Microbiological safety of ready-to-eat foods in Republic of Palau

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Abstract:
Microbiological quality of 45 ready-to-eat foods in Palau was investigated. About half (23 foods) were categorized as "unsatisfactory" quality with the results of aerobic bacteria counts, while 4% (2 foods) with the E. coli counts. Proportion of "unsatisfactory" quality foods did not vary by food type. Inappropriate storage conditions may be the factor that allowed bacterial growth, which need to be considered in future policy of food administration in Palau.
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Introduction

Ready-to-eat foods, such as rice ball (musubi), rice rolled in laver (sushi), packed lunch and sandwiches, comprises important part of Palauan diet. The ready-to-eat foods are usually prepared by individual caterers (mostly house wives) in the morning and consigned to retail stores for sale for lunch. Because the foods are usually displayed in room temperature, time length of display in retail stores may be essential factors that determine the microbiological safety. Despite the potential health risk of bacterial contamination, microbiological assessment of the ready-to-eat foods has not been conducted in Palau. In the present paper, we will report the preliminary findings for the microbiological quality of ready-to-eat foods sold in Palau.

Subjects and methods

The principle goal of the present study was to investigate the bacterial safety level of the ready-to-eat foods in the conditions when they were usually eaten by the people. The foods were purchased in 16 representative stores/markets in Koror (the capital city) between 11:00 and 12:00 AM during the period between 25 August and 1st September 2004. They were immediately brought back to the laboratory, and analyzed between 12:00 AM and 13:00 PM. Totally 45 ready-to-eat foods were investigated, of which 22 were sold in the shops with air condition (average temperature 28.1 Celsius degree) and 23 in those without (average temperature 32.1 Celsius degree). Ready-to-eat foods analyzed in the present study included packed lunch (N=9), fast foods/snacks (N=18), tubers (N=6), sea foods (N=10), and others (N=6).

In the stores or markets, a thermometer was placed on the food shelf for 5 minutes for recording the room temperature. Then, the information sheet was filled with the name of food, ingredients, operation of air condition machine, whether the foods were placed in refrigerator or not, time cooked, time purchased, name of shop, name of caterer, and residential place (name of State) of caterer. Food samples were photographed.

Petrifilm aerobic count plate (AC plate) and Petrifilm E. coli/coliform count plate (EC plate) were used for the assessment of bacterial contamination. Petrifilm methods are simple and accurate alternatives to the Association of Official Analytical Chemists' (AOAC)

standard enumeration methods in a wide range of foods [1] [2], and usually suitable for use in the countries with insufficient laboratory equipments.

In the laboratory, 5 g of each food item was sampled and homogenized with 45ml of dilution water (10 times dilution sample). Then the sample was successively diluted to 10000. On AC plate, 1 ml of 1000 and 10000 samples were dropped, while 1 ml of 10 and 100 samples were dropped on EC plate. The AC plates were incubated for 48 hours and EC plates for 24 hours at 35 Celsius degree.

Table 1. PHLS guidelines for the microbiological quality (on the basis of aerobic colony count) for ready-to-eat foods in Palau

Food category	Microbiological quality (CFU per gram)		
	Satisfactory	Acceptable	Unsatisfactory
1	$<10^3$	10^3-$<10^4$	10^4_
2	$<10^4$	10^4-$<10^5$	10^5_
3	$<10^5$	10^5-$<10^6$	10^6_
4	$<10^6$	10^6-$<10^7$	10^7_

Source: Gilbert et al. (2000), Table 1. Food category (1) was applied to beef burger, and raw sea foods; (2) Packed lunch, hot dog, sushi, musubi (rice ball), tubers, salad, soup, and cooked vegetables; (3) cooked sea foods; (4) sandwiches, and smoked fish. See Table 2 for the criteria for each ready-to-eat-food investigated in the present study.

After the incubation, on EC plate, colonies of E coli (blue colonies with gas) and those of coliform except E coli (red colonies with gas) were counted separately (EC counts and CC counts, respectively). On AC plate, red colonies were counted (AC counts). In case that more than 300 colonies existed on plate, we counted the colonies in randomly selected two meshes (2 cm^2) and multiplied by 10 (total area of plate is 20 cm^2). If more than 500 colonies existed on plate or colonies overlapped each other, it was judged as "too numerous to count (TNTC)". Per gram bacteria counts in the original food items was calculated.

Food handlers in Palau undergo physical examination every year and are issued health certificate from physician.

The association of bacterial counts with air conditioning or residential State of caterer (Koror or others) was investigated for the ready-to-eat foods displayed on food shelf with t-test. The correlations between bacterial counts and time of cooking, duration from cooking to purchasing, or indoor temperature of the shops were examined with Spearman correlation coefficient.

Microbiological quality of ready-to-eat foods was evaluated on the basis of Microbiological Guidelines for ready-to-eat foods sampled at the point-of-sale by a working group of the PHLS Advisory Committee for Food and Dairy Products [3]. The PHLS Guidelines graded that "unsatisfactory" as that test results indicating that further sampling may be necessary and that environmental health officer may wish to undertake a further inspection of the premises concerned to determine whether hygiene practices for food production or handling are adequate or not, while "satisfactory" indicates the good microbiological quality. "Acceptable" reflects a borderline limit of microbiological quality. For aerobic colony count, five different criteria were set depending on the type of foods (Table 1).

Results

Table 2 shows the results of AC counts, EC counts, and CC counts for 45 ready-to-eat foods in Republic of Palau. Major ingredients for each food are also listed. Of 45 samples, AC counts of 15 foods exceeded 5,000,000 (judged as "too numerous to counts" on the 10000 dilution sample). EC counts exceeded 50,000 in one Musubi sample and 1,000 in one sushi sample. CC counts showed the similar pattern with AC counts. Air conditioning, residential State of caterer, time of cooking, duration from cooking to purchasing, and indoor temperature of the shops did not explain the variation in bacterial counts (results not shown).

Table 3 shows the categorization of microbiological quality on the basis of PHLS guidelines. Of 45 ready-to-eat foods, 23 (51%) were categorized as "unsatisfactory" quality with the results of AC counts, while 2 (4%) were categorized as "unsatisfactory" with the results of EC counts. Percentage of "unsatisfactory" quality foods did not vary by food type.

Discussion

Of the 45 ready-to-eat foods surveyed in Palau, about half were judged as "unsatisfactory" bacterial quality with AC counts, the indicator of contamination by general bacteria flora, while only two were "unsatisfactory" quality with EC counts, the indicator of fecal origin bacteria contamination. The reasons of poor bacterial quality in half of the ready-to-eat foods may be either the foods were contaminated in the process of

Table 2. Microbiological results for 45 ready-to-eat foods in Republic of Palau

Category	Name in Palau	Major ingredients	Aerobic Colony Count	*E.coli* Colony Count	*Coliform* Colony Count	PHLS Guidelines[1]
Packed lunch						
	Bento	Rice, fried fish, pickles	212,000	0	210	2
	Bento	Rice, chicken, rice, potato croquette, spring rolls	710,000	0	0	2
	Bento	Rice, fried fish, fried chicken, carrot salad, cabbage	5,000,000+	50	50,000+	2
	Bento	Rice, egg, water spinach, chicken	5,000,000+	0	50,000+	2
	Bentou	Rice, chicken, bamboo shoot, *konnyaku* , mushroom, onion	0	0	0	2
	Bentou	Rice, chicken, beef,	2,860,000	0	50,000+	2
	Bentou	Rice, chicken, Spam, egg	5,000,000+	0	50,000+	2
	Fried rice	Rice, egg, spam, carrot, peas	4,000	0	10	2
	Steamed rice	Rice	9,000	0	610	2
Fast Foods/snack						
	Beef burger	Bread, beef, lettuce	10,000	0	0	1
	Beef burger	Bread, beef, lettuce	340,000	0	300	1
	Egg roll	Egg, fried rice	1,170,000	0	230	2
	Hot dog	Bread, sausage	2,000	0	0	2
	Maki-sushi	Rice, laver, cucumber, egg, Spam	5,000,000+	1,100	50,000+	2
	Maki-sushi	Rice, laver, tuna, pickled radish, Spam	5,000,000+	0	360	2
	Musubi	Rice, laver, Spam, egg	18,000	0	110	2
	Musubi	Rice, laver, Spam, egg	356,000	0	1,010	2
	Musubi	Rice, laver, Spam, egg	5,000,000+	50,000+	50,000+	2
	Musubi	Rice, laver, Spam, egg	5,000,000+	0	10,400	2
	Sandwiches	Bread, egg, mayonnaise	340,000	0	10,600	4
	Sandwiches	Bread, cheese, bread, ham, mayonnaise, lettuce	5,000,000+	0	0	4
	Sandwiches	Bread, tuna, egg, mayonnaise	5,000,000+	0	50,000+	4
	Sandwiches	Bread, egg, mayonnaise	5,000,000+	0	2,400	4
Tubers						
	Belsiich	*Kukau* -taro, salad oil	620,000	0	45,200	2
	Billum	Cassava, coconut milk, sugar	0	0	0	2
	Elbis diokeang	Cassava, coconut milk, sugar	5,000,000+	0	2,400	2
	Sweet potato	Sweet potato	48,000	0	560	2
	Taro	Taro	23,000	0	0	2
	Telledou	Taro, coconut	790,000	0	5,500	2
Sea foods						
	Eremrun	Fresh of sea cucumber (raw)	5,000,000+	0	0	1
	Fried fish	Tuna, flour	0	0	0	3
	Fried fish	Tuna, flour	1,000	0	0	3
	Giant Clam chowder	Giant clam, coconut milk, bean, leafy vegetables	0	0	0	3
	Irimd	Fresh of sea cucumber (raw)	5,000,000+	30	9,900	1
	Molech	Fresh of sea cucumber (raw)	112,000	0	70	1
	Ngims	Intestine of sea cucumber (raw)	14,000	10	430	1
	Sashimi	Sliced raw fish (tuna), lemon	88,000	20	270	1
	Smoked fish	Tuna	0	0	0	4
	Smoked fish	Tuna	0	0	0	4
Others						
	Macaroni Salad	Macaroni, mayonnaise, Spam	530,000	0	1,100	2
	Demok	Taro leaf, coconut milk, tin-mackerel, onion, salt	0	0	30	2
	Osiruko	Brown bean, rice powder, sugar	4,000	0	20	2
	Potato Salad	Potato, Egg	9,000	0	0	2
	Potato salad	Potato, egg, carrot, corn, peas, mayonnaise	43,000	0	2,280	2
	Pumpkin soup	Pumpkin, rice, coconut milk, sugar	0	0	0	2

1 PHLS Guidelines have been used for the standardized evaluation of microbiological quality of ready-to-eat foods in UK. The guideline categorizes the microbiological quality of ready-to-eat foods into "satisfactory", "acceptable", or "unsatisfactory" on the basis of Aerobic Colony Count (ACC), count of indicator organisms, or counts of pathogens. For the evaluation of ACC, different cut-off-points were assumed for different food category. See Table 1 for the criteria in each category.

Table 3. Microbiological quality of 45 ready-to-eat foods evaluated on the basis of PHLS guidelines

Food type	Aerobic Colony Count			*E.coli* Colony Count		
	Satisfactory	Acceptable	Unsatisfactory[1]	Satisfactory	Acceptable	Unsatisfactory[1]
Packed lunch	4	1	6 (55%)	10	1	0
Fast Foods/snack	2	4	8 (57%)	12	0	2 (14%)
Tubers	1	2	3 (50%)	6	0	0
Sea foods	4	0	5 (56%)	7	2	0
Other foods	4	0	1 (20%)	5	0	0
Total	15	7	23 (51%)	40	3	2 (4%)

[1] Percentage of "unsatisfactory samples" in each food category are given in parentheses.

cooking/packing or the bacterial grew during the period for which the foods were displayed in shops, or both.

Regarding the possibility of contamination during the process of cooking/packing, the study of street foods in 13 towns in different continents may be suggestive because that showed that the lack of training for venders increased the risk of unsatisfactory biological quality in ice creams or sorbets [4]. Yet it does not seem to be the case in Palau. Division of Environmental Health in Palau government has already started several measures for the food safety issues. They included Food Safety training workshop, registration of individual caterers and obligation for caterers to label foods with the name of caterer, food handler permit number and time of cooking. Food handlers in Palau undergo physical examination every year and are issued health certificate from physician. Food Safety Unit of the Division monitors and inspects household kitchens before health permit is issued. These conditions are contrasting with "street-foods" universally sold by individual venders in the world that have frequently been regarded as potentially hazardous for human health[5 6 7] because of the absence of surveillance of vender's health, their lack of appreciation of basic food safety issues, poor conditions of food preparation in the absence of food control, and improper storage conditions (time-temperature) [4].

Thus, it will be reasonable to judge that the contamination of ready-to-eat foods in Palau was mainly due to the inappropriate storage during display in shops. Refrigerator was rarely used for the display of ready-to-eat foods except for raw sea foods. The foods were displayed on the shelf in room temperature (average=30, minimum=27, maximum=37 Celsius degree) from 6:00-7:00 AM (time of cooking) until time of purchasing (11:00-12:00 AM), which allowed the exponential growth of bacterial for 4 to 6 hours. Variation in room temperature or duration for which the foods were displayed in shops did not explain the variation in AC counts in the present study probably because statistical power may be lacking with small sample size or the conditions were relatively homogenous among the stores or markets for the aspect of bacterial growth.

Improvement of storage conditions by introducing refrigerator may be one ideal measure for the food safety problems in Palau. Yet it can increase the economic burden of the venders and, eventually, the individual caterers. Discussion among different sectors of the government as well as individual shops/markets and caterers is necessary for the practical and effective decisions for food safety issues. Also, bacterial investigation of each process of cooking/packing as well as that for swab samples for every part in kitchen will help us to confirm whether all individual caters are really following the instructions given in their training. Longitudinal and continuous accumulation of monitoring data on biological quality of ready-to-eat foods will be of importance as the scientific evidences to be used for policy-making not only in Palau but also in other Micronesia nations that share similar needs in food administration.

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He who smiles rather than rages is always the stronger
(Japanese Wisdom)