

ACT HEALTH PROTECTION SERVICE

MICROBIOLOGICAL

QUALITY OF

SAVOURY BAKERY PRODUCTS



January 2012 - March 2012

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OBJECTIVE

- To determine the microbiological status of savoury bakery products in the ACT
- Determine the compliance of these products to the Food Standards Australia New Zealand (FSANZ) Guidelines for the Microbiological Examination of Ready to Eat (RTE) Foods

BACKGROUND

While previous surveys of bakeries have been performed, the focus was generally on sweet bakery products rather than savoury. The increased popularity of Ready to Eat foods such as pies, sausage rolls, pasties, pork buns, beef buns, pizza slices etc warrants a survey concentrating on the savoury goods produced. Several outbreaks have been linked to consumption of savoury baked products (Queensland and NSW). There is little data about savoury bakery products in the ACT and whether such products are stored appropriately.

The savoury bakery survey was undertaken

- to assess its microbiological safety and quality
- to provide microbiological data on new bakery retailers,
- provide an integrated approach to food issues and benefit not only the HPS but also advise individual food businesses on the risk factors and best practice in preparation of savoury bakery products, based on survey results.

SURVEY

The survey ran from January 2012 until March 2012 inclusive. During this period a total of 55 samples were collected and processed by the Health Protection service laboratory. Six of these samples, however, were sweet and were not included in the survey. The savoury bakery samples were collected from 13 different establishments. The samples consisted of a wide variety of savoury baked products such as rolls, pizza, sausage rolls, savoury buns, frittatas, pies, quiche etc. The samples were tested (Table 1) for a variety of different organisms in line with the ready to eat food standards.

Where the HPS identifies non compliance issues in food businesses, corrective actions are addressed through a graduated and proportionate response. Unsatisfactory results, excluding those for SPC are re-sampled. Marginal results may be re-sampled; this is dependent on resources as these foods are still considered compliant. Unsatisfactory SPC results are not re-sampled unless pathogens are also isolated.

STANDARDS

Savoury bakery products are categorised as a Ready-to-Eat food. The FSANZ Guidelines for the Microbiological Examination (RTE) Foods as shown in Table 1 are applicable to this food.

Table 1

Test	Microbiological Quality (colony forming units per gram (cfu/g))			
	Satisfactory	Marginal	Unsatisfactory	Potentially Hazardous
Standard Plate Count (SPC)				
Level 1*	<10 ⁴	<10 ⁵	≥10 ⁵	
Level 2*	<10 ⁶	<10 ⁷	≥10 ⁷	
Level 3*	N/A	N/A	N/A	
Indicators				
<i>Escherichia coli</i>	<3	3-100	>100	**
Pathogens				
Coagulase positive staphylococci	<10 ²	10 ² -10 ³	10 ³ -10 ⁴	≥10 ⁴ SET +ve
<i>Bacillus cereus</i>	<10 ²	10 ² -10 ³	10 ³ -10 ⁴	≥10 ⁴
Salmonella spp.	not detected in 25g			detected
<i>Listeria monocytogenes</i>	not detected in 25g	detected but <10 ² #		≥10 ² ##

NOTE:

*Pathogenic strains of *E. coli* should be absent.

** Foods with a long shelf life stored under refrigeration should have no *L. monocytogenes* detected in 25g.

The detection of *L. monocytogenes* in ready-to-eat-foods prepared specifically for "at risk" population groups (the elderly, immunocompromised and infants) should also be considered as potentially hazardous.

SET +ve: Staphylococcus enterotoxin positive.

Level 1 – applies to ready-to-eat foods in which all components of the food have been cooked in the manufacturing process/preparation of the final food product and, as such, microbial counts should be low i.e. fried chicken.

Level 2 – applies to ready-to-eat foods which contain some components which have been cooked and then further handled (stored, sliced or mixed) prior to preparation of the final food or where no cooking process has been used i.e. custard slice.

Level 3 – SPC not applicable. This applies to foods such as fresh fruits and vegetables (including salad vegetables), fermented foods and foods incorporating these (such as sandwiches and filled rolls). It would be expected that these foods would have an inherent high SPC because of the normal microbial flora present

Note: An examination of the microbiological quality of a food should not be based on SPC alone. The significance of high (unsatisfactory) SPC cannot truly be made without identifying the predominant microorganisms or other microbiological testing.

Microbiological Method of Analysis

Samples were tested for the presence of:

- *Escherichia coli* using ISO16649-2
- coagulase positive *Staphylococci* using AS 1766.2.4(modified)
- *Bacillus cereus* using AS 5013.2
- *Clostridium perfringens* AS5013 - 2006
- Standard Plate Count (SPC) using AS 5013.5
- *Salmonella* species using AS: 5013.10–2004 (modified)
- *Listeria monocytogenes* using AS 1766.2.16.1 (modified).

The sample preparation for *Escherichia coli*, coagulase positive *Staphylococci*, *Bacillus cereus* and SPC consisted of 25g of sample being homogenised with 225mL of 0.1% peptone diluents with subsequent serial dilutions were prepared for use in enumeration.

- ***Escherichia coli* enumeration:** Pour plates of 1ml of 10⁻¹ dilution were prepared in triplicate on TBX medium and incubated at 37°C/4 h followed by 44°C/20 h. Confirmed *E. coli* colonies appear blue/green after incubation.
- **Coagulase positive *Staphylococci* enumeration:** Pour plates (using a 1ml of 10⁻² and 10⁻⁴ dilutions) of Baird Parker medium with RPF supplement were prepared in

duplicate and incubated at 37°C/48h. Typical black colonies, with a halo of precipitation surrounding the colony were indicative of coagulase activity.

- ***Bacillus cereus* enumeration:** Spread plates (using a 100µl of each dilution) on a solid selective medium containing egg yolk and mannitol. Typical large, pink colonies, with or without lecithinase action were counted and a proportion of the colonies confirmed by a haemolysis test and spore staining. *B. cereus* cells are rods 4-5 µm long and 1-1.5 µm wide and stain red. The cells contain black-stained lipid globules. The spores stain green, are ellipsoidal in shape, central to sub central in position, and do not swell the sporangium.
- ***Clostridium perfringens*:** Pour plates of 1ml of 10⁻² and 10⁻⁴ dilution prepared in duplicate using TSCNE agar, overlaid and incubated anaerobically at 37°C for 18-22 h. Black colonies are presumptive, with confirmation performed using API 20A test kit.
- **Standard Plate Count:** Pour plates (using a 1ml of each dilution) of plate count agar were incubated at 30 °C/72h. Plates from the dilution on which there are greater 15 and less than 300 colonies visible were counted. Counts outside this range were considered estimate counts only.
- ***Salmonella* detection:** 25g of sample was weighed out aseptically and homogenised with 225mL buffered peptone water (non-selective enrichment) and incubated at 37°C/16-20 h. Aliquots were then transferred into Brain Heart Infusion broth (BHI) and incubated for 4h. DNA was extracted from 200uL of enriched BHI. This was screened for the presence of salmonella using a BAX System Salmonella PCR Assay. No confirmation testing was performed as there were no samples that screened positive.
- ***Listeria monocytogenes* detection:** 25g of sample was weighed out aseptically and homogenised with 225mL half Fraser broth (selective enrichment) and incubated at 30°C/24h. Aliquots were then transferred into a single tube of Fraser broth incubated for 37°C/48h and MOPS BLEB broth incubated for 37°C/24h. DNA was extracted from 200uL of enriched MOPS BLEB broth. This was screened for the presence of *Listeria monocytogenes* using a BAX System Listeria monocytogenes PCR Assay. No confirmation testing was performed as there were no samples that screened positive.

RESULTS

Escherichia coli

No *Escherichia coli* were isolated from any of the 49 savoury bakery samples tested in this survey. The results indicate that 100% of the samples were satisfactory.

Coagulase positive *Staphylococci*

All 49 samples showed a satisfactory microbiological quality for coagulase positive *Staphylococci* ie. <100 cfu/g.

Bacillus cereus

All 49 samples showed a satisfactory microbiological quality for *Bacillus cereus* ie. <100 cfu/g.

Clostridium perfringens

No *Clostridium perfringens* were isolated from any of the 49 savoury bakery products tested in this survey.

Standard Plate Count

All samples (49) were tested for SPC. The results for the samples ranged between <50 and 2.1×10^7 cfu/g. A total of 36 of the food samples were assessed as applying to Level 1 SPC criterion, with counts ranging between <50 and 40,000 cfu/g. Thirty four (94.4%) samples were in the satisfactory category i.e. contained counts of less than 10,000 cfu/g while two samples (5.6%) were in the marginal category. There were no samples in the unsatisfactory category .

A total of eight samples were assessed as applying to Level 2 SPC criterion. The results ranged between 200 and 2.1×10^7 cfu/g. Seven (87.5%) samples were in the satisfactory category i.e. contained counts of less than 100,000 cfu/g while one sample (12.5%) was in the unsatisfactory category i.e. $>10^7$ cfu/g. No re-sample was taken as no pathogens were isolated.

A total of five samples were assessed as applying to Level 3 SPC criterion. The SPC test is not applicable to these products. The results for these products ranged from 50 to 8.0×10^6 cfu/g. This is to be expected as these foods, may include raw fruits and vegetables or fermented products which have an inherently high SPC because of their normal microbial flora.

Salmonella spp.

No *Salmonella* were isolated from any of the 49 savoury bakery samples in this study. The results indicate that 100% of the samples were satisfactory.

Listeria monocytogenes

No *Listeria monocytogenes* were isolated from any of the 49 savoury bakery samples in this study. The results indicate that 100% of the samples were satisfactory.

CONCLUSION

Overall, the microbiological quality of savoury bakery products surveyed was of a very good standard. In conclusion, the results of this survey show a very high level of compliance with the FSANZ RTE Guidelines

BIBLIOGRAPHY

1. Guidelines for the Microbiological Examination of Ready-to-eat-Foods (December 2001) FSANZ

	SPC Level	SPC result (cfu/g)	<i>E. coli</i> count in food (cfu/g)	Coag Pos Staph (cfu/g)	<i>B. cereus</i> result (cfu/g)	<i>Salmonella</i> in food (P/A in 25g)	<i>L. monocytogenes</i> in food (P/A in 25g)	<i>C. perfringens</i> (cfu/g)	Assessment
Savoury Roll	1	50*	<3	<50	<50	Absent	Absent	<50	S
Ham and Cheese Roll	3	50*	<3	<50	<50	Absent	Absent	<50	S
Supreme Pizza	1	150*	<3	<50	<50	Absent	Absent	<50	S
Steak and Onion Pie	1	50*	<3	<50	<50	Absent	Absent	<50	S
Sausage Roll	1	700*	<3	<50	<50	Absent	Absent	<50	S
Sausage Roll	1	500*	<3	<50	<50	Absent	Absent	<50	S
Meat Pie	1	2600	<3	<50	<50	Absent	Absent	<50	S
Spinach Pie	1	<50	<3	<50	<50	Absent	Absent	<50	S
Pastie	1	1000	<3	<50	<50	Absent	Absent	<50	S
Quiche	1	900*	<3	<50	<50	Absent	Absent	<50	S
Curry Chicken Bun	1	100*	<3	<50	<50	Absent	Absent	<50	S
Crabmeat Bun	1	<50	<3	<50	<50	Absent	Absent	<50	S
Bacon Bun	1	<50	<3	<50	<50	Absent	Absent	<50	S
Ham Bun	1	50*	<3	<50	<50	Absent	Absent	<50	S
Chicken, Mushroom Donut	1	5800	<3	<50	<50	Absent	Absent	<50	S
Potato Pie	1	<50	<3	<50	<50	Absent	Absent	<50	S
Frittata	1	100*	<3	<50	<50	Absent	Absent	<50	S
Roast Beef Roll	3	8000000*	<3	<50	<50	Absent	Absent	<50	S
Smoke Salmon Roll	3	5400000*	<3	<50	<50	Absent	Absent	<50	S
Potato Frittata	1	50*	<3	<50	<50	Absent	Absent	<50	S
Steak and Onion Pie	1	<50	<3	<50	<50	Absent	Absent	<50	S
Steak and Curry Pie	1	450*	<3	<50	<50	Absent	Absent	<50	S
Steak, Egg and Mushroom Pie	1	<50	<3	<50	<50	Absent	Absent	<50	S
Steak, Bacon and Cheese Pie	1	<50	<3	<50	<50	Absent	Absent	<50	S
Sausage Roll	1	100*	<3	<50	<50	Absent	Absent	<50	S
Sausage Croissant	2	15000	<3	<50	<50	Absent	Absent	<50	S
Tuna Bun	2	200*	<3	<50	<50	Absent	Absent	<50	S

	SPC Level	SPC result (cfu/g)	<i>E. coli</i> count in food (cfu/g)	Coag Pos Staph (cfu/g)	<i>B. cereus</i> result (cfu/g)	<i>Salmonella</i> in food (P/A in 25g)	<i>L. monocytogenes</i> in food (P/A in 25g)	<i>C. perfringens</i> (cfu/g)	Assessment
Crab meat Bun	2	400*	<3	<50	<50	Absent	Absent	<50	S
Pork Chop bun	2	8800	<3	<50	<50	Absent	Absent	<50	S
Ham and Cheese Sandwich	3	900*	<3	<50	<50	Absent	Absent	<50	S
Crabmeat Bun	2	1600	<3	<50	<50	Absent	Absent	<50	S
Tuna Bun with Egg	2	21000000	<3	<50	<50	Absent	Absent	<50	U
Ham and Cheese sandwich roll	3	29000*	<3	<50	<50	Absent	Absent	<50	S
Pizza supreme	2	500*	<3	<50	<50	Absent	Absent	<50	S
Pizza Mexicana	2	300*	<3	<50	<50	Absent	Absent	<50	S
Pepper Pie	1	40000*	<3	<50	<50	Absent	Absent	<50	S
Sausage Roll	1	250*	<3	<50	<50	Absent	Absent	<50	S
Spinach and Ricotta Roll	1	700*	<3	<50	<50	Absent	Absent	<50	S
Pepper Pie	1	13000*	<3	<50	<50	Absent	Absent	<50	S
Cheese and Bacon Pie	1	<50	<3	<50	<50	Absent	Absent	<50	S
Plain Beef Pie	1	<50	<3	<50	<50	Absent	Absent	<50	S
Angus Pie	1	500*	<3	<50	<50	Absent	Absent	<50	S
Sausage Roll	1	100*	<3	<50	<50	Absent	Absent	<50	S
Beef Meat Pie	1	1600	<3	<50	<50	Absent	Absent	<50	S
Ham and Cheese Quiche	1	300*	<3	<50	<50	Absent	Absent	<50	S
Spinach and Mushroom Quiche	1	550*	<3	<50	<50	Absent	Absent	<50	S
Focaccia	1	250*	<3	<50	<50	Absent	Absent	<50	S
Ham, Tomato Quiche	1	200*	<3	<50	<50	Absent	Absent	<50	S
Spinach Tomato Quiche	1	<50	<3	<50	<50	Absent	Absent	<50	S

* = estimate count only.