Prevalence and Levels of Spore-forming Pathogens in Spices

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April 2021

Introduction

Food companies use various ingredients in their products, some of which can be harmful to the consumers if they are contaminated with pathogenic microorganisms. Spices, as minimally processed plant material, are a category of ingredients which can contain pathogens in unsafe amounts. Spore-forming pathogens are of particular concern with spices because their spores may be significantly more resistant to treatments such as irradiation than their vegetative counterparts.

Bacterial spores in a product can lead to foodborne disease outbreaks. For example, in 2016 there was an outbreak of gastrointestinal illnesses from a catered lunch shared by many. These illnesses stem from the *C. perfringens* and its enterotoxin (1).

The purpose of this research is to provide food companies with microbiological profiles of different spices that have been reported previously in the scientific literature. We focused on three key spore-forming bacteria: *Bacillus cereus, Clostridium perfringens, and Clostridium botulinum.* This information may be of use to the food industry when they evaluate potential hazards associated with spices used in their products as part of their Food Safety Plans.

Methods

To collect data for this analysis, scientific literature databases including Google Scholar and Web of Science were used to search for relevant studies. Terms relating to either the spice or pathogen were searched to find relevant literature. Studies that were not written in English were not analyzed, but papers from all parts of the world were reviewed. Documents published from 1979 to 2021 were evaluated in the process.

Once identified, full-length articles were retrieved and reviewed for data concerning the microbiological profiles of spices, mainly pertaining to *B. cereus* and *C. perfringens*. The information found in the documents were organized into categories in a Microsoft Excel[®] spreadsheet for ease of use. Geographic region was recorded for studies that indicated a particular region of origin and omitted for those spices that made no mention of geographic region.

None of the literature reviewed explicitly stated that the cells they were measuring were spores or vegetative cells, therefore the levels stated in this paper could be either.

Results

Black Pepper

Nine studies were identified which measured the prevalence and/or levels of spore-forming pathogens in black pepper. In five out of nine studies, at least 50% of the black pepper samples tested positive for *B. cereus*. The range of *B. cereus* levels found ranged from 0 to 6 log colony forming units per gram (CFU/g) (1-8).

In contrast to the *B. cereus* findings, smaller proportions (0 to 40%) of black pepper samples tested positive for *C. perfringens*, with levels ranging from 0.9 to 5 log CFU/g (spores or vegetative cells). Samples from India were 17 to 40% positive for *C. perfringens* (2-4).

Anise seeds

Two studies found a high percentage of anise seed samples contaminated with *B. cereus*, with 20% to 80% of samples containing the pathogen (2, 5). Levels of *B. cereus* in these samples ranged from 2 to 6.2 log CFU/g.

Comparatively, anise seed samples containing *C. perfringens* ranged from 1.0 to 2.0 log CFU/g with only 20% testing positive for this pathogen. Anise seeds tend to contain *B. cereus* more frequently at a higher level than *C. perfringens* (2).

Cumin

Of the eight studies identified that measured the prevalence of spore-forming pathogens in cumin, eight tested positive for *B. cereus* (2, 3, 6-11). The percentage of the samples that tested positive for the pathogen ranged from 8 to 100%. Samples with cumin originating from Australia and India tested at least 86% positive for *B. cereus*. The levels of *B. cereus* found ranged from 367 to 6 log CFU/g.

Eight studies measured *C. perfringens* prevalence in cumin. Up to 38% of samples tested positive for *C. perfringens*. The range of *C. perfringens* levels found ranged from 0 to 2 log CFU/g (2, 3, 12, 13).

Chili

India had the highest percentage of samples testing positive for detecting *B. cereus* at 80%. Across all studies, *B. cereus* levels ranged from 0 to 6 log CFU/g (3, 5, 6, 14, 15), with the higher levels from chili powder samples from retail stores in Pakistan. Meanwhile while testing for *C. perfringens*, only 20% of samples tested positive with levels ranging from 0 to 3 log CFU/g (3, 6, 16).

Turmeric

A wide range of *B. cereus* levels (from a total absence up to a maximum of 6 log CFU/g) were found in turmeric samples across various studies (2, 3, 5, 17). When testing turmeric samples for *C. perfringens*, various studies found microbiological levels ranging from 0 to 3 log CFU/g (3, 4).

Cinnamon

Of the studies identified that measured the prevalence of spore-forming pathogens in cinnamon, ten measured *B. cereus* content. In five of those studies, at least 57% of samples tested positive for *B. cereus*. The range of *B. cereus* levels found ranged from 1 to 5.3 log CFU/g (2, 3, 5, 7, 8, 14, 15, 18, 19).

Only two studies tested for *C. perfringens* content in cinnamon samples. Up to 40% of samples tested positive for containing *C. perfringens*, however, samples sold in India had a higher percentage test

positive than samples from other regions. The range of microbiological levels of *C. perfringens* in cinnamon was from 0 to 4 log CFU/g (2, 3, 16).

Basil

Eight studies measured the prevalence and/or levels of spore forming pathogens present in basil samples. Seven studies measured *B. cereus*; three of the studies had 100% of their samples test positive for *B. cereus*. The range of *B. cereus* levels found varied from 1 to 4 log CFU/g (*3, 5, 8, 14, 18-20*).

Coriander

Ten studies identified measured the prevalence of spore-forming pathogens in coriander, while seven investigated *B. cereus*. A study from Poland reported samples with the highest percentage of positive samples at 100%. Three of the studies included samples testing positive at a prevalence of 40% or higher. *B. cereus* levels found ranged from 1 to 5.1 log CFU/g (*2*, *3*, *7*, *9*, *10*, *14*, *20*).

Comparatively, three studies measured the prevalence and/or levels of *C. perfringens* in samples of coriander. In contrast to samples testing for *B. cereus*, these samples exhibited a lower prevalence (\leq 40%) for *C.* perfringens, with levels ranging from 0 to 3 log CFU/g (2, 3, 13).

Mustard Seed

Four studies tested for *B. cereus* prevalence and/or levels in samples of mustard seed. Up to 80% of samples tested positive for *B. cereus* across studies. *B. cereus* levels found in the samples ranged from 2 to 5.7 log CFU/g (2, 3, 5, 7).

Two studies looked for the prevalence and/or levels of *C. perfringens* in samples of mustard seed. In those studies, fewer than 20% of samples tested positive for *C. perfringens*. The levels of *C. perfringens* in mustard seed ranged from 0 to 2.7 log CFU/g (2, 3).

Thyme

Six studies measured the prevalence and/or levels of *B. cereus* in samples of thyme. The three that reported prevalence all found that 100% of samples contained *B. cereus*. The microbiological levels of *B. cereus* found in thyme ranged from 2 to 2.4 log CFU/g across these six studies (*3, 5, 8, 11, 14, 20, 21*).

One of the studies reported that 50% of thyme samples contained *C. perfringens*. The mean microbiological levels of *C. perfringens* in the thyme samples were 3.6 log CFU/g (3, 13).

Oregano

Nine studies measured the prevalence of spore-forming pathogens in oregano, including seven which measured *B. cereus*. Across these studies, 43%-100% of the samples tested positive for the pathogen. The range of levels of *B. cereus* found in the oregano samples varied from 1 to 2 log CFU/g (*3*, *5*, *7*, *14*, *15*, *18-20*).

Two studies measured the prevalence and/or levels of *C. perfringens* in samples of oregano. Up to 53% of samples were reported to test positive for the pathogen, with levels of *C. perfringens* ranging from 2 to 4 log CFU/g. Mexico reported samples sourced from their retail stores as having the lowest prevalence of *C. perfringens*, reporting 101 to 300 CFU/g (*3, 16*).

Cloves

Of seven studies measuring the prevalence and/or levels of spore-forming pathogens in samples of cloves, five of them tested for *B. cereus*. The range of *B. cereus* levels reported in the samples tested ranged from 0 to 4 log CFU/g. Samples tested up to 75% positive for *B. cereus* (2, 3, 8, 14).

Two studies measured the prevalence and/or levels of *C. perfringens* in cloves. Levels of the pathogens were reported to range from 1 to 2 log CFU/g. However, in contrast to *B. cereus* prevalence in cloves, $\leq 17\%$ of cloves samples were positive for *C. perfringens* (2, 3).

Paprika

Five studies measured the prevalence and/or levels of *B. cereus* in paprika. Across studies, 38% of paprika samples tested positive for *B. cereus*. Samples of paprika demonstrated a wide range of *B. cereus* levels, from 1 to 6 log CFU/g (7, 14, 18, 19, 22).

The studies reviewed did not analyze *C. perfringens* content in samples of paprika. In the future, more research is needed with respect to the prevalence and/or levels of *C. perfringens* in paprika.

Onion Powder

Three studies measured the prevalence and /or levels of *B. cereus* in onion, with levels ranging from 0 to 5 log CFU/g. Across studies, 23% of onion samples tested positive for *B. cereus* content. Onion powder samples sourced from Australia had the highest prevalence of *B. cereus* (5, 7, 22).

More studies need to be published studying the prevalence and/or levels of *C. perfringens* in onion. There is not enough information to make any conclusion on the relationship between onion and *C. perfringens*.

Conclusions

Originally, this study was meant to assess the microbiological profiles of *B. cereus*, *C. botulinum*, and *C. perfringens* in spices. However few studies were identified which examined the prevalence and/or levels of *C. botulinum* in spices. More studies need to be published to come to any conclusion regarding *C. botulinum* and its relationship with spices. *C. botulinum*'s toxin and its status as a select agent has made studying this organism more difficult in recent years.

We were, generally, able to assess *B. cereus* and *C. perfringens* content in multiple spices because they had sufficient data. In general, among all the spices, *B. cereus* was more prevalent than *C. perfringens* in samples of spices.

B. cereus was found at the highest levels in samples of black pepper, cumin, chili, turmeric, and paprika. *C. perfringens* was found in, relatively, higher quantities in samples of black pepper, cinnamon, and oregano.

Frequent, high levels of spore-forming pathogens in spices are a significant issue the food industry needs to understand when spices are used as ingredients in their products so that effective but not unnecessary preventive controls are in place. Control over spore-forming pathogens in spices helps keep society safe and may reduce production costs, increase yield, and increase shelf life of products. Existing assessments of spore-forming pathogenic content are, however, irregular, and inconsistent. More work may be necessary to properly assess the risk assumed when using spices.

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