Microbiological Investigation of Fresh Edible Raw Sea Urchin and its Expiration Date

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(Received February 5, 2007; Accepted May 18, 2007; Published online May 23, 2007)

Since no definitive standard exists for setting the expiration date of edible raw sea urchins, it is up to the importer or domestic processing manufacturer to assign an expiration date. In the present study, we performed a microbiological examination of 126 raw sea urchin samples collected from distributors at Tsukiji Market (Tokyo Metropolitan Central Wholesale Market) and evaluated their expiration dates. The expiration dates were set at 5 to 8 days after processing in approximately 80% of the products; however, microbiological examination of the samples indicated that the dates were not sufficiently accurate. Of the products, 27% did not meet the microbiological standards for freshness established by the Tokyo metropolitan government during the period preceding their expiration dates. Our results suggest that the expiration date of edible fresh raw sea urchins should be based on a microbiological examination.

Key words —— sea urchin, expiration date, microbiological examination

INTRODUCTION

Raw sea urchins (uni in Japanese) are often contaminated with *Vibrio parahaemolyticus*, which is responsible for numerous cases of food poisoning in Japan. In response, the Food Sanitation Law (Shokuhin-eisei-hou in Japanese) was revised in 2001,[1)] and a preservation standard for perishable seafood was established. In addition, beginning in 2002, it became mandatory to display an expiration date;[2)] however, since expiration dates are set by the importer or domestic processing manufacturer alone, a standard method for determining the expiration date of raw sea urchins has not yet been established.

In the Tokyo area, raw sea urchins are marketed by wholesalers through distributors at Tsukiji Market (Tokyo Metropolitan Central Wholesale Market) from the initial processing stage (Fig. 1), and are subsequently distributed to consumers via retailers.

In the present study, we investigated the expiration date and viable bacterial cell count of raw sea urchins to evaluate and improve the information being provided to traders at Tsukiji Market (The Japanese terms Shohi-kigen and Shomi-kigen are not distinguished in this article. Both are expressed as "the expiration date.").

MATERIALS AND METHODS

Sample Collection —— One hundred and twenty-six raw sea urchins were collected from a distributor in Tsukiji Market between April 2005 and February 2006, including 47 from the United States, 30 from China, 14 from North Korea, 13 from Chile, 8 from Russia, 7 from Japan, 4 from Canada, and 3 from Mexico.

Viable Bacterial Cell Count —— The numbers of general viable bacteria and coliform bacteria in the raw sea urchins were counted by the "standard determination assay for viable cell count on a plate"[3)]

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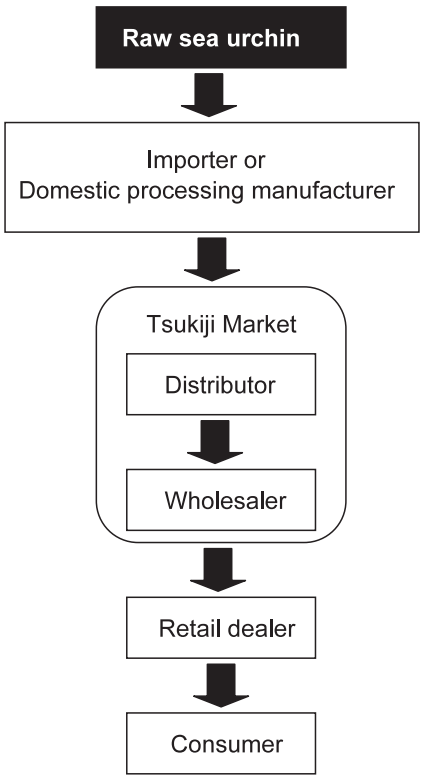

Fig. 1. Circulation Process of Raw Sea Urchins

on days 0, 5, 9, and 14 after collection. The samples were also incubated at 7 + 1°C for 10 days to detect the presence of psychrotrophic bacteria.

Evaluation Criteria —— We evaluated the food quality of the sea urchins in accordance with the "Standard of the Tokyo Metropolitan Government."[4] A sample was judged to be "inappropriate for food" when greater than 10^6/g general viable bacteria or 3×10^3/g coliform bacteria were detected, or if fungi were visually detected.

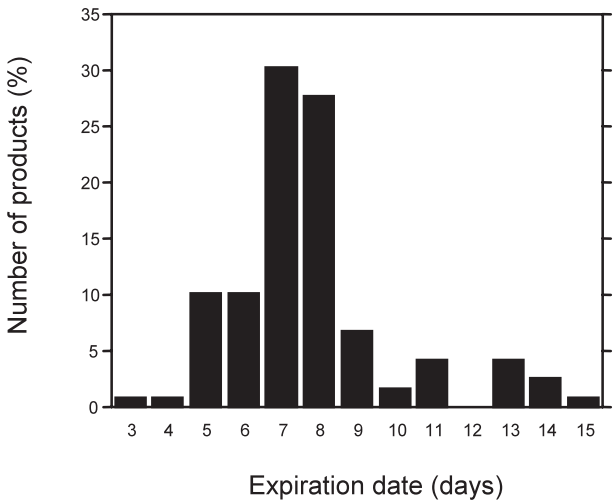

Fig. 2. The Expiration Dates Displayed on the Products

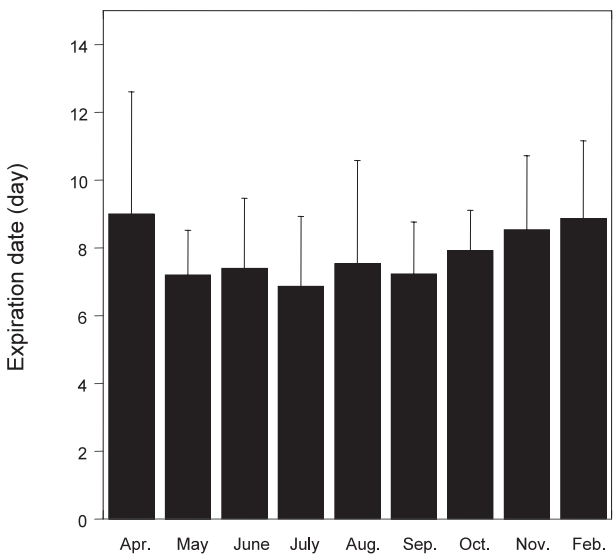

Fig. 3. The Expiration Dates of Products Collected During Different Months
The data show mean + standard deviation.

RESULTS

Expiration Dates Displayed on the Products

The expiration dates were set from 3 to 15 days after processing, with approximately 80% of the samples set 5 to 8 days after processing (Fig. 2). The relationship between the month a sample was collected and its expiration date is shown in Fig. 3. There was no correlation between the expiration date and the month a sample was collected.

Viable Bacterial Cell Count

The number of general viable bacteria on days 0, 5, 9 and 14 after collection increased 3033 + 180 (mean + standard deviation), 14421 + 615, 39246 + 1062, and 169902 + 1260, respectively.

The numbers of psychrotrophic bacteria and general viable bacteria at day 0 in each month are shown in Fig. 4. There was no correlation between the number of psychrotrophic bacteria or general viable bacteria and the month a sample was collected.

The Expiration Date and Viable Bacterial Cell Count

The number of products that had become substandard, as judged by a general viable bacteria count of more than 10^6/g or a coliform group count

Table 1. The Number of Products with General Viable Bacteria Present on the Day of Collection and Their Expiration Dates

The number of products with general viable bacteria present on the day of collection	Average expiration dates displayed on the products	The percentage of products that became inferior prior to their expiration date	The percentage of products that became inferior within 5, 9, and 14 days		
			5 days	9 days	14 days
$< 10^2$/g (34)[a)]	8.1 ± 2.0[b)]	0.0	0.0	0.0	2.9
$< 10^3$/g (24)	8.4 ± 2.6	4.2	0.0	0.0	12.5
$< 10^4$/g (28)	7.6 ± 2.3	10.7	10.7	17.9	39.3
$< 10^5$/g (24)	7.1 ± 1.6	45.8	37.5	45.8	79.2
$< 10^6$/g (14)	7.1 ± 1.9	92.9	92.9	92.9	100.0

a) number of products. *b*) mean ± standard deviation. Two products which exceeded 10^6/g were excluded from Table 1.

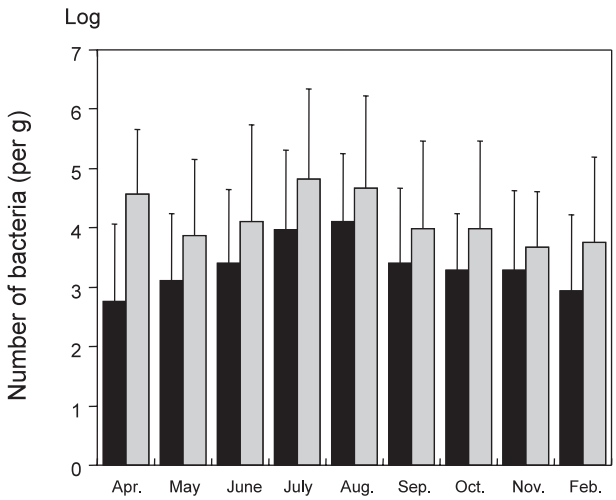

Fig. 4. Numbers of General Viable Bacteria and Psychrotrophic Bacteria Detected During Different Months

Closed square, general viable bacteria; dotted square, psychrotrophic bacteria. The data show mean + standard deviation.

of more than 10^3/g, on the day of collection was 8 of 126, or 6.3%. The number of products judged to be inferior prior to their expiration date was 34 of 126, or 27.0%. Of those 34 products, 32 (94.1%) exceeded the standard values for general viable bacteria or coliform bacteria within 5 days. The detection rate of coliform bacteria was low (34/126, or 27.0%); only 8 of the 126 products contained more than 10^3/g.

The number of general viable bacteria present on the day of collection (day 0) and on the expiration date was further analyzed (Table 1). For those products with less than 10^2/g general bacteria on the day of collection (day 0), none became substandard within the period preceding the expiration date set by the importer or manufacturer. On day 14, only 1 of 34 products (2.9%) exceeded 10^6/g. For the 14 products with 10^5 to 10^6/g general bacteria, 13 (92.9%) became inferior prior to their expiration date. By day 5, 13 of the 14 products were judged to be inferior goods. As the number of bacteria present on the day of collection increased, so did the frequency of inferior goods.

DISCUSSION

In food microbiology, bacteria that can grow under general refrigeration are called psychrophilic bacteria. Most Gram-negative psychrophilic bacteria are associated with rotting or with unpleasant odors from food products. Given that psychrophilic bacteria were also detected with general viable bacteria in raw sea urchins, we postulated that psychrophilic bacteria influence the quality degradation of foods such as fresh fish and shellfish, which are circulated or stored at low temperatures. Current guidelines do not include a standard for the acceptable number of psychrophilic bacteria in raw sea urchins; however, a quality standard should be set. Coliform bacteria were rarely detected in the sea urchins (8/126), indicating good hygiene management. Of the products that became substandard prior to their expiration date, most (32/34, or 94.1%) did so within 5 days. The Food Sanitation Law (Sekoukisoku in Japanese) states that the expiration date of foods that rapidly and easily decrease in quality should be set within 5 days of processing.[5)] Based on our results, this requirement is reasonable for fresh sea urchins from a legal and hygienic standpoint. The number of viable bacteria present on the day of collection should also be considered when setting expiration dates. As the initial number of bacteria increased, so did the frequency of foods becoming inferior prior to their expiration date (Table 1); therefore, by examining the number

of viable bacteria on the day of collection, reasonable expiration dates for sea urchins can be determined.

Setting a 5-day expiration date may be inconvenient for distributors or retailers, considering the subsequent distribution period (*i.e.*, from landing to auction sale in the market); however, it is necessary to provide the products to consumers quickly, without a circulation delay.

In conclusion, we identified a relationship between the expiration date of raw sea urchins and the results of a bacteriological examination. These data will assist manufacturers and distributors in establishing reasonable expiration dates for their products.

REFERENCES

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3) Japan Food Hygiene Association (2004) *Guideline for Food Hygienic Examination* (*Shokuhin-eisei-kensa-shishin in Japanese*), pp.116–145, Tokyo.
4) Standard-based collection examination of the Tokyo Metropolitan Wholesale Market Sanitation Inspection Station (1 October 1998).
5) *The enforcement regulation* (*Sekoukisoku in Japanese*) *of the Food Sanitation Law*, Law number 23 (2006, revised), Tokyo.