

Microbial Contamination of the Food Materials for Manufacturing Korean Laver Roll (*Kimbab*) and the Effect of Gamma Irradiation

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Abstract

Microbial contamination of ready-to-eat ingredients for *Kimbab* manufacturing and the effect of irradiation to reduce the microbial contamination of the products were investigated. Among 9 food items tested, there were no viable cells in the ham, seasoned and cooked beef, imitation crab leg, fried egg, and seasoned burdoc. Cucumber, surimi gel, and seasoned and blanched spinach were counted at 5.07 ± 0.97 , 3.50 ± 0.14 , and 5.41 ± 0.51 log CFU/g, respectively. Irradiation at 1 kGy reduced the number of microorganism in these ready-to-eat foods to an undetectable level. However, the dried laver showed an 8.83 ± 0.10 log CFU/g and an irradiation at 3 kGy reduced the level to only 7.14 ± 0.23 . Sensory evaluation of the irradiated *Kimbab* prepared from these food materials indicated that the measure of the control of the sensorial quality should be provided before applying an irradiation to the prepared *Kimbab*.

Key words: *Kimbab*, ready-to-eat, microorganism, irradiation

INTRODUCTION

A laver roll (*Kimbab*, steamed rice rolled in dried laver) is a major ready-to-eat food in Korea. Many countries have similar types of ready-to-eat foods even though the materials used are different. *Kimbab* is commonly used in a lunch menu or for picnics, but the recent development of improved distribution systems, enlargement of the catering industry, and the eating-out life style has dramatically increased the demand for this type of prepared ready-to-eat food (1).

However, there is a great potential for *Kimbab* to become contaminated from the different materials used during the manufacturing process of the laver roll (2). In addition, water activity of the product is high, and most importantly it cannot be stored in a refrigerator because of the retrogradation of rice starch (3). Statistics of foodborne disease outbreaks in Korea in 2000 indicated that composite seasoned products, including *Kimbab*, accounted for 24% of the incidences. Meat products and fish or shellfish products were 27.9% and 26.0%, respectively (4). A recent report from the Korea Food and Drug Administration (KFDA) also indicated that the foodborne disease outbreaks in Korea in 2003 included 36 cases attributed to composite seasoned products and the majority of these cases were from *Kimbab* (5). An HACCP model study provided evidence that the major contam-

ination came from the personal hygiene conditions and the major materials added to the *Kimbab* (1). Rice is usually steamed at over 100°C, but because of a high water activity, cross contamination can easily occur and it also has the potential hazard of pathogenic *Bacillus cereus* (6). *Salmonella* is distributed in egg shells and thus a cross-contamination during the frying of eggs was observed (7,8). Raw vegetables and fruits also had a high level of bacterial contamination (9,10). Dried laver is known to be contaminated by pathogens in seawater, air, and cross-contamination from other materials; a microbial load of about a 10^6 of CFU/g of microorganisms was observed (11).

Gamma irradiation technology effectively prevents decay by sterilizing the microorganisms and by improving the safety and shelf-stability of food products without compromising the nutritional or sensory quality (12,13). Recently, the US Center for Disease Control (CDC) estimated that if half of the ground beef, pork, poultry, and processed luncheon meats in the US were irradiated, there would be over 880,000 fewer cases of foodborne illness, 8,500 fewer hospitalizations, 6,660 fewer catastrophic illness, and 352 lives saved every year (14).

The objective of this study was to investigate the microbial contamination of the major food materials in manufacturing *Kimbab* and the effects of an irradiation on the reduction of microbial contamination.

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MATERIALS AND METHODS

Sample preparation

Among the commonly used food materials for manufacturing *Kimbab*, ham, seasoned and blanched spinach, seasoned and cooked beef, cucumber, surimi gel, imitation crab leg, and seasoned burdock were prepared and used for this study. Ham (CJ Co., Ltd., Icheon, Korea) was purchased from a local store, sliced into about $1 \times 1 \times 1$ cm cubes, and used without any further treatment. Fresh egg was purchased from a local store and fried in a frying pan. Frying was done by placing the broken whole egg (approximately 2 mm in thickness) after mixing it thoroughly in a preheated pan (about 170°C), then 1 and half minutes later the fried egg was turned over and cooked on the other side for another 30 sec. Seasoned beef in a raw state was also purchased from a local store and cooked in a frying pan (about 170°C) for about 4 min. Surimi gel, imitation crab leg, and dried laver were purchased from a local store and sliced into $1 \times 1 \times 1$ cm cubes for surimi gel and imitation crab leg into about 1×1 cm pieces; the dried laver was used without any further treatment. Cucumber was purchased from a store, washed with tap water, and sliced to be about $1 \times 1 \times 1$ cm cubes. Seasoned burdock and seasoned and blanched spinach were also cut into $1 \times 1 \times 1$ cm cubes and 1×1 cm pieces, respectively. All the samples were obtained from 3 different local stores and pooled. Prepared *Kimbab* for the sensory analysis was purchased from a local store. Ten grams of each of the different samples was packed in oxygen-impermeable nylon bags ($2 \text{ mL O}_2/\text{m}^2/24 \text{ h}$ at 0°C , 0.09 mm thickness; Sunkyung Co., Ltd., Seoul, Korea).

Irradiation

Packed samples were irradiated in a cobalt-60 irradiator (Point source AECL, IR-79, MDS Nordion International Co., Ltd., Ottawa, ON, Canada) at the Korea Atomic Energy Research Institute, Daejeon, Korea. The source strength was approximately 100 kCi with a dose rate of 10 kGy h^{-1} at $12 \pm 0.5^{\circ}\text{C}$. Dosimetry was performed using 5 mm diameter alanine dosimeters (Bruker Instruments, Rheinstetten, Germany), and the free-radical signal was measured using a Bruker EMS 104 EPR Analyzer. The dosimeters were calibrated against an International standard set by the International Atomic Energy Agency (Vienna, Austria). The applied doses in this study were 0, 1, 2, and 3 kGy for the food materials. After irradiation, the samples were transferred to designated incubators (10°C) and a microbiological analysis was performed during storage for 24 hrs. The prepared *Kimbab* for the sensory analysis was irradiated at 0, 0.5, 1, 2, and 3 kGy and the sensory analysis was performed

within 3 hrs after irradiation.

Microbiological analysis

A sample (10 g) was aseptically homogenized for 2 min in a sterile stomacher bag containing 90 mL of sterile peptone water using a stomacher lab blender (Model 400, Tekmar Co., Cincinnati, OH, USA). Media for the enumeration of the microbes was prepared by a total plate count agar (Difco Laboratories, Detroit, MI, USA). The plates were incubated at 37°C for 48 h, and colony forming units (CFU) per gram were counted at a dilution of 30 to 300 CFU per plate. Experiments with each bacteria culture were independently conducted twice.

Sensory evaluation

The non-irradiated control and irradiated samples (20 g each) were served to the panelists individually within 3 hrs after irradiation. The sensory test was performed at 3 different irradiation times and the data was pooled. The sensory parameters for this study were appearance, texture, flavor, overall acceptability, and the intensity of an irradiation odor. The sensory scores were evaluated independently by the panelists for each of the 3 different times with different individuals. A 9 point hedonic scale was provided to the panelists as follows; like extremely (9), like very much (8), like moderately (7), like slightly (6), neither like nor dislike (5), dislike slightly (4), dislike moderately (3), dislike very much (2), and dislike extremely (1). For the intensity of the irradiation odor, extremely intense (9) and none (1) were used.

Statistical analysis

Mean values and the standard deviation are reported for the microbial analysis. Analyses of variance were performed using SAS software (15) for the sensory analysis and the Student Newman Keul's multiple range test was used to compare the differences among the mean values. Mean values and the pooled standard errors of the mean (SEM) are reported and significance defined as $p < 0.05$.

RESULTS AND DISCUSSION

Total plate count

A total of 9 food materials, prepared in a ready-to-eat form, were tested for total plate count. Ham, seasoned and cooked beef, imitation crab leg, fried egg, and seasoned burdock did not initially have detectable levels of microorganisms (10^2 CFU/g). These food materials for manufacturing *Kimbab* are processed foods including a heat pasteurization step, except for seasoned burdock. The root of burdock (*Arctium lappa* L.) possesses various pharmaceutical activities including an antibacterial activity (16), antioxidant activity (17) and an anti-inflam-

matory activity (18). The absence of initial contamination in the above mentioned coprocessed foods suggest that good hygienic quality can be maintained when good manufacturing practices are properly utilized in additional production processes.

Surimi gel was another processed food evaluated, but it was contaminated by microorganisms with 3.5 log CFU/g initially (Table 1). After 24 hrs of refrigerated storage at 10°C, the level of microorganisms increased to 5.64 log CFU/g. Irradiation at 1 kGy reduced the level of microorganisms to an undetectable level. Once the surimi gel was irradiated, there were no viable cells after refrigerated storage for 24 hrs at 10°C. Using 4 different pathogens, *Escherichia coli*, *Salmonella typhimurium*, *Staphylococcus aureus*, and *Listeria ivanova*. Jo et al. (19) inoculated surimi gel and the radio-sensitivity was reported as 0.31, 0.42, 0.22 and 0.63 kGy, respectively. This indicates that a low dose irradiation can eliminate the pathogens contaminating surimi gel.

Cucumber, often used for *Kimbab* manufacturing, was contaminated by microorganisms at a substantial level (5 log CFU/g) after being washed by tap water (Table 2). The seasoned and blanched spinach also had a similar level of microorganisms to the cucumber even after washing, seasoning, and blanching (Table 3). However, 1 kGy irradiation resulted in no viable cell counts. These data suggest that vegetables could be the main source of the microbial contamination in *Kimbab* processing. It has been reported that raw vegetables may harbor potential foodborne pathogens (10,21). *Aeromonas* spp. was also isolated from 41% of the organic vegetables eval-

Table 1. Effect of irradiation on the microbial population in surimi gel during storage at 10°C

Irradiation dose (kGy)	Viable cell counts (log CFU/g)		
	0 hr	8 hr	24 hr
0	3.50 ± 0.14	5.57 ± 0.16	5.64 ± 0.07
1	ND ¹⁾	ND	ND
2	ND	ND	ND
3	ND	ND	ND

¹⁾Viable cells were not detected at the detection limit of <10² CFU/g.

Table 2. Effect of irradiation on the microbial population in cucumber during storage at 10°C

Irradiation dose (kGy)	Viable cell counts (log CFU/g)		
	0 hr	8 hr	24 hr
0	5.07 ± 0.92	4.98 ± 0.80	5.10 ± 0.77
1	ND ¹⁾	ND	ND
2	ND	ND	ND
3	ND	ND	ND

¹⁾Viable cells were not detected at detection limit of <10² CFU/g.

Table 3. Effect of irradiation on the microbial population in seasoned and blanched spinach during storage at 10°C

Irradiation dose (kGy)	Viable cell counts (log CFU/g)		
	0 hr	8 hr	24 hr
0	5.41 ± 0.51	5.35 ± 0.44	5.43 ± 0.49
1	ND ¹⁾	ND	ND
2	ND	ND	ND
3	ND	ND	ND

¹⁾Viable cells were not detected at the detection limit of <10² CFU/g.

uated (20).

The major microbial contaminant was revealed to be the dried laver. The dried laver initially had 8.83 log CFU/g of microorganisms and an irradiation of 1, 2, and 3 kGy only reduced the level to 7.60, 7.48, and 7.14 log CFU/g, respectively (Table 4). During 24 hrs of storage at 10°C, the level of the microbial contamination was suspended. Ahn et al. (11) reported that the number of total aerobic bacteria in a commercial laver was 3.2 × 10⁶ CFU/g and an irradiation of 10 kGy resulted in only a 2 decimal point reduction. A significant shoulder line was shown until 20 kGy and the D₁₀ value for the total aerobic bacteria present in the laver was 11.27 kGy. The authors also identified a radiation-resistant bacterium isolated from the laver and reported it potentially as *Micrococcus roseus* spp. (11).

When the *Kimbab* was processed using the food materials tested, the initial microbial load was 8.73 ± 0.05 log CFU/g. After an irradiation of 1, 2, and 3 kGy, the number of microorganisms were reduced to 5.55 ± 0.71, 4.33 ± 0.46, and 3.29 ± 0.59, respectively (data not shown). These results indicate that commercially processed *Kimbab* may be highly contaminated by the microorganisms originally present in the dried laver, other vegetables and ingredients, and by additional cross-contamination during manufacturing. Therefore, a minimum hygienic quality requirement for pre-treatment to reduce the microbial load before manufacturing *Kimbab* is highly recommended.

Sensory evaluation

From the results of the microbial test, it was confirmed that irradiation might be an effective pre-treatment of highly contaminated ready-to-eat foods. Therefore, a sen-

Table 4. Effect of irradiation on the microbial population in dried laver during storage at 10°C

Irradiation dose (kGy)	Viable cell counts (log CFU/g)		
	0 hr	8 hr	24 hr
0	8.83 ± 0.10	8.80 ± 0.06	8.83 ± 0.09
1	7.60 ± 0.51	7.63 ± 0.48	7.60 ± 0.51
2	7.48 ± 0.19	7.48 ± 0.19	7.98 ± 0.52
3	7.14 ± 0.23	7.14 ± 0.23	7.14 ± 0.23

Table 5. Sensory evaluation of irradiated *Kimbab*

Irradiation dose (kGy)	Appearance	Texture	Flavor	Overall acceptability	Irradiation odor
0	6.40 ^{a1)}	6.45 ^a	6.75 ^a	7.00 ^a	1.65 ^c
0.5	5.30 ^{abc}	5.35 ^{abc}	4.60 ^{bc}	4.85 ^{bc}	3.65 ^b
1	5.00 ^{bc}	5.60 ^{ab}	5.55 ^b	5.55 ^b	3.05 ^b
2	4.55 ^c	4.25 ^c	3.40 ^c	3.45 ^d	5.45 ^a
3	6.05 ^{ab}	4.65 ^{bc}	3.85 ^c	4.10 ^{cd}	4.35 ^{ab}
SEM	0.385	0.404	0.417	0.409	0.450

¹⁾Means with the same letter in each column are not significantly different ($p < 0.05$).

sory evaluation was performed immediately after irradiation at 3 different times with the prepared *Kimbab* using the microbiologically tested food ingredients. All the sensory parameters tested including: appearance, texture, flavor, and overall acceptance were negatively-correlated with the irradiation dose applied (Table 5). These data suggests that the prepared *Kimbab* is not suitable for irradiation by itself, and methods need to be developed to preserve sensory quality. However, a low dose of irradiation significantly reduced the microbial contamination of the materials for *Kimbab* and increased the safety of the product. Intensity of the irradiation odor was progressively increased by increases in the irradiation dose (Table 5).

In conclusion, irradiation was an effective tool to reduce the highly contaminated microorganisms in the ready-to-eat materials for manufacturing *Kimbab*. However, irradiation is not suitable for preserving prepared *Kimbab* until optimum conditions for preserving sensory quality can be established.

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REFERENCES

- Kwak TK, Kim SH, Park SJ, Cho YS, Choi EH. 1996. The improvement of the sanitary production and distribution practices for packaged meals (*Kimbab*) marketed in convenience stores using hazard analysis critical control point (HACCP) system. *J Food Hyg Safety* 11: 17-187.
- Lee HS, Ryu SY. 1998. The seasonal microbiological quality assessment of *Kimbab* (seaweed roll) production flow in food service facilities for university students. *J Food Sci Nutr* 14: 367-374.
- Zhou Z, Robards K, Helliwell S, Blanchard C. 2003. Effect of rice storage on pasting properties of rice flour. *Food Res Int* 36: 625-634.
- Kang YS, Yoon SK, Jwa SH, Lee DH, Woo GJ, Park YS, Kim CM. 2002. Prevalence of *Staphylococcus aureus* in *Kimbab*. *J Food Hyg Safety* 17: 31-35.
- Food Information Korea. 2004. Foodborne disease last year: *Kimbab* and lunch set was the most. Feb. 9. <http://www.foodikorea.com>
- Finlay WJJ, Logan NA, Sutherland AD. 2002. *Bacillus cereus* emetic toxin production in cooked rice. *Food Microbiol* 19: 431-439.
- Jang KI, Park JH, Kim KY. 1999. Studies on *Salmonella enteritidis* contamination in chicken egg using confocal scanning laser microscopy. *Korean J Food Sci Technol* 31: 771-777.
- Kim JW, Kim HC, Hur JW. 1998. Quality changes of egg products during storage. *Korean J Food Sci Technol* 30: 1480-1483.
- Jo C, Kim DH, Shin MG, Kang IJ, Byun MW. 2003. Irradiation effect of *bulgogi* sauce for manufacturing Korean traditional meat product, *bulgogi*. *Radiat Phy Chem* 68: 851-856.
- Nguyen-the C, Carlin F. 1994. The microbiology of minimally processed fresh fruits and vegetables. *Crit Rev Food Sci Nutr* 34: 370-401.
- Ahn HJ, Yook HS, Kim DH, Kim S, Byun MW. 2001. Identification of radiation resistant bacterium isolated from dried laver (*Porphyra tenera*). *J Food Sci Nutr* 30: 193-195.
- Abu-Tarboush HM, Al-Kahtani HA, Abou-Arab AA, Baij-aber AS, El-Mojadid MA. 1996. Sensory and microbial quality of chicken as affected by irradiation and post-irradiation storage at 4°C. *J Food Prot* 60: 761-770.
- World Health Organization. 1999. High-dose irradiation: Wholesomeness of food irradiated with doses above 10 kGy. WHO Technical Report Series 890. Geneva.
- Anonymous. 2003. Irradiation enhances food safety and quality. *Food Prot Trends* 23: 573-574.
- SAS Institute Inc. 1985. *SAS User's Guide*. SAS Institute Inc., Cary, NC.
- Chow LW, Wang SJ, Duh PD. 1997. Antibacterial activity of burdoc. *Food Sci* 24: 195-202.
- Duh PD. 1998. Antioxidant activity of burdoc (*Arctium lappa* Linne): Its scavenging effect on free-radical and active oxygen. *J Am Oil Chem Soc* 75: 455-461.
- Lin CC, Lin JM, Yang JJ, Chuang SC, Ujiie T. 1996. Anti-inflammatory and radical scavenging effect of *Arctium lappa*. *Am J Chinese Med* 24: 127-137.
- Jo C, Lee NY, Kang HJ, Hong SP, Kim YH, Kim JK, Byun MW. 2004. Inactivation of pathogens inoculated into prepared seafood products for manufacturing *Kimbab*, steamed rice rolled by dried seaweed, by gamma irradiation. *J Food Prot* In Press.
- Beuchat LR. 1996. Pathogenic microorganisms associated with fresh produce. *J Food Prot* 59: 204-216.
- Johannessen GS, Loncarevic S, Kruse H. 2002. Bacteriological analysis of fresh produce in Norway. *Int J Food Microbiol* 77: 199-204.

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