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Microbial Profiles of On-line–Procured Sprouting Seeds and Potential Hazards Associated with Enterotoxigenic *Bacillus* spp. in Homegrown Sprouts†

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ABSTRACT

We examined the microbiological quality of sprouting seeds sold through the Internet. Overall, five types of seeds each from six organic and six conventional sources were evaluated. The growth and toxin production of naturally occurring *Bacillus* spp. in sprouts produced using home-scale sprouting devices also were investigated. For alfalfa, broccoli, lentil, mungbean, and radish seeds, the average microbial counts were 3.3, 4.0, 2.8, 3.5, and 3.6 log CFU/g, presumptive *B. cereus* counts were 0.7, 1.0, 0.8, 1.0, and 0.9 log CFU/g, and total coliform counts were −0.3, −0.4, −0.5, 0.0, and −0.4 log of the most probable number per gram, respectively. No *Salmonella*, *Escherichia coli* O157, other fecal coliforms, or *Staphylococcus aureus* was found on seeds. Compared with conventional seeds, the organic seeds had lower or equivalent counts for total microorganisms, presumptive *B. cereus*, and total coliforms. When seeds were sprouting using a glass jar, the growth of presumptive *B. cereus* was significant for radish and broccoli but not for alfalfa, lentil, and mungbean sprouts; the counts exceeded 5.0 log CFU/g in radish sprouts. When sprouts were grown using an automatic sprouting device, presumptive *B. cereus* showed slight growth (<3.0 log cycles) in radish, broccoli, and mungbean sprouts but no growth in alfalfa and lentil sprouts. Although the presumptive *B. cereus* isolates were enterotoxigenic, they did not produce or accumulate detectable levels of diarrheal toxins in freshly produced sprouts.

Sprout contamination has caused numerous foodborne disease outbreaks in recent years. In 2000, a *Salmonella* Enteritidis outbreak in The Netherlands was linked to eating contaminated bean sprouts *(31)*. In 2001, bean sprouts contaminated with *Salmonella* Enteritidis caused an outbreak of gastroenteritis in Canada *(11)*. In the same year in the United States, a multistate outbreak of *Salmonella* Kottbus infection was associated with eating alfalfa sprouts *(15)*. A number of seed sanitizing methods, including chlorine treatments, have been used for industry-scale sprout production to enhance food safety *(2, 4, 7, 20)*. However, the potential hazards associated with contamination of retail seed and homegrown sprouts remain to be investigated.

Bacillus cereus is an etiological agent in foodborne disease outbreaks associated with produce *(8, 28)*. Portnoy et al. *(19)* suggested that *B. cereus* caused a food poisoning outbreak resulting from contaminated vegetable sprouts, and enterotoxigenic *B. cereus* was isolated from raw soybean sprouts by Kim et al. *(13)*. However, information on the fate of *B. cereus* and its actual toxin production capability during sprouting is limited. Typically, homegrown sprouts are produced in jars or on trays using either manual or automatic watering systems. Although the design of one tray-sprouting kit was thought to have an influence on the multiplication of *B. cereus*, other types of home-sprouting kits have not been examined in previous studies *(10, 11)*.

In the United States, sprouting seeds can be purchased at local stores or through the Internet for at-home sprout production. Some seed suppliers believe that certified organic seeds offer greater food safety because the certification process assures that the seeds have been grown and handled in a manner that helps minimize possible sources of contamination. However, results of a recent study suggest that organic produce is more susceptible to fecal contamination than is conventional produce at the preharvesting stage *(16)*, although the results of this study do not support allegations that organic produce poses a substantially greater risk of pathogen contamination. Contaminated organic sprouts recently triggered a product recall in the United Kingdom *(6)*.

In this study, we examined the microbiological quality of organic and conventional sprouting seeds sold through the Internet. Trials also were conducted to evaluate the growth and toxin production of naturally occurring *Bacillus* spp. in sprouts grown in two commonly used home-scale sprouting devices.

MATERIALS AND METHODS

Seed preparation. Five types of sprouting seeds (alfalfa, broccoli, lentil, mungbean, and radish) were procured through Internet communication from a total of 19 companies in the United States during February and March 2004 (Table 1). For each seed type, the procurement included six organic and six conventional (nonorganic) sources as verified by their Internet postings and

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TABLE 1. *Type and source of sprouting seeds procured through the Internet*

Company no.	State	Seed type	Seed source
1	Arizona	Radish	Organic
2	California	Alfalfa, broccoli, mungbean	Organic
3	California	Lentil, radish	Organic
4	Iowa	Alfalfa, broccoli, lentil, mungbean, radish	Organic
5	Idaho	Broccoli, lentil, mungbean, radish	Conventional
6	Illinois	Broccoli, radish	Conventional
7	Illinois	Broccoli, radish	Conventional
8	Illinois	Mungbean, radish	Conventional
9	Maine	Alfalfa, broccoli, mungbean, radish	Conventional
10	New York	Alfalfa, broccoli, lentil, mungbean, radish	Organic
11	New York	Alfalfa, lentil, mungbean	Conventional
12	Oregon	Alfalfa, broccoli, lentil, mungbean	Organic
13	Oregon	Alfalfa, lentil	Conventional
14	Tennessee	Alfalfa, broccoli, lentil, mungbean, radish	Organic
15	Texas	Broccoli, radish	Conventional
16	Utah	Alfalfa, lentil, mungbean	Conventional
17	Utah	Alfalfa, broccoli, lentil, mungbean	Conventional
18	Utah	Alfalfa, lentil	Conventional
19	Washington	Alfalfa, broccoli, lentil, mungbean, radish	Organic

product labels. For the conventional seeds, additional telephone contacts were made to confirm that the seeds were not from organic sources. Upon receipt in the mail, seed packages were refrigerated at 4°C and used for seed evaluation within 2 weeks and for the sprouting study within 3 months. To open seed bags for sampling, one corner of each bag was wiped with ethanol, air dried, and cut with flame-sterilized scissors.

Microbial detection. For seed evaluation, each seed sample (100 g) was mixed and washed with 100 ml of peptone water (0.1%) in a laboratory blender (Masticator Silver, IUL Instruments, Barcelona, Spain) for 60 s. Unless otherwise stated, all culture media were purchased from BioPro (Bothell, Wash.). For total microbial counts, appropriate dilutions of the wash water were pour plated on standard method agar and incubated for 48 h at 35°C. The wash water was surface plated on Baird-Parker agar and incubated for 48 h at 35°C for presumptive *Staphylococcus aureus* counts. Wash water also was surface plated on mannitol–egg yolk–polymyxin agar (MYP; Difco, Becton Dickinson, Sparks, Md.) and incubated for 24 h at 30°C for presumptive *B. cereus* counts *(29).* To differentiate the presumptive *B. cereus* from culturally similar species, representative colonies found on the MYP plates were isolated using nutrient agar, incubated at 30°C, and examined microscopically for positive cell motility and Gram staining on day 1 and for the absence of bipyramidal-shape crystal formation on day 4 with basic fuchsin staining (Fisher Scientific, Fair Lawn, N.J.) *(1).*

Total coliform, fecal coliform, and *Escherichia coli* counts were determined using three-tube most-probable-number (MPN) evaluation with appropriate dilutions of each sample *(29).* After incubation for 24 to 48 h at 35°C, a loopful of culture from positive gassing lauryl sulfate broth tubes was transferred to brilliant green bile broth (BG) and EC-mug broth. After incubation for 24 to 48 h, BG tubes with growth and gas production (at 35°C) confirmed the presence of coliforms and EC-mug tubes with growth (at 45.5°C) and fluorescence under long-wave UV light (336 nm) confirmed the presence of *E. coli (17).*

Salmonella detection was performed using an AOAC-approved method (998.09) for raw foods *(27).* The seed wash water (25 ml) was preenriched in lactose broth (225 ml) at 36°C for approximately 20 h, enriched in Rappaport-Vassiliadis broth at 42°C for approximately 18 h, and postenriched in M broth at 36°C for approximately 7 h. An enzyme-linked immunoassay was performed using a *Salmonella* visual immunoassay (VIA) test kit (Tecra, Frenchs Forest, Australia). For *E. coli* O157 detection, an AOAC performance tested VIA and its manufacturer-recommended enrichment protocol for environmental samples *(26)* were used. The seed wash water (25 ml) was enriched in modified EC broth (225 ml) with novobiocin (Sigma, St. Louis, Mo.) at 42°C for 24 h before the *E. coli* O157 VIA was performed.

Sprouting procedures. For glass-jar sprouting, 50 g of seeds was placed in a sterilized glass jar (1 liter) and soaked with 150 ml of sterilized tap water (from Matoca, Va.) for 8 h at 25°C. After draining, the glass jar was placed horizontally for seed germination at 25°C for 6 days. The glass jar was loosely capped to allow air exchange and covered with a plastic sheet to reduce light exposure. Twice each day, the germinating seeds in the jars were gently rinsed with 250 ml of sterilized tap water for about 30 s. The sprouting samples (10 g) were taken consecutively from the jars at hour 8 (immediately after soaking) and before rinsing on days 2, 4, and 6. Appropriate dilutions of each macerated sample were analyzed for total plate counts and presumptive *B. cereus* counts using methods described above. To evaluate the effect of rinsing during seed sprouting, additional jars of seeds were irrigated daily by adding (without draining) sterilized tap water (25 ml) instead of twice-daily rinsing.

For automatic sprouting, the sprouter (Freshlife Model 2000, Tribest, Santa Fe Springs, Calif.) was disassembled and sanitized with 80°C hot water. After reassembling, 80 g of seeds was placed on a sprouter tray within the sprouter barrel and the water barrel was filled with about 2.5 liters of sterilized tap water. The sprouter was then powered to intermittently sprinkle and recycle water from the water barrel to the sprouter tray for 5 days at 25°C; water in the barrel was replaced daily. Before the daily water changing, water samples (0.1 ml) were obtained from the water barrel on days 1, 3, and 5, and sprout samples (10 g) were obtained from the sprouter tray on day 5. Appropriate dilutions of the water and

TABLE 2. *Microbiological quality of sprouting seeds sold on the Internet*[a]

Microbes	Microbial counts by seed type				
	Alfalfa	Broccoli	Lentil	Mungbean	Radish
Total microorganisms (log CFU/g)					
Organic	3.3 ± 0.4 AB	3.8 ± 0.1 A	2.3 ± 0.2 B[b]	3.1 ± 0.2 AB[b]	3.7 ± 0.5 A
Conventional	3.4 ± 0.1 A	4.1 ± 0.3 A	3.3 ± 0.1 A[b]	4.0 ± 0.2 A[b]	3.5 ± 0.2 A
Presumptive *B. cereus* (log CFU/g)					
Organic	0.6 ± 0.3 A	0.9 ± 0.2 A	0.7 ± 0.2 A	0.8 ± 0.2 A	0.8 ± 0.2 A
Conventional	0.8 ± 0.4 A	1.1 ± 0.2 A	0.8 ± 0.2 A	1.2 ± 0.2 A	1.0 ± 0.2 A
Total coliforms (log MPN/g)					
Organic	−0.5 ± 0.0 A	−0.5 ± 0.0 A	−0.5 ± 0.0 A	−0.4 ± 0.1 A[b]	−0.3 ± 0.2 A
Conventional	−0.2 ± 0.3 AB	−0.3 ± 0.1 A	−0.5 ± 0.0 A	0.5 ± 0.5 B[b]	−0.5 ± 0.0 A

[a] Data are means ± standard errors of testing results with seeds from six vendors. In the same row, means not followed by at least one identical letter are significantly different ($P < 0.05$).

[b] Significant difference between organic and conventional seeds ($P < 0.05$).

sprout samples were individually analyzed for total plate counts and presumptive *B. cereus* counts using methods described above.

***Bacillus* diarrheal toxin detection.** The *Bacillus* diarrhea enterotoxin VIA (Tecra) was used to detect the enterotoxins produced by the presumptive *B. cereus.* To determine the enterotoxin-producing capability of the isolated presumptive *B. cereus* strains, a loopful of each isolate was enriched with 10 ml of brain heart infusion plus 0.1% glucose for 16 to 18 h at 36°C before immunoassay according to the manufacturer's instructions *(9, 25).* To determine the presence of *Bacillus* diarrheal enterotoxins in sprouts, each sprout sample (10 g) was processed with 30% sodium azide (Fisher) to inhibit the naturally occurring peroxidase activities from the macerated sprouts before the assay *(25).*

Statistical analysis. Microbial populations were assessed by two-way analysis of variance using SigmaStat software (version 3.0, SPSS Inc., Chicago, Ill.). Differences were considered significant at $P \leq 0.05$.

RESULTS

Seed survey. The average total plate counts for alfalfa, broccoli, lentil, mungbean, and radish seeds were 3.3 ± 0.2, 4.0 ± 0.2, 2.8 ± 0.2, 3.5 ± 0.2, and 3.6 ± 0.2 log CFU/g, respectively. Lentil seeds had lower total microbial counts ($P < 0.01$) than did broccoli, mungbean, and radish seeds. Conventional lentil and mungbean seeds had higher total microbial counts ($P < 0.05$) than did their organic counterparts (Table 2). When the results from the five types of seeds were grouped together, the average total microbial count of organic seeds was about 0.4 log cycles lower ($P < 0.05$) than that of the conventional seeds. There was no significant interaction ($P = 0.13$) between seed source (organic or conventional) and seed type.

The presumptive *B. cereus* counts were 0.7 ± 0.2, 1.0 ± 0.1, 0.8 ± 0.1, 1.0 ± 0.2, and 0.9 ± 0.1 log CFU/g for alfalfa, broccoli, lentil, mungbean, and radish seeds, respectively. Overall, presumptive *B. cereus* was found in 57 of the 60 seed samples at ≥1 CFU/g. Significant difference in the presumptive *B. cereus* counts was not found among the five seed types or between the two seed sources (Table 2). Microscopic observations confirmed that the representative colonies (two from each positive seed sample) on MYP plates were exclusively gram-positive bacilli. No isolates resembling *Bacillus anthracis* (a nonmotile bacterium) were observed, and only 3 of 114 isolates resembling *Bacillus thuringiensis* produced bipyramidal crystals during sporulation. Immunoassays on the presumptive *B. cereus* isolates positively confirmed the presence of *Bacillus* diarrheal toxin-producing *Bacillus* spp. in each of the seed samples that were gram positive in MYP culture.

The average total coliform counts (which included counts from both organic and conventional seeds) were −0.3 ± 0.1, −0.4 ± 0.1, −0.5 ± 0.0, 0.0 ± 0.3, and −0.4 ± 0.1 log MPN/g for alfalfa, broccoli, lentil, mungbean, and radish seeds, respectively. A significant difference in the counts ($P < 0.05$) was found between lentil and mungbean seeds. Conventional mungbean seed had about 1.0 log cycle higher most probable numbers of coliforms ($P = 0.01$) than did its organic counterpart (Table 2). When the results from the five types of seeds were grouped together, the average coliform population in organic seeds was about 0.3 log cycles less ($P < 0.05$) than that in the conventional seeds. However, there was a significant interaction ($P < 0.05$) between seed source (organic or conventional) and seed type. No *Salmonella, E. coli* O157, other fecal coliforms, or *S. aureus* was found on any seeds.

Sprout evaluation. When seeds from three organic and three conventional venders were sprouted in glass jars, no growth of presumptive *B. cereus* was found during the entire sprouting process in alfalfa, lentil, or mungbean sprouts (Fig. 1). However, the average counts of presumptive *B. cereus* increased about 3.8 and 3.5 log cycles, respectively, for radish and broccoli sprouts after 2 days. The presumptive *B. cereus* numbers in radish and broccoli sprouts remained constant ($P > 0.05$) after day 2. Using microscopy and immunoassay, we confirmed that all representative isolates from the radish and broccoli sprouts sampled at day 6 were enterotoxigenic *Bacillus* spp. However, no *Bacillus* diarrheal toxin was detectable in any radish or broccoli sprouts produced using the glass-jar method.

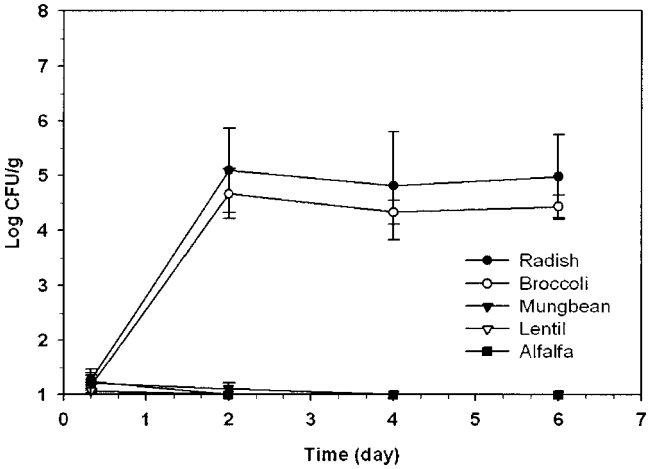

FIGURE 1. *Concentrations of presumptive* Bacillus cereus *in sprouts during glass-jar sprouting. Seeds were soaked in water for 8 h before sprouting at 25°C and then rinsed with water twice daily. Data represent means ± standard errors (*n = 6*).*

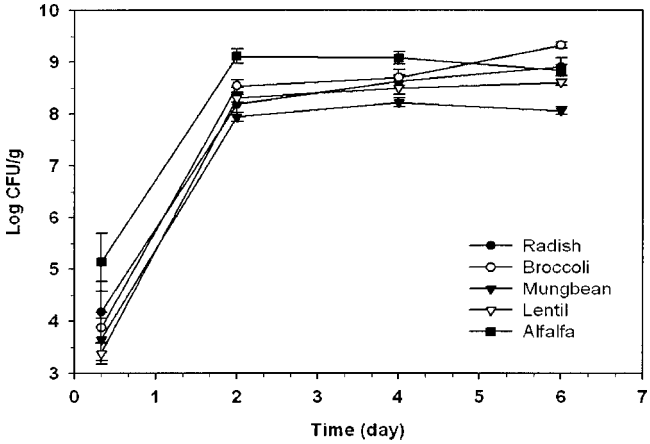

FIGURE 2. *Concentrations of total microbes in sprouts during glass-jar sprouting. Seeds were soaked in water for 8 h before sprouting at 25°C and then rinsed with water twice daily. Data represent means ± standard errors (*n = 6*).*

The total microbial counts increased about 4.0 to 5.5 log cycles during seed sprouting (Fig. 2). Major increases in the total plate counts occurred during the first two sprouting days. At day 6, mungbean sprouts had significantly lower total microbial counts (8.1 log CFU/g) than did broccoli sprouts (9.3 log CFU/g).

Similar to the results of glass-jar sprouting, no growth ($P > 0.05$) of presumptive *B. cereus* was found when sprouting alfalfa or lentil seeds in the automatic system (Table 3). The growth of presumptive *B. cereus* on the radish, broccoli, and mungbean sprouts was significant, and presumptive *B. cereus* reached to 3.3 ± 0.4, 3.3 ± 0.2, and 2.5 ± 0.2 log CFU/g, respectively, by day 5. Rinse water in the sprouting system had about 0.9 log cycle lower presumptive *B. cereus* counts than did the sprouts. Using microscopy and immunoassay, we confirmed that all representative isolates from the radish, broccoli, and mungbean sprouts sampled at day 5 were enterotoxigenic *Bacillus* spp. However, *Bacillus* diarrheal toxin was not detected in radish, broccoli, or mungbean sprouts produced using the automatic sprouting system. The total microbial counts of the rinse water in the sprouting system reached 7.9 log CFU/g in the first sprouting day (Fig. 2). At day 5, mungbean sprouts had significantly lower total microbial counts (approximately 7.6 log CFU/g) than did all other sprouts (approximately 8.7 log CFU/g).

When the sprouting seeds in glass jars were irrigated not by rinsing but by daily irrigation with a small amount of water (25 ml), the growth of total microorganisms and enterotoxigenic *Bacillus* spp. in the sprouts was greater (Table 4), but *Bacillus* diarrheal toxin was not detected in the radish sprouts.

DISCUSSION

More people are using e-commerce for purchasing goods, including food and horticultural items. In this study, we examined seeds for home-sprouting use that were sold in the United States through the Internet. Despite the fact that contaminated sprouts have caused numerous *Salmonella* and *E. coli* O157:H7 outbreaks *(3, 4, 7, 15, 20, 24)*, we did not find fecal coliforms (including *Salmonella* and

TABLE 3. *Total microbial and presumptive* Bacillus cereus *counts during seed sprouting in an automatic sprouting system*[a]

Microbes	Counts by seed type				
	Alfalfa	Broccoli	Lentil	Mungbean	Radish
Total microorganisms					
Water at day 1	8.3 ± 0.1 A	7.8 ± 0.1 A	7.8 ± 0.2 A	7.9 ± 0.2 A	7.9 ± 0.2 A
Water at day 3	8.0 ± 0.3 AB	8.2 ± 0.2 A	7.3 ± 0.2 BC	6.9 ± 0.2 C	7.8 ± 0.1 AB
Water at day 5	7.4 ± 0.2 AB	7.8 ± 0.3 A	7.5 ± 0.3 AB	6.9 ± 0.1 B	7.8 ± 0.4 A
Sprouts at day 5	8.8 ± 0.1 A[b]	8.7 ± 0.2 A[b]	8.8 ± 0.2 A[b]	7.6 ± 0.2 B[b]	8.6 ± 0.1 A[b]
Presumptive *B. cereus*					
Water at day 1	1.3 ± 0.2 A	1.1 ± 0.1 A	1.0 ± 0.0 A	1.4 ± 0.2 A	2.4 ± 0.2 B
Water at day 3	1.1 ± 0.1 A	1.7 ± 0.2 A	1.0 ± 0.0 A	1.1 ± 0.1 A	2.5 ± 0.4 B
Water at day 5	1.3 ± 0.2 A	2.4 ± 0.2 B	1.1 ± 0.1 A	1.5 ± 0.2 A	2.6 ± 0.2 B
Sprouts at day 5	1.3 ± 0.2 A	3.3 ± 0.2 C[b]	1.0 ± 0.0 A	2.5 ± 0.2 B[b]	3.3 ± 0.4 C[b]

[a] Data are means ± standard errors (log CFU per gram) of testing results with seeds from six vendors. In the same row, means not followed by at least one identical letter are significantly different ($P < 0.05$).

[b] Microbial counts in sprouts were significantly higher than those in nonsprouted seeds ($P < 0.05$).

TABLE 4. *Effect of rinsing on the microbial populations of radish sprouts sprouted in a glass jar*[a]

Microbes	Seeds A		Seeds B	
	Rinsing	Nonrinsing	Rinsing	Nonrinsing
Total microorganisms	9.2 ± 0.1[b]	9.7 ± 0.1[b]	8.1 ± 0.0	8.3 ± 0.3
Presumptive *B. cereus*	4.9 ± 0.2[b]	6.1 ± 0.1[b]	7.1 ± 0.1[b]	8.1 ± 0.1[b]

[a] Data are means ± standard errors of testing results (log CFU/g) with seeds from two vendors: seeds A from company 14 and seeds B from company 5.

[b] Significant difference for the same seeds between rinsing and nonrinsing treatments ($P < 0.05$).

E. coli O157:H7) or *S. aureus* in any of the 60 survey samples (including five types of sprouting seeds, each from 12 different vendors). In general, the concentration of total coliforms was low (≤1 CFU/g) in Internet-procured sprouting seeds. Sprouting seeds could be produced without major contamination using current good agricultural practices for seed production and handling. Nevertheless, previous reports indicate that pathogens can be dispersed heterogeneously on seeds and capable of rapid growth during sprouting *(7, 18).* Consequently, seed testing alone is inadequate for assuring sprout safety.

Although human pathogens may be transferred to adjacent crops via a variety of routes before or during organic soil amendment *(30),* data in the literatures do not support the idea that organic produce poses a greater risk of pathogen contamination than does conventional produce *(12, 14, 16, 22).* The results of our microbial survey on sprouting seeds sold on the Internet indicate that organic seeds in general are less contaminated with total microorganisms and coliforms than are conventional seeds. Additional research at seed production and handling stages is needed to confirm that organic practices can help minimize seed surface contamination. Because no fecal coliforms were detected and about the same concentration of presumptive *B. cereus* was found on organic and conventional seeds, the current study provides no evidence the organic and conventional seeds differ in risk posed to the consumer.

B. cereus is a spore-forming foodborne pathogen widely distributed in the agricultural environment and food crops *(23).* In our seed survey, we found high prevalence (≥1 CFU/g on 95% of the 60 samples) of presumptive *B. cereus* in sprouting seeds sold by Internet businesses for home sprouting. In a previous survey of health food stores in the Washington, D.C., area, *B. cereus* was found at ≥3 MPN/g in 69% of the 98 samples tested *(10).* Other investigations also revealed the association between *B. cereus* and plants that produce sprouting seeds *(5, 8, 28).* Kim et al. *(13)* reported the presence of enterotoxigenic *B. cereus* on mungbean sprouts in Korea.

In the present study, only one (<2%) sample from the Internet vendors had a presumptive *B. cereus* concentration of 100 CFU/g, in contrast to 13% of the seed samples from the local health food stores surveyed by Harmon et al. in 1987, who confirmed *B. cereus* at concentrations greater than 100 CFU/g. The concentrations of presumptive *B. cereus* we found in the sprouting seeds were much lower (10^5 CFU/g) than that considered sufficient to cause food poisoning directly *(10).*

In the sprouting study, the concentrations of presumptive *B. cereus* in the radish and broccoli seeds increased significantly during sprouting when using either a glass jar or the automatic sprouting device. The growth of enterotoxigenic *Bacillus* spp. in radish sprouts exceeded 10^5 CFU/g in 2 days when sprouted in the glass jar, and the populations continued to be high despite the twice-daily rinsing. However, no significant growth was observed for alfalfa and lentil sprouts during the entire sprouting period in either the glass jar or automatic sprouting system. Enterotoxigenic *Bacillus* spp. growth in mungbean sprouts was observed for the automatic sprouting system, but no growth was found for the glass jar system. These results suggest that both seed type and sprouting process can affect the safety of sprouts in terms of contamination with enterotoxigenic *Bacillus* spp. Previous studies have indicated that seed type may influence the growth of *Salmonella* on sprouting seeds *(18).* Previous research also revealed that a natural compound, canavanine, extracted from seeds of alfalfa and many legumes (but not broccoli and radish) inhibits the growth of *B. cereus (5, 21).* The presence of antimicrobial agents (such as canavanine) in selected seeds offers a possible explanation for the observed difference in *Bacillus* growth during sprouting. Because the automatic sprouting system uses and drains more water each day than the glass jar system, the dilution effect on the antimicrobial agent(s) released from seeds may explain why mungbean sprouts grown in the automatic sprouting system supported the growth of enterotoxigenic *Bacillus* spp. but those sprouts grown in the glass jar did not. Both radish and broccoli seeds belong to the family *Brassicaceae,* and it would be interesting to determine whether enterotoxigenic *Bacillus* spp. also grow strongly in other sprouting seeds in the same family. We did not find *Bacillus* diarrheal enterotoxin in any of the sprouts that showed significant growth of enterotoxigenic *Bacillus* spp., even radish sprouts grown in glass jars without adequate rinsing. Additional research is needed to determine the factors affecting the production or accumulation of *Bacillus* diarrheal and emetic toxins on sprouts.

The microbial quality of sprouting seeds can be influenced by seed type and source. The organic sprouting seeds we purchased from Internet sources were less or equally loaded with total microorganisms and coliforms compared with the conventional seeds. Enterotoxigenic *Bacillus* spp. were highly prevalent in all types and sources of sprouting seeds. However, its growth during hydroponic sprouting was influenced by both seed type and sprouting method.

Although the enterotoxigenic *Bacillus* spp. did not produce or accumulate appreciable amounts of diarrheal toxins in the home-sprouting devices tested in this study, the potential for growth of these bacteria in radish and broccoli seeds is evident and populations may reach dangerous levels (10^5 CFU/g) when glass jars are used for sprouting.

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