Canadian Food Inspection Agency | Agence canadienne d'inspection des aliments

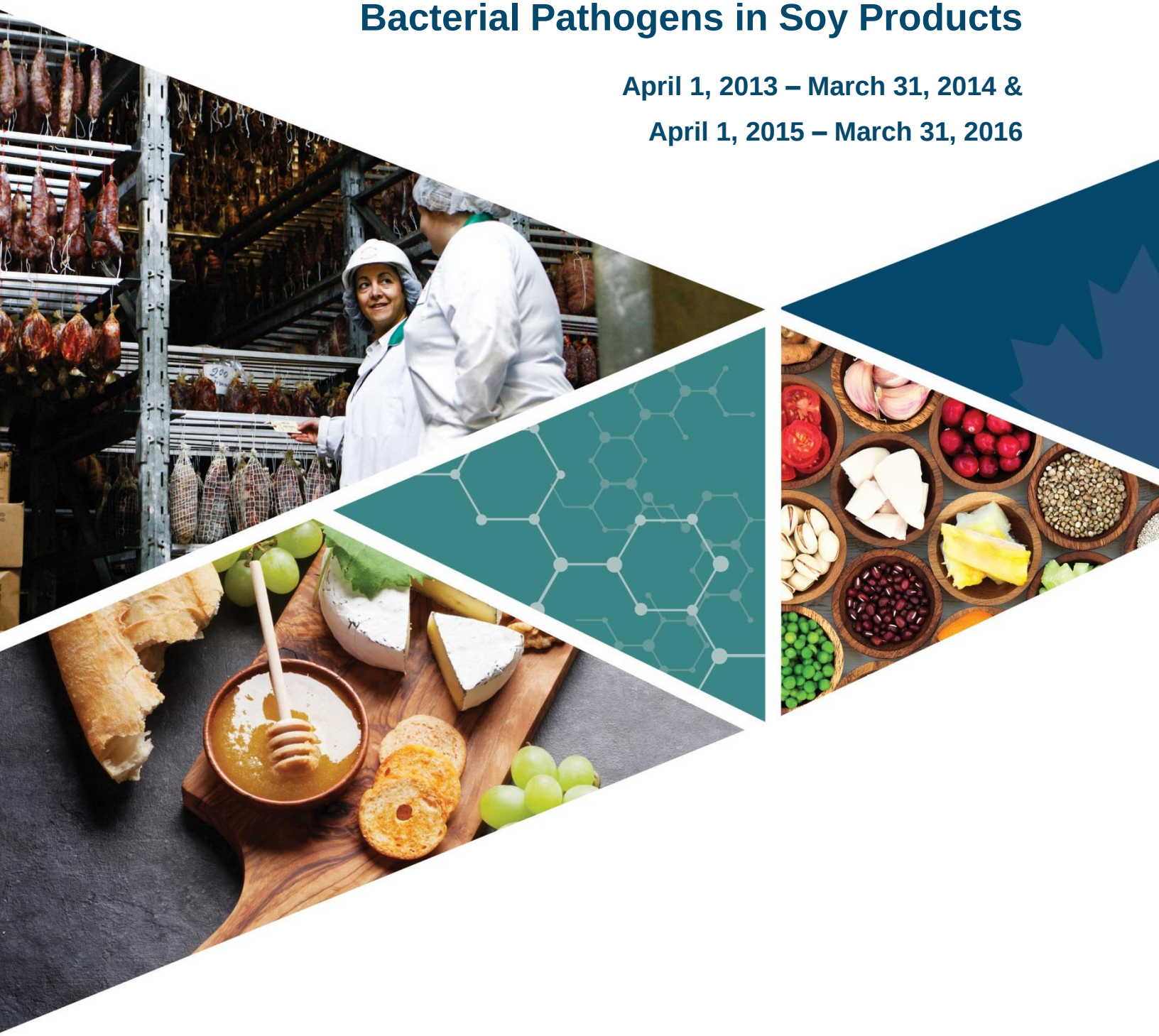
Food Microbiology – Targeted Surveys

FINAL REPORT

Bacterial Pathogens in Soy Products

April 1, 2013 – March 31, 2014 &

April 1, 2015 – March 31, 2016



Canada

Summary

The demand for soy products has been growing in Canada and around the world due to the health benefits associated with their consumption. However, there are few guidelines regulating the soy food industry. A few reported foodborne illness outbreaks associated with soy products have been reported worldwide in the past 30 years. Soy products, such as tofu and compressed tofu, are highly perishable ready-to-eat (RTE) foods. Since these products are often consumed as-is or lightly cooked, the presence of pathogens can cause foodborne illnesses.

Considering the factors mentioned above and their relevance to Canadians, soy products were selected for targeted surveys. The purpose of the survey was to generate baseline information on the occurrence of bacterial pathogens in soy products. Over the course of two fiscal years (April 1, 2013 to March 31, 2014, and April 1, 2015 to March 31, 2016), a total of 3157 samples of soy products were collected from retail locations in 11 cities across Canada and tested for bacterial pathogens of concern: *Salmonella, Listeria monocytogenes* (*L. monocytogenes*), and *Staphylococcus aureus* (*S. aureus*), as well as generic *Escherichia coli* (*E.coli*). Generic *E. coli* is an indicator organism which is used to assess the overall sanitation conditions under which the soy products were produced.

All samples (100%) of soy products including fresh tofu, compressed tofu, soy-based meat analogues, tempeh and soy paste, were found to be safe for consumption, as *Salmonella* and *L. monocytogenes* were not found in any samples, and levels of *S. aureus* were below 100 colony forming unit (CFU)/g in all samples tested for. Samples where the level of *S. aureus* is below 100 CFU/g are considered to have been produced under sanitary conditions and are safe for consumption. Elevated levels (100 to 1000 most probable number (MPN)/g) and high levels (>1000 MPN/g) of generic *E. coli* were found in three and one compressed tofu samples, respectively. As these samples were processed, pre-packaged RTE foods, the results indicate the possibility of inadequate sanitation controls of the processing facility. The Canadian Food Inspection Agency (CFIA) conducted appropriate follow-up activities including follow-up sampling and facility inspections. No product recalls were issued as follow-up sampling yielded satisfactory test results in two cases, and in the other two cases samples were no longer available on the market as the products were at the end of their shelf life. Improvements were recommended to the facilities regarding their sanitation controls.

The results indicate that almost all of the soy products appear to have been produced under Good Manufacturing Practices, as there were very few occurrences of high levels of generic *E. coli*.

What Are Targeted Surveys?

Targeted surveys are used by the Canadian Food Inspection Agency (CFIA) to focus its surveillance activities on areas of highest health risk. The information gained from these surveys provides support for the allocation and prioritization of the Agency's activities to areas of greater concern. Originally started as a project under the Food Safety Action Plan (FSAP), targeted surveys have been embedded in the CFIA's regular surveillance activities since 2013. Targeted surveys are a valuable tool for generating information on certain hazards in foods, identifying and characterizing new and emerging hazards, informing trend analysis, prompting and refining health risk assessments, highlighting potential contamination issues, as well as assessing and promoting compliance with Canadian regulations.

Food safety is a shared responsibility. The Canadian Food Inspection Agency works with federal, provincial, territorial and municipal governments and provides regulatory oversight of the food industry to promote safe handling of foods throughout the food production chain. The food industry and retail sectors in Canada are responsible for the food they produce and sell, while individual consumers are responsible for the safe handling of the food they have in their possession.

Why Did We Conduct This Survey?

The demand for soy products, such as tofu and soy-based foods has been growing in Canada and around the world due to the health benefits associated with their consumption. Tofu is a highly perishable manufactured food and various sanitation controls may be used to ensure food safety, such as pasteurization, rapid cooling of the product after pasteurization, refrigeration, aseptic packaging and storage at refrigeration temperatures [1, 2]. Current information regarding the microbial quality and safety of tofu and soy-based products on the Canadian and the U.S. market is limited. A few studies from other countries have revealed food safety concerns related to the processing conditions and sanitary practices under which soy products are produced [3].

There have been a few reported outbreaks associated with soy products worldwide in the past 30 years. The reported outbreaks were associated with *Yersinia* in 1981 [4], *Shigella* in 1988 [5], *Salmonella* in 2012 [6, 7], and *Clostridium botulinum* in 2006 [8] and 2012 [9]. There also have been recalls of soy products in the U.S. due to potential contamination by *Listeria monocytogenes* (*L. monocytogenes*) in 2007 and 2015 [10]. In addition, the majority of tofu products are highly perishable ready-to-eat (RTE) and therefore consumed as-is, or after only being lightly cooked and therefore the presence of bacterial pathogens can cause foodborne illnesses.

Based on the above information and in an effort to fill the current information gap regarding the microbial safety of soy products in Canada, tofu and other soy products were selected for targeted surveys over two fiscal years (2013-2014 and 2015-2016). Samples were tested for bacterial pathogens of concern: *Salmonella, L. monocytogenes*, and *Staphylococcus aureus* (*S. aureus*), as well as generic *E. coli*. *S. aureus* was only tested in the samples collected during fiscal year 2015-2016. This report details results of the entire survey period of April 1, 2013 to March 31, 2014 and April 1, 2015 to March 31, 2016.

What Did We Sample?

For this survey, a sample consisted of a single unit (e.g., individual consumer-size package(s)) from a single lot. These samples were randomly collected including organic and conventional products, imported and domestic products. The samples were collected from national retail chains, local/regional grocery stores, and ethnic stores located in 11 major cities across Canada. These cities encompassed four geographical areas: Atlantic (Halifax and Saint John), Quebec (Quebec City, Montreal), Ontario (Toronto, Ottawa), and the West (Vancouver, Kelowna, Calgary, Saskatoon and Winnipeg). The number of samples collected from these cities was in proportion to the relative population of the respective areas. These samples were collected in fiscal year 2013/14 (April 1, 2013 to March 31, 2014) and fiscal year 2015/16 (April 1, 2015 to March 31, 2016).

A variety of soy products were sampled during this study and can be divided into two broad categories: tofu and soy-based products. The tofu category can be further divided into two subcategories: fresh tofu and compressed tofu. The soy-based products category can be further divided into two subcategories: meat analogues and fermented soybean products.

Fresh tofu, also known as fresh bean curd, is a processed product made by coagulating soy milk, and pressing the resulting curd into soft white blocks. The use of different coagulants such as acid (e.g., glucono delta-lactone) or salt (e.g., magnesium chloride, calcium sulfate) can produce a variety of tofu (soft, regular, firm, and extra firm), which are classified based on the water content. All types of fresh tofu are highly perishable, even at refrigerated temperatures due to its high water content and high protein content. Compressed tofu (moisture ≤75%) is derived from fresh tofu by reducing moisture through frying or a compressing process. Examples of compressed tofu products are fried tofu, marinated or smoked tofu-curd, tofu-noodle, tofu-sheets, and five-spices tofu.

Soy-based products such as meat analogues contain soy protein isolates, soy protein concentrates or soy flour, but do not contain animal meat. Samples of soy-based meat analogues such as soy-based bacon, sausage, hotdog, meat strips, burger patties, nuggets,

and other flavored or seasoned soy-based mock meats were collected under this survey. Another subcategory of soy-based products is fermented soy-based products, such as tempeh and soy pastes.

What Analytical Methods Were Used and How Were Samples Assessed?

Samples were analyzed using analytical methods published in Health Canada's *Compendium of Analytical Methods for the Microbiological Analysis of Foods* [11] The assessment criteria used in this survey (Table 1) are based on the principles of the *Health Products and Food Branch Standards and Guidelines for Microbiological Safety of Foods* [12] and associated methods published in Health Canada's *Compendium of Analytical Methods* [11].

Table 1 Assessment Criteria for Bacteria in Samples of Soy Products

Bacterial Analysis	Method Identification Number*	Assessment Criteria		
		Satisfactory	Investigative	Unsatisfactory
Salmonella	MFLP-29 MFHPB-20	Absent in 25g	Not Applicable (NA)	Present in 25g
Staphylococcus aureus	MFHPB-21	≤ 100 CFU/g	$10^2 < x \leq 10^4$ CFU/g	$>10^4$ CFU/g
*L. monocytogenes***	MFLP-28 MFHPB-30	Absent in 25 g	Not applicable (Category 1) $\leq$ 100 CFU/g (Category 2)	Present in 25 g (Category 1) or >100 CFU/g (Category 2)
Generic *E. coli*	MFHPB-19 MFHPB-27	≤ 100 MPN/g	$100 < x \leq 1000$ MPN/g	> 1000 MPN/g

*The methods used were the published versions at the time of analysis.

** The assessment criteria for *Listeria monocytogenes* (*L. monocytogenes*) are based on Health Canada's Policy on *Listeria monocytogenes in Ready-to-Eat foods* [13], and depend upon the sample type analyzed (i.e., Category 1 or Category 2A or 2B products).

S. aureus is commonly found in the environment and is a bacterium that can produce protein toxins in contaminated food, which can cause foodborne illness. Elevated levels of *S. aureus* (10^2 colony forming unit (CFU)/g $< x \leq 10^4$ CFU/g) indicate that food may have been produced under unsanitary conditions. Therefore, an investigative assessment which may result in further

follow-up actions is associated with elevated levels of *S. aureus*. As the results are based on the analysis of one unit (n=1) instead of five units (n=5), further sampling is required to verify the levels of *S. aureus* of the lot. The presence of high levels of *S. aureus* (>10^4 CFU/g) is indicative of high enough levels of *S. aureus* toxins to cause foodborne illnesses. Therefore, samples with high levels of *S. aureus* (>10^4 CFU/g) are assessed as unsatisfactory indicating that follow-up activities are warranted.

Unlike harmful bacterial pathogens (e.g. *Salmonella, L. monocytogenes*), generic *E. coli* is commonly found in the intestines of humans and most strains are harmless. It is considered to be an indicator organism and levels of generic *E. coli* found in a food product are used to assess the overall sanitation conditions throughout the food chain from production to the point of sale. An investigative assessment is associated with elevated levels of generic *E. coli* ($100 < x \leq 1000$ most probable number (MPN)/g), which may result in further follow-up actions. As the results are based on the analysis of one unit (n=1), further sampling is required to verify the levels of generic *E. coli* of the lot. An unsatisfactory assessment is associated with high levels of generic *E. coli* (> 1000 MPN/g) as it may indicate a breakdown in Good Manufacturing Practices (sanitation practices), and therefore possibly warranting the initiation of follow-up activities.

What Were the Survey Results?

A total of 3157 samples of soy products were collected and analysed for *Salmonella*, *L. monocytogenes* and generic *E. coli.* Of these samples, a total of 1063 samples (collected in fiscal year 2015/16) were additionally tested for *S. aureus*. Results indicate that *Salmonella* and *L. monocytogenes* were not detected in any samples tested. *S. aureus* were below 100 CFU/g in all samples tested for. Generic *E. coli* at elevated levels (100 to 1000 MPN/g) and high levels (>1000 MPN/g) were found in three samples (0.1%) and one sample (0.03%), respectively (Table 2). The assessment results of the samples are summarized in Table 2.

Table 2 Assessment Results of Samples of Soy Products

Product Type	Number of Samples	Satisfactory Assessment	Investigative Assessment	Unsatisfactory Assessment
			Generic *E. coli* 100 - 1000 MPN/g	Generic *E. coli* > 1000 MPN/g
Tofu	1604	1600	3	1
Soy-based products	1553	1553	0	0
Total	**3157**	**3153**	**3 (0.1%)**	**1 (0.03%)**

A variety of tofu and soy-based products were collected under this survey. Almost an equal number of samples were collected of each product type with tofu making up 50.8% of the samples and soy-based products making up 49.2% of the samples (Table 3). Samples with elevated and high levels of generic *E. coli* were found to be compressed tofu (1.1%, 4/373) (Table 3).

Table 3 Sample Distribution by Product Types

Product Type	Subtype	No. Samples (%)	No. Samples with Generic *E. coli* > 100 MPN/g (%)
Tofu	Fresh tofu (soft, regular, firm and extra firm)	1231	0
	Compressed tofu	373	4 (1.1)
	Subtotal	*1604 (50.8)*	*4 (0.7)*
Soy-based products	Meat analogues	1497	0
	Fermented soy-based products	56	0
	Subtotal	*1553 (49.2)*	*0 (0)*
Total		**3157 (100)**	**4 (0.13)**

A majority (62.5%) of the samples were produced in Canada (Table 4). Imported samples and those of unknown country of origin accounted for less than 20% each of the samples, respectively. Domestic samples were comprised of a higher proportion of tofu (70.6%, 1393/1973) as compared to the soy-based products (29.4%, 580/1973). Conversely, imported

samples were comprised of a higher proportion of soy-based products (67.1%, 390/581) as compared to tofu products (32.9%, 191/581).

Table 4 Sample Distribution by Country of Origin

Country of Origin	Total No. Samples (%)	Tofu	Soy-based Products
		No. Samples	No. Samples
Canada	1973 (62.5)	1393	580
Imported	581 (18.4)	191	390
China	18	18	0
Czech Republic	47	0	47
Israel	50	0	50
Japan	17	13	4
Korea, Republic	24	24	0
Taiwan	16	15	1
United States	409	121	288
Unknown	603 (19.1)	20	583
Total	**3157 (100)**	**1604**	**1553**

What Do the Survey Results Mean?

In this survey, all samples (3157 samples) of soy products were determined to be safe for consumption, as *Salmonella* and *L. monocytogenes* were not detected in any samples, and levels of *S. aureus* were below 100 CFU/g in all samples tested for. Samples where the level of *S. aureus* is below 100 CFU/g are considered to have been produced under sanitary conditions and are safe for consumption.

Elevated levels (100 to 1000 MPN/g) and high levels (>1000 MPN/g) of generic *E. coli* were found in three and one compressed tofu samples (1.1%), respectively. Generic *E. coli* is an indicator organism used to assess the overall sanitary conditions under which the compressed tofu was produced. As these samples were processed and pre-packaged RTE foods, the results indicate the possibility of inadequate sanitation controls of the processing facility. The CFIA conducted food safety investigations including follow-up sampling and facility inspections.

No product recalls were issued as follow-up sampling yielded satisfactory test results in two cases, and in the other two cases samples were no longer available on the market as the samples were at the end of shelf life. Improvements were recommended to the facilities regarding their sanitation controls.

A few studies were conducted regarding the microbial quality and safety of soy products at the retail level and in processing facilities. In a 2010-2012 study [14] of pre-packaged tofu sampled at retail in Thailand, generic *E. coli* and *Staphylococcus species* were found (quantification results are not available) in 2% (1/59) and 24% (14/59) of samples and *S. aureus* and *Salmonella* were not detected. In another study conducted in 2012 in Shaanxi province, China[15], *S. aureus* was isolated from 32.7% (17/52) of RTE compressed tofu retail samples, however quantification of *S. aureus* was not performed. A 2015 study conducted in Brazil noted poor sanitation conditions in local tofu processing facilities [16]. An early (1999) study conducted in the state of Illinois (U.S.) also revealed poor sanitary conditions of packaged tofu in the U.S. market [17].

Our survey results provide an indication of the current state of the microbial quality and safety of soy products on the Canadian market. Overall, our survey results suggest that almost all soy products on the Canadian market are safe for consumption.

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