

**Risk Assessment Studies**

**Report No. 52**

Microbiological Hazard Evaluation

**MICROBIOLOGICAL QUALITY OF  
PREPACKAGED LONG SHELF LIFE  
REFRIGERATED PRODUCTS**

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Centre for Food Safety

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PREPACKAGED LONG SHELF LIFE  
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## EXECUTIVE SUMMARY

Listeriosis is a primarily foodborne disease caused by *Listeria monocytogenes*. Asymptomatic infection of listeriosis probably occurs in most healthy people, but it can pose serious health risks for the susceptible population including pregnant women, the elderly and immunocompromised individuals. There is an increasing trend of reported listeriosis in Hong Kong in recent years.

This study aims to assess the microbiological quality, particularly the level of *L. monocytogenes*, of prepackaged long shelf life refrigerated products at the end of shelf life.

From mid-September 2013 to February 2014, the Centre for Food Safety (CFS) obtained a total of 100 ready-to-eat samples, including (i) 28 cheeses, (ii) 28 smoked seafood, (iii) 28 processed meats and (iv) 16 salads from different retail stores. Samples were stored in a monitored laboratory refrigerator ( $4\pm 3^{\circ}\text{C}$ ) until they were ready for testing, i.e. within the week which the sample to be expired. Laboratory analysis for *L. monocytogenes* count, aerobic colony count (ACC) and *Escherichia coli* count were conducted by the Public Health Laboratory Services Branch of the Centre for Health Protection, Department of Health.

Results showed that the *L. monocytogenes* counts in all 100 samples at the end of shelf life were less than 20 cfu/g i.e. none of the samples contained excessive *L. monocytogenes*. The microbiological quality

(both ACC and *E. coli* count are quality but not safety indicators) of 89 samples (89%) were either of satisfactory or borderline at the end of shelf life. Regarding the samples with unsatisfactory microbiological quality, one smoked seafood and seven processed meat samples were found to contain excessive ACC (ranged from  $1.7 \times 10^8$  –  $3.9 \times 10^8$  cfu/g). Post-processing contamination and/or inadequate temperature control might result in the unsatisfactory microbiological quality of these samples. Three cheese samples were found to contain high *E. coli* count (one sample contained 240 MPN/g while the other two contained more than 1100 MPN/g); it might be due to the use of raw/ unpasteurised milk contaminated with *E. coli* in these products. In response to the concerned samples of unsatisfactory microbiological quality, the CFS gave health advice to relevant premises and took follow-up samples. All follow-up samples were satisfactory.

### **Conclusion and recommendations**

Majority (89%) of the prepackaged long shelf life refrigerated products available in the local retail stores were of satisfactory or borderline microbiological quality even at the end of shelf life. Post-processing contamination and/or inadequate temperature control as well as the use of contaminated raw material in products were the possible causes of the unsatisfactory results.

Results of the study highlight the importance of the trade to adopt

measures to ensure prepackaged long shelf life refrigerated products are safe throughout their shelf life and provide sufficient information on label for consumers to make informed food choices. Even excessive pathogens i.e. *L. monocytogenes* were not detected in this study, high level of *E. coli* in the concerned raw milk cheese samples concur with the recommendation that susceptible populations should avoid high risk foods.

### **Advice to public**

- Read food labels carefully to make informed food choices.
- Follow the storage instructions e.g. "keep in a refrigerator" provided by the manufacturer and avoid cross-contamination.
- Do not use food after the "use by" date as shown on the food label.

### **Advice to susceptible populations including pregnant women, the elderly and immunocompromised individuals**

- Avoid high risk foods especially refrigerated ready-to-eat foods with long shelf life.
- Choose cheeses carefully before consumption.
  - Hard and extra hard cheeses are generally safe.
  - Avoid soft cheeses such as Feta, Brie, Camembert, blue cheeses (e.g. Danish blue, Gorgonzola and Roquefort).
  - For other types of cheeses, choose only those made from pasteurised milk.
  - Do not eat if in doubt.

- Cook all food thoroughly and use ready-to-eat food as soon as possible.

### **Advice to trade**

- Maintain good food and personal hygiene at all food processing stages.
- Provide an adequately refrigerated environment (at or below 4 °C) throughout the supply chain for refrigerated products.
- Provide sufficient information on food label for the consumers to make informed food choices.
  - Properly label whether the dairy products are made from raw milk or pasteurised milk.
- Ensure all information provided on food label complies with the legal requirements.



# Microbiological Quality of Prepackaged Long Shelf Life Refrigerated Products

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## OBJECTIVES

The objective of this study is to assess the microbiological quality, particularly the level of *Listeria monocytogenes*, of prepackaged long shelf life refrigerated products at the end of shelf life.

## INTRODUCTION

2. In recent years, there has been an increasing trend of reported human listeriosis, a relatively uncommon but can be serious disease caused by a bacterium called *Listeria monocytogenes*, in Hong Kong.

3. Prepackaged long shelf life refrigerated ready-to-eat products e.g. cheeses, deli meats, smoked seafood are generally considered as of higher *L. monocytogenes* risk. Since *L. monocytogenes* is ubiquitous in the environment; even when it is initially present at low level in a contaminated food, it can multiply slowly to significant levels at refrigerated temperatures in foods that support its growth and may lead to infection when the food is consumed without further cooking.<sup>1</sup>

4. In order to evaluate the microbiological quality, particularly the level of *L. monocytogenes*, of prepackaged long shelf life refrigerated

ready-to-eat products at the end of shelf life, a study was conducted.

### *Listeria monocytogenes*

5. *L. monocytogenes* is a Gram-positive, facultatively anaerobic, non-spore forming rod-shaped bacterium. It is widely distributed in the environment and has been found in different sources e.g. soil, vegetation, faecal material, sewage and water.<sup>2</sup>

6. As compared with other non-spore forming foodborne pathogens, *L. monocytogenes* is resistant to various environmental conditions e.g. it can grow at pH levels between 4.4 and 9.4, and at water activities ( $a_w$ )  $\geq 0.92$ . *L. monocytogenes* also grows at low oxygen conditions and over a temperature range of 0°C to 45°C, with an optimum around 37°C, and survives for long periods in the environment, on foods, in the processing plant and in the refrigerator. However, this bacterium is killed by cooking and pasteurisation<sup>a 1,2</sup>

7. Even *L. monocytogenes* is also a transitory resident in the human intestinal tract, with 2-10% of the general population being carriers of the bacterium without any apparent health effects, consuming food contaminated with excessive *L. monocytogenes* may lead to the development of a disease called listeriosis.<sup>1,2</sup>

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<sup>a</sup> Pasteurisation is a heat treatment (e.g. not less than 15 seconds at a temperature of not less than 72°C) aimed at reducing the number of any pathogenic microorganisms in food including milk, if present, to a level at which they do not constitute a significant health hazard.

([http://www.codexalimentarius.org/download/standards/10087/CXP\\_057e.pdf](http://www.codexalimentarius.org/download/standards/10087/CXP_057e.pdf))

## Listeriosis

8. Listeriosis is primarily a foodborne disease. It can also be transmitted through placenta from mother to foetus or through an infected birth canal to her newborn.<sup>3</sup>

9. At present, the infective dose of human listeriosis is still undetermined; however, it is believed to be dependent on a variety of factors including the strain, susceptibility of the host and the food matrix.<sup>4</sup> Based on the currently available epidemiological data, the levels of *L. monocytogenes* in the implicated food generally exceeded 1,000 cfu/g.<sup>2</sup>

10. Incubation period of listeriosis may range from 3 to 70 days (3 weeks on average).<sup>3</sup> Most healthy people infected with listeriosis are asymptomatic or have only flu-like symptoms.<sup>2,5</sup> However, listeriosis often leads to severe consequences in susceptible populations e.g. pregnant women, unborn or newly delivered infants, the elderly and immunocompromised individuals.<sup>2</sup> For pregnant women, they may experience a fever and other non-specific symptoms such as fatigue and aches, followed by miscarriage or stillbirth, or her newborn resulting in septicaemia or meningitis. In the elderly and immunocompromised individuals, septicemia and meningitis are the most common symptoms.<sup>6</sup> Listeriosis can be life threatening with fatality rates of 20-30% in the world.<sup>2</sup>

11. In Hong Kong, the Department of Health has made listeriosis notifiable under the Prevention and Control of Disease Ordinance (Cap. 599) since July 2008.<sup>7</sup> Local listeriosis cases were largely sporadic and the infection occurred throughout the year with no particular seasonal pattern. From 2009 to 2013, a total of 85 listeriosis cases were recorded by the Centre for Health Protection of the Department of Health, with an annual number of cases ranged from 6 to 26. The number of listeriosis cases has increased in recent years that the cases in 2012 (26 cases) and 2013 (26 cases) have accounted for 61% of the total number of cases in the past five years. All cases required hospitalisation with 7 of them died from listeriosis, which gave an overall case fatality rate of 8% in Hong Kong.

#### High risk food items

12. Although *L. monocytogenes* is frequently present in raw foods of both plant and animal origin, foods most often implicated in major listeriosis outbreaks are those ready-to-eat foods that (i) support *L. monocytogenes* growth, (ii) have a long recommended refrigerated shelf life i.e. sufficient time is given to allow the pathogen to grow slowly to significant numbers at refrigerated temperatures and (iii) are consumed without further listericidal treatment e.g. cooking. This also includes products that receive a listericidal treatment but are subject to post-processing recontamination and cross-contamination in both the retail and home setting.<sup>2</sup>

13. Cheese made from either raw or pasteurised milk, fish and seafood products such as smoked mussels, gravlax, cold-smoked trout and shrimp, meat and poultry products like pate, frankfurters and deli meats as well as salad are some of the food sources previously linked to foodborne *L. monocytogenes* cases/ outbreaks occurred around the world.<sup>2,8</sup>

14. Overseas studies have also demonstrated the presence of *L. monocytogenes* in certain ready-to-eat food. For instance, in the EU, a baseline survey on was carried out in 2010 and 2011 to estimate the prevalence of *L. monocytogenes* in certain ready-to-eat foods at retail. The prevalence of *L. monocytogenes* in the EU in packaged (not frozen) hot or cold smoked or gravad fish samples at the end of shelf-life was 10.3 %, while those for heat treated meat products and soft or semi-soft cheese samples at the end of shelf-life were 2.07 % and 0.47 % respectively. In the same study, the proportion of samples with a *L. monocytogenes* count exceeding the level of 100 cfu/g at the end of shelf life in the EU was 1.7 %, 0.43 % and 0.06 % for fish, meat and cheese samples respectively.<sup>9</sup>

## **SCOPE OF STUDY**

15. Due to the diversity of prepackaged long shelf life refrigerated ready-to-eat products available in the local market, this study focused on

four food categories which may be of higher *L. monocytogenes* risk –

(i) Cheese

- *L. monocytogenes* in cheese can originate either from the raw material i.e. the milk used for making cheese or the processing plant environment. When the conditions permit, including the processing/ storage temperature as well as the acidity and water content of the cheese, the bacterium can grow to high levels after prolonged storage.
- Soft cheeses and semi-soft cheeses especially those made from milk that do not undergo any listericidal treatment i.e. raw/unpasteurised milk, are recognised as high risk food items for foodborne listeriosis.
- Hard and extra hard cheeses, cream cheeses and processed chesses were excluded from this study as they are generally of lower *L. monocytogenes* risk. The processing and packaging used as well as their inherent characteristics e.g. high salt content, low pH and low moisture content generally inactivate the growth of *L. monocytogenes*.<sup>5</sup>

(ii) Smoked seafood

- Some studies have demonstrated that *L. monocytogenes* is frequently isolated from vacuum packed cold-smoked salmon and other fish products as these products can have a long shelf life and may support the growth of *L. monocytogenes* during prolonged refrigerated storage.<sup>2</sup>

(iii) Processed meat

- Deli meats usually have an extended shelf life and may be contaminated during handling e.g. slicing subsequent to the listericidal step (if any). Some of these products support the growth of *L. monocytogenes* during storage, even under refrigeration.
  - Dry/ semi-dry fermented sausages are excluded in this study as they do not support the growth of *L. monocytogenes*.<sup>5</sup>
- (iv) Salad
- Fresh fruit and vegetables may be contaminated with pathogens including *L. monocytogenes* in the production stage and through cross-contamination during processing. Since it is not supposed to have any lethal process to eliminate *L. monocytogenes* in salad before consumption, prolonged refrigerated storage of these products may also of *L. monocytogenes* risk. However, it is noted that freshly cut vegetables do not support as rapid growth of *L. monocytogenes* as some other foods such as milk or deli meats.<sup>5</sup>

## **DEFINITIONS**

16. For the purpose of this study, the terms below are defined as follows-

“Prepackaged”: any food packaged, whether completely or partially, in such a way that (a) the contents cannot be altered without opening or changing the packaging; and (b) the food is ready for presentation to the

ultimate consumer or a catering establishment as a single food item.<sup>10</sup>

“Long shelf life”: The period during which the product maintains its microbiological safety and sensory qualities at a specific storage temperature is greater than 5 days.

“Refrigerated food”: perishable food stored in a refrigerator (excluding freezer) to maintain its safety, quality and suitability, for the intended shelf life.

“Ready-to-eat”: food intended by the producer or the manufacturer for direct human consumption without the need for cooking or other processing effective to eliminate or reduce to an acceptable level the microorganisms of concern.<sup>11</sup>

## **METHODOLOGY**

### Sampling

17. The sampling was conducted from mid-September 2013 to February 2014.

18. A total of 100 prepackaged ready-to-eat samples (each sample consisted of two sets from the same batch; each set weighed at least 200 grams) displayed in refrigerator, including (i) 28 cheeses, (ii) 28 smoked



seafood, (iii) 28 processed meats and (iv) 16 salads, were purchased as consumer from different local retail stores.

19. To prevent prolonged storage of samples at the laboratory before analysis, the period between the sampling date and the expiry date as shown on the package of the concerned sample taken for analysis was at least 5 days but less than 4 weeks.

#### Sample treatment and laboratory analysis

20. All samples were stored at 4°C or below during transport and they were delivered to the Public Health Laboratory Services Branch of the Centre for Health Protection, Department of Health, within four hours of sampling.

21. Since *L. monocytogenes* can grow slowly at low temperature and may reach a high level after prolonged storage, products in this study were analysed right before their expiry date so as to assess if there was any outgrowth of microorganisms at the end of shelf life. As such, samples purchased were stored in a monitored laboratory refrigerator at 4±3°C until they are ready for testing, i.e. within the week which the sample expired.

22. To avoid possible cross-contamination, one set of sample was used to analyse for *L. monocytogenes* count while the other was used to

analyse for aerobic colony count (ACC) and *Escherichia coli* count.

23. The *L. monocytogenes* count was performed according to the Health Protection Agency (2004) National Standard Method F19 issue 1. The enumeration of ACC in samples was performed according to the Health Protection Agency (2004) National Standard Method F11 Issue 1 (Spiral Plate Method at 30°C). *E. coli* count in samples was performed using the AOAC Official Methods 991.14 (Revised: March 1998) (Petrifilm Method). If the *E. coli* count could not be determined by the Petrifilm method due to interference of mould and/or other bacteria present, the *E. coli* count would be analysed by the most probable number (MPN) method specified in ISO/TS 16649-3:2005. For the purpose of this study, the results obtained from the MPN method were compared directly with the *E. coli* criterion expressed as colony-forming unit (cfu)/g.

### Result analysis

24. Results of the samples were analysed by the Risk Assessment Section of the CFS.

### *L. monocytogenes* criteria used in this study

25. The Microbiological Guidelines for Food stipulate *L. monocytogenes* criteria for various ready-to-eat foods (Table 2).<sup>11</sup>

**Table 2. *L. monocytogenes* criteria stipulated under the Microbiological Guidelines for Food**

	<b>Microbiological quality</b>		
	Result (colony-forming unit (cfu/g unless otherwise specified))		
	<b>Satisfactory</b>	<b>Borderline</b>	<b>Unsatisfactory: Potentially injurious to health and/or unfit for human consumption</b>
For refrigerated food* (excluding frozen food) or food intended for infants	Not detected in 25g	N/A	Detected in 25g
For other ready-to-eat food	<10	10-≤100	>100

N/A= not applicable

\* This criterion applies to all refrigerated food (excluding frozen food) unless there is scientific evidence supporting that the food concerned does not support the growth of *Listeria monocytogenes* under refrigeration. Reference can be made to the Codex Guidelines on the Application of General Principle of Food Hygiene to the Control of *Listeria monocytogenes* in Food (CAC/GL 61-2007).

26. As shown in Table 2, different *L. monocytogenes* criteria, which made reference to the international practices, are established for (i) refrigerated food (excluding frozen food) or food intended for infants and (ii) other ready-to-eat food. It is based upon the current knowledge that the risk of *L. monocytogenes* increases in ready-to-eat food i.e. refrigerated food (excluding frozen food) which supports the growth of the pathogen. Establishing the limit of “not detected in 25g” for high risk foods is to provide a specific degree of confidence that *L. monocytogenes* will not be present in food at levels that represent a risk to consumers. For other ready-to-eat food as well as any refrigerated food with scientific evidence demonstrating that it does not support the growth

of *L. monocytogenes* under refrigeration, a less stringent limit ( $\leq 100$  cfu/g) is applied.

27. Since this study aims at assessing if there is any outgrowth of *L. monocytogenes* in the samples at the end of shelf life, the pathogen count in each sample was analysed. For the purpose of this study, the microbiological quality of the sample is considered “Unsatisfactory: Potentially injurious to health and/or unfit for human consumption” if the *L. monocytogenes* count is greater than 100 cfu/g.

#### ACC

28. ACC is the total number of bacteria found in food. It includes those naturally occur and those as a result of contamination. ACC is an indicator of quality but not safety. The level of ACC in food depends on the type and duration of processing that the food has received during production as well as how the food is handled and stored thereafter.<sup>12</sup>

29. However, ACC is not applicable to food such as raw salad vegetables and fermented foods. In general, raw vegetables are expected to contain microorganisms present from the environment; ACCs are likely to be much higher in these products. For many types of cheeses, since they are produced by adding starter cultures of bacteria; apart from the starter bacteria which are predominant, other bacteria are usually present in low numbers due to the acidity produced during fermentation.<sup>12</sup>

## *E. coli*

30. *E. coli* is a bacterium commonly found in the gastrointestinal tract of humans and warm-blooded animals. It is commonly used as an indicator organism to reflect the hygienic quality of food. Its presence in food generally indicates direct or indirect faecal contamination. In general, substantial number of the bacterium in food suggests a general lack of cleanliness in handling and improper storage.<sup>11</sup>

31. Under the local Microbiological Guidelines for Food, the *E. coli* criterion for ready-to-eat food in general does not apply to cheeses made from raw milk<sup>b, 11</sup>. Generally speaking, low levels of *E. coli* may be present in raw milk as well as their products, including cheeses made from raw milk, even when they are properly produced. It is because of the close association of raw milk with the animal environment. Insanitary conditions, including poor employee hygiene practices, improperly sanitised utensils and equipment or contaminated raw materials may also result in *E. coli* in cheeses made from raw milk.<sup>13</sup>

32. Currently, Codex has not established any *E. coli* criteria for cheese made from raw milk. However, as stipulated in the Australia New Zealand Food Standards Code, the *E. coli* criterion of n=5, c=1,

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<sup>b</sup> As defined under Codex General Standard for the Use of Dairy Terms (CODEX STAN 206-1999), raw milk means milk which has not been heated beyond 40°C or undergone any treatment that has an equivalent effect.

([http://www.codexalimentarius.org/input/download/standards/332/CXS\\_206e.pdf](http://www.codexalimentarius.org/input/download/standards/332/CXS_206e.pdf))

n=10/g, M=100/g is set for all cheese, including those produced from raw milk.<sup>14,15</sup> In the UK, even currently there is no legislative or guidance criteria for *E. coli* in raw milk cheeses, a Health Protection Agency survey (2012) suggested establishing microbiological criteria including *E. coli* for raw milk cheeses placed on the market, with the proposed level of *E. coli*  $\geq 100/g$  i.e. same as any other ready-to-eat food to be introduced as a guidance level.<sup>16</sup>

*ACC and E. coli criteria used in this study*

33. The ACC and *E. coli* results were assessed against the criteria listed in Table 1. These criteria were extracted from the Microbiological Guidelines for Food, except the *E. coli* criterion for cheeses made from raw/ unpasteurised milk i.e. it is considered unsatisfactory if the *E. coli* level was greater than 100 cfu/g.

**Table 1. ACC and *E. coli* criteria used in this study**

	<b>Microbiological quality</b>		
	Result (colony-forming unit (cfu/g))		
	Satisfactory	Borderline	Unsatisfactory
<b>Aerobic colony count (ACC) [30°C/48 hours]</b>			
Food category 8. Extended shelf life food products requiring refrigeration†	<10 <sup>6</sup>	10 <sup>6</sup> -<10 <sup>8</sup>	≥10 <sup>8</sup>
Food category 12. Fresh fruit and vegetables, products containing raw vegetables‡	N/A	N/A	N/A
Food category 13. Fermented, cured and dried meats, fermented vegetables, ripened cheeses§	N/A	N/A	N/A

	<b>Microbiological quality</b>		
	Result (colony-forming unit (cfu/g))		
	Satisfactory	Borderline	Unsatisfactory
<b>Hygiene indicator organisms</b>			
<i>Escherichia coli</i> #	<20	20 - $\leq 10^2$	$>10^2$

N/A= not applicable

For the purpose of this study when assessing the ACC results -

† Smoked seafood and processed meat samples were grouped under Food category 8.

‡ Salad samples were grouped under Food category 12.

§ Cheese samples were grouped under Food category 13.

# In this study, the microbiological quality of cheese made from raw/ unpasteurised milk is considered unsatisfactory if the *E. coli* level was greater than 100 cfu/g.

## RESULTS

34. All samples contained *L. monocytogenes* count less than 20 cfu/g.

35. Among the samples (n=56) applicable for ACC assessment, 48 of them (86%) were found to contain ACC less than  $10^8$  cfu/g. One smoked fish and seven processed meat samples contained ACC ranged from  $1.7 \times 10^8 - 3.9 \times 10^8$  cfu/g (Table 3).

**Table 3. ACC of prepackaged long shelf life refrigerated products at the end of shelf life (Number of applicable samples=56)**

	Microbiological results (cfu/g)						
	Satisfactory				Borderline		Unsatisfactory
	< $10^3$	$10^3$ -< $10^4$	$10^4$ -< $10^5$	$10^5$ -< $10^6$	$10^6$ -< $10^7$	$10^7$ -< $10^8$	$\geq 10^8$
Smoked seafood	3	1	4	8	8	3	1

(n=28)							
Processed meat (n=28)	14	1	1	1	1	3	7

36. All, except three cheeses, samples contained *E. coli* count less than 20 cfu or MPN/g. Two cheese samples contained *E. coli* count greater than 1,100 MPN/g and the other one contained *E. coli* count at 240 MPN/g.

## DISCUSSION

37. This study showed that majority (89%) of the samples was of satisfactory or borderline microbiological quality even they were tested at the end of shelf life. The eleven samples of unsatisfactory microbiological quality were due to excessive ACC (one smoked fish and seven processed meat samples) or *E. coli* count (three cheese samples).

38. In response to the concerned samples of unsatisfactory microbiological quality, the CFS gave health advice to relevant premises and took follow-up samples. All follow-up samples were satisfactory.

### Listeria monocytogenes

39. None of the samples contained excessive *L. monocytogenes* i.e. the count in all samples were less than 20 cfu/g.



40. In general, it is assumed that growth of *L. monocytogenes* can occur in ready-to-eat foods if they fall outside the specified condition below-

- a pH below 4.4 regardless of  $a_w$ ,
- a  $a_w < 0.92$  regardless of pH,
- combination of factors (e.g. pH <5.0 and  $a_w < 0.94$ ),
- freezing (during that period when the product remains frozen),
- presence of inhibitors e.g. preservatives, including nitrite, potassium lactate, sodium acetate, sodium diacetate and sodium lactate.<sup>1,8,17</sup>

41. Control of *L. monocytogenes* growth in ready-to-eat food can be achieved by various approaches. For instance, reformulation of the product such that one or more of the parameters influencing the growth of *L. monocytogenes* e.g. pH,  $a_w$ , presence of inhibitory compounds is affected can result in food no longer supports *L. monocytogenes* growth.<sup>1</sup>

42. In addition, basic cleaning and disinfection programmes are critical to control *L. monocytogenes*. *L. monocytogenes* has the ability to form biofilms on a variety of surfaces; food contact surfaces should be cleaned and disinfected.<sup>1</sup>

43. Besides, temperature abuse may allow the growth of *L. monocytogenes*, which could result in a reduction of product shelf life. Strict control of temperature so that ready-to-eat foods never exceed 6°C (preferably 2°C - 4°C) is essential to assure that growth to any significant

degree does not occur before the product is consumed.<sup>1</sup>

44. The length of the shelf life is also an important factor contributing to the risk associated with foods that support *L. monocytogenes* growth. The shelf life of such food should be consistent with the need to control the growth of *L. monocytogenes*. Since *L. monocytogenes* is able to grow under refrigerated temperatures, the length of the shelf life should be based on appropriate studies that assess the growth of *L. monocytogenes* in the food.

#### ACC

45. ACC is a quality but not safety indicator; high level of ACC alone does not indicate an immediate risk to public health. In general, it is expected that samples analysed at the end of shelf life, their ACC may approach the upper “borderline” limit.<sup>12</sup> Excessive ACC found in the concerned samples indicated possible post-processing contamination e.g. during slicing and/or the length of time and temperature control in storage or facilitates was inadequate to prevent bacterial growth.

46. In order to maintain the microbiological quality of prepackaged refrigerated products throughout their stated shelf life, it is essential to keep them at designated temperature throughout the supply chain until they are consumed or prepared for consumption. It is also important to minimise any possible post-processing contamination and maintain cleanliness of utensils and personal hygiene.

*E. coli*

47. Three Camembert<sup>c</sup> cheese samples (two specifically labelled as “Camembert de Normandie”<sup>d</sup>) were found to contain excessive *E. coli* count. This may be due to the use of raw milk contaminated with *E. coli*.

48. Raw milk can harbour pathogens, such as *Salmonella*, pathogenic *E. coli* and *L. monocytogenes*, that can pose serious health risks to consumers. According to the local Milk Regulation (Cap. 132AQ), no person shall sell for human consumption any milk or any milk beverage which has not been heat-treated. However, such prohibition does not apply to cheeses made from milk which has not been heat-treated i.e. raw milk cheeses.

49. A Food Standards Australia New Zealand study had predicted by quantitative modelling that there were no steps resulted in an

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<sup>c</sup> According to Codex Standard for Camembert (CODEX STAN 276-1973, amendment 2010), Camembert is a soft surface ripened, primarily mould ripened cheese. The body has a near white through to light yellow colour and a soft-textured. A rind is to be developed that is soft and entirely covered with white mould but may have red, brownish or orange coloured spots. Rind formation and maturation from the surface to the centre is predominantly caused by *Penicillium candidum* and /or *Penicillium camembertii* and *Penicillium caseicolum*. For Camembert ready for consumption, the ripening procedure to develop flavour and body characteristics is normally from 10 days at 10-16°C depending on the extent of maturity required.

([http://www.codexalimentarius.org/download/standards/218/CXS\\_276e.pdf](http://www.codexalimentarius.org/download/standards/218/CXS_276e.pdf))

<sup>d</sup> ‘Camembert de Normandie’ is a lightly-salted soft cheese made from raw milk of Normande cows. It is characterised by a flat cylindrical shape with a so-called ‘blooming’ rind, white in colour, with a surface mould creating a white covering with possibly some red spots visible. The colour of the cheese varies from ivory to pale yellow. Ripened throughout, the cheese is smooth and soft.

(<http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:C:2013:140:0020:0026:EN:PDF>)

inactivation of microorganisms during the production of raw milk Camembert cheese, leading to a substantial increase in microorganisms during the manufacturing process.<sup>18</sup>

50. The ripening stage of Camembert cheese production affects the survival of microorganisms including *E. coli* that may be present in the cheese. Due to the growth of the surface moulds, there are changes on the physicochemical composition of Camembert cheese, resulting from growth inhibiting to growth permissive of microorganisms at different stages of ripening.<sup>18</sup>

51. During the first 14 days of ripening, the temperature is maintained between 12 - 13°C; growth of all microorganisms is possible. However, the concentration of lactic acid inhibits the growth of *E. coli* until day 11 when it reduces to a level where *E. coli* growth is possible at the surface. After day 14, the temperature is reduced to 4°C, which prevents the growth of *E. coli*, however, it is assumed that there is limited inactivation or reduction in numbers during that time.<sup>18</sup>

52. Although the *E. coli* detected in this study only acts as an indicator for the presence of other microorganisms of intestinal origin, it highlighted an increased likelihood of the presence of pathogens e.g. *L. monocytogenes* in the concerned raw milk cheeses, even it was not shown in the results of this study.

53. Due to the survival and possible increase of microorganisms

including pathogens during the manufacture of raw milk Camembert cheese, the microbiological quality of the raw milk and prevention of any contamination during manufacture is vital to ensure the safety of these products. It is also important for the susceptible populations to avoid consuming relevant products i.e. Camembert and other soft cheeses made from raw milk by making informed food choices.

*Labelling of cheeses (from pasteurised milk or raw milk)*

54. In this study, out of the 28 cheese samples, 16 of them were labelled that pasteurised milk or microfiltered<sup>e</sup> milk was one of the ingredients while four of them were labelled that they contained raw or unpasteurised milk. However, seven samples did not explicitly label whether the milk used in the product was pasteurised or not i.e. only labelled milk was one of the ingredients. One sample labelled that the product was made from fresh milk; however, there is no legal definition for the word “fresh” for milk products in the existing food safety legislation.

55. The local Food and Drugs (Composition and Labelling) Regulations (Cap. 132W) stipulates that prepackaged food shall be legibly marked or labelled with a list of ingredients, however, there is no

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<sup>e</sup> Microfiltration of milk is used in the dairy industry to produce milk with an extended shelf life, greater than that achieved by pasteurisation alone. The milk is separated into skimmed milk and cream, then the skimmed milk is subject to microfiltration to remove bacteria and the cream is heat treated, before the skimmed milk and cream are recombined and pasteurised.  
(<http://multimedia.food.gov.uk/multimedia/pdfs/committee/acm995mbovis.pdf>)

specific requirement to indicate whether the milk ingredient is pasteurised or not. However, in the EU, the label of cheese made from raw milk must clearly show the words “made with raw milk”.<sup>19</sup> Similarly, in the US, in the case of varieties of cheeses made from unpasteurised milk or its wrapper or immediate container shall be affixed a removable tag reading, “(the applicable name of the variety of cheese) cheese made from unpasteurised milk”.<sup>20</sup>

56. In order to protect public health and provide sufficient information for the consumers to make informed food choice, the trade is advised to properly label whether the dairy products i.e. cheeses are made from raw milk or pasteurised milk. Susceptible populations should read food labels carefully when selecting food so as to avoid high risk foods, including soft cheeses from raw/ unpasteurised milk and/or choosing safer alternatives e.g. hard and extra hard cheeses and other cheeses from pasteurised milk.

#### *Indication of “use by” or “best before” date*

57. As stipulated in Cap 132W, prepackaged food shall be legibly marked or labelled with the appropriated durability indication: a “best before” date - the food can reasonably be expected to retain its specific properties if properly stored or a “use by” date - from the microbiological point of view, the product is highly perishable and is therefore likely after a short period to constitute an immediate danger to human health.

58. Out of 100 samples in this study, 71 of them (71%) carried a “use by” date; while 27 (27%) including two (one cheese and one processed meat samples) unsatisfactory samples due to excessive ACC or *E. coli* count carried a “best before” date. It is also noted that two samples, including one processed meat sample containing excessive ACC, carried both “use by” and “best before” dates on the package (Table 4). Follow up inspections revealed that one of the concerned samples showed only the "use by" date on the package, while the other questioned product was not detected during the follow up inspections.

**Table 4. Number of samples carried a “use by” date and/or “best before” date on package**

	“Use by” date	“Best before” date	Both “Use by” and “Best before” date
<b>Cheese (n=28)</b>	18	10	-
<b>Smoked seafood (n=28)</b>	22	6	-
<b>Processed meat (n=28)</b>	23	4	1
<b>Salad (n=16)</b>	8	7	1
<b>Total (n=100)</b>	71 (71%)	27 (27%)	2 (2%)

59. Currently, there is no definitive list of which foods should carry a particular type of date mark.<sup>21</sup> It is the responsibility of the trade to set the appropriate durability indication or date mark, together with the storage instructions required to achieve the stated shelf life.

60. In order to provide non-misleading information to the consumer, when establishing shelf life for the products, traders should consult with technical experts on the microbiological risks posed by the products and give a “use by” or “best before” date mark on the package where appropriate.

61. In general, food products that are microbiologically perishable, e.g. chilled ready-to-eat foods and may consequently, after a short period of time, pose a risk to public health should have their shelf-life indicated by a “use-by” date.

## **LIMITATIONS**

62. In this study, only 100 prepackaged long shelf life refrigerated ready-to-eat samples were taken, covering selected types of products. Non-prepackaged products were not covered in this study.

63. Due to limited availability of salad products, which could fully fulfil with the criteria set out in this study, identified in the local retail stores, only 16 salad samples were incorporated. The outstanding sample quotas were evenly distributed to other food categories i.e. cheese, smoked seafood and processed meat to make up to 100 samples in this study.

64. Samples were stored in a monitored laboratory refrigerator at



4±3°C. There might be differences between the storage condition at the laboratory and at the marketplace. The day of analysis was within the week which the sample expired, which might not fall on the exact expiry date of the product.

65. For the purpose of this study, the microbiological quality of the sample is considered “Unsatisfactory: Potentially injurious to health and/or unfit for human consumption” if the *L. monocytogenes* count is greater than 100 cfu/g. Samples contained *L. monocytogenes* count less than 20 cfu/g was therefore considered of either satisfactory or borderline microbiological quality.

66. Depending on the method used, the *E. coli* results in this study were expressed as in either cfu/g or MPN/g and are used to compare directly with the microbiological criteria expressed as cfu/g. In addition, the maximum reporting limit for the MPN method used in this study is >1100 MPN/g.

67. Further investigations on whether the *E. coli* detected in the concerned unsatisfactory cheese samples are pathogenic fall outside the scope of this study.

## **CONCLUSION AND RECOMMENDATIONS**

68. Results showed that the *L. monocytogenes* counts in all 100 samples at the end of shelf life were less than 20 cfu/g i.e. none of the

samples contained excessive *L. monocytogenes*. The microbiological quality of 89 samples (89%) were either of satisfactory or borderline at the end of shelf life. Regarding the samples with unsatisfactory microbiological quality, one smoked seafood and seven processed meat samples were found to contain excessive ACC (ranged from  $1.7 \times 10^8$ – $3.9 \times 10^8$  cfu/g). Post-processing contamination and/or inadequate temperature control might result in the unsatisfactory microbiological quality of these samples. Three cheese samples were found to contain high *E. coli* count (one sample contained 240 MPN/g while the other two contained more than 1100 MPN/g); it might be due to the use of raw/unpasteurised milk contaminated with *E. coli* in these products.

69. Below are some advices for public and trade in relation to prepackaged long shelf life refrigerated products

#### **Advice to public**

- Read food labels carefully to make informed food choices.
- Follow the storage instructions e.g. "keep in a refrigerator" provided by the manufacturer and avoid cross-contamination.
- Do not use food after the "use by" date as shown on the food label.

#### **Advice to susceptible populations including pregnant women, the elderly and immunocompromised individuals**

- Avoid high risk foods especially refrigerated ready-to-eat foods with long shelf life.

- Choose cheeses carefully before consumption.
  - Hard and extra hard cheeses are generally safe.
  - Avoid soft cheeses such as Feta, Brie, Camembert, blue cheeses (e.g. Danish blue, Gorgonzola and Roquefort).
  - For other types of cheeses, choose only those made from pasteurised milk.
  - Do not eat if in doubt.
- Cook all food thoroughly and use ready-to-eat food as soon as possible.

### **Advice to trade**

- Maintain good food and personal hygiene at all food processing stages.
- Provide an adequately refrigerated environment (at or below 4 °C) throughout the supply chain for refrigerated products.
- Provide sufficient information on food label for the consumers to make informed food choices.
  - Properly label whether the dairy products are made from raw milk or pasteurised milk.
- Ensure all information provided on food label complies with the legal requirements.

## REFERENCES

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- <sup>1</sup> Codex, 2009. Guidelines on the Application of General Principles of Food Hygiene to the Control of *Listeria monocytogenes* in Ready-to-Eat Foods. (CAC/GL 61-2007).  
[http://www.codexalimentarius.org/download/standards/10740/CXG\\_061e.pdf](http://www.codexalimentarius.org/download/standards/10740/CXG_061e.pdf)  
(Accessed 10 September 2014).
- <sup>2</sup> World Health Organization Food and Agriculture Organization of the United Nations, 2004. Risk assessment of *Listeria monocytogenes* in ready-to-eat foods. Microbiological risk assessment series 5.  
<ftp://ftp.fao.org/docrep/fao/010/y5394e/y5394e.pdf> (Accessed 10 September 2014).
- <sup>3</sup> Centre for Health Protection, 2008. Listeriosis.  
<http://www.chp.gov.hk/en/content/9/24/14450.html> (Accessed 10 September 2014).
- <sup>4</sup> US Food and Drug Administration, 2012. Bad Bug Book (Second Edition) Foodborne Pathogenic Microorganisms and Natural Toxins Handbook. *Listeria monocytogenes*. P. 100-104.  
<http://www.fda.gov/downloads/Food/FoodborneIllnessContaminants/UCM297627.pdf>  
(Accessed 10 September 2014).
- <sup>5</sup> US Food and Drug Administration, 2003. Quantitative Assessment of the Relative Risk to Public Health from Foodborne *Listeria monocytogenes* Among Selected Categories of Ready-to-Eat Foods.  
<http://www.fda.gov/downloads/Food/FoodScienceResearch/UCM197330.pdf>  
(Accessed 10 September 2014).
- <sup>6</sup> US Centers for Disease Control and Prevention, 2013. *Listeria* (Listeriosis). Definition.  
<http://www.cdc.gov/listeria/definition.html> (Accessed 10 September 2014).
- <sup>7</sup> Centre for Health Protection, 2010. Scientific Committee on Enteric Infections and Foodborne Disease. Epidemiology of Listeriosis and Prevention Strategies.  
[http://www.chp.gov.hk/files/pdf/epidemiology\\_of\\_listeriosis\\_and\\_prevention\\_strategies\\_r.pdf](http://www.chp.gov.hk/files/pdf/epidemiology_of_listeriosis_and_prevention_strategies_r.pdf) (Accessed 10 September 2014).
- <sup>8</sup> Health Canada, 2011. Policy on *Listeria monocytogenes* in Ready-to-Eat Foods.  
[http://www.hc-sc.gc.ca/fn-an/alt\\_formats/pdf/legislation/pol/policy\\_listeria\\_monocytogenes\\_2011-eng.pdf](http://www.hc-sc.gc.ca/fn-an/alt_formats/pdf/legislation/pol/policy_listeria_monocytogenes_2011-eng.pdf) (Accessed 10 September 2014).
- <sup>9</sup> The EFSA Journal, 2013. Analysis of the baseline survey on the prevalence of *Listeria monocytogenes* in certain ready-to-eat foods in the EU, 2010-2011 Part A: *Listeria monocytogenes* prevalence estimates. 11(6):3241.  
<http://www.efsa.europa.eu/en/efsajournal/doc/3241.pdf> (Accessed 10 September 2014).
- <sup>10</sup> Food and Drugs (Composition and Labelling) Regulations (Cap 132W).

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Schedule 3. Marking and Labelling Prepackaged Foods.

[http://www.legislation.gov.uk/blis\\_ind.nsf/CURALLENGDOC/35B32379D1420DD2482577520022790C?OpenDocument](http://www.legislation.gov.uk/blis_ind.nsf/CURALLENGDOC/35B32379D1420DD2482577520022790C?OpenDocument) (Accessed 10 September 2014).

<sup>11</sup> Centre for Food Safety, 2014. Microbiological Guidelines for Food.

[http://www.cfs.gov.hk/english/food\\_leg/files/food\\_leg\\_Microbiological\\_Guidelines\\_for\\_Food\\_e.pdf](http://www.cfs.gov.hk/english/food_leg/files/food_leg_Microbiological_Guidelines_for_Food_e.pdf) (Accessed 10 September 2014)

<sup>12</sup> Health Protection Agency, 2009. Guidelines for Assessing the Microbiological Safety of Ready-to-Eat Foods Placed on the Market.

[http://www.hpa.org.uk/web/HPAwebFile/HPAweb\\_C/1259151921557](http://www.hpa.org.uk/web/HPAwebFile/HPAweb_C/1259151921557) (Accessed 10 September 2014).

<sup>13</sup> US Food and Drug Administration, 2010. Guidance for FDA Staff Compliance Policy Guide Sec. 527.300 Dairy Products - Microbial Contaminants and Alkaline Phosphatase Activity.

<http://www.fda.gov/downloads/ICECI/ComplianceManuals/CompliancePolicyGuidanceManual/UCM238465.pdf> (Accessed 10 September 2014).

<sup>14</sup> Australia New Zealand Food Standards Code - Standard 1.6.1 - Microbiological Limits for Food.

<http://www.comlaw.gov.au/Details/F2012C00862> (Accessed 10 September 2014).

<sup>15</sup> User guide to Standard 1.6.1 – Microbiological Limits for Food with additional guideline criteria. July 2001.

[http://www.foodstandards.gov.au/code/userguide/documents/Micro\\_0801.pdf](http://www.foodstandards.gov.au/code/userguide/documents/Micro_0801.pdf) (Accessed 10 September 2014).

<sup>16</sup> Cheshire, Cumbria, Greater Manchester, Lancashire, Merseyside, North Wales FLGs & HPA FW&E Microbiology Laboratory, Preston. Microbiological Examination of Retail Cheeses made from Raw Milk with a Focus on Pathogens. Coordinated Survey Report. Survey Number 1004001. January 2012.

<http://www.salford.gov.uk/d/1004001report.pdf> (Accessed 10 September 2014).

<sup>17</sup> The EFSA Journal, 2003. Opinion of the Scientific Panel on Biological Hazards on a request from the Commission related to the effects of Nitrites/Nitrates on the Microbiological Safety of Meat Products. 14, p 1-34.

<http://www.efsa.europa.eu/fr/scdocs/doc/14.pdf> (Accessed 10 September 2014).

<sup>18</sup> Food Standard Australia New Zealand, 2009. Microbiological Risk Assessment of Raw Milk Cheese.

<http://www.foodstandards.gov.au/code/proposals/documents/P1007%20PPPS%20for%20raw%20milk%201AR%20SD3%20Cheese%20Risk%20Assessment.pdf>

(Accessed 10 September 2014).

<sup>19</sup> Regulation (EC) No 853/2004 of the European Parliament and of the Council of 29 April 2004 laying down specific hygiene rules for food of animal origin.

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<http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=CONSLEG:2004R0853:20130701:EN:PDF> (Accessed 10 September 2014).

<sup>20</sup> US Food and Drug Administration. Code of Federal Regulations Title 21 – Food and Drugs. Part 101- Food Labelling. Subpart G - Exemptions From Food Labeling Requirements. Sec. 101.100 Food; exemptions from labeling. <http://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/CFRSearch.cfm?fr=101.100> (Accessed 10 September 2014).

<sup>21</sup> UK Department for Environment, Food and Rural Affairs, 2011. Guidance on the Application of Date Labels to Food. [https://www.gov.uk/government/uploads/system/uploads/attachment\\_data/file/69316/pb132629-food-date-labelling-110915.pdf](https://www.gov.uk/government/uploads/system/uploads/attachment_data/file/69316/pb132629-food-date-labelling-110915.pdf) (Accessed 10 September 2014).