Effectiveness of Radiation Processing in Elimination of Salmonella Typhimurium and Listeria monocytogenes from Sprouts

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ABSTRACT

The effectiveness of radiation treatment in eliminating Salmonella Typhimurium and Listeria monocytogenes on laboratory inoculated ready-to-eat sprouts was studied. Decimal reduction doses ($D_{10}$ values) for Salmonella Typhimurium and L. monocytogenes in dry seeds of mung (green gram), matki (dew gram), chana (chick pea), and vatana (garden pea) ranged from 0.189 to 0.303 kGy and 0.294 to 0.344 kGy, respectively. In sprouts made from these seeds, the $D_{10}$ values ranged from 0.192 to 0.208 kGy for Salmonella Typhimurium and from 0.526 to 0.588 kGy for L. monocytogenes. Radiation treatment with a 2-kGy dose resulted in complete elimination of $10^4$ CFU/g of Salmonella Typhimurium and $10^5$ CFU/g of L. monocytogenes from all four varieties of sprouts. No recovery of Salmonella Typhimurium and L. monocytogenes was observed in the irradiated samples stored at 4 and 8°C up to 12 days. Radiation treatment with 1 kGy and 2 kGy resulted in a reduction of aerobic plate counts and coliform counts by 2 and 4 log CFU/g, respectively; the yeast and mold counts and staphylococci counts decreased by 1 and 2 log CFU/g, respectively. However, during postirradiation storage at 4 and 8°C, aerobic plate counts, coliform counts, yeast and mold counts, and staphylococci counts remained constant throughout the incubation period. This study demonstrates that a 2-kGy dose of irradiation could be an effective method of processing to ensure microbial safety of sprouts.

The increasing appreciation by consumers regarding the nutritional benefits of raw foods of plant origin has resulted in an increased consumption of raw, fresh, produce-like vegetables, fruits, and sprouts. The consumption of sprouts has increased due to their high nutritional quality, fiber, vitamins and mineral content, and low caloric value (5, 14, 38). In India, the sprouts are mainly prepared at home or produced on small scale and sold in local markets in open condition by small units. However, in recent years, due to the increased demand, the shopping malls in big cities have started selling packaged sprouts in refrigerated condition. The sprouts made from mung, matki, chana, and vatana are generally consumed in India. A typical sprout-making process in India involves soaking of seeds for 12 h and spraying for 24 h, while in other countries such as the United States and the European Union, sprout production is a 3- to 7-day process (14). Traditionally, the sprouts were eaten after cooking; however, recently the consumption of uncooked sprouts in salads has increased.

Seed sprouts can have a high bacterial count, possibly including pathogens (19). A variety of pathogenic bacteria, such as Salmonella serotypes, E. coli O157:H7, Bacillus cereus, Aeromonas hydrophila, and Staphylococcus aureus have been isolated from sprouts (19). Studies have shown attachment and growth of Salmonella, Escherichia coli O157:H7, and Listeria monocytogenes in alfalfa sprouts (1, 17). The sprouts marketed in Mumbai and its suburbs were found to have poor microbiological quality with a high bacterial load and a high percentage of samples were found to be contaminated with pathogens such as Salmonella, E. coli and coagulase-positive S. aureus (unpublished data).

Since 1988, the number of reported fresh produce, including sprout borne, outbreaks has steadily increased (19). Between 1995 and 2003, there were 13 outbreaks of Salmonella and E. coli O157:H7 infections associated with commercial sprouts (7, 8, 11, 19, 29, 37). In 11 of the 13 outbreaks, the pathogen was found to be Salmonella. Salmonella is considered a zero-tolerance pathogen in sprouts (34). L. monocytogenes has been isolated from sprouts (13), but no outbreaks related to it have been reported. However, an increasing body of data supports and suggests that salad vegetables, such as cabbage, celery, lettuce, cucumber, onion, radish, and tomatoes, can have a high incidence of L. monocytogenes, and some of them have been implicated in outbreaks of foodborne listeriosis (38). The U.S. Food and Drug Administration and U.S. Department of Agriculture, Food Safety and Inspection Service, have established a zero-tolerance policy for L. monocytogenes in ready-to-eat foods, including minimally processed fresh and precut fruits and vegetables (3).

The contamination of fresh produce could occur from any combination of the following: contaminated seeds, con-
taminated equipment, contaminated water sources, or poor hygienic handling (19). With sprouts, seeds are considered the most likely source of contamination. To date, there are no chemical or water-rinse treatments that will effectively decontaminate fresh produce. Taormina and Beuchat (30) were able to reduce the population of *E. coli* O157:H7 in germinated seeds, but could not eliminate the pathogens on the sprouts. In many cases, it is difficult, if not impossible, to either wash these pathogens from the sprouts or inactivate them by chemical treatments because the pathogens may be located in inaccessible areas or they may have penetrated the product (4, 6, 20, 39). Scanning electron microscopy studies have shown formation of biofilms on cotyledons, hypocotyls, and roots of alfalfa seeds (12). Such biofilms may afford protected colonization sites for human pathogens such as *Salmonella* and *L. monocytogenes* making their eradication with antimicrobial compounds difficult. Studies have shown association of *L. monocytogenes* with root hairs of alfalfa sprouts and washing treatment was not sufficient to remove them (17). Since there is no other way to guarantee pathogen-free raw sprouts, the U.S. government issued a warning in 1999 regarding the hazard of eating raw sprouts (19).

Because of their high penetration power, ionizing radiations can inactivate pathogens that may have gained entry in tissues of sprouts. Radiation processing is a cold process; therefore, changes occurring in food constituents are minimum and insignificant (31). Gamma radiation has been shown to be effective to eliminate *Salmonella* and *E. coli* from radish sprouts and *L. monocytogenes* from broccoli sprouts (3, 23, 24). The most common sprouts consumed in India are mung, matki, vatana, and chana, and no work has been carried out on these sprouts to ensure their safety. Therefore, there is a need to study radiation sensitivity of different pathogens in sprouts used in India and to find the dose for their elimination without affecting the nutritional and organoleptic qualities.

The objectives of the present study were (i) to determine the *D*<sub>10</sub>-value for *Salmonella* Typhimurium and *L. monocytogenes* inoculated in different sprouts, (ii) to determine the radiation dose for a 5-log elimination of *Salmonella* Typhimurium and *L. monocytogenes* from sprouts, (iii) to study the survival and recovery, if any, of these pathogens in inoculated and radiation-treated sprout samples, during storage at 4 and 8°C, and (iv) to study the microbial profile of both radiation-treated and untreated market sprout samples during storage.

**MATERIALS AND METHODS**

**Bacterial strains.** *Salmonella* Typhimurium MTCC 98 was obtained from Microbial Type Culture Collection (Chandigarh, India). *L. monocytogenes* NCIMB-B-01442, an avirulent strain, was kindly supplied by Dr. C. Mohacsi-Farkas (Szent Istvan University, Budapest, Hungary). *E. coli* ATCC 35218 and *S. aureus*, a coagulase-positive strain isolated from poultry meat, were used as standards for biochemical tests.

**Samples.** Seed and sprout samples of mung, matki, chana, and vatana were obtained from local market in Mumbai and Suburbs.

**Chemicals and media.** Microbiological media used in the studies were from Hi Media Laboratories (Mumbai, India). Blood used for blood agar was obtained from human volunteers. Rabbit plasma and *E. coli* H antisemur H7 were obtained from Becton Dickinson and Company (Sparks, Md.).

**Microbiological analysis.** The microbiological analysis was performed as per the standard methods adopted from the online *Bacteriological Analytical Manual* for the detection, enumeration, and identification of individual organisms (33).

**Aerobic plate count (APC).** The sample (25 g) was homogenized in 225 ml of sterile physiological saline. After appropriate serial dilutions, the samples were pour plated on plate count agar (PCA). The colonies were counted after 24 h of incubation at 35°C.

**Coliform count.** Dilutions made for APC were pour plated on violet red bile agar (VRBA). After the plates were solidified, they were overlaid with a layer of VRBA. Typical dark red colonies were counted after 24 h of incubation at 35°C.

**Yeast and mold count (YMC).** YMC was determined on yeast and mold agar using spread plate technique. The colonies were counted after 48 h of incubation at room temperature (≈26°C).

**Staphylococci count.** Spread plate technique was used to determine staphylococci count on Baird Parker agar (BPA). Typical black colonies were counted after 48 h of incubation at 35°C.

**Salmonella serotypes.** The sample (25 g) was homogenized in lactose broth and incubated overnight at 35°C. After the initial preenrichment step, samples were further enriched in tetraphionate broth and Rapport Vassiliadis medium at 35°C. A loopful of growth from each of these media was streaked on bismuth sulphite agar, xylose lysine deoxycholate agar and Hektoen enteric agar. Both typical and atypical colonies from each of these plates were purified and identified by subsequent biochemical tests.

**E. coli.** *E. coli* was isolated by preenrichment in lauryl sulphate tryptose broth at 35°C, selective enrichment at 45.5°C in *E. coli* broth, and selective plating on eosin methylene blue agar. Purple colonies with metallic sheen were selected and the isolates confirmed by the indole, methyl red, Voges-Proskauer, and citrate tests.

**E. coli** O157:H7. Biochemically confirmed *E. coli* isolates were plated on 4-methylumbrelliferol-d-glucuronide (MUG) sorbitol agar and incubated at 35°C for 24 h. Colonies not showing fluorescence were further tested using Difco *E. coli* O antisemur O157 and *E. coli* H antisemur H7.

**L. monocytogenes.** The sample (25 g) was homogenized in 225 ml of *Listeria* enrichment broth and incubated at 35°C for 48 h. A loopful of enriched broth was streaked on *Listeria* identification agar (PALCAM). Characteristic grayish black colonies were picked up for further biochemical confirmation. Isolates showing typical biochemical tests were further tested for hemolysin production using blood agar.

**Coagulase-positive** *S. aureus.* Characteristic black colonies with peripheral clearance zone from BPA were selected. Confirmation of the coagulase activity was done by checking the clot formation in coagulate salt mannitol broth supplemented with 12% plasma.

**Preparation of inocula.** Either *Salmonella* Typhimurium or *L. monocytogenes* was inoculated in 25 ml of brain heart infusion
broth and incubated at 35°C at 100 rpm. Twenty-hour-old cultures were used in all the experiments.

Irradiation of samples. Packaged samples were kept in ice and radiation processing was carried out at 0 to 4°C in a cobalt-60 irradiator (Gamma cell 5,000, Board of Radioisotopes and Technology, Mumbai, India) at a dose rate of 0.083 kGy/min for decontamination. For other studies, radiation processing was carried out in a cobalt-60 irradiator (Gamma cell 220, Atomic Energy of Canada Ltd., Ontario, Canada) at a dose rate of 0.011 kGy/min. Dose rate of all the radiation sources was measured using the Frickes method (28). Variations in doses absorbed by experimental samples were minimized by placement within a uniform area of the radiation field.

Determination of decimal reduction dose ($D_{10}$). Seed and sprout samples were packed in low-density polyethylene bags (LDPE) of 35 μm thickness (ACE packaging Ltd., Mumbai, India), sealed using heat sealer (Sevana, Mumbai, India), and exposed to 3 kGy and 6 kGy dose, respectively, for the elimination of native microbial flora. Conditions necessary to get a desired number of cells of *Salmonella* Typhimurium and *L. monocytogenes* attached to sprouts were standardized. Seed and sprout samples were dipped in sterile tap water containing 10^6 CFU/ml of *Salmonella* Typhimurium culture for 2 min or 10^6 CFU/ml of *L. monocytogenes* culture for 5 min and dried on sterile blotting paper to remove excess water under aseptic conditions. Samples were then packed in LDPE bags (25 g each) and sealed using a heat sealer (Sevana, Mumbai, India). Seed and sprout samples inoculated with *Salmonella* Typhimurium were exposed to a radiation dose of 0.1, 0.2, 0.3, 0.4, 0.5, and 0.6 kGy. Seed samples inoculated with *L. monocytogenes* were exposed to a radiation dose of 0.25, 0.5, 0.75, and 1 kGy. A radiation dose of 0.5, 1, 1.5, and 2 kGy was given to sprout samples inoculated with *L. monocytogenes*. The samples were aseptically homogenized for 1 min in a sterile stomacher bag (stomacher lab blender, model 400, Seward, London, UK) containing 225 ml of sterile saline. Serial dilutions of the homogenate were prepared and appropriate dilutions were used to determine viable counts with plate count agar. The plates were incubated at 35°C for 48 h and CFU values were determined. Each study included three samples per dose and was repeated three times. The average number of surviving viable cells (CFU per gram) in the samples was plotted against the radiation dose. The slopes of the individual survivor curves were calculated by linear regression with a computer program (Origin 6.1 version 6.1052 B232, Origin Lab Corporation, Northampton, Mass.). The $D_{10}$-value was calculated by taking the negative reciprocal of the survival curve slope.

Determination of the dose required to eliminate 5 log CFU/g of the inoculated cells. Sprout samples were packed in LDPE bags and exposed to a 6-kGy dose for the elimination of native microbial flora and then inoculated with *Salmonella Typhimurium* or *L. monocytogenes* as described above. The inoculated samples (10^5 to 10^6 CFU/g of *Salmonella* Typhimurium or *L. monocytogenes*) in triplicate were irradiated at 0.5, 1, 1.5, 2, 2.5, and 3 kGy, and the surviving population was determined by the plating of serial dilutions on PCA after an incubation of 48 h at 35°C. Enrichment and selective plating were carried out to confirm the complete elimination of these pathogens. Each experiment was repeated three times.

Storage studies of sprout samples inoculated with *Salmonella Typhimurium* or *L. monocytogenes*. A radiation dose of 6 kGy was given to sprouts for decontamination and then dipped in sterile tap water containing 10^5 CFU/ml of *Salmonella Typhimurium* for 2 min or 10^4 CFU/ml of *L. monocytogenes* for 5 min and dried on sterile blotting paper to remove excess suspension under aseptic conditions. Samples were packed in LDPE bags (25 g each) and exposed to a radiation dose of 2 kGy. These samples were then stored at 4 and 8°C for 12 days and were screened for *Salmonella Typhimurium* and *L. monocytogenes* on day 4, 8, and 12 of storage. Enrichment and selective plating were carried out to confirm the complete elimination of these pathogens. Each experiment was repeated three times.

Storage studies of un inoculated samples. Market samples of sprouts were exposed to a radiation dose of 1 kGy and 2 kGy. These samples along with unirradiated samples were stored at 4 and 8°C for 12 days. APC, coliform counts, YMC, and staphylococci counts of these samples were determined at intervals of 4 days. Also, the samples were screened for the presence of *Salmonella, E. coli, E. coli* O157:H7, *L. monocytogenes*, and coagulase-positive *S. aureus*. Each experiment was repeated three times.

Statistical analysis. All the data for $D_{10}$-values of *Salmonella Typhimurium* and *L. monocytogenes* in seeds and sprouts were analyzed statistically using Origin 6.1 software version 6.1052 B232. Significant differences in $D_{10}$-values for each seed and each sprout were evaluated by the Student’s t test while the significant differences between seeds and sprouts were analyzed by one-way ANOVA.

RESULTS AND DISCUSSION

Although treatment with a 2-kGy dose of gamma radiation for the elimination of *Salmonella* and *E. coli* O157: H7 from mung, radish, alfalfa, and broccoli sprouts has been reported (2, 23, 24), this is the first report on the effectiveness of irradiation to eliminate *Salmonella* and *L. monocytogenes* from mung, matki, chana, and vatana sprouts, which are commonly used in India. Gupta and Verma (15) reported *Salmonella Typhimurium* as the most common *Salmonella* serotype in India; therefore, this serotype was used to study the radiation dose necessary for the elimination of *Salmonella* from sprouts.

To decontaminate the seeds and sprouts, they were irradiated at different doses and aerobic plate counts were determined. The treatment with a 3- and 6-kGy dose was found sufficient for the destruction of the native flora from seeds and sprouts, respectively (data not shown). The higher dose of radiation needed for sprout decontamination is due to high bacterial counts in sprouts (>10^8 to 10^9 CFU/g). Similar high doses were reported for decontamination of radish, alfalfa, and broccoli sprouts (25).

The $D_{10}$-values for *Salmonella Typhimurium* were found to be 0.189 ± 0.003, 0.212 ± 0.007, 0.303 ± 0.008, and 0.294 ± 0.007 kGy in mung, matki, chana, and vatana seeds, respectively (Fig. 1). The radiation sensitivity of *Salmonella Typhimurium* was found to vary in different seeds significantly ($P < 0.05$). Thayer et al. (32) reported 0.97 ± 0.003 kGy as the $D_{10}$-value for *Salmonella* in alfalfa seeds while Rajkowski et al. (23) reported 0.74 kGy in broccoli seeds. The lower $D_{10}$-values observed in the present study may be due to the different bacterial strain used as well as to the differences in seeds. $D_{10}$-values for *L. monocytogenes* in mung, matki, chana, and vatana seeds were not significantly different ($P > 0.05$) and the values
were 0.294 ± 0.03, 0.312 ± 0.031, 0.344 ± 0.024, and 0.312 ± 0.001 kGy, respectively (Fig. 2). To date, there are no reports on radiation sensitivity of \( L.\ monocytogenes \) in seeds; however, survival of this pathogen on seeds and growth during sprouting has been reported (27). The treatment of seeds with low-dose ionizing radiation has been tested for the elimination of pathogens. Rajkowski et al. (23) reported that at a dose of 3 kGy, only a 2.7-log kill for \( Salmonella \) was observed for the dried inoculated broccoli seeds. Although the germination percentage was not affected, the yield ratio (wt/wt) was reduced at a dose of 2 kGy. Also, even if the seeds are made free from pathogen, any contamination during the germination process can lead to the presence of pathogen in sprouts. The aerobic plate counts of sprouts made from seeds irradiated at a 2-kGy dose were high (data not shown). Therefore, there is a need for radiation processing of final product (sprouts).

In all the sprouts studied, the \( D_{10} \)-value for \( Salmonella \) Typhimurium was found to be around 0.2 kGy. No significant difference \( (P > 0.05) \) in \( D_{10} \)-values was observed in different sprouts (Fig. 3). Dhokane et al. (9) observed \( D_{10} \)-values of 0.164 kGy in carrot and 0.178 kGy in cucumber by using the same strain of \( Salmonella \) Typhimurium. \( D_{10} \)-values of 0.46 kGy and 0.54 kGy for \( Salmonella \) isolates from vegetable and meat inoculated in alfalfa sprouts have been reported (24). \( D_{10} \)-values presented here were found to be much lower than the reported values which could be due to the differences in the \( Salmonella \) Typhimurium strains used as well as to the differences in the sprouts under study. \( D_{10} \)-values for \( L.\ monocytogenes \) inoculated in sprouts were found to be between 0.526 ± 0.033 to 0.588 ± 0.05 kGy (Fig. 4) and no significant difference in \( D_{10} \)-values in different types of sprouts were observed at \( P > 0.05 \). The radiation resistance of \( L.\ monocytogenes \) was found to be significantly higher in sprouts as compared to that in seeds which might be due to changes in the chemical composition of seeds during the sprouting process. The sprouts have a more complex chemical composition than that of seeds due to the hydrolysis of complex polymers and synthesis of vitamins and other chemicals during germination.
minimization of seeds (36). Bari et al. (4) reported $D_{10}$ values of 0.20 and 0.22 kGy for $L. \text{monocytogenes}$ in broccoli and mung bean sprouts, respectively. Niemira et al. (21) reported 0.21 kGy as the $D_{10}$ value for $L. \text{monocytogenes}$ in the leafy salad vegetable, endive, and Farkas et al. (10) reported a $D_{10}$ value of 0.4 kGy in bell pepper.

The wide variation in $D_{10}$ values is probably due to intrinsic properties of the sprouts as well as irradiation conditions. Niemira et al. (22) demonstrated that radiation sensitivity of inoculated $E. \text{coli}$ O157:H7 is influenced by the type of lettuce in which it is suspended. The $D_{10}$ values of bacteria in food are affected by a number of factors such as the water activity, composition, irradiation temperature, and presence of oxygen (18). Also, in a complex food system, some of the constituents, such as proteins, are thought to compete with cells for interaction with radiolytic free radicals, thereby reducing the net effect of radiation damage and making organisms more radiation resistant (35).

Since the $D_{10}$ values observed for each of the pathogens in all different sprouts were similar (Fig. 3 and 4), studies for optimization of the radiation dose to achieve 5-log kill were carried only with inoculated mung sprouts. Treatment with 2 kGy was sufficient for the total elimination of $10^4$ CFU/g of $\text{Salmonella Typhimurium}$; however, treatment with 3 kGy was needed for the elimination of $L. \text{monocytogenes}$ (Table 1). Because the $D_{10}$ value for $L. \text{monocytogenes}$ was around 0.550 kGy (Fig. 4), a higher dose of 3 kGy was necessary for 5-log reduction of this pathogen. These studies confirm the observation by Bari et al. (2) that radiation processing of mung sprouts with a 2-kGy dose can eliminate $5 \log$ CFU/g of $\text{Salmonella}$. Bari et al. (4) reported a reduction in $L. \text{monocytogenes}$ by 4.88 and 4.57 $\log$ CFU/g in broccoli and mung bean sprouts, respectively, at a 1-kGy radiation dose. Inactivation of a higher number of $L. \text{monocytogenes}$ by Bari et al. (3) using a 1-kGy radiation dose might be due to the lower $D_{10}$ values of $L. \text{monocytogenes}$ strains used in their study.

Treatment with 2 and 3 kGy could eliminate $5 \log$ CFU/g of $\text{Salmonella Typhimurium}$ and $L. \text{monocytogenes}$, respectively, when the survival of the pathogens was analyzed immediately after irradiation (Table 1). However, recovery of these pathogens was observed when these treated samples were stored at 4 and 8°C (data not shown). This could be due to repair of radiation-induced damage during storage. Thus, a higher dose of radiation, $>2$ kGy for $\text{Salmonella Typhimurium}$ and $>3$ kGy for $L. \text{monocytogenes}$, is required to achieve a 5-log kill.

Although $\text{Salmonella}$ was detected in 24% of the samples of sprouts marketed in Mumbai, all of the tested 124 sprout samples were found to be free from $L. \text{monocytogenes}$ (unpublished data). Usually, the cell number of these pathogens, if present, is $<10^2$ CFU/g. Therefore, radiation treatment using a low dose of 2 kGy was tested for the elimination of these pathogens from inoculated sprouts and their survival during storage at 4 and 8°C was studied. Radiation treatment of 2 kGy resulted in complete elimination of $10^4$ CFU/g of $\text{Salmonella Typhimurium}$ and $10^3$ CFU/g of $L. \text{monocytogenes}$. There was no recovery of these pathogens, even after enrichment, from the samples stored up to 12 days at 4 and 8°C. Similar studies have shown effective elimination of $\text{Salmonella}$ from alfalfa and radish sprouts and other vegetables after radiation treatment with 2 kGy (2, 23, 26).

The treatment of sprouts even with a low dose of 1 kGy resulted in significant reduction in the number of both pathogens (Table 1). Therefore, the effect of radiation treatment (1 and 2 kGy) on the native microbial flora of different market sprouts during storage at 4 and 8°C was studied. Microbial profile of both radiation-treated and untreated mung sprouts is shown in Figure 5 through 8. Similar microbial profiles were observed for matki, chana, and vatana sprouts (data not shown). Treatment with 1 kGy resulted in a 2-log reduction in APC and coliform counts and a 1-log

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**FIGURE 4.** Radiation sensitivity of $L. \text{monocytogenes}$ in sprouts. Mung (■), Matki (●), Chana (▲), and Vatana (▼). Each symbol represents plate counts at each dose. Average values of three experiments are plotted along with standard deviation.

**TABLE 1.** Effect of irradiation on the growth of $\text{Salmonella Typhimurium}$ and $L. \text{monocytogenes}$ inoculated in mung sprout samples

<table>
<thead>
<tr>
<th>Irradiation dose (kGy)</th>
<th>$\log$ CFU per gram</th>
<th>Enrichment&lt;sup&gt;a&lt;/sup&gt;</th>
<th>$\log$ CFU per gram</th>
<th>Enrichment&lt;sup&gt;b&lt;/sup&gt;</th>
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<tr>
<td>0.0</td>
<td>5.85</td>
<td>+</td>
<td>5.74</td>
<td>+</td>
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<tr>
<td>0.5</td>
<td>3.74</td>
<td>+</td>
<td>4.92</td>
<td>+</td>
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<td>+</td>
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<td>+</td>
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<td>+</td>
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<td>NVC&lt;sup&gt;a&lt;/sup&gt;</td>
<td>−</td>
<td>NVC&lt;sup&gt;a&lt;/sup&gt;</td>
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<sup>a</sup> NVC, no viable counts detected.  
<sup>b</sup> Enrichment was carried out by pre-enrichment, enrichment, and selective plating.
reduction in YMC and staphylococci counts, whereas treatment with 2 kGy reduced APC and coliforms by 4 log and YMC and staphylococci counts by 2 log. Rajkowski and Thayer (25) also reported a decrease of 4 log in APC and coliform counts in alfalfa sprouts after treatment with 2 kGy. There was no significant increase in APC, coliforms, YMC, and staphylococci counts during storage up to 12 days at 4 and 8°C. One of the mung market samples was found to be positive for Salmonella, E. coli, and coagulase-positive S. aureus. The samples of the same lot treated with a 1- as well as a 2-kGy dose were found to be negative for all the pathogens screened and no recovery was observed during storage at 4 and 8°C for 12 days. This observation clearly indicates that although the dose needed for a 5-log kill of these pathogens is >2 to 3 kGy, a dose as low as 1 kGy is effective in the elimination of a low number of these pathogens which could naturally be present in sprouts. Rajkowski et al. (23) have also observed that treatment with a 0.5-kGy dose can eliminate Salmonella from naturally contaminated alfalfa seeds. The unirradiated control samples of matki and chana were spoiled after 8 days of storage at 4 and 8°C, as seen by off-odor as well as sliminess, while irradiated samples were found to be in good condition even on day 12 of storage (data not shown). Rajkowski and Thayer (25) have also shown increase in shelf life of alfalfa sprouts by treatment with a 2-kGy dose.

Studies in our laboratory have shown that treatment with a 2-kGy dose of radiation does not affect textural, nutritional, and organoleptic qualities of sprouts (unpublished data). These observations are in agreement with the previous reports that a 2-kGy treatment has no undesirable effect on the nutritional (in terms of total vitamin C and total carotenoids) as well as sensory quality (in terms of color, odor, texture, and flavor) of minimally processed carrot, cucumber, tomato, cabbage, broccoli, and mung sprouts (2, 3, 9, 16). The present studies show that radiation processing is useful to reduce bacterial pathogens from sprouts thereby ensuring their safety.
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