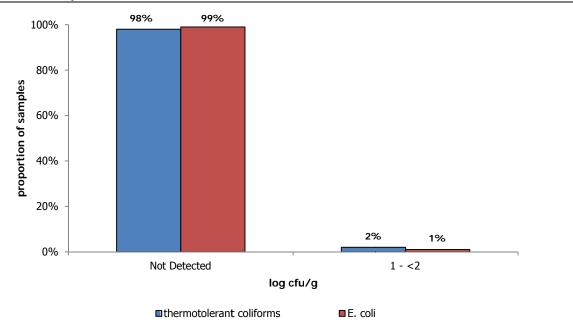
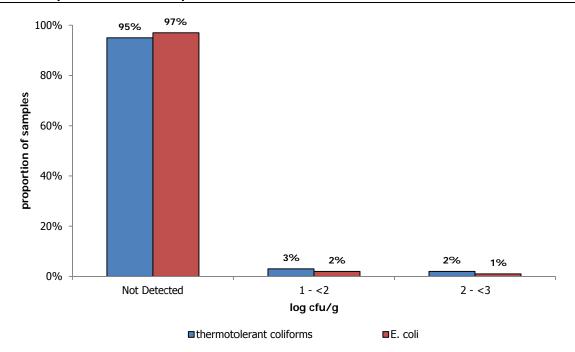




Figure 3. Frequency distribution for thermotolerant coliforms and *E. coli* in vegetable-based dips and sauces samples





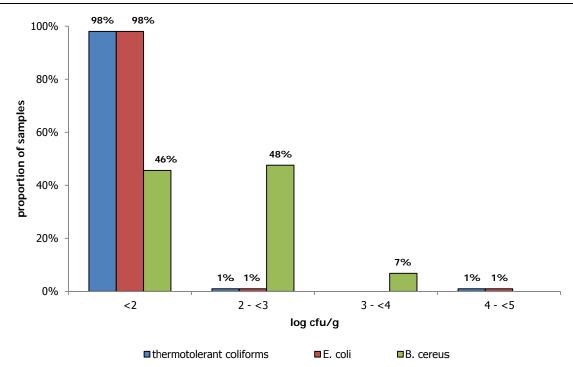
3.2 Microbiological results - fresh herbs

The scope of this survey was limited to readily available bunched fresh herbs, excluding those that are commonly intended to be cooked before use. A total of 103 samples were tested, including chives, parsley, coriander, chervil, mint, basil, and dill.

No *Salmonella* was detected in any of the samples. Also, no sample contained CPS or *L. monocytogenes* above the limit of detection (200 cfu/g and 20 cfu/g, respectively). Frequency distributions for thermotolerant coliforms, *E. coli* and *B. cereus* are presented in **Error! Reference source not found**. The graph shows that the majority of samples contained thermotolerant coliforms and *E. coli* at low level or under the limit of detection. A number of samples contained *B. cereus* at higher levels.

Elviss et al (2009) examined *E. coli* levels in fresh herbs and compared results to Public Health Laboratory Service guidelines and an EC regulation. The *E. coli* levels reported for 99% of samples in this study would be classed as 'Acceptable' or better using either the guideline or the regulation. Guidance was not provided for *B. cereus*, which is an expected contaminant in fresh herbs.

Figure 4. Frequency distribution for thermotolerant coliforms, *E. coli* and *B. cereus* in fresh herbs





3.3 Chemical properties

pH and water activity

Some of the food categories were also tested for pH and/or water activity (a_w) to determine the intrinsic properties of the samples. Chemical properties can assist with determining whether a product is potentially hazardous, ie able to support the growth of pathogenic microorganisms. The summary of the pH and water activity for each product category is presented in Table 7.

Table 7. The pH and water activity for each product category

		р	Н	Water activity ⁷					
	Mean	Median	Min	Max	Mean	Median	Min	Max	
Tofu	5.9	5.9	5.0	7.5	0.99	1.0	0.96	1.0	
Mixed salads	4.7	4.6	4.0	6.6	N/T	N/T	N/T	N/T	
Vegetable- based dips & sauces ⁸	4.4	4.4	3.5	5.8	0.97	0.98	0.86	0.99	

Food acids

In this survey, levels of four commonly used food acids in vegetable-based dips were measured (Table 8). Vegetable-based dips can be broken down into different types based on their main ingredients.

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⁷ N/T: Not Tested

 $^{^{8}}$ Only 93/100 and 94/100 samples were tested for pH and $a_{\rm w}$



Table 8. Levels of food acids in vegetable-based dips

Product No of		Citric acid (mg/kg)			Malic acid (mg/kg)			Lactic acid (mg/kg)			Acetic acid (mg/kg)						
types	samples	Mean	Median	Min	Max	Mean	Median	Min	Max	Mean	Median	Min	Max	Mean	Median	Min	Max
Hommus	32	7,928	7,855	691	14,875	1,477	1,375	351	4,128	3,238	4,484	408	5,380	1,183	995	238	3,085
Baba ghanoush	10	6,492	6,667	1,981	10,541	1,737	1,606	541	3,187	ND	ND	ND	ND	1,112	735	530	2,447
Guacamole	5	2,414	1,985	894	6,089	2,008	2,144	1,336	2,408	2,981	2,981	1,225	4,737	326	278	201	609
Pesto	15	2,442	1,328	526	8,842	600	530	161	1,061	1,596	1,484	1,035	2,836	2,222	1,798	410	5,078
Salsa	2	3,469	3,469	3,088	3,849	8,097	8,097	5,386	10,807	ND	ND	ND	ND	1,890	1,890	606	3,173
Other	26	4,466	3,012	340	10,391	1,895	1,559	580	4,701	2,253	1,607	140	5,692	3,180	2,292	138	11,234



4. DISCUSSION

The microbiological testing illustrated that the plant products tested were, in general, of a good or acceptable microbiological standard, with 96.7% of samples tested categorised as such.

4.1 Tofu

In this survey, 96% of tofu samples tested was either good or acceptable. This is comparable to results obtained in a previous survey conducted by van Kooij & de Boer (1985).

The initial cooking of soybeans and soy milk assists in eliminating the initial vegetative microflora of the ingredients of tofu. However further processing after cooking, such as pressing and packaging introduces the potential for contamination. Three samples (3%) were found to be unsatisfactory due to elevated level of CPS (3400 cfu/g); *B. cereus* (1600 cfu/g); *B. cereus* and *E. coli* (4700 and 2400 cfu/g respectively). Another sample was also unsatisfactory due to very low level of *Salmonella* (only positive by PCR and not by cultural method).

Bacillus cereus is known to be present on soybeans and its heat resistant spores can germinate in the products. Food Science Australia considered tofu as medium risk in relation to this organism because it can grow in the products during storage (FSA, 2000). *E. coli* and *S. aureus* can be present in tofu due to its high protein content and the amount of handling subsequent to the heating process. Prolonged storage at improper temperature can also lead to *S. aureus* proliferation. In this survey, all samples were stored under refrigeration at the retail shops. However, their storage condition prior to purchase is unknown.

4.2 Mixed salads

Of 101 mixed salads samples tested, three (3%) samples were found to be unsatisfactory due to elevated level of *B. cereus* (1200 cfu/g, 2100 cfu/g, and 3800 cfu/g). The products were red quinoa with tomato salad (pH=4.4); tomato, corn and tuna pasta salad (pH=5.2); and sundried tomato pesto pasta salad (pH=5.0) respectively. The presence of *B. cereus* in these types of products is not unexpected due to the presence of raw vegetables. *B. cereus* is present in soil and may contaminate vegetables used in the salad.

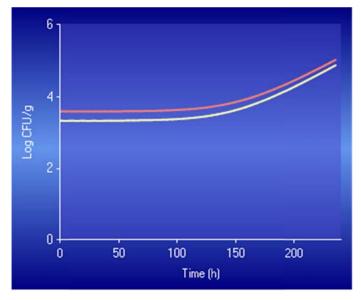
The pH limit for growth of *B. cereus* is listed as 5.0 under ideal conditions (ICMSF, 1996). Therefore it is expected that no growth would occur in the red quinoa with tomato salad that had a pH of 4.4. The refrigerated storage of these products also means that the other products with pH values of 5.0 and 5.2 may also be unlikely to support the growth of *B. cereus*, as reductions in storage temperature below the optimum will increase the minimum pH value for growth.

Using the ComBase growth model for *B. cereus*, it was predicted that growth may occur in the latter two products at pH 5.0 and 5.2, however the lag phase is likely to last between 4-5 days before the organism may start to grow very slowly with a doubling time of around 16-17 hours.

Figure 5 shows the predicted growth over the course of a 10 days shelf life with approximately a 1.5 log increase in both products. Given how close this is to the growth no-growth boundary, in reality it is likely that growth will not occur.



Figure 5. Predicted growth of B. cereus in the salads with pH value of 5.0 and 5.29



Note: the red line represents *B. cereus* predicted growth in salads with pH value of 5.0 and the yellow line represents that with pH value of 5.2

Five studies conducted overseas on the microbiological quality of mixed salads (pre-packaged or unpackaged) are summarised in Appendix 2. In four of the studies, *L. monocytogenes* was detected with prevalence rates from 2% to 21%. While in this survey, no sample was found to contain *L. monocytogenes* above the limit of the enumeration test (greater than 10 cfu/g). It is important to note that the presence/absence test is a much more sensitive than the enumeration test. The presence/absence test uses 25g of sample, while the enumeration test effectively uses 0.1g of sample. A sample with low levels of contamination can be positive in a presence/absence test, but produce a 'not detected' result in an enumeration test.

4.3 Vegetable-based dips & sauces

Three (3%) vegetable-based dips and sauces tested in this survey were found to be unsatisfactory due to elevated level of *B. cereus* (1300 and 2400 cfu/g) and *E. coli* (190 cfu/g). No sample contained pathogenic organisms such as *Salmonella* or *L. monocytogenes*. The unsatisfactory samples were hommus topped with paprika, hommus, and beetroot dip (containing tahini and chickpeas).

Hommus has a high water activity, high nutrient content and relatively neutral pH, which makes for a favourable microbial media. The presence of olive oil and citric acid in the products may have bactericidal or baterialstatic effects (Al-Holy et al., 2006; Almualla et al., 2010), however, hommus has been linked to foodborne illness caused by *Salmonella* and *Clostridium perfringens*. Blakey & Priest (1980) found that *B. cereus* is naturally present in various pulses and could multiply during the soaking step. At room temperature this would not lead to excessive growth of *B. cereus*, but it is important to note that subsequent boiling was regarded as insufficient to destroy the activity of emetic toxin which is stable at 126°C for at least 90 minutes. Blake & Priest also found that *B. cereus* flora on red lentils and kidney beans survived cooking and grew rapidly in the stored, cooked food at both 22 and 37°C. Thus, it is important that food either kept hot or cooled quickly and refrigerated within two hours of cooking.

⁹ This graph uses the values found in the survey as the starting numbers at day 0. The remaining shelf lives of these products were 5 days.



Food acids are added to food to make flavours "sharper", and can also act as preservatives or antioxidants. Common food acids include vinegar (acetic acid), citric acid, tartaric acid, malic acid, fumaric acid, and lactic acid. Food acids may occur naturally in food, or be added.

This survey found that hommus and baba ghanoush contained high level of citric acid. This is expected since all of them listed lemon juice or citric acid (330) in their ingredients. Citric acid is commonly used in food industry because it is easily obtained and relatively cheap. It can act as a preservative by increasing the acidity of the products, making it difficult for bacteria to survive or grow. It also helps to stabilise emulsions such as vegetable-based dips.

This survey found salsa products to contain high level of malic acid. Malic acid is naturally occurring in tomatoes at the level of 20 to 30% of their total acid. Malic acid has similar applications to citric acid.

4.4 Fresh herbs

In this survey, seven samples were found to contain *B. cereus* at the level above 1000 cfu/g and one garlic chives sample was found to contain 22,000 cfu/g of *E. coli* and 1600 cfu/g of *B. cereus*. The results are comparable to results obtained in previous surveys (Appendix 2). In addition, a number of previous studies found *Salmonella* in few of the samples, while in this survey, no sample was found to contain *Salmonella*.

Fresh herbs may easily become contaminated with *E. coli* and *B. cereus*. These organisms are common environmental bacteria that are found in soil and water. The process of cutting may introduce additional contaminants, and the availability of moisture and nutrients on the cut surfaces creates conditions that generally favour microbial growth (Doyle & Erickson, 2008; Elviss et al., 2009). However, it is important to note that all products tested in this survey were sold in a bunch, wrapped loosely in plastic sleeve or unpackaged. This type of packaging or the lack of packaging does not create the environment that would favour the growth of pathogens. This is quite different to sprouts processing where seed is held under warm moist conditions and the potential for pathogen growth is very high.

Chopped fresh herbs, other than in mixed salads, were not found in the market place and were not included in the survey. There does appear to be a market in the food service industry for chopped parsley for use in kebab preparation. Further work on food service products, such as chopped parsley, is likely to result as a consequence of an investigation of a foodborne illness outbreak which occurred in early 2014.

5. CONCLUSION

The microbiological testing illustrated that the plants products tested were, in general, of a good or acceptable microbiological standard. The survey supported the finding of the CSIRO scoping study and literature review that the products are generally of lower risk when compared to those covered by the Scheme.



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APPENDICES

Appendix 1: Foodborne illness attributed to plant products considered in this survey

Appendix 2: Selected studies on microbiological quality of plant products considered in this survey



Appendix 1. Foodborne illness attributed to plant products considered in this survey

Outbreaks linked to tofu

Year	Country	No of cases	Pathogen	Vehicle	Reference
2012	USA (New York)	2	C. botulinum	Home fermented tofu	Chai, Choi, Guitierrez et al., 2013
2006	USA (California)	2	C. botulinum	Home fermented tofu	Meyers et al., 2007
2003	Australia (NSW)	20	S. Typhimurium 170	Restaurant tofu dish	OzFoodNet, 2004
1988	USA (Michigan)	3175	Shigella sonnei	Festival tofu salad	Lee et al., 1991
1981/82	USA	50	Y. enterocolitica	Tofu	Tacket et al., 1985

Outbreaks linked to pre-packaged mayonnaise-based salad

Year	Country	No of cases (hospitalised)	Pathogen	Vehicle	Reference
1987	USA	(4)	C. botulinum	Coleslaw	Sewell & Farber, 2001
1981	Canada	41 (17 deaths)	L. monocytogenes	Coleslaw	Sewell & Farber, 2001

Outbreaks linked to vegetable-based dips and sauces

Year	Country	No of cases (hospitalised)	Pathogen	Vehicle	Reference
2013	Italy	(50)	C. botulinum	Pesto	Roberts, 2013
2002/03	Australia & NZ	68	S. Montevideo	Tahini	Unicomb et al., 2005
2000	USA	30	Shigella sonnei	5-layer party dip (bean, salsa, guacamole, nacho cheese, sour cream)	CDC, 2000



Outbreaks linked to kimchi

Year	Country	No of cases	Pathogen	Vehicle	Reference
2012	Korea	1642	<i>E. coli</i> O169	Radish & cabbage kimchi	Hu, Seo & Choe, 2013
2001	Japan	Unknown	<i>E. coli</i> O157:H7	Locally made kimchi	Inatsu et al., 2004
1991	Japan	3	E. coli 0169:H41	Kimchi purchased during a trip to Korea	Nishikawa et al., 1995

Outbreaks linked to fresh herbs

Year	Country	No of cases (hospitalised)	Pathogen	Vehicle	Reference
2007	USA, North Europe, & Russia	74 (3)	S. Senftenberg	Fresh basil	Pezzoli et al., 2007
2006	Denmark	200	S. Anatum & enterotoxigenic E. coli	Basil used in pesto	Pakalniskiene et al., 2008
1999	USA	35	S. Thompson	Coriander	Campbell et al., 2001
1999	USA	62	Cyclospora	Basil	Lopez et al., 2001
1998	USA	300	Shigella sonnei	Coriander	Naimi et al., 2003
1998	USA & Canada	486 (163)	Shigella sonnei	Imported chopped uncooked parsley	Crowe et al., 1999
1998	USA	77 (12)	ETEC	Imported parsley	Naimi et al., 2003
1997	USA	48	Cyclospora	Basil used in pesto	Pritchett et al., 1997



Appendix 2. Selected studies on microbiological quality of plant products considered in this survey

Surveys on microbiological quality of tofu at retail

Year	Country	No of samples	Findings	Reference
Unknown	The Netherlands	23	 Samples: sufu (fermented tofu) Three samples had <i>B. cereus</i> at the level of greater than 10⁵ CFU/g One sample contained <i>C. perfringens</i> at ~10⁵CFU/g <i>S. aureus</i> was not detected in any of the sample, but <i>S. aureus</i> enterotoxin A was detected in some of the white and grey sufu <i>L. monocytogenes</i> was not detected in any of the samples 	Han et al., 2001b
1999	Japan	29	 Samples include: fresh and fried tofu SPC was higher in fried tofu compared to fresh tofu (3.6 log and 2.6 log respectively) one fresh tofu sample contained Listeria murrayi (now grayi) 	Kaneko et al., 1999
1999	USA	60	 Temperature of tofu at time of sale ranged from 1.7°C to 17°C SPC ranged from 3.33x10¹ to 7.73x10² CFU/g pH ranged from 4.68 to 6.36 two samples were bulging in the packet at time of purchase (stored at 16°C & 13.9°C) 	Ashraf, 1999
1985	The Netherlands	154	 No Salmonella was detected in any of the samples B. cereus was detected in one sample at the level of >10⁵ CFU/g S. aureus was detected in one sample at the level of >10⁵ CFU/g E. coli present in 36% of samples Mould present in 14% of samples Yeast present in 59% of samples at levels greater than 10³ CFU/g 95% had SPC >10⁶ CFU/g 35% of samples were sold at temperatures greater than 7°C pH of samples ranged from <4.5 to ≥6.0, with the majority of samples at pH of 5.0 to 5.4. 	van Kooij & de Boer, 1985



Surveys on microbiological quality of mayonnaise-based salad at retail

Year	Country	Food tested (retail establishment)	No of samples tested	Organisms tested	Result	Reference
2005 - 2007	Belgium	Mayonnaise- based deli salads (retail)	1187	L. monocytogenes ¹⁰	80 (6.7%)	Uyttendaele et al., 2009
2005 - 2006	South Africa	Assorted salads e.g. fruit & vegetables, mixed with mayonnaise (delicatessens)	35	APC (CFU/g) S. aureus (CFU/g) B. cereus (CFU/g) L. monocytogenes ¹⁰ Salmonella ¹¹	10 ⁷ 10 ² 10 ² 1 (3%) 4 (11%)	Christison et al., 2008
2000 - 2001	USA	Seafood salads Deli salads – potato, tuna, pasta & coleslaw	2446 8549	L. monocytogenes ¹⁰	115 (4.7%) 202 (2.4%)	Gombas et al., 2003
1997 - 1998	Belgium	Mayonnaise based salad – ham, chicken, seafood, vegetables (retail)	874	L. monocytogenes ¹⁰	186 (2 1 .3%)	Uyttendaele et al., 1999b
1996	Japan	Cream based salad – potato, macaroni, coleslaw (packaged)	71	APC (>10 ⁵ cfu/g) Listeria spp ¹¹	13 (18.3%) 3 (4.2%)	Kaneko et al., 1999

¹⁰ detected in 25g

¹¹ detected in 10g



Surveys conducted on the microbiological quality of vegetable-based dips

Year	Country	Food tested	No of samples	Findings	Reference
2004	Australia (National)	Sesame seeds, tahini, halva, hommus, baba ghanoush	40	1 sample was positive for <i>Salmonella</i> Richmond	FSANZ, 2005
1997	Australia (ACT)	Dairy and non- dairy dips (packaged & loose)	76 (35 non- dairy)	For non-dairy samples: - 1 sample had pH of 4.6 (unacceptable) - 8 samples contained SPC at the level of >1,000,000 CFU/g (3 packaged & 5 unpackaged) - No samples contained coagulase positive Staphylococcus 3 samples contained <i>E. coli</i> at low level (unpackaged) but there was no indication of the types of dip.	Christen et al., 1997

Previous studies on kimchi

There was a study conducted in Taiwan on microbiological quality of kimchi and their histamine content. Thirty seven retail kimchi were tested for aerobic plate count (APC), total coliform and *E. coli*. The study found that all samples had pH between 3.6 and 5.1, with salt content ranging from 1.5 to 16%. Eighteen samples were unacceptable due to their APC content, fourteen samples were unacceptable in term of total coliform, and one sample contained *E. coli* at the level of 20 MPN/g.

Twenty three samples contained detectable level of histamine (LOD is 5mg/100g), with one sample contained histamine at the level of 535 mg/100g. Histamine is formed mainly through the decarboxylation of histadine released by many bacterial species known to possess histadine decarboxylase. Several species of histamine-producing LAB belonging to the *Lactobacillus*, *Leuconostoc* and *Pediococcus* have been isolated from fermented food (Tsai et al., 2005).



Surveys conducted on the microbiological quality of fresh herbs

Year	Country	Food tested	No of samples	Findings Reference
2008	Lebanon	Parsley	25	 Samples were collected from area using irrigation water from local river and ground water wells Four samples (13.8%) were positive for <i>E. coli</i> and ten samples (38%) were positive for <i>S. aureus</i>
2008	England	Fresh herbs	298	 Samples were tested for Salmonella only Five samples (1.7%) were positive. They were coriander, curry leaves and holy basil
2007	UK	Various herbs e.g. basil, parsley, coriander, mint	3760	 Samples are RTE and include loose or in a bunch, pre-packed or grown in a pot 18 samples (0.5%) were positive for <i>Salmonella</i> 137 samples (3.6%) contained <i>E. coli</i> at the level greater than 100cfu/g
2005- 2007	Australia	Parsley Basil	15 2	 Samples were collected from field, farm gate, and retail Samples were tested for <i>E. coli, E. coli</i> O157:H7 or VTEC, <i>Salmonella</i>, and <i>Listeria</i> spp One parsley sample (6.7%) contained <i>E. coli</i> at the level of 3.6 MPN/g and one sample (6.7%) was positive for VTEC One basil sample (50%) contained <i>E. coli</i> at the level of 23 MPN/g
2005 & 2007	Norway	Basil, mint, coriander	Unknown	 Samples were pre-cut and imported from SE Asia 28% and 15% of samples were contaminated with Salmonella Cited by Elviss et al., 2009
2002- 2003	USA	Coriander Parsley Dill	141 72 9	 Samples included domestically produced and imported Samples were collected from different step of processing Samples were tested for APC, E. coli, E. coli O157:H7, Salmonella, Shigella, and L. monocytogenes No pathogenic organisms were detected on the samples
2000-	USA	Coriander	94	- Samples were tested for Johnston et



Year	Country	Food tested	No of samples	Findings	Reference
2002		Dill Parsley	12 78	Salmonella, L. monocytogenes, & E. coli O157:H7 No pathogenic organisms were detected in any of the samples	al., 2005
2000	USA	Coriander Parsley (domestically produced)	85 90	 Samples were collected from packinghouses, repackers, or wholesellers Samples were tested for <i>E. coli</i> O157:H7, <i>Salmonella</i> & <i>Shigella</i> spp (parsley only) No sample was positive for <i>E. coli</i> O157:H7 One coriander sample (1.2%) was positive for <i>Salmonella</i> One parsley sample (1.1%) was positive for <i>Shigella</i> 	FDA, 2003
1999- 2001	Norway	Various herbs Parsley & dill	130 100	 Three samples of various herbs (2.3%) and five samples of parsley/dill (5%) contained thermotolerant coliform No sample contained <i>E. coli</i> O157:H7, <i>Salmonella</i>, <i>L. monocytogenes</i>, <i>Staphylococcus</i> spp or <i>Y. enterocolitica</i> 	Johannessen et al., 2002
1999	USA	Coriander Culantro Parsley (imported)	177 12 84	 Samples were tested for <i>E. coli</i> O157:H7, <i>Salmonella</i> & <i>Shigella</i> spp (parsley only) 16 coriander samples (9%) were positive for <i>Salmonella</i> Six culantro samples (50%) were positive for <i>Salmonella</i> One parsley sample (1.2%) was positive for <i>Salmonella</i> and one (1.2%) was positive for <i>Shigella</i> 	FDA, 2001
1983	Spain	Parsley	23	 Samples were collected during four seasons and from different sources (field & retail) 13 samples (56.5%) contained <i>E. coli</i> at a level greater than 1000cfu/100g One sample (4.3%) contained <i>S.</i> Typhimurium 	Ruiz et al., 1987

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