

Contamination of Potatoes with Spore-forming Bacteria

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Question:

We appreciate if you find any initial contamination levels for potatoes, tubers and root crops, preferably if you can find information related to *Clostridium botulinum* and spore forming microorganisms.

FRI response:

As detailed below (and depending on where they are grown and other factors), potatoes and other tubers or root crops can carry a variety of spore-forming microorganisms (including *Clostridium botulinum*) that are present in soils (Sugiyama et al., 1981; Doan and Davidson, 2000a; Kõiv et al., 2019).

CLOSTRIDIUM BOTULINUM

C. botulinum types A or B may be found on root vegetables (Austin, 2016).

The type of *C. botulinum* spores in soils varies by geography, with type A spores found in soils west of the Rocky Mountains and type B spores (usually proteolytic) found in the Eastern U.S. (Austin, 2016).

Potato products have been associated with numerous *C. botulinum* outbreaks, as discussed in an earlier review (Doan and Davidson, 2000b). More recently, *C. botulinum* outbreaks have been associated with commercial produced roasted potatoes (Gayou et al., 2022) and potato salad made from improperly canned potatoes (McCarty et al., 2015).

Prevalence and contamination levels of root vegetables

A 2016 review commented that prevalence rates for *C. botulinum* in foods are not readily available, as assessing the presence of the organism in foods (usually by enriching for spores, broth culture, and then detection of botulinum toxin in the culture supernatant) is expensive and difficult (Austin, 2016). However, this review did say that when *C. botulinum* is present in food, it is typically present at low levels (<1 to 1000 spores/kg) (Austin, 2016).

A 1981 study from the Food Research Institute found type A *C. botulinum* contamination (assessed by presence of toxin in enrichment cultures) on 1 of 2, 1 of 3, and 1 of 4 potatoes (peels) in three separate trials using potatoes from one supermarket (Sugiyama et al., 1981). In a separate part of the same study, as little as 10 spores of *C. botulinum* inoculated on potatoes was sufficient to make them botulinogenic.

A 1994 report found no botulinum positive samples present in 48 samples of refrigerated raw carrots and 78 samples of refrigerated cooked potatoes purchased at retail (Gibbs et al., 1994). Similarly, a 1995 study found no *C. botulinum* spores isolated from MAP fresh carrots or onions (7 and 4 packages, respectively)(Lilly et al., 1996).

A 2001 French study used a PCR-based assay to look for *C. botulinum* type A, B, or E in a variety of vegetables, including fresh vegetables (including potatoes and other root vegetables) and in cooked potato purees (Braconnier et al., 2001). All samples in this study were negative for *C. botulinum*, which the authors concluded meant the prevalence of *C. botulinum* in these raw vegetables and vegetable purees was probably low. The sample numbers in this study were not large (2-16 for each type of sample), and the limits of detection for their assays were 8 spores/g for type A, <1 spores/g for proteolytic type B, <21 spores/g for nonproteolytic type B, and <0.1 spores/g for type E.

A 2012 French paper found 1 of 128 raw carrot samples to contain *C. botulinum* spores by the PCR-based BoNT GeneDisc array assay (Sevenier et al., 2012). This study also enumerated mesophilic anaerobic spores (minimum 1.7 log cfu/g; maximum 6.1 log cfu/g, mean 3.8 log cfu/g) and thermophilic anaerobic spores (minimum <1 log cfu/g; maximum 4.6 log cfu/g, and mean 2.7 log cfu/g) for 121 raw carrot samples.

A 2016 paper explored statistical properties of nonproteolytic *C. botulinum* spores in foods using a Bayesian model approach based on data collected from an extensive literature review (Barker et al., 2016). The authors commented that the actual *C. botulinum* spore loads in food materials are generally very small and very difficult to detect, which leads to observations that fail to detect the spores. Their modeling results suggest that the probability that the nonproteolytic *C. botulinum* spore load in plant foods exceeded 1 spore/kg was 0.82; that it exceeded 10 spores/kg was 1.1×10^{-2} , and that it exceeded 30 spores/kg was 5.3×10^{-15} spores/kg (Barker et al., 2016).

A detailed 2000 review of the microbiology of potatoes cited an older Swedish study that found type E *C. botulinum* in 27 of 40 potato peel samples collected from various parts of Sweden and in each of three fresh potato samples from Israel (Doan and Davidson, 2000a). The review also discusses older studies in which processed raw potatoes were found to harbor Gram-positive bacilli as well as a pectolytic clostridia (presumed to be *Clostridium butyricum*).

CLOSTRIDIUM PERFRINGENS

Soil attached to commercial potatoes was assessed by PCR for the presence of *C. perfringens* spores in a 2022 Japanese study (Hashimoto et al., 2023). A total of 288 *C. perfringens* isolates were collected from

25 potato samples, and 83% of potato samples contained *C. perfringens* spores. Of the 288 *C. perfringens* isolates, 22% contained *cpe*, the toxin gene associated with *C. perfringens* food poisoning.

BACILLUS SPP.

The 2000 review on potato microbiology states that *B. cereus* is the main foodborne pathogen associated with dehydrated potato products (Doan and Davidson, 2000a). The review cites a study in which 40% of dried potato samples contained *B. cereus*, with counts ranging from 2 to 3 log CFU/g. More recent studies have also noted high rates of *B. cereus* group species contamination in mashed potato powder (up to 92%) (Heini et al., 2018).

A 2007 paper from New Zealand found dehydrated potato products contained *Bacillus cereus* at a prevalence of 10-40% and at numbers generally less than 3 log CFU/g (Turner et al., 2006; King et al., 2007).

A 2016 paper from Korea found a mean of 3 log CFU/g *B. cereus* in samples of retail raw potatoes (Luo et al., 2016).

A 2010 paper from Argentina found *B. cereus*, *B. licheniformis*, and *B. mycoides* in raw potato samples at 1×10^2 to 7×10^2 , 1×10^2 to 2×10^2 , and 1×10^2 to 2.8×10^3 log CFU/g (Fangio et al., 2010).

A Swiss study found that among 20 purchased samples of mashed potato powders (representing 7 different brands), at least one *B. cereus sensu lato* isolate was found in each tested brand (Burtscher et al., 2021). *B. cytotoxicus* was found in 19 of the 20 samples. However, the hazardous potential of the *B. cytotoxicus* isolates was not clear.

A German study also found a high prevalence of *Bacillus cytotoxicus* (which the authors describe as a thermotolerant “potential foodborne pathogen”) in various commercial potato products, including 71% of dehydrated potato products but only 10% of raw potatoes (Contzen et al., 2014). Similar high prevalence of *B. cytotoxicus* was observed in a 2018 Swiss study (Heini et al., 2018).

A 2021 Colombian study found *Bacillus cereus sensu lato* present in 57% of cassava (a tuber) starch samples (Sánchez-Chica et al., 2021).

GENERAL MICROBIOTA OF POTATOES AND ROOT VEGETABLES

The diversity and number of bacteria found in the peel of root vegetables is higher than that in the pulp (Kõiv et al., 2019).

The dominant bacterial phyla in all plant root compartments are often the same, with the most abundant being Proteobacteria, followed by Actinobacteria, Bacteroidetes, and Firmicutes (Kõiv et al., 2019).

A 2012 paper quantitate the numbers of spores (thermophilic anaerobes, thermophilic aerobes, and mesophilic aerobes in various unspecified dehydrated vegetables, but no quantitation of individual isolates was noted (Postollec et al., 2012).

Numerous other papers which explore potato microbiomes including those in the following references: (Lauridsen and Knøchel, 2003; van Overbeek and van Elsas, 2008; Manter et al., 2010; Kõiv et al., 2015; Luo et al., 2016; Kõiv et al., 2019; Cangioli et al., 2022; Li et al., 2022; Faist et al., 2023)

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