Risk Assessment Studies
Report No. 75

Microbiological Hazard Evaluation

MICROBIOLOGICAL QUALITY OF PACKAGED RICE BALLS

December 2024
Centre for Food Safety
Food and Environmental Hygiene Department
The Government of the Hong Kong Special Administrative Region

Correspondence:
Risk Assessment Section
Centre for Food Safety
Food and Environmental Hygiene Department
43/F, Queensway Government Offices,
66 Queensway, Hong Kong
Email: enquiries@fehd.gov.hk

Table of Contents

Risk Assessment Studies
Report No. 75

MICROBIOLOGICAL QUALITY OF PACKAGED RICE BALLS

EXECUTIVE SUMMARY

The popularity of rice balls (onigiri), a type of rice product with various ingredients embedded inside some cooked rice, has increased over the past decade in Hong Kong. The convenient package design of packaged rice balls may encourage consumers to consume rice balls on-the-go, potentially exposing them to improper temperature for an extended period before consumption. Certain rice balls may be displayed under conditions above 4°C. Rice balls are typically prepared by cooking rice with added salt for flavor and preservation purpose. A sufficient amount of salt, along with other factors, may be used to control microbial growth, particularly *Bacillus cereus*, which is a known hazard in cooked rice products. However, during the preparation of ready-to-eat rice ball fillings, post-cooking contamination with *Staphylococcus aureus* may result if food handlers do not observe Good Hygiene Practices (GHPs). *Listeria monocytogenes*, a ubiquitous pathogen in the environment, can also potentially contaminate the ingredients during preparation. Considering the intrinsic microbiological risk of packaged rice balls and their storage practices, the Centre for Food Safety (CFS) conducted a study to assess the change in microbiological quality of packaged rice balls after leaving them under ambient condition for a period of time.

Methodology

Between February and March 2024, 63 pairs of samples of packaged rice balls were collected from four takeaway chains and five supermarket chains/convenience store chains in different regions of Hong Kong. To simulate the scenario where packaged rice balls were left under ambient condition for a prolonged period of time, one sample in each pair was kept under ambient condition after purchase for four hours before refrigeration (i.e. “simulated

sample") while the other one was kept under refrigeration at 4°C or below immediately after purchase (i.e. "control sample"). The microbiological quality of the packaged rice ball samples was assessed against the criteria stipulated in the Microbiological Guidelines for Food (the Guidelines). The microbiological criteria include (a) aerobic colony count (ACC, a quality indicator)[i], (b) *Escherichia coli* (a hygiene indicator), and (c) specific foodborne pathogens. In addition, the change in microbiological quality after leaving packaged rice balls under ambient condition for a period of time was assessed by comparing the microbiological quality of the simulated samples to that of the corresponding control samples.

Results and Discussion

All samples complied with the microbiological food safety criteria for pathogenic bacteria (i.e. *Listeria monocytogenes*, *Staphylococcus aureus,* and *Bacillus cereus*). The overall microbiological quality of control samples (i.e. those stored under refrigeration at 4°C or below immediately after purchase) of packaged rice balls was found satisfactory except that one rice ball sample with braised salmon was detected with excessive level of *E. coli*, resulting in an unsatisfactory rating for hygienic quality but there was no food safety concern. It indicated that there may be room for improvement in the food manufacturing and handling process. The CFS has provided advice on the food preparatory process to the concerned manufacturer. A follow-up sample was taken and the result was satisfactory.

Among the 63 simulated samples, ACC criterion is applicable to 58 of them and only one sample was found borderline for ACC while all the others were found to be satisfactory. However, among these 58 samples, three samples had an ACC more than ten times higher than those control samples. ACC is an indicator of

[i] ACC is not applicable to rice balls containing raw ingredients or fermented ingredients.

quality rather than safety and hence does not indicate food safety concern. Nevertheless, ACC can provide useful information about the general quality and remaining shelf life of the food. As regard the hygienic quality, only the simulated sample of the braised salmon rice ball that was detected with excessive level of *E. coli* was 1.1-fold higher in *E. coli* compared to the corresponding control sample. As mentioned above, *E. coli* is a hygienic quality and does not indicate food safety concern. Nonetheless, the food handlers should observe good personal, food and environmental hygiene in the food preparatory process.

Time/temperature control is commonly used to control microbial growth of perishable ready-to-eat food, while other physical parameters, such as pH and water activity, could be applied in combination to create an environment resistant to spoilage. The modification of ingredients or addition of food additive to change the physical parameters can be used as control measures for limiting microbial growth so that the rice balls could be kept at temperatures above 4ºC for a certain period of time. Nevertheless, these control measures should be validated to see if they are sufficient to control relevant hazards. In addition, vendors should provide appropriate information, e.g. storage condition and/or duration, about their products to consumers for the safe handling of rice balls.

Conclusion

In this study, the overall microbiological quality of packaged rice balls was satisfactory. All samples complied with the microbiological food safety guideline levels for pathogenic bacteria. However, one control sample was found to have the hygiene indicator organism (i.e. *E. coli*) that exceeded the hygiene quality criterion in the Guidelines indicating that there may be room for improvement in the food manufacturing and handling process, but there was no food safety concern. Ready-to-eat ingredients are usually prepared in advance for use as rice ball fillings, manufacturers should follow the GHPs when handling

these ready-to-eat ingredients.

Compared to the control samples which had not been subject to the same condition, more than ten-fold higher in microbial count was noted in a few simulated samples left under ambient condition for four hours after purchase, but the microbial counts still complied with the microbiological criterion as specified in the Guidelines. Apart from time/temperature control, modification of ingredients or the addition of food additive to change the physical parameters of food (e.g. pH and water activity) can be used as control measures. Nevertheless, these control measures should be validated to see if they are sufficient to safeguard food safety. In addition, vendors can provide correct and appropriate information, e.g. storage condition and/or duration, about their products to consumers for the safe handling of rice balls.

Below is advice for the public and trade regarding the safe handling of packaged rice balls.

Advice to Trade

GHPs for preventing contamination in food and proper storage of food, particularly ready-to-eat food, should be followed at all times. Food businesses also have the responsibility to provide sufficient ongoing food safety/hygiene training to their staff (i.e. food handlers in their businesses). Furthermore, a preventive food safety management system (such as the Hazard Analysis and Critical Control Point (HACCP) system) should be established to ensure that effective control measures are in place to minimise potential contamination of the products during the manufacturing process. Some GHPs are highlighted below:

- Food handlers should receive proper training to prepare rice balls properly and prevent contamination of rice balls. The most important points are: wash hands with correct hand washing method and change disposable gloves properly.

- The preparation areas for raw food should be separated from areas for handling ready-to-eat food, as far as possible. For packaged ready-to-eat ingredients, where appropriate, wipe clean and dry the packaging before opening to minimise potential cross-contamination.
- The schedule or sequence of rice ball preparation should be planned to avoid leaving perishable/ready-to-eat ingredients under ambient conditions for too long.

Storage and Display

- In general, rice balls intended to be display for a prolonged period should be refrigerated after preparation;
- Control measures for limiting microbial growth so that the rice balls could be kept at temperatures above 4ºC for a certain period of time, should be validated to see if they are sufficient to control relevant hazards.
- Recommendations on storage conditions should be provided for the safe handling of rice balls by consumers.

<u>Advice to Public</u>

- Consume rice balls as soon as possible after purchase.
- For packaged rice balls, follow the storage instructions available on the packaging and consume them before the use-by date.
- Refrigerate packaged rice ball at 4°C or below if they are not to be consumed immediately.

OBJECTIVES

The purpose of the study is to assess the change in microbiological quality of packaged rice balls after leaving them under ambient condition for a period of time.

INTRODUCTION

2. The popularity of rice balls (onigiri), a type of rice product with various ingredients embedded inside some cooked rice, has increased over the past decade in Hong Kong. The wide variety of embedded ingredients provides consumers with a diverse range of flavours. Packaged rice balls are usually sold in shops or small kiosks for takeaway consumption. The convenient package design of packaged rice balls, which is easy to open, makes rice balls readily accessible for on-the-go consumption. Similar to sushi, rice balls are usually non-hot served though they may be reheated briefly during wintertime.

3. Rice ball is different from its counterpart, sushi, in that vinegar is not added to make the cooked rice acidic and thus less favourable to support the growth of microorganisms. Instead, salt is added when cooking the rice to make rice balls. Salt reduces the water activity of foods, thereby aiding in their preservation. A sufficient amount of salt, along with other factors such as pH, temperature, or food additives, may be used to effectively control microbial growth.[1] In a study investigating the inhibitory effect on toxin production by *Staphylococcus aureus* in rice balls, toxin production was found to be reduced in

the presence of glycine and 2% of NaCl for short incubation period (i.e. four hours) but not for long incubation period (i.e. eight to 24 hours) at 37°C.[2]

4. Besides the rice, the fillings and sauces inside the rice balls, such as ready-to-eat meats and ready-to-eat sauces like mayonnaise, could also contribute to the microbial load or even be a source of contamination. These ingredients are typically prepared in advance for subsequent incorporation into the rice balls. The handling and storage of ready-to-eat ingredients can increase the microbiological risk if Good Hygiene Practices (GHPs) are not observed. For instance, *Listeria monocytogenes*, a ubiquitous pathogen in the environment, can potentially contaminate the ingredients during preparation if GHPs are not observed.

5. Rice balls may be left under ambient condition for a period of time after purchase, but similar to other perishable ready-to-eat food, they should be discarded after being kept in temperature danger zone (i.e. 4°C to 60°C) for four hours.[3] After purchasing packaged rice balls, consumers may impose the risk of storage at improper temperature for several hours before consumption, such as when purchasing them in advance for a subsequent meal, during outdoor activities such as picnics, or during extended transportation from purchase location to the final destination. In addition, according to information provided by one of the manufacturers of packaged rice balls, it is noted that some rice balls with the addition of salt may be stored or displayed at temperature above 4°C for extended durations, as low temperatures (e.g. at 4°C or below) may cause the change in texture of rice balls. Low temperatures can cause retrogradation of starch, leading to a hardened texture of the cooked rice and a lower eating quality of the rice balls.[4,5]

6. Storage without temperature control may allow microorganisms to thrive. Two rice ball samples collected by local media exhibited a more than 10-

log increase in total bacterial counts after being left under 25°C for four hours.[6] This could allow contaminated pathogens, such as *S. aureus* and *Bacillus cereus*, to grow to high levels and causing foodborne disease. Post-cooking contamination with *S. aureus* may result if food handlers do not observe GHPs. Cooked rice is one of the commonly incriminated foods of *B. cereus* food poisoning, as the endospores of this pathogen can survive the cooking process, germinate into vegetative cells, then proliferate and produce heat-stable toxin under prolonged storage at ambient conditions.[7] Rice meals that are not freshly prepared are of particular concern in this regard.[8]

7. Taking into account the intrinsic microbiological risk of packaged rice balls and the risk related to their storage practices, the Centre for Food Safety (CFS) conducted a study to assess the change in microbiological quality of packaged rice balls after leaving them under ambient condition for a period of time. The results of this study can provide the basis for raising the awareness of the general public about the proper handling of packaged rice balls, and reviewing whether additional recommendations or advice can be formulated for the general public and the food trade.

SCOPE OF STUDY

8. This study focused on packaged rice balls sold at the local retail outlets, which contain ingredients such as undercooked eggs, fish roe, salad dressing, cooked meat, cooked seafood, or cooked eggs.

METHODOLOGY

Sampling

9. Between February and March 2024, 63 pairs of samples of packaged rice balls with different types of ingredients were collected from the local retail outlets by the CFS of the Food and Environmental Hygiene Department (FEHD) (Table 1). The samples were collected from four takeaway chains and five supermarket chains/convenience store chains in different regions of Hong Kong. In order to collect wide variety of samples from different shops, only one type was collected from a shop of the same brand/chain as far as possible.

Table 1: Distribution of the samples of packaged rice balls

Types	**Ingredient**	**Numbers**		**Total**
		Supermarket/ Convenience store	**Chain store**	
A	Egg (undercooked)	1	2	3
B	Fish roe	4	6	10
C	Mixed with salad sauce or dressing	4	5	9
D	Cooked meat	10	7	17
E	Cooked seafood	15	8	23
F	Egg (cooked)	0	1	1
Total		34	29	63

10. The inclusion criteria for the sampling were those packaged rice balls that were kept or displayed for sale at either cold temperature or under ambient condition at the sampled retail outlets. Hot or non-packaged rice balls displayed for sale were excluded.

11. The study simulated the scenario after purchase where packaged rice balls were left under ambient condition for a prolonged period of time. For each set, two samples were collected. One sample (around 200 g) in each pair was kept under refrigeration at 4°C or below immediately after purchase (i.e. “control sample”), while the other one was left under ambient condition after purchase for four hours, i.e. the time limit for keeping perishable ready-to-eat food in the

temperature danger zone (i.e. between 4°C and 60°C), before refrigeration (i.e. "simulated sample"). The change in microbiological quality of ready-to-eat packaged rice balls after leaving them under ambient condition for a period of time after purchase was assessed by comparing the microbiological quality of the paired simulated and control samples.

12. For samples with seaweed packaged separately, the seaweed was combined with the rice balls for testing. The information on packing of seaweed were recorded.

Laboratory analysis

13. Both simulated and control samples were stored at 4°C or below, and were delivered to the laboratory of the contractor commissioned by the CFS to provide the microbiological testing services, on the day following sampling. Aerobic colony count (ACC), *Escherichia coli* count, *S. aureus* and other coagulase-positive staphylococci count (*S. aureus* count), *B. cereus* count and *L. monocytogenes* count were used to reflect the microbiological quality of rice ball samples.

14. The enumeration of ACC in samples was performed using the AOAC official method 990.12 (18th ed. 2005) (3M Petrifilm™ Plate). *E. coli* count in samples was performed using the AOAC Official Methods 991.14 (18th ed. 2005) (3M Petrifilm™ Plate). *S. aureus* count was enumerated according to Bacteriological Analystical Manual Online Chapter 12 (Jan 2001) (Direct Plate Count Method) of the U.S. Food and Drug Administration (FDA). *B. cereus* determination was conducted with reference to ISO 7932 (3rd ed., 2004 (E)), colony technique at 30°C. Enumeration of *L. monocytogenes* was conducted according to Bacteriological Analytical Manual Online Chapter 10 (Apr 2022) of FDA.

Result analysis

15. The microbiological results of the packaged rice balls were analysed by the Risk Assessment Section of the CFS, and assessed against the criteria in CFS' Microbiological Guidelines for Food (the Guidelines).[9] The relevant criteria are extracted from the Guidelines and listed in Tables 2 and 3. The changes in bacterial counts of the samples were evaluated.

Quality and Hygiene – Aerobic Colony Count and E. coli

16. Aerobic colony count is the total number of bacteria found in food. 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.[9] The number of bacteria can increase significantly over time as a result of lack of temperature control of the product. Hence, ACC can highlight potential storage and handling problems since production and provide clues for early detection of problems and how the problems may be solved. It is worth noting that ACC is an indicator of food quality but not food safety.[9]

17. The Guidelines classify ready-to-eat food products[ii] into 14 categories because several factors (such as the types of raw ingredients used and the nature of processing) may affect the ACC of a product at the point of sale. For example, heat processes such as cooking will result in low ACC, and products containing raw ingredients such as fresh vegetables will have much higher ACC due to the natural flora present. Handling after heat processing such as slicing, portioning, packaging may increase the ACC, although this should be minimised by GHPs.

[ii] "Ready-to-eat food" means 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.

18. Taking the ingredients and the nature of processing into consideration, packaged rice ball samples in this study were classified according to the Guidelines as shown in Table 2. The ingredients of packaged rice balls are usually cooked in advance and likely to be further handled (e.g. mixing with rice) during preparation; hence, they generally belong to category 5. For those containing raw seafood or salad dressing, the satisfactory limit is less stringent as ACC are expected to be higher. As for rice ball samples mixed with raw vegetables (such as shredded cucumber or salad vegetables), they are classified into category 12, as raw vegetables can contain high levels of bacteria as part of their normal micro-flora and hence, are likely to have high ACC and ACC is not applicable to rice balls containing raw ingredients i.e. category 12. ACC is also not applicable to fermented foods, i.e. category 13.

Table 2. ACC and *E. coli* criteria used in this study

Food category in the Guidelines	**Microbiological quality** Result (colony-forming unit (cfu/g))		
	Satisfactory	Borderline	Unsatisfactory
Aerobic colony count (ACC) [30°C /48 hours]			
5. Cooked foods chilled but with some handling prior to sale or consumption, e.g. cooked meat, cooked egg, etc.	$<10^5$	10^5-$<10^7$	$\geq 10^7$
9. Raw ready-to-eat meat and fish, cold smoked fish, including fish roe	$<10^6$	10^6-$<10^7$	$\geq 10^7$
12. Fresh fruit and vegetables, products containing raw vegetables	N/A	N/A	N/A
13. Fermented, cured and dried meats, fermented vegetables, ripened cheeses			
Hygiene indicator organisms			
E. coli	<20	20 - $\leq 10^2$	$>10^2$

Remarks:
1. Category numbers as marked for the food categories under ACC are directly adapted from the CFS' Microbiological Guidelines for Food.
2. For ACC, the detection limit is 100 cfu/g. For *E. coli*, the detection limit is 10 cfu/g.

19. *E. coli* is a bacterium found in the gastrointestinal tract of humans and is commonly used as a faecal indicator to reflect the hygienic quality of a food product. *E. coli* are killed by the heat processes used in food production and should be readily removed from the equipment and surfaces by appropriate

cleaning procedures. Its presence in food indicates direct or indirect faecal contamination, and a substantial number of *E. coli* in food suggests a general lack of cleanliness in handling and improper storage.[9]

Microbiological safety – Pathogens

20. Taking into account the nature of rice balls and the preparatory practices, *S. aureus*, *B. cereus*, and *L. monocytogenes* were used to assess the safety of packaged rice balls in this study.

21. *S. aureus* is one of the common food poisoning microorganisms in Hong Kong. The most common way of contamination of food is by contact with food handlers' hands, especially in the cases where the food is handled after cooking, and once contaminated with *S. aureus,* prolonged storage without refrigeration allows *S. aureus* to grow to high numbers and form enterotoxins. Although staphylococcal enterotoxins can also be produced in food by some other coagulase-positive staphylococci such as *Staphylococcus intermedius*, most coagulase-positive staphylococci which cause foodborne illness are *S. aureus*.[9] *B. cereus* is a spore-forming bacterium that is ubiquitous in the environment and readily isolated from soil cereal crops and vegetables, etc.[9] The spores, if present, can survive cooking temperature and become vegetative cells in cooked rice. If the rice is left under ambient condition for a prolonged period after cooking, vegetative cells can proliferate into a large number. *L. monocytogenes* is universally present in the environment. It can survive and multiply at temperature as low as 0°C but can be easily destroyed under normal cooking temperature. Growth of this bacterium following contamination of cooked foods will result in disease transmission.[9] Rice balls may be contaminated during the handling process and subsequent storage can allow the proliferation of the bacterium.

22. The microbiological safety of the samples was determined using the criteria for pathogens of the Guidelines (Table 3).[9]

Table 3. Pathogen criteria of the Guidelines

Criterion	**Result (cfu/g unless otherwise specified)**		
	Satisfactory	**Borderline**	**Unsatisfactory:** potentially injurious to health and/or unfit for human consumption
Listeria monocytogenes ● For other ready-to-eat food	< 10	10 - ≤ 100	> 100
S. aureus and other coagulase-positive staphylococci	< 20	20 - $\leq 10^4$	> 10^4
Bacillus cereus	< 10^3	10^3 - ≤ 10^5	> 10^5

Remark: The detection limit is 10 cfu/g.

RESULTS

Specific foodborne pathogens

23. All samples complied with the microbiological food safety criteria for pathogenic bacteria (i.e. *L. monocytogenes*, *S. aureus,* and *B. cereus*).

Quality and hygiene indicators – ACC and E. coli count

24. Among the 63 control samples taken, 49 samples were classified into category 5 while nine were classified into category 9. None of the 58 samples in categories 5 and 9 were unsatisfactory (Table 4). Five control samples were classified into category 12 or category 13 as they contained raw ingredients or fermented ingredients, such as fresh scallion and miso, which inherently contained high ACC as natural flora and hence, the ACC criterion does not apply to these samples.

Table 4. ACC results of control samples (Number of applicable samples=58)

	Microbiological results (cfu/g)					
	$<10^3$	10^3-$<10^4$	10^4-$<10^5$	10^5-$<10^6$	10^6-$<10^7$	$\geq 10^7$
Microbiological quality (Food category 5)	**Satisfactory**			**Borderline**		**Unsatisfactory**
Cooked foods chilled but with some handling prior to sale or consumption (Total no. of sample=49)	46	2	1	0	0	0
Microbiological quality (Food category 9)	**Satisfactory**				**Borderline**	**Unsatisfactory**
Raw ready-to-eat meat and fish, cold smoked fish, including fish roe (Total no. of sample=9)	8	0	1	0	0	0

25. Regarding the hygienic quality, one control sample of packaged rice ball with braised salmon was rated as unsatisfactory, with 540 cfu/g *E. coli* detected (Table 2). All other control samples were rated as satisfactory because their *E. coli* counts were less than 20 cfu/g.

Change in microbiological quality of packaged rice balls after leaving them under ambient condition for a period of time

26. Among the 63 simulated samples, ACC criterion is applicable to 58 of them and only one sample was found borderline for ACC while all the others were found to be satisfactory (Table 5). However, nine out of these 58 samples were found to have higher ACC ranging from 1.2-fold to 540-fold (median: 2.6-fold) in comparison with the corresponding control samples.[iii] Among these nine simulated samples, three were found to have more than 10-fold higher in ACC count: the grilled chicken rice ball (540-fold), the teriyaki salmon rice ball (88-fold), and the tuna fish rice ball (36-fold) compared to the corresponding control samples. For the remaining simulated samples, the ACC count was not higher

[iii] For samples with ACC below detection limit (i.e. <10 cfu/g), the ACC was assumed to be 10 cfu/g for the purpose of calculation of ACC changes.

compared to the corresponding control samples or the counts were found to be <10 cfu/g.

Table 5. ACC results of simulated samples (Number of applicable samples=58)

	Microbiological results (cfu/g)					
	$<10^3$	10^3-$<10^4$	10^4-$<10^5$	10^5-$<10^6$	10^6-$<10^7$	$\geq 10^7$
Microbiological quality (Food category 5)	**Satisfactory**			**Borderline**		**Unsatisfactory**
Cooked foods chilled but with some handling prior to sale or consumption (Total no. of sample=49)	45	2	1	1	0	0
Microbiological quality (Food category 9)	**Satisfactory**				**Borderline**	**Unsatisfactory**
Raw ready-to-eat meat and fish, cold smoked fish, including fish roe (Total no. of sample=9)	8	0	1	0	0	0

27. For the packaged rice ball with braised salmon that was rated as unsatisfactory for *E. coli* count, the *E. coli* count of the simulated sample (i.e. 600 cfu/g) was 1.1-fold higher compared to that of the corresponding control sample (i.e. 540 cfu/g). All other simulated samples were rated as satisfactory, with *E. coli* counts were less than 20 cfu/g, after leaving them under ambient condition for four hours after purchase.

Storage/display temperature of control samples collected at sampled retail outlets

28. Based on the collected information, packaged rice balls were usually kept refrigerated in fridge at 4ºC or below. A few samples were kept above 4ºC but still at or below 8ºC at the sampled retail outlets (Table 6). Among the sampled retail outlets, one chain was found to keep rice balls under ambient condition for sale, with a time limit of display established as eight hours while two chains were found to display packaged rice balls in display fridges at around 14ºC to 18ºC, without a specified time limit for display.

Table 6. Sample temperature of control samples (n=63)

Sample Temperature	No. of Samples	No. of Chains
≤4ºC to ≤ 8 ºC	40	6
9 ºC to ≤ 20 ºC	15	2
≥21 ºC (ambient conditions)	8	1

DISCUSSION

General microbiological quality of packaged rice balls

29. The microbiological quality of control samples (i.e. those stored under refrigeration at 4°C or below immediately after purchase) of packaged rice balls, was generally satisfactory, except for one sample with potential hygiene issue. All samples complied with the microbiological food safety guidance levels for pathogenic bacteria, as well as the ACC levels, as stipulated in the Guidelines.

30. Only one control sample of rice ball with braised salmon was detected with excessive level of *E. coli*, resulting in an unsatisfactory rating for hygienic quality but there was no food safety concern. It indicated that there may be room for improvement in the food manufacturing and handling process. The CFS has provided advice on the food preparatory process to the concerned manufacturer. A follow-up sample was taken and the result was satisfactory.

31. Rice balls are usually prepared with freshly cooked rice and mixed with different types of ready-to-eat ingredients in a moulding machine before packaging. These ingredients are prepared in advance and they are subject to manual handling and stored for a period of time before using as rice ball fillings. Manufacturers should limit the amount of time these potentially hazardous

ingredients are left out of temperature control. The preparation time should be limited to two hours before storing in the refrigerator, whereas the products can be left out of time control after having been taken out from the refrigerator for two hours until the four-hour limit. The total allowable time of ready-to-eat food can stay out of temperature control is four hours, so the time taken in each stage during transport, storage or preparation of rice balls between 4ºC and 60 ºC must be taken into account.

32. Good personal and environmental hygienic practices should be observed to minimise the chance of cross contaminations and the growth of bacteria. Food businesses also have the responsibility to provide sufficient ongoing food safety/hygiene training to their staff, including reminding them to observe personal hygiene while handling food, especially during the preparation of ready-to-eat ingredients in the production process.

Change in microbiological quality of packaged rice balls after leaving them under ambient condition for a period of time

33. Among the 63 simulated samples, ACC criterion is applicable to 58 of them and only one sample was found borderline for ACC while all the others were found to be satisfactory. However, among these 58 samples, three samples had an ACC more than ten times higher than those control samples. ACC is an indicator of quality rather than safety and hence does not indicate food safety concern. Nevertheless, ACC can provide useful information about the general quality and remaining shelf life of the food. Although the quality indicator had not increased to unsatisfactory level for the packaged rice balls, microbial growth in the food can increase over time under ambient condition. As regard the hygienic quality, only the simulated sample of the braised salmon rice ball that was detected with excessive level of *E. coli* was 1.1-fold higher in *E. coli* compared to the corresponding control sample. As mentioned above, *E. coli* is a

hygienic quality and does not indicate food safety concern. Nonetheless, the food handlers should observe good personal, food and environmental hygiene in the food preparatory process.

34. The Food Hygiene Code of FEHD states that a ready-to-eat potentially hazardous food may be displayed or stored at ambient temperature for a period of not more than four hours (i.e. 4-hour rule).[3] *B. cereus* and *S. aureus* are microbiological hazards relevant to rice balls. Spore-forming *B. cereus* can survive the cooking temperature during the cooking of rice and become vegetative cells. If cooked rice is left at room temperature for a prolonged period, the vegetative cells can multiply into large numbers and/or eventually produce emetic (cause vomiting) toxin.[9] *S. aureus* is a bacterium commonly found on the hair and skin and in the nasal cavity, throat, and wounds of humans. Typically, *S. aureus* contaminates food through contacts with the hands of food handlers, especially in scenarios where the food is handled manually after cooking.[9] This may happen during the handling of ready-to-eat ingredients for rice balls in particular, if hand hygiene is not observed. Combined with prolonged storage or handling at room temperature, the bacterium can grow exponentially and form enterotoxin toxins. To minimise the risk of proliferation of pathogenic bacteria, it is recommended to adopt time/temperature control for rice balls.

35. Time/temperature control is commonly used to control microbial growth of perishable ready-to-eat food, there are instances of rice balls being displayed at temperatures around 14ºC to 18ºC without a time limit of display. To control microbial growth, multiple hurdles, such as temperature, pH, and water activity, could be applied together. For example, the combination of addition of salt and adjusting the pH with acid regulator could create an environment resistant to spoilage and unfavourable for survival of pathogens, while they may not be sufficient when applied alone at certain levels.[10] The modification of ingredients or addition of food additives in order to change the physical parameters can be

used as control measures for microbial growth, allowing the rice balls to be kept at temperatures above 4ºC for a certain period of time. Previous study showed that combination of 1% glycine and 2% NaCl was partly effective in the reduction of toxin production by *S. aureus*.[2] Control measures should be validated to ensure they are sufficient to safeguard food safety, i.e. obtaining evidence that a control measure or combination of control measures, if properly implemented, is capable of controlling the hazard to a specified outcome.[11] For example, laboratory challenge testing designed to evaluate if the product formulation could prevent the microbial growth within the shelf life.

36. In addition, vendors can provide appropriate information, e.g. storage condition and/or duration, about their products to consumers for the safe handling of rice balls. The preparation and storage time of the rice balls is not known by consumers, vendors may provide advice to consumers on their products, such as "Store under XX°C for consumption within the day", where necessary for protection of food safety.

Limitations

37. In this study, only 63 pairs of samples were taken. Due to the wide variety of packaged rice balls available in the market and the changing menu options, only selected types were covered. The microbiological quality of the final products might be affected by other ingredients used to prepare the rice balls.

38. Packaged rice ball samples were collected in pairs, one of which was left under ambient condition after purchase by sampling officers to assess the microbiological changes by comparing the testing results of control and simulated samples. Although samples were collected in pairs, there could be variation in microbiological quality among individual packaged rice balls of the same type, where the initial loads could be different for the sample pairs. As noted from the

result, one control sample was found to contain <10 cfu/g but the corresponding simulated sample was found to contain 5,400 cfu/g. Such large difference was not noted in other paired samples and hence such large difference could be due to differences in the initial bacterial load of the control sample and the corresponding simulated sample. Unlike a challenge test under a set of well-controlled laboratory conditions where a pre-defined number of bacteria is inoculated evenly onto the food, bacteria were likely unevenly distributed in the rice ball samples in this study.

CONCLUSION AND RECOMMENDATIONS

39. In this study, the overall microbiological quality of packaged rice balls was satisfactory. All samples complied with the microbiological food safety guideline levels for pathogenic bacteria, as well as the ACC levels, as stipulated in the Guidelines. However, one control sample was found to have the hygiene indicator organism (i.e. *E. coli*) that exceeded the hygiene quality criterion in the Guidelines indicating that there may be room for improvement in the food manufacturing and handling process, but there was no food safety concern. The hygiene issue highlights the importance of good personal and environmental hygienic practices in the food manufacturing and handling process.

40. Compared to the corresponding control samples, a few simulated samples had ACC more than ten-fold higher after having been left under ambient condition for four hours after purchase but the microbial counts still complied with the microbiological criterion as specified in the Guidelines. Rice balls have potential food safety risks, as they are subject to handling after cooking and may be kept under ambient condition for a prolonged period. In general, the 4-hour rule for perishable ready-to-eat food, is also applicable to packaged rice balls. Proper time and temperature control should be implemented to ensure food safety.

Modification of rice ball ingredients could allow the display of temperature above 4ºC for a certain period of time, but any control measure, such as product formulation, should be validated to ensure they are sufficient to safeguard food safety.

41. Advice for trade and the public about the safe handling of packaged rice balls are provided as follows:

Advice to Trade

GHPs for preventing contamination in food and proper storage of food, particularly ready-to-eat food, should be followed at all times. Food businesses also have the responsibility to provide sufficient ongoing food safety/hygiene training to their staff (i.e. food handlers in their businesses). Furthermore, a preventive food safety management system (such as the Hazard Analysis and Critical Control Point (HACCP) system) should be established to ensure that effective control measures are in place to minimise potential contamination of the products during the manufacturing process. Some GHPs are highlighted below:

- Food handlers should receive proper training to prepare rice balls properly and prevent contamination of rice balls. The most important points are: wash hands with correct hand washing method and change disposable gloves properly.
- To facilitate proper and efficient hand washing, food establishments should provide the following items at the washing basin for their staff: liquid soap, tap with sensor or non-touch design (e.g. operated with foot or elbow), a continuous supply of clean running water (warm water can encourage hand washing in cold weathers), disposable tissues for drying hands and rubbish bin with a hands-free lid.
- The preparation areas for raw food should be separated from areas for handling ready-to-eat food, as far as possible. If raw, cooked and ready-to-

eat ingredients need to be handled in the same preparation area, disinfect the area thoroughly between uses.

- The schedule or sequence of rice ball preparation should be planned to avoid leaving perishable/ready-to-eat ingredients under ambient conditions for too long.
- For packaged ready-to-eat ingredients, where appropriate, wipe clean and dry the packaging before opening to minimise potential cross-contamination.
- Keep only the required amount of fillings / ingredients on the counter for the preparation of rice balls. Fillings for rice balls should be covered and stored at 4°C or below.

Storage and Display

- In general, rice balls intended to be display for a prolonged period should be refrigerated after preparation.
- If food businesses choose to display rice balls at ambient temperature, they should keep the rice balls for no longer than 4 hours, taking into account all durations during transport, storage or preparation of rice balls between 4°C and 60°C.
- Control measures for limiting microbial growth so that the rice balls could be kept at temperatures above 4ºC for a certain period of time, should be validated to see if they are sufficient to control relevant hazards.
- Recommendations on storage conditions should be provided for the safe handling of packaged rice balls by consumers.

<u>Advice to Public</u>

- Consume rice balls as soon as possible after purchase.
- For packaged rice balls, follow the storage instructions on the packaging carefully and consume them before the use-by date.
- Refrigerate packaged rice ball at 4°C or below if they are not to be consumed immediately.

REFERENCES

[1] Institute of Medicine (US) Committee on Strategies to Reduce Sodium Intake; Henney JE, Taylor CL, Boon CS, editors. Strategies to Reduce Sodium Intake in the United States. Washington (DC): National Academies Press (US); 2010. Chapter 4, Preservation and Physical Property Roles of Sodium in Foods. Available from: URL: https://www.ncbi.nlm.nih.gov/books/NBK50952/ [Accessed 24 May 2024]

[2] Tsutsuura S., Hayashida N., and M. Murata, 2018. Effect of Glycine on Production of Staphylococcal Enterotoxin A in Cooked Rice, and Limitation of Its Organoleptic Detection. Journal of Home Economics of Japan. 69:12:799-810. Available from: URL: https://www.jstage.jst.go.jp/article/jhej/69/12/69_799/_pdf/-char/ja [Accessed 24 May 2024]

[3] Food and Environmental Hygiene Department, 2021. Food Hygiene Code. Chapter 4 Safe Food Handling. Available from: URL: https://www.fehd.gov.hk/english/publications/code/code_all.pdf [Accessed 24 May 2024]

[4] Ngan F, 2019. 為何在日本買的飯糰比香港好吃？告訴你 8 個飯糰小秘密. Available from: URL: https://www.cosmopolitan.com.hk/lifestyle/japanese-rice-onigiri-secrets [Accessed 24 May 2024]

[5] NSW Department of Primary Industries, 2017. Rice based desserts survey. Available from: URL: https://www.foodauthority.nsw.gov.au/about-us/science/market-analysis/rice-based-desserts [Accessed 24 May 2024]

[6] TOpick，2019。飯糰室溫存放含菌量比拼　理大教授：1 小時內食用減食物中毒風險（2019 年 8 月 1 日）。https://topick.hket.com/article/2178011/%E3%80%90%E7%90%86%E5%A4%A7%E5%AF%A6%E6%B8%AC%E3%80%91%E9%A3%AF%E7%B3%B0%E5%AE%A4%E6%BA%AB%E5%AD%98%E6%94%BE%E5%90%AB%E8%8F%8C%E9%87%8F%E6%AF%94%E6%8B%BC%E3%80%80%E7%90%86%E5%A4%A7%E6%95%99%E6%8E%88%EF%BC%9A1%E5%B0%8F%E6%99%82%E5%85%A7%E9%A3%9F%E7%94%A8%E6%B8%9B%E9%A3%9F%E7%89%A9%E4%B8%AD%E6%AF%92%E9%A2%A8%E9%9A%AA ，瀏覽日期：2024 年 5 月 24 日。

[7] New Zealand Food Safety, 2021. Risk Profile: Bacillus cereus in rice and starchy foods. Available from: URL: https://www.mpi.govt.nz/dmsdocument/44125-Risk-Profile-Bacillus-cereus-in-rice-and-starchy-foods [Accessed 24 May 2024]

[8] Schreiber N., Hackl G., Reisinger A.C., Zollner-Schwetz I., Eller K., Schlagenhaufen C., Pietzka A., Czerwenka C., Stark T.D., Kranzler M., Fickert P., Eller P., and M. Ehling-Schulz M, 2021. Acute Liver Failure after Ingestion of Fried Rice Balls: A Case Series of Bacillus cereus Food Poisonings. Toxins (Basel). 23;14(1):12. Available from: URL: https://pubmed.ncbi.nlm.nih.gov/35050989/ [Accessed 24 May 2024]

[9] CFS, 2014. Microbiological Guidelines for Food. Available from: URL: http://www.cfs.gov.hk/english/food_leg/files/food_leg_Microbiological_Guidelines_for_Food_e.pdf [Accessed 24 May 2024]

[10] Institute of Medicine (US) Committee on Strategies to Reduce Sodium Intake, 2010; Henney J.E., Taylor C.L., Boon C.S., editors. Strategies to Reduce Sodium Intake in the United States. Washington (DC): National Academies Press (US); 2010. 4, Preservation and Physical Property Roles of Sodium in Foods. Available from: URL: https://www.ncbi.nlm.nih.gov/books/NBK50952/ [Accessed 24 May 2024]

[11] Codex, 2008. Guidelines for the Validation of Food Safety Control Measures (CXG 69-2008). Available from: URL: https://www.fao.org/fao-who-codexalimentarius/sh-proxy/en/?lnk=1&url=https%253A%252F%252Fworkspace.fao.org%252Fsites%252Fcodex%252FStandards%252FCXG%2B69-2008%252FCXG_069e.pdf [Accessed 24 May 2024]