Effect of Storage at 5°C on the Bacterial Flora and Physical Properties of Malaysian Fishballs

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Abstract

Fishballs from six local factories were stored at 5°C for 1, 2, 3, 4, 7 and 10 d. There were no changes in texture at 3 d of storage but bacterial spoilage rendered the fishballs unacceptable by the fourth day. Most of the bacteria were non-halophilic. The main genera isolated were *Aerococcus, Acinetobacter, Pseudomonas, Staphylococcus, Corynebacterium, Micrococcus, Streptococcus* and *Enterobacteria.*

Introduction

Fishballs are a popular food in much of the Asian region. Made by mixing fish mince with starch, salt, sugar, monosodium glutamate and spices into a ball-shaped dough, they are cooked to a gel which is consumed with rice, noodles or as a snack. They are usually sold in local wet markets, unchilled and with little or no packaging. Being a high moisture food (>80%), they are highly perishable with a short shelf-life. Thus, data on their spoilage characteristics is necessary before monitoring and control systems can be implemented.

This study reports the microbial and physical changes occurring in fishballs from six Malaysian factories during storage at 5°C.

Materials and Methods

Raw Materials

Freshly produced fishballs were purchased from the six largest factories in Kuala Lumpur. The samples were packed in aseptic plastic bags and transported immediately to the laboratory. Upon arrival at the laboratory, the fishballs were transferred using aseptic forceps into sterile plastic bags (200 g per bag), sealed in a laminar-flow chamber, and stored at 5°C. Samples were analyzed at 0, 1, 2, 3, 4, 7 and 10 d of storage. Two samples from each factory were tested for each parameter on each day except for the folding test, when five samples were tested.

Physical Tests

The Folding and Teeth-Cutting Tests of Hasegawa (1987) were used. The Folding Test measures resilience, and the Teeth-Cutting Test measures springiness of the fishballs. Samples were boiled for 5 minutes and then cooled to room temperature (28°C) before testing.

FOLDING TEST

Five slices, each 5 mm thick and 20 mm in diameter, were cut from five fishballs. Each was then folded in half and, if there was no tear or breakage, further folded into quarters. The grading was as follows: AA, no breakage in any of five samples when folded in quarters; A, slight tear in any one of five samples when folded in quarters; B, slight tear in any one of five samples when folded in half; C, breakage (but two pieces still connected) when folded in half; D, breaks completely into two pieces when folded in half.

TEETH-CUTTING TEST

This test gives a subjective assessment of the resistance experienced by a trained panel of 10 when the test piece is bitten between the upper and lower incisors. Two slices of 5 mm in thickness and 20 mm diameter were tested. Scores were attributed as follows: 10, extremely strong springiness; 9, very strong springiness; 8, strong springiness; 7, quite strong springiness; 6, acceptable springiness; 5, acceptable, slight springiness; 4, weak springiness; 3, quite weak springiness; 2, very weak springiness; 1, mushy texture, no springiness.

pH Measurement

A 10 g sample was homogenized in 90 ml distilled water and the pH measured using a combination electrode with an Ag/Ag Cl reference system (Model HI 1911B, Hanna Instruments SpA. Padova, Italy).

Microbial Analyses

AEROBIC PLATE COUNT

Whole fishballs were ground in a sterile Waring blender flask, using two samples from each factory. The method as described by APHA (1976) was used. Plates containing 25-250 colonies were counted after incubation at 35°C for 48 h, 20°C for 96 h and 5°C for 144 h.

HALOPHILIC AND NON-HALOPHILIC BACTERIA

Samples from the plate count were inoculated onto nutrient agar containing 0, 5, 10 and 15% NaCl and incubated at 5°C; 10 and 15% NaCl and incubated at 5°C, for 48 and 24 h, respectively. Results were obtained by counting the number of colonies on the media.

GENERIC CHARACTERIZATION

Colonies from the plate count and nutrient agar were isolated and identified using the Primary Characterization Tests of Cowan (1975).

Results

Physical Changes in Fishballs During Storage

No significant changes in the pH and the resilience measured by a Folding Test were detected after 10 d of storage at 5°C (Table 1). All samples were graded to AA. There were no changes in the teeth-cutting score, after 3 d. However, in terms of appearance and odor, the fishballs became unacceptable on the fourth day of storage. An unpleasant odor associated with spoilage was detected and the surface of the fishballs were covered with slime. The presence of a turbid and viscous exudate was also noticed.

Table 1. Physical changes of fishballs during storage at 5°C*.

Day	pH	Teeth Cutting Test	Appearance/Odor
0	7.23 (±0.09)	8(±0.7)	Fresh, shiny and glossy, smooth. Fishy.
1	7.27 (±0.12)	8(±0.5)	Fresh, shiny and glossy, smooth. Fishy.
2	7.25 (±0.13)	8(±0.4)	Slight exudate, a bit milky, less glossy and shiny. Fishy, slightly pungent odor.
3	7.24 (±0.08)	8(±0.5)	Slight milky exudate, glossy and wet. Stronger fishy odor.
4	7.22 (±0.11)	-	Slimy, milky and viscous exudate, glossy and wet. Slightly unpleasant odor.
7	7.18 (±0.10)	-	Slimy. Yellow-milky exudate. Patches of yellow on fishballs. Unpleasant odor.
10	7.30 (±0.08)	-	Slimy. Yellow-milky exudate. Very unpleasant odor.

* Average values of samples from six factories.
  Numbers in brackets are standard deviations of the mean.

Effect of Storage on Bacterial Flora

The average number of mesophiles (measured at 35°C) after 1 d storage (Table 2) would be considered unsafe for human consumption if they were consumed without re-cooking. By day four the mesophile numbers reached an unacceptable level. The number of psychrotrophs (measured at 5°C), at day four, also indicated spoilage of the product.

No halophilic psychrotrophs and few halophilic mesophiles were detected (Table 3) indicating that most of the spoilage organisms were of land origin. The non-halophilic psychrotrophs increased with time, while the non-halophilic mesophiles remained virtually constant. Initially, there were twice as many mesophiles as psychrotrophs, but by day three they were approximately equal and thereafter the psychrotrophs predominated.

Table 2. Aerobic plate counts (organisms g^{-1} sample) of fishballs during storage at 5°C*.

Day	Aerobic plate count (APC)·g^{-1} sample	
	5°C	35°C
0	2.2 x 10^4 (±1.0)	2.6 x 10^5 (±0.9)
1	7.5 x 10^5 (±2.1)	5.4 x 10^6 (±1.8)
2	2.6 x 10^6 (±1.7)	6.6 x 10^6 (±2.0)
3	2.2 x 10^7 (±1.5)	3.6 x 10^7 (±1.7)
4	8.1 x 10^7 (±2.7)	1.0 x 10^8 (±1.5)
7	1.1 x 10^8 (±2.3)	2.0 x 10^8 (±1.8)
10	1.2 x 10^8 (±1.5)	2.2 x 10^8 (±2.2)

* Average values of samples from six factories. Numbers in brackets are standard deviations of the mean.

Table 3. Halophilic and non-halophilic bacteria during storage of fishballs at 5°C.

Days of storage	Number of colonies			
	Halophilic		Non-halophilic	
	Psychro. (5°C)	Meso (35°C)	Psychro. (5°C)	Meso (35°C)
0	-	1	8	14
1	-	-	9	19
2	-	-	11	21
3	-	1	19	18
4	-	-	21	17
7	-	1	23	16
10	-	-	22	15
Total %	0	3 (1.3%)	113 (47.9%)	120 (50.8%)

* Average values of samples from six factories.

Generic Characterization Tests

Results of the characterization tests are shown in Table 4. The eight dominant species isolated were classified under the terms of genus *Aerococcus, Acinetobacter, Pseudomonas, Staphylococcus, Corynebacterium, Micrococcus, Streptococcus* and *Enterobacteria*.

Discussion

Fishballs produced in Malaysia have a high initial bacterial contamination and although there were no changes in texture, bacterial spoilage rendered the fishballs unacceptable by the fourth day of storage. Most of the fishballs are produced by cottage-scale industries dependent upon manual labor and using a minimum of equipment. The short shelf-life of fishballs may be due to a combination of some or all of the following points: raw materials that are not fresh and are heavily contaminated due to poor handling after catch; slow processing at ambient temperatures (30-35°C) and poor personal hygiene result in further deterioration of raw material; use of dirty processing equipment; cooked products are left to cool in rattan baskets on the floor for long periods; packaging is done manually; packed products are often distributed in non-refrigerated vans and lorries; fishballs are sold at ambient temperatures in the market.

Malaysian law does not permit the use of preservatives in fishballs. Despite this, many processors have been known to use common preservatives such as sodium benzoate to extend the shelf life of their products. This illegal practice is also harmful to consumers. The shelf-life of fishballs might be improved by reducing the initial and subsequent microbial counts through the use of simple refrigerators and more sanitary working conditions.

Table 4. Generic characterization of bacterial flora isolated from fishballs (based on analyses of samples from six factories).

Genera	Shape	Gram stain	Motility	Catalase test	Oxidase test	Oxidative fermentation
Aerococcus	C	+	-	W	-	F
Acinetobacter	C	-	-	+	-	O
Pseudomonas	R	-	+	+	+	O
Staphylococcus	C	+	-	+	-	F
Corynebacterium	R	+	-	+	-	F
Micrococcus	C	+	-	+	-	O
Streptococcus	C	+	+	-	-	F
Enterobacteria	R	-	+	+	-	F

C = Cocci; R = Rod; W = Weak reaction

In view of the high bacterial contamination and short shelf life of fishballs produced in Malaysia, a system of quality control must be implemented to improve product quality. A study leading to control of all factors related to contamination, survival and growth of microorganisms at all stages of fishball processing, known as the Hazard Analysis Critical Control Point (HACCP) approach, is currently the best system available for improving microbiological safety. This study will form the basis for future research in fish processing in Malaysia.

Although many large factories have been set up to produce surimi-based products in recent years, there is still a niche for small-scale processors of fresh fishballs to be sold in the wet markets on the day of production. Consumers should be advised not to store fishballs for more than 3 d and should cook them thoroughly before consumption.

Acknowledgment

References

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