



Food Microbiology – Targeted Surveys

FINAL REPORT

Bacterial Pathogens in Sprouts

April 2012 – October 2014



Summary

Sprouts, such as mung bean sprouts and alfalfa sprouts, have been responsible for numerous outbreaks of foodborne illnesses around the world over the years. Contamination in sprouts often originates from their seeds, which are agricultural products. The sprouting process requires warm and humid conditions that are ideal for the proliferation of bacterial pathogens. Since sprouts are often consumed raw or lightly cooked, the presence of pathogens in sprouts can cause foodborne illnesses.

Considering the factors mentioned above and their relevance to Canadians, sprouts were selected for targeted surveys. Over the course of this study (April 1, 2012 to October 31, 2014), a total of 3221 samples of bean sprouts and seed sprouts were collected from retail locations in 11 cities across Canada and tested for bacterial pathogens of concern: *Salmonella*, *Escherichia coli* (*E. coli*) O157:H7, as well as generic *E. coli*. Generic *E. coli* is an indicator of the overall sanitation conditions throughout the food production chain. A subset (925) of these 3221 samples was also tested for non-O157 Verotoxigenic *E. coli* (VTEC).

Salmonella was found in three (0.09%) samples. *E. coli* O157:H7 was not found in any samples, however six (0.65%) samples were found to contain non-O157 VTEC of which none were priority non-O157 VTEC (O26, O103, O111, O117, O121, and O145). Priority non-O157 VTEC are those serotypes that have been identified as causing the majority of non-O157 VTEC infections in Canada. Generic *E. coli* at levels of >1000 Most Probable Number (MPN)/g were found in 17 (0.53%) samples. The Canadian Food Inspection Agency (CFIA) conducted appropriate follow-up activities for the *Salmonella* contaminated samples resulting in product recalls and/or the implementation of corrective actions by the processing facilities. No reported illnesses were in association with any of the *Salmonella* positive samples.

The results indicate that *Salmonella* contamination can sporadically occur in sprouts. In addition, the majority of the products sampled appear to have been processed under good agricultural and manufacturing practices, as there were very few occurrences of high levels of generic *E. coli* (>1000 MPN/g).

Introduction

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.

The CFIA 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. However, it is important to note that the food industry and retail sectors in Canada are ultimately 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.

Rationale

Sprouts are a low-calorie, healthy ingredient that are commonly consumed by Canadians. Unfortunately, they can sometimes become contaminated with potentially harmful bacteria. Sprouts have been responsible for numerous foodborne illness outbreaks around the world. In 2007 a joint Food and Agriculture Organization of the United Nations and World Health Organization (FAO/WHO) Expert Committee classified sprouts as the second highest priority group of concern among fresh fruits and vegetables in terms of microbial hazards¹. *Salmonella* and *E. coli* O157:H7 have been identified as the primary bacterial pathogens of concern in sprouts.

Given the above information, sprouts were selected for targeted surveys over a 3.5 year period starting in 2011 to gather baseline information on the occurrence of pathogens in fresh sprouts available at retail in Canada. This report details results of the survey period from April 1, 2012 to October 31, 2014. Results of the first year of the survey (April 1, 2011-March 31, 2012) are detailed in a [separate report](#) and all samples were assessed as satisfactory.

Sample Collection and Size

For this survey, a sample consisted of a single sample unit (e.g., individual consumer-size package(s) from a single lot) with a total weight of at least 150 g. All samples were collected from national retail chains and local/regional grocery 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. Samples were collected between April 1, 2012 and October 31, 2014. Sprout samples were randomly collected including products of domestic and imported origin, organic and conventional, as well as a variety types of bean and seed sprouts.

Analytical Methods and Assessment Criteria

Samples were analyzed using analytical methods published in Health Canada’s *Compendium of Analytical Methods for the Microbiological Analysis of Foods*². All methods are used for regulatory testing by the CFIA and are fully validated for the analysis of fresh sprouts. 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*³ and associated methods published in Health Canada’s *Compendium of Analytical Methods for the Microbiological Analysis of Foods*².

Table 1 Assessment Criteria for Bacteria in Sprouts

| Bacterial Analysis | Method Identification Number* | Assessment Criteria | | |
|------------------------|-------------------------------|---------------------|--|--|
| | | Satisfactory | Investigative | Unsatisfactory |
| <i>Salmonella</i> | MFLP-29, MFHPB-20 | Absent in 25 g | Non applicable (NA) | Present in 25 g |
| <i>E. coli</i> O157:H7 | MFLP-30, and MFLP-80 | Absent in 25 g | NA | Present in 25 g |
| VTEC | MFLP-52 | Absent in 25 g | Present non-priority non-O157 VTEC in 25 g | Present priority non-O157 VTEC in 25 g |
| Generic <i>E. coli</i> | MFHPB-19 | ≤ 100 MPN/g | 100 < x ≤ 1000 MPN/g | > 1000 MPN/g |

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

No assessment guidelines had been established in Canada for the presence of *E. coli* O157:H7 and non-O157 VTEC in sprouts (i.e., sprouted seeds)³ at the time of writing this report. In the absence of assessment guidelines, the presence of *E. coli* O157:H7 or priority non-O157 VTEC (O26, O103, O111, O117, O121, and O145)⁴ in sprouts is considered to be a violation of the *Food and Drugs Act* (FDA) Section 4(1)a⁵ and is therefore assessed by the CFIA as unsatisfactory. The detection of non-priority non-O157 VTEC is assessed as investigative.

An investigative assessment is associated with elevated levels of generic *E. coli* (100-1000 MPN/g) as the results are based on the analysis of one unit (n=1), and further sampling is required to verify the levels of generic *E. coli* of the lot.

Results

A total of 3,221 sprout samples were analysed for *Salmonella*, *E. coli* O157:H7, and generic *E. coli*. Some of these samples (Group II samples, Table 2) were also tested for non-O157 VTEC. *Salmonella* was found in three (0.09%) samples. *E. coli* O157:H7 was not found in any samples. Non-O157 VTEC was found in six (0.64%) samples and none of the samples were found to contain priority non-O157 VTEC (O26, O103, O111, O117, O121, and O145)⁴. Priority non-O157 VTEC are those serotypes that have been identified as causing the majority of non-O157 VTEC infections in Canada. Generic *E. coli* at levels of >1000 MPN/g and 100 to 1000 MPN/g were found in 17 (0.53%) and 37 (1.15%) samples, respectively. The assessment results of the samples are summarized in Table 2.

Table 2 Assessment Results of Sprout Samples

| Analysis group | Number of Samples | Satisfactory Assessment | Investigative Assessment | | Unsatisfactory Assessment | |
|------------------|-------------------|-------------------------|-----------------------------|---|--------------------------------------|-------------------|
| | | | Non-priority non- O157 VTEC | Generic <i>E. coli</i> (100-1000 MPN/g) | Generic <i>E. coli</i> (>1000 MPN/g) | <i>Salmonella</i> |
| Group I | 2296 | 2253 | NA | 29 | 12 | 2 |
| Group II | 925 | 905 | 6 | 8 | 5 | 1 |
| Total (%) | 3221 (100%) | 3158 (98.0%) | 6 (0.65%) | 37 (1.15%) | 17 (0.53%) | 3 (0.09%) |

A variety of bean sprouts (40.0%) and seed sprouts (60.0%) were analyzed for *Salmonella*, *E. coli* O157:H7 and generic *E. coli*, of which mung bean sprouts (30.6%) and alfalfa based seed sprouts (35.7%) were the predominant types (Table 3). All *Salmonella* positive results were associated with seed sprouts containing alfalfa (alfalfa or alfalfa mixed with radish). High levels of generic *E. coli* (>1000 MPN/g) were mainly found in mung bean sprouts of which nine mung bean samples contained generic *E. coli* over >1,600 MPN/g. Over 95% of the samples analyzed were domestically produced and all aforementioned unsatisfactory samples were of domestic origin.

Non-O157 VTEC was additionally analyzed in a subset of samples consisting of bean sprouts (44.1%) and seed sprouts (55.9%) of which mung bean sprouts (29.9%) and alfalfa based seed

sprouts (36.3%) were the predominant types. Non-priority non-O157 VTEC was found in six samples of which four samples were alfalfa sprouts and the other two samples were bean sprouts and broccoli sprouts. The product types of the samples are summarized in Table 3.

Table 3 Samples Distribution by Product Type

| Product type | Samples Analysed for <i>Salmonella</i>, <i>E. coli</i> O157:H7 and generic <i>E. coli</i> (%) | <i>E. coli</i> O157:H7 | <i>Salmonella</i> | <i>E. coli</i> (>1000 MPN/g) | <i>E. coli</i> (100-1000 MPN/g) | Samples Analysed for non-O157 VTEC (%) | Non-O157 VTEC |
|----------------------------------|--|-------------------------------|--------------------------|--|--|---|----------------------|
| Bean sprouts | 1289 (40.0%) | 0 | 0 | 14 | 26 | 408 (44.1%) | 1 |
| -Mung bean sprouts | 986 (30.6%) | 0 | 0 | 14 | 25 | 277 (29.9%) | 0 |
| -Bean sprouts (not specified) | 111 | 0 | 0 | 0 | 0 | 69 | 1 |
| -Soy bean sprouts | 138 | 0 | 0 | 0 | 1 | 42 | 0 |
| -Fenugreek sprouts | 21 | 0 | 0 | 0 | 0 | 5 | 0 |
| -Mixed bean sprouts | 33 | 0 | 0 | 0 | 0 | 15 | 0 |
| Seed sprouts | 1932 (60%) | 0 | 3 | 3 | 11 | 517 (55.9%) | 5 |
| Alfalfa based | 1150 (35.7%) | | | | | 336 (36.3%) | |
| -Alfalfa sprouts | 849 | 0 | 1 | 0 | 5 | 266 | 4 |
| -Alfalfa with radish | 178 | 0 | 2 | 0 | 2 | 39 | 0 |
| -Alfalfa with other seed sprouts | 123 | 0 | 0 | 0 | 3 | 31 | 0 |
| -Radish sprouts | 71 | 0 | 0 | 1 | 0 | 18 | 0 |
| -Broccoli sprouts | 243 | 0 | 0 | 2 | 0 | 66 | 1 |
| -Snow pea sprouts | 97 | 0 | 0 | 0 | 0 | 19 | 0 |
| -Pea sprouts | 66 | 0 | 0 | 0 | 0 | 9 | 0 |
| -Mixed seed sprouts | 224 | 0 | 0 | 0 | 1 | 56 | 0 |
| -Sunflower seed sprouts | 67 | 0 | 0 | 0 | 0 | 11 | 0 |
| -Grain seed sprouts | 14 | 0 | 0 | 0 | 0 | 2 | 0 |
| Total (%) | 3221 (100%) | 0 | 3 | 17 | 37 | 925 (100%) | 6 |

The serotypes of the non-O157 VTEC samples are listed in Table 4. None of the serotypes were identified as priority non-O157 VTEC (O26, O103, O111, O117, O121, and O145).

Table 4 Serotypes of Non-O157 VTEC Sample

| Product type | Sample type | Non-O157 VTEC Serotype |
|---------------------|--|-------------------------------|
| Seed sprouts | Alfalfa sprouts | O100, O6, O118 |
| | Alfalfa sprouts | O128 |
| | Alfalfa sprouts | O128 |
| | Alfalfa sprouts | Strain was not typeable |
| | Broccoli sprouts | O174 |
| Bean sprouts | Bean sprouts (product type was not identified) | O177:NM,O154:H31 |

Discussion and Conclusion

In this survey, over 98 % of the sprout samples analyzed were determined to be free of specific pathogenic microorganisms. *E. coli* O157:H7 and priority non-O157 VTEC (O26, O103, O111, O117, O121, and O145) were not found in any of the sprout samples. Six (0.65%) samples were found to contain non-priority non-O157 VTEC.

However, *Salmonella*, a common bacterial pathogen associated with foodborne illnesses, was found in three (0.09%) samples. The CFIA conducted appropriate follow-up activities resulting in product recalls and/or implementation of corrective actions by the processing facilities. No reported illnesses were found to be in association with the *Salmonella* contaminated products. All *Salmonella* contaminations were detected in alfalfa based seed sprouts; therefore, the incidence of *Salmonella* in alfalfa based seed sprouts was 0.26% (3 in 1150 alfalfa based seed sprouts). A similar incidence of *Salmonella* in alfalfa sprouts was found to be 0.07% - 0.44% in the United States Department of Agriculture surveys conducted between 2006 and 2009⁶.

E. coli are commonly found in the intestines of humans and most strains are harmless. Generic *E. coli* is an indicator used to assess the overall sanitation conditions under which the sprouts were produced. High levels of generic *E. coli* (>1000 MPN/g) were mainly found in mung bean sprouts (14 out of 17 were mung bean sprouts) of which nine mung bean sprouts contained generic *E. coli* over 1600 MPN/g. In addition, elevated levels of generic *E. coli* were found in a total of 37 samples (1.15%). Therefore, 1.67% (54 samples) of the samples were found to have high and elevated levels of generic *E. coli*. Up to 2.7% of sprout samples were reported as having generic *E. coli* at >100 MPN/g in a survey conducted in Western Australia in 2002⁷.

These generic *E. coli* results indicate that sprouting process and conditions could sustain microbial growth.

No trend was observed with respect to seasonality in the occurrences of *Salmonella* or high and elevated levels of generic *E. coli* in sprouts in this CFIA survey.

Overall, our survey results suggest that a majority of sprouts are safe for consumption. Sporadically, sprouts can become contaminated by *Salmonella*. Sprouts are considered a high risk food, as such, safe handling of the sprouts are strongly recommended to producers, retailers and consumers^{8, 9, 10}.

References

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