

Quantification of Temperature Fluctuations in Restaurant Coolers and Modelled *Listeria monocytogenes* Growth

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ABSTRACT

Background: Coolers in food service establishments should ideally operate at 4°C or less. However in restaurant environments cooler doors are continually being opened and closed as food workers gather and store items. These actions may lead to temperature fluctuations in coolers which may pose a health risk towards the storage of potentially hazardous foods. This study measured and analyzed temperature fluctuations in coolers and quantified the risk they presented by modelling *Listeria monocytogenes* growth in response to these temperatures.

Method: *ACR Systems Inc. Smart Buttons* were placed near the opening of restaurant coolers and recorded temperatures over a 1-week span. *Food Spoilage and Safety Predictor (FSSP)* was used to model *L. monocytogenes* growth in response to the collected cooler temperatures.

Results: Coolers spend significantly less than 50% of the time above 4°C. The magnitude of temperature fluctuations during open business hours was found to be insignificant in comparison to fluctuations during closed business hours. However, fluctuations were significantly greater in reach-in coolers than in walk-in coolers. With respect to modeled *L. monocytogenes* growth, it was inconclusive on whether growth would be more or less than Health Canada's 100cfu/g policy in smoked salmon. However growth was significantly less than this limit in ready-to-eat ham.

Conclusions: More restaurant coolers need to be analyzed to confirm whether the defrost cycles of coolers have a greater impact on temperature fluctuations above 4°C than the daily activities of staff members. In addition, more coolers need to be analyzed to determine whether *L. monocytogenes* growth in smoked salmon stored in coolers for a week grow significantly more than 100cfu/g. However, it can be concluded *L. monocytogenes* growth will be significantly less than 100cfu/g in ready-to-eat ham and will pose a lower risk for listeriosis than smoked salmon.

Keywords: restaurant, coolers, temperature, fluctuation, *Listeria monocytogenes*, growth, smoked salmon, ready-to-eat ham.

INTRODUCTION

This research project seeks to determine whether minor temperature fluctuations in commercial refrigeration units pose a significant health risk by promoting *Listeria monocytogenes* to grow to unsafe levels for human consumption. This question was originally proposed by Lorraine McIntyre of the British Columbia Centre for Disease Control as *L. monocytogenes* is an ongoing food safety concern due to the numerous outbreaks it has caused. The most prominent of these outbreaks are Maple Leaf Foods and their luncheon meats (1) and cantaloupes from Jensen Farms in the United States (2), which resulted in 23 and 33 deaths, respectively. Also, the most recent outbreak currently involves a grocery store in Richmond, B.C. which has resulted in at least six hospitalizations and one death (3).

Clearly, *L. monocytogenes* has a large impact on public health. Therefore, this review aims to explore the scientific literature to gain an understanding of the basic biological characteristics of this pathogen, where it is found in the environment, and how it can contaminate food. In addition, surveys of refrigeration temperatures will be analyzed along with studies that evaluated the growth of *L. monocytogenes* in response to a variety of temperatures on different food products. Together, these findings will depict the current knowledge status of *Listeria* on food safety and identify any gaps of understanding in the pathogen's nature.

EVIDENCE REVIEW

Characteristics of Listeria monocytogenes and why it is a health concern:

Listeria monocytogenes is a Gram positive, non-spore forming bacterial pathogen that has various implications that make it a serious food safety concern. Firstly, even though it is an infrequent cause of foodborne illness, it has a 20-40% mortality rate among vulnerable populations. These individuals are primarily the elderly, pregnant women and their fetuses, and other persons with weakened immune systems (4). *L. monocytogenes* causes the disease listeriosis where the initial symptoms include

fever, muscle aches, and diarrhea (5). However, the pathogen also has the ability to cross the epithelial layer of the intestine, the blood-brain barrier, and the placental barrier which can cause life-threatening meningitis, spontaneous abortions, and stillbirths (6). As such, outbreaks of listeriosis have can have serious consequences on human life.

The second implication to consider is that *L. monocytogenes* is ubiquitous in the natural environment where it can be commonly found in soil, vegetation, water, sewage, and animal feces (7). In addition, this pathogen can also be prevalent in food processing plants and can persist on equipment for up to 12 years (4). Furthermore, the food processing workers themselves may also be a source of *L. monocytogenes* contamination where recent study has shown that 37% of workers in a cured meat plant were positive for the pathogen on their hands (8). Together, these reservoirs of *L. monocytogenes* allows for easy contamination of food products both during and after food processing.

Consequently, these food products are sold to stores and studies have attempted to measure the prevalence of contaminated foods at the retail level. However, due to the vast amount of foods that are sold every day and the relatively small sample sizes researchers were able to take, there cannot be a definitive statement on the overall presence of *L. monocytogenes* in retail foods. With regards to luncheon meats for example, Reda et al. (9) found that 5% of the meats that were surveyed contained *L. monocytogenes*. In contrast, Kovacevic et al. (4) found no presence of the pathogen in the deli-meats that they surveyed. With regards to seafood however, both Kovacevic et al. (4) and Gonzalez et al. (10), found that 5% of their smoked salmon samples contained *L. monocytogenes*.

The factor that makes the presence of *L. monocytogenes* in these foods such a concern is that these foods are ready-to-eat. This means that the foods are able to be consumed right after purchase without a cooking step in order to kill any pathogens that may be present. This is an important implication in that *L. monocytogenes*

has the ability to grow at temperatures as low as 1°C (11) so even small amounts of *L. monocytogenes* on foods can potentially grow to hazardous levels in refrigeration units. In addition, this pathogen can grow at a wide pH range (4.7-9.2) and at high salt concentrations (7).

However, the exact infectious dose of *L. monocytogenes* is uncertain as it can range from 0.1 to 100 million colony forming units (11) as it may depend on the strain or *L. monocytogenes* and the immune status of the individual (12). Nevertheless, the number of *L. monocytogenes* that are associated with outbreaks typically range from 10⁶-10⁸ colony forming units (13). Currently, Health Canada has implemented a policy in 2011 which states that ready-to-eat foods that are known to occasionally have low levels of *L. monocytogenes*, such as smoked salmon, (Category 2A foods) should not allow the pathogen to grow to more than 100cfu/g (15) during its shelf life. The European Commission has taken this a step further by making this 100cfu/g limit a regulation (14). So far, the only critical safety control for ready-to-eat foods that can suppress the growth of *L. monocytogenes* is maintaining strict refrigeration temperatures in order to slow the growth of *L. monocytogenes* so its numbers do not reach to high levels. This temperature is 4°C or below (16).

Refrigeration temperatures:

Even though many health inspectors and restaurant operators measure and record the temperature of refrigeration units, this data is limited in the scientific literature. However, studies have been completed on domestic refrigerators which revealed wide temperature ranges and averages that are above 4°C. For example, Rossvoll et al. (13) found that the refrigerators they measured were between 2.9°C and 12.0°C with an average of 6.2°C. Similarly, other studies also report means of 6°C (cited by 17, p2-4). With regards to the proportion of domestic refrigerators that are above 4°C, numerous studies indicate that approximately 70% are operating above this level (cited by 17, p3) and a study in Greece found that 55% of refrigerators were above 9°C (Sergelidis, et al, 1997). With regards to warm sections of refrigerators, there is a general trend for the top

shelf to have higher temperatures but this is not true in all cases as it can sometimes be warmer on middle or bottom shelves (cited by 17, p4-5). Altogether, these non-ideal refrigeration temperatures pose a microbiological health risk in that there is a potential for *L. monocytogenes* to grow faster to hazardous levels in these warmer temperatures.

Listeria monocytogenes-growth in response to different temperatures:

Numerous studies have measured the growth of *L. monocytogenes* in response to different temperatures on various food products. Their results generally agreed with each other but also contained some differences. As expected, exposure to higher temperatures led to more rapid bacterial multiplication. With initial inoculations of 100 CFU/g, one study demonstrated that it only took 3.5 days at 8°C for *L. monocytogenes* populations to reach outbreak levels of 10⁶ CFU/g on cold cuts, compared to 7 to 8 days at 4°C (13). This same growth trend was also observed in another study with ready-to-eat ham where 5 days of storage at 9°C led to 10⁵ CFU/g. However, at 5°C the populations of *L. monocytogenes* were not close to those associated with outbreaks and only reached to 10³ CFU/g, but this is still above Health Canada's limit of 100cfu/g (18). Similarly, this slower growth trend was also seen in fresh-cut cantaloupes where it took a week for *L. monocytogenes* to multiply to 10⁶ CFU/g at 8°C as opposed to the 3.5 days seen in cold cuts. Also, bacteria levels did not reach the 10⁶ CFU/g-outbreak thresholds when the cantaloupes were held at 4°C, but were above 100cfu/g (19). This limited growth was also seen in other tropical fruits where *L. monocytogenes* could not grow at 5°C in fresh-cut pitaya, mango, and papaya (20). With regards to smoked salmon however, levels of *L. monocytogenes* surpassed outbreak-associated levels at 5°C as the pathogen was found to grow to 10⁸ CFU/g in the absence of competitive spoilage bacteria. However, this was over a course of 193 days as opposed to a week (21). In all, these studies show that *L. monocytogenes* have the ability to grow at temperatures at or around 4°C and surpass Health Canada's

category 2A limit depending on the food product the pathogen is growing on.

Altogether, this research is valuable in that they help predict the growth of *L. monocytogenes* at fixed temperatures over specified times. However there is a gap in these studies in that, while they measured the constant average temperature over a period of time and its effect on the growth of *L. monocytogenes* in a controlled environment, they did not analyze the potential of minor temperature fluctuations which may have an impact on *L. monocytogenes*' growth. In a restaurant environment there may be temperature spikes due to the opening and closing of doors in addition to routine defrost cycles that refrigerators undergo. Furthermore, the initial placement of large batches of hot foods, such as soups, into coolers may also contribute temporary increases of temperature. Therefore there is a possibility for refrigeration temperatures to be much higher than 4°C for short periods of time, thus potentially allowing the growth of *L. monocytogenes* and other pathogens to multiply faster to unsafe levels.

Conclusion:

Listeria monocytogenes is a unique pathogen as it can easily contaminate foods, can survive a wide pH range, is able to live in high salt concentrations, and has the ability to grow at refrigeration temperatures. In addition, studies have shown that many refrigerators do not operate at optimal conditions which can allow for faster multiplication of *L. monocytogenes* to potentially hazardous levels in ready-to-eat foods. Other studies have provided evidence that support where *L. monocytogenes* was shown to be able to grow to outbreak-associated levels if food products are left in the fridge for a little over a week.

However, the studies relating to refrigeration temperatures were only conducted in a home environment as opposed to a busier restaurant environment and did not analyze minor temperature fluctuations that these coolers may have undergone. For example, there were no studies that specifically investigated the total

time coolers spend above 4°C or analyzed the magnitude of these fluctuations.

Therefore, this study measured and analyzed temperature variations in refrigeration units in restaurant establishments. This was accomplished two ways: i) determining whether the proportion of time above 4°C is greater or less than 0.5 and ii) comparing the area under the temperature-time curve above 4°C and seeing whether there are any differences between when a food establishment is open or closed for business. In addition, the gathered temperature measurements were entered into a modelling program to predict the amount of *L. monocytogenes* growth there would be if the pathogen was exposed to these conditions.

METHODS

Time Span of the Study:

Three different food service establishments participated in the study and the temperatures of 10 different coolers (eight reach-in coolers & two walk-in coolers) were logged over a 7-day span. This time span of seven days was chosen in order to obtain a representation of the “busy” and “slow” periods that food establishments experience in a typical week. This data collection occurred between January 12, 2017 and January 20, 2017.

SmartButtons in reach-in coolers: 4D 21, 4F 21, 6D 21, 6O 21, 71 21, 88 21, 91 21, C0 21.
SmartButtons in walk-in coolers: 70 21, EE 21.

Inclusion and Exclusion Criteria:

24-hour food establishments were excluded because a goal of the study investigates whether the intensity of temperature fluctuations above 4°C is greater when a food establishment is open compared to when they are closed. In addition, open display coolers were also excluded as this study explores whether the opening or closing of cooler doors has an impact on temperatures.

Setup and Placement of SmartButtons:

Since each SmartButton only has enough memory for 2,048 consecutive temperature measurements (22), measuring at time intervals that are less than five minutes would have resulted in the SmartButtons' memory being

filled before the 7-day period was complete. Therefore, each SmartButton was programmed to record temperatures every five minutes.

In addition each SmartButton was placed near the opening of each cooler. The rationale for this is that areas near the opening would experience most of the temperature fluctuations due to restaurant workers opening cooler doors to grab food products (23). To ensure that the SmartButtons did not become lost, each device was placed in a small mesh pouch and tied to a shelf post or the shelf's wiring. A mesh material was chosen to ensure that the SmartButtons would receive adequate airflow for proper temperature recording.

Calibration of SmartButtons:

Each SmartButton was set to record temperatures at 1-minute intervals. At the beginning of each time interval, a calibrated *Traceable® Ultra Waterproof Food Thermometer* was used to record the current air temperature. The collected SmartButton temperatures were then compared to the temperatures recorded from the probe thermometer and any discrepancies were noted and corrected for after the data was imported into Microsoft Excel for analysis.

Determining the Proportion of Time Spent Above 4°C & the Area Under the Curve Above 4°C:

The drafting program, AutoCAD was used to determine these values (24).

Modelling Listeria monocytogenes growth in FSSP:

Within FSSP, the model “*Listeria monocytogenes* in chilled seafood and meat products” was chosen and the subcategory, “Growth of *L. monocytogenes*: Effect of temp., atmosphere, salt, smoke, pH, nitrite and organic acids” was chosen. To simulate food products contaminated with *L. monocytogenes*, the program parameters were adjusted to mimic smoked salmon and ready-to-eat ham. These parameters were set as follows:

	Smoked Salmon	RTE Ham
<i>L. monocytogenes</i> initial cell level (cfu/g)	10 cfu/g	10 cfu/g
NaCl in water phase %	3.5 (0.98 A _w)	7.0 (0.96 A _w)
pH	6.2	6.7
Smoke components – phenol (ppm)	0	0
% CO₂ in headspace gas at equilibrium	0	0
Nitrite, mg/kg	0	0

For both food products, the initial cell contamination was chosen to be 10cfu/g. This was based on studies by Beaufort et al.(25) and Gonzalez et al.(10) which found that retail ready-to-eat products contaminated with *L. monocytogenes* typically had less than 10cfu/g. With regards to smoked salmon, the values for percentage of NaCl and pH are characteristic of smoked salmon products (26). With respect to the ready-to-eat ham, the NaCl percentage in water/water activity and pH were based on values found by Garrido et al. (18). Finally, for the final three parameters, a zero value was assigned for simplicity as discussed with Ms. McIntyre, Mr. Shyng, and Mr. Barrios (27).

With regards to the input of data into FSSP, the program only accepts time intervals in hours so the raw times collected by the SmartButtons were converted to hours from minutes in Excel.

ETHICAL CONSIDERATIONS

All food establishments that participated in the study were and will be kept anonymous.

RESULTS

Microsoft Excel 2007 was used to gather descriptive statistics and NCSS 11 was used to

conduct one-sample t-tests and two-sample t-tests. The statistical analysis was done on the three parts of the project:

- i) Analysis of the total time the temperature was above 4°C.
- ii) Analysis of the total area/hr. above 4°C between open and closed business hours of all coolers.
- iii) Analysis of *L. monocytogenes* growth in simulated smoked salmon and ready-to-eat ham.

1) Analysis of the Proportion of Time Spent Above 4°C:

It was investigated whether coolers spend more than half the time above 4°C. This was accomplished by comparing the proportion of time each cooler was above 4°C during 7 days to 0.5. Two analyses were conducted:

- i) Proportion of time above 4°C during open business hours.
- ii) Proportion of time above 4°C during closed business hours.

Inferential Statistics:

A one-sample t test was conducted. The standard that the numerical data was compared to was 0.5.

Interpretation:

i) Open Hours

H₀:	The proportion of time spent above 4°C is greater than or equal to 0.5 during open hours.
H_a:	The proportion of time spent above 4°C is less than 0.5 during open hours.
Test Used:	Wilcoxon Signed-Rank Test
Result:	P = 0.0372; Power = 0.804
Conclusion:	Reject H ₀ and conclude that coolers spend less than half the time above 4°C. Power is greater than 0.80 indicating it is a powerful study. Since P > 0.01, there is a potential α error.

ii) Closed Hours

H₀:	The proportion of time spent above 4°C is greater than or
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	equal to 0.5 during closed hours.
H_a:	The proportion of time spent above 4°C is less than 0.5 during open hours.
Test Used:	Wilcoxon Signed-Rank Test
Result:	P = 0.0293; Power = 0.840
Conclusion:	Reject H ₀ and conclude that coolers spend less than half the time above 4°C. Power is greater than 0.80 indicating it is a powerful study. Since P > 0.01, there is a potential α error.

2) Analysis of Area/hr. above 4°C between Open Business Hours vs. Closed Business Hours:

In addition to determining the amount of time spent above 4°C, another goal of the study was to quantify the magnitude the temperatures rose above 4°C during a one-week span. To do this, the area under the curve above 4°C was calculated. Here, comparisons of areas of when food establishments were open or closed for business were performed. Since different food establishments have different operating hours, the areas under the curve were converted to unit-area/hour-open (°C*minute/hour) and unit-area/hour-closed (°C*minute/hour) so comparisons between different restaurants could be conducted. Three different analyses were conducted:

- i) Comparison of areas/hr. between open and closed business hours for all ten coolers.
- ii) Comparison of areas/hr. between open and closed business hours for reach-in coolers.
- iii) Comparison of areas/hr. between open and closed business hours for walk-in coolers.

Inferential Statistics:

Two-sample t-tests were conducted on the continuous numerical data for each of the three analyses.

Interpretation:

H₀:	The area/hr. above 4°C during open business hours is less than or equal to the area/hr. when the business is closed.
H_a:	The area/hr. above 4°C during open business hours is greater than the area/hr. when the business is closed.
Test Used:	Mann-Whitney U or Wilcoxon Rank-Sum Test
Result:	All Coolers: P = 0.0993; Power = 0.0714 Only Reach – In Coolers: P = 0.147; Power = 0.0710 Only Walk – In Coolers: P = 0.0512; Power = 0.190
Conclusion:	All Coolers: Cannot reject H ₀ and cannot conclude that the area/hr. above 4°C was greater when restaurants were open for businesses than closed. P-value is close to 0.05 and power is less than 0.80 so there is a potential β-error. More coolers need to be sampled. Only Reach – In Coolers: Cannot reject H ₀ and cannot conclude that the area/hr. above 4°C was greater during open business hours than closed hours for reach-in coolers. Power is less than 0.80 so more reach-in coolers need to be sampled to increase the power of the study. Only Walk – In Coolers: Cannot reject H ₀ and cannot conclude that the area/hr. above 4°C was greater when restaurants were open for businesses than closed. P-value is close to 0.05 and power is less than 0.80 so there is a potential β-error. More coolers need to be sampled.

3) *Analysis of Area/hr. above 4°C between Reach-in Coolers vs Walk-in Coolers During Open & Closed Business Hours:*

The above statistical interpretations indicated that that area/hr. above 4°C was insignificant between a restaurant’s open and closed business hours. However, considering that the p-value for the walk-in coolers was very close to the 0.05 threshold of significance, there is possibility of a beta-error. Therefore, it was investigated whether reach-in coolers experienced more fluctuations for both open and closed business hours than walk-in coolers.

Inferential Statistics:

A two-sample t-test was conducted on the continuous numerical data.

Interpretation:

H₀:	The area/hr. above 4°C in reach-in coolers is less than or equal to the area/hr. above 4°C in walk-in coolers.
H_a:	The area/hr. above 4°C in reach-in coolers is greater than the area/hr. above 4°C in walk-in coolers.
Test Used:	Mann-Whitney U or Wilcoxon Rank-Sum Test
Result:	During Open Business Hours: P = 0.0184; Power = 0.483 During Closed Business Hours: P = 0.0181; Power = 0.345
Conclusion:	During Open Business Hours: Reject H ₀ and conclude that the area/hr. above 4°C was greater in reach-in coolers than in walk-in coolers during open business hours. Power is less than 0.80 so more coolers need to be sampled. Since P > 0.01, there is potential for α- error. During Closed Business Hours: Reject H ₀ and conclude that the area/hr.

	<p>above 4°C was greater in reach-in coolers than in walk-in coolers during closed business hours.</p> <p>Power is less than 0.80 so more coolers need to be sampled. Since $P > 0.01$, there is potential for α- error.</p>
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4) Analysis of *L. monocytogenes* growth in smoked salmon and ready-to-eat ham:

In this study, having *L. monocytogene* growth exceeding 100cfu/g was labelled as a health risk. This is based on the policy implemented by Health Canada in 2011 which stated that ready-to-eat foods that are known to occasionally have low levels of *L. monocytogenes* (Category 2A foods) should not have *L. monocytogenes* growth greater than 100 cfu/g during the prescribed shelf life (28). To model *L. monocytogenes* growth over 7 days, *Food Spoilage and Safety Predictor (FSSP)*, was used to determine the total growth of the pathogen if it was subjected to the temperatures gathered from the SmartButtons.

Inferential Statistics:

A one-sample t-test was conducted for both *L. monocytogenes* growth on smoked salmon and ready-to-eat ham with 100cfu/g being the comparison standard.

Interpretation:

H₀:	The growth of <i>L. monocytogenes</i> is equal to or exceeds 100cfu/g.
H_a:	The growth of <i>L. monocytogenes</i> is less than 100cfu/g.
Test Used:	Wilcoxon Rank-Sum Test; One Sample T Test (for Walk-in Coolers Only)
Result:	<p>Smoked Salmon in All Coolers: $P = 0.207$; Power = 0.00820</p> <p>Smoked Salmon in Only Reach-In Coolers: $P = 0.444$; Power = 0.00678</p>

	<p>Smoked Salmon in Only Walk-In Coolers: $P = 0.0197$; Power = 0.992</p> <p>Ready-to-Eat Ham in All Coolers: $P = 0.0415$; Power = 0.723</p> <p>Ready-to-Eat Ham in Only Reach-In Coolers: $P = 0.0805$; Power = 0.444</p> <p>Ready-to-Eat Ham in Only Walk-In Coolers: $P = 0.00582$; Power = 1.00</p>
Conclusion:	<p>Smoked Salmon in All Coolers: H_0 cannot be rejected and it cannot be concluded that <i>L. monocytogenes</i> growth would not be significantly less than 100 cfu/g. Power is much less than 0.80 so more coolers need to be modelled for <i>L. monocytogenes</i> growth to increase the power of the study.</p> <p>Smoked Salmon in Only Reach-In Coolers: H_0 cannot be rejected and <i>L. monocytogenes</i> growth above the 100 cfu/g limit cannot be ruled out in reach-in coolers if smoked salmon was stored for one week. Power is less than 0.80 so more coolers need to be sampled to increase the power of the study.</p> <p>Smoked Salmon in Only Walk-In Coolers: Reject H_0 and conclude that <i>L. monocytogenes</i> growth would be significantly less than 100 cfu/g if smoked salmon were stored in walk-in coolers for a week. Power is greater than 0.80 meaning that the data is powerful. Since, $P > 0.01$, there's potential for α error.</p> <p>Ready-to-Eat Ham in All Coolers: Reject H_0 and conclude that growth of <i>L. monocytogenes</i> will be significantly less than 100 cfu/g if stored for one week. Power is less than 0.80 so more coolers need to be tested to model</p>

	<p><i>L. monocytogenes</i> growth. $P > 0.01$ so there's potential for α error.</p> <p>Ready-to-Eat Ham in Only Reach-In Coolers: Cannot reject H_0 and cannot conclude that the growth of <i>L. monocytogenes</i> will be significantly less in ready-to-eat ham. P-value is close to 0.05 and power is less than 0.80 so there is a potential β-error. More coolers need to be sampled.</p> <p>Ready-to-Eat Ham in Only Walk-In Coolers: Reject H_0 and conclude that <i>L. monocytogenes</i> growth would be significantly less than 100 cfu/g if ready-to-eat ham were stored in walk-in coolers for a week.</p> <p>Power is greater than 0.80 meaning that the data is powerful.</p>
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With respect the areas underneath the 4°C curve, it was found that the magnitude of temperature fluctuations during open business hours were insignificant from closed business hours ($p = 0.0993$). These insignificancies were also found when reach-in coolers and walk-in coolers were analyzed separately from each other ($p = 0.147$ and $p = 0.0512$, respectively). These results indicate that the defrost cycles of coolers contribute more to temperature fluctuations above 4°C than restaurant activities such as the opening and closing of cooler doors during business hours. If the latter were true, then the areas underneath the curves above 4°C would have been greater during open hours, but this is not the case. However, considering that the powers of these statistical tests were much less than 0.80, it is very possible that the resulting insignificancies were due to the high variability of the small sample size. As mentioned above, some coolers, specifically walk-in coolers, underwent almost no temperature fluctuations above 4°C while some reach-in coolers experienced more spikes in temperature. Therefore, a larger sample size would need to be collected in order to differentiate fluctuations due to defrost cycles and a restaurant's daily activities.

Nevertheless, when comparing the areas of reach-in coolers versus walk-in coolers, it was concluded that reach-in coolers significantly undergo more temperature fluctuations than walk-in coolers for both open and closed business hours ($p = 0.0184$ and $p = 0.0181$). This could be due to a number of reasons. Firstly, considering that there is a high potential for an α -error, it is possible that the greater amount of fluctuations in reach-in coolers during open hours could have been due to the fact that these coolers tend to be opened and closed much more often than walk-in coolers as restaurant staff take and put away ingredients during their daily routines. Walk-in coolers on the other hand are primarily used for long-term storage, and are usually only opened when receiving shipments of supplies or taking out bulk items. This was reflected in cooler EE 21 where single, prominent spikes in temperature were seen on Jan. 13th, 16th, 17th, and 18th. This is confirmed by this particular restaurant operator when they

DISCUSSION

This report sought to investigate three aspects of temperature fluctuations in restaurant coolers:

- i) Whether coolers spend more than 50% of the time above 4°C.
- ii) The total magnitude of temperature fluctuations above 4°C by determining the area underneath the time-temperature curve.
- iii) Predicting the amount of *L. monocytogenes* growth in response to the collected temperatures.

With regards to the proportion of time spent above 4°C, it was found that restaurant coolers spent significantly less than half the time above this threshold. This was true for both open and closed business hours where p-values were 0.0372 and 0.0293, respectively. Considering that these values are greater than 0.01, there is the possibility of an α -error. This possibility can be attributed to the variability of the collected data where some of the coolers, such as 70 21, operated almost exclusively below 4°C while other coolers fluctuated above 4°C more often. In fact, coolers 6D 21 and 88 21 were operating at temperatures above 4°C for most of the week.

said that they receive shipments almost daily. With regards to the second walk-in cooler (70 21), a spike in temperature was seen on Jan. 17th when they received their shipments. Another contributing factor was that the walk-in coolers were normally operating at much lower temperatures (~0-2.5°C) compared to the reach-in coolers which were operating closer to 4°C. This could be due to the walk-in coolers' much larger volume which allows these units to maintain colder temperatures for longer periods of time. By operating at a slightly colder temperature, this would create a temperature buffer which would lessen temperature fluctuations as cooler doors open and close during business hours and during daily defrost cycles. It is also important to note that neither walk-in cooler had plastic strip curtains at their entrances meaning that walk-in coolers that do contain them may experience even less temperature fluctuations during open restaurant hours.

In addition, it was noticed that the reach-in coolers had more of their volumes filled more with food products while the walk-in coolers had more air space. This would probably impact a reach-in cooler's ability to keep colder temperatures, as revealed in cooler 60 21 for example. This reach-in cooler held multiple large bins of warm pinto and black beans, which were made daily, in a relatively small space. This alludes to another point on whether cold or hot foods stored in a cooler have an effect. For example, cooler 91 21, which belongs to the same restaurant as cooler 60 21, was only filled with cold products such as pre-made BBQ sauces, fresh vegetables, and cheese and experienced less temperature fluctuations and maintained an overall colder temperature than cooler 60 21.

However, how often a cooler is open and closed and the types of foods that are stored in reach-in coolers are not the sole contributors to the overall temperature of reach-in coolers. It also depends how well the coolers are operating or the correct temperature restaurant operators set their coolers at. For example, coolers 6D 21 and 88 21 were reach-in coolers which mainly held colder products and still experienced a vast

majority of their temperatures above 4°C, even at night when the restaurant was closed for business.

With regards to the modelled *L. monocytogenes* growth in response to the recorded cooler temperatures, it was inconclusive on whether this growth would be greater than or less than Health Canada's policy of 100cfu/g in smoked salmon, indicating that more coolers need to be analyzed. However, *L. monocytogenes* growth is significantly less than 100 cfu/g in ready-to-eat ham due to the ham's lower water activity. Also, the reach-in coolers had more *L. monocytogenes* growth compared to the walk-in coolers due to their greater fluctuations in temperature.

When compared to a previous study, Garrido et al. (18) found that an initial inoculum of ~1-cfu/g on ready-to-eat ham reached to 1000cfu/g at 5°C at the end of 5 days. This is much higher than the maximum 229cfu/g in simulated ready-to-eat ham found in this study. This could be attributed to the fact that Garrido et al. (18) ensured a relatively constant 5°C temperature in their experimental refrigerator while most of the coolers in this study were found to be operating below 4°C for less than fifty percent of the time (i.e. an average of 80% of time below 4°C) which would slow *L. monocytogenes* growth.

It is important to note that the parameters that were used to model *L. monocytogenes* growth in this study were set at ideal growth conditions to represent a worse-case scenario. For instance, competitive bacteria were not included in the simulation and nor were smoke components (i.e. phenol) and nitrites. In addition, the lag time for *L. monocytogenes* was also excluded in the simulation. Altogether, these aspects would have further inhibited *L. monocytogenes* growth in addition to colder temperatures. Therefore, there would be less growth in typical smoked salmon and ready-to-eat ham than seen in this study after one week, presenting an even lower risk for listeriosis.

LIMITATIONS

A limitation of the study was the small number of coolers that were monitored. There was a lot

of variability in the data, especially in the reach-in coolers. Differences in temperature fluctuations can be attributed to how often the coolers were used, what sort of items they stored, how full they were, and how well the coolers operated which were not controlled for. Therefore, in order to obtain a better representation of temperature fluctuations in reach-in coolers, more coolers need to be surveyed. In addition, only two walk-in coolers were analyzed. However, considering that walk-in coolers typically experience less opening and closing of doors and that many walk-in coolers contain plastic strip curtains, the collected temperature fluctuation data on walk-in coolers can be seen as representative for walk-in coolers as a whole.

In addition to the limited number of coolers that were analyzed, another limitation of the project was that the SmartButtons were set to take temperatures every 5 minutes. Therefore, there may have been temperature fluctuations above or below 4°C within the 5 minute intervals that would not have been detected by the SmartButtons. In addition, the *L. monocytogenes* modelling program, FSSP, assumed that the inputted temperatures were constant for those 5 minutes which may have led to an over- or underestimate of *L. monocytogenes* growth.

Another constraint the study presents is that changes in air temperature do not necessarily reflect temperature changes in stored food products. It may take prolonged periods of time for air temperatures to change the surface temperature of food products, meaning that brief temperature fluctuations may not have an effect. Therefore, in order to measure any discrepancies between air and food temperatures, a simultaneous comparison should be performed between SmartButtons that are exposed to the air and those that are inserted into food items.

KNOWLEDGE TRANSLATION

To prevent exposing foods to temperature fluctuations throughout the day, food products should be placed away from doorways and closer to the back of coolers. Also, since temperature fluctuations are more likely to occur

in reach-in coolers, foods that will be stored for long periods of time should be placed in walk-in coolers. Furthermore, installation of plastic strip curtains will help keep temperatures constant in walk-in coolers. Finally, restaurant operators should set their coolers to temperatures to below 4°C in order to create a temperature buffer to lessen fluctuations as cooler doors are being opened and closed and increases in temperature during defrost cycles.

FUTURE RESEARCH

- Determine the extent of air temperature fluctuations' effects on the temperature of food products.
- Measure more walk-in coolers and determine whether they actually do experience less temperature fluctuations.
- Conduct a controlled experiment to determine how long cooler doors need to be opened for in order to change the air temperature of coolers.

CONCLUSION

This study concluded that there were no significant differences in temperature fluctuations between open and closed business hours. This may indicate that the defrost cycles of coolers have a greater impact on temperature fluctuations above 4°C than the daily activities of a restaurant. However, only a small sample size of coolers was analyzed and the statistical power of these tests were much less than 0.80 so future research with more coolers should be conducted to fully confirm whether the daily operations of a restaurant do not have a significant impact on temperature fluctuations. When comparing reach-in coolers with walk-in coolers however, it was found that reach-in coolers experience greater temperature spikes above 4°C. This is most likely due to their smaller volumes of space which makes reach-in coolers less resistant to temperature spikes. In order to minimize temperature fluctuations, operators should place food away from cooler

entrances, install plastic strip curtains, and adjust cooler temperatures to below 4°C to minimize the effects of defrost cycles. Finally, the storage of smoked salmon presents a greater health risk when stored for 7 days while ready-to-eat ham presents a very low health risk for listeriosis.

ACKNOWLEDGEMENTS

This research was completed with the support of Helen Heacock, Vanessa Karakilic, and Fred Shaw of BCIT and Lorraine McIntyre, Sion Shyng, and Pablo Romero Barrios of the BCCDC. Special thanks to Daniel Wong for his instruction on how to use AutoCAD.

COMPETING INTEREST

The authors declare that they have no competing interests.

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