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Clostridium botulinum and Its Control in Low-Acid Canned Foods

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Abstract *Clostridium botulinum* spores are widely distributed in nature. Type A and proteolytic type B bacteria produce heat-resistant spores that are primarily involved in most of the food-borne botulism outbreaks associated with low-acid canned foods. Food-borne botulism results from the consumption of food in which *C. botulinum* has grown and produced neurotoxin. Growth and toxin production of type A and proteolytic type B in canned foods can be prevented by the use of thermal sterilization alone or in combination with salt and nitrite. The hazardousness of *C. botulinum* in low-acid canned foods can also be reduced by preventing post-process contamination and introducing hazard analysis and critical control point (HACCP) practices during production. Effectiveness of non-thermal technologies such as high pressure processing with elevated process temperatures on inactivation of spores of *C. botulinum* will be discussed.

Keywords: *Clostridium botulinum*, food-borne botulism, low-acid canned foods

Introduction

C. botulinum spores and toxins are the causative agents of a neuroparalytic disease called 'botulism'. The word 'botulism' comes from the Latin word for sausage (*botulus*). In 1896, the causative organism of botulism was first isolated by E.P.M. Van Ermengen (1) from a large outbreak (involved 23 cases with 3 deaths) in Ellezelles, Belgium involving inadequately cured ham. Van Ermengen named the causative organism *Bacillus botulinus* because the patients of this outbreak showed the same symptoms as those of blood-sausage poisoning. In the 1920s, the organism became known as *C. botulinum*. It is a Gram-positive, spore-forming, rod-shaped obligate anaerobe. To this day, only the *C. botulinum* and *C. butyricum* types have been associated with food-borne botulism. Processors and consumers must take control measures to eliminate these species or inhibit their growth and toxin production in food(s) to prevent food-borne botulism. This paper presents a brief overview of the background, distribution, related outbreaks, and control of *C. botulinum* in low-acid canned foods. Several books, book chapters, and reviews have been published about *C. botulinum*, i.e., Hauschild, 1989 (2); Rhodehamel *et al.* 1992 (3); Hauschild and Dodds, 1993 (4); ICMSF, 1996 (5); Bell and Kyriakides, 2000 (6); and Lund and Peck, 2000 (7).

Clostridium botulinum Types

Seven types (A, B, C, D, E, F, and G) of *C. botulinum* are recognized based on the antigenic specificity of their toxins. However, all types of *C. botulinum* produce protein neurotoxins that have similar effects on an affected host. The types of *C. botulinum* differ in their tolerance to salt and water activity (a_w), minimum growth temperatures, and heat resistance of their spores (Table 1) (2, 3, 8). The *C. botulinum* types are divided into four groups, designated by Roman numerals and based on their metabolic and serological similarities. This grouping is further supported by the morphology of inducible temperate phages, the activity spectra of boticins, and the distinct host ranges of wide-type phages (3). Group I consists of the strongly proteolytic strains and includes all type A and proteolytic strains of type B and F. Another *Clostridium* species, *C. sporogenes*, is culturally identical to group I strains of *C. botulinum*, but is non-toxigenic. Group II is comprised of nonproteolytic strains and includes all type E and nonproteolytic strains of B and F. Group III strains are nonproteolytic and culturally complex; these include all strains of type C and D. Group IV consists of weakly proteolytic and nonsaccharolytic type G strains. Types A, B, E, and F (i.e., Groups I and II) are involved in human botulism; type C causes botulism in birds, turtles, cattle, sheep, horses, mink, and other animals; and type D is associated with forage poisoning of cattle and sheep in Australia and South Africa. No outbreaks of type G have been reported; however, type G has been isolated from soils in Argentina and from cases of sudden, unexpected death in humans and infants (9, 10). In addition to these types, *C. baratii* (produces type F toxin) and *C. butyricum* (produces type E toxin) have been isolated and reported to be implicated in human botulism. Hall *et al.* (11) isolated an organism associated with infant botulism that produced type F toxin, but phenotypically resembled *C. baratii*. *C. butyricum* was first identified in 1986 in association with two cases of type E infant botulism in Rome, Italy based on phenotypic resemblance (12-14).

Distribution of *C. Botulinum*

Spores of *C. botulinum* are found in all parts of the world. They are widely distributed in cultivated and forest soils, a variety of raw agricultural products including fruits and vegetables as a result of their contact with soil, shore, and

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Table 1. Growth characteristics and foods involved in outbreaks of *Clostridium botulinum* types A, B, E, and F and *Clostridium butyricum*[1)]

Characteristics	Proteolytic *C. botulinum* types A, B, and F	Nonproteolytic *C. botulinum* types B, E, and F	Neurotoxigenic *C. butyricum*
Neurotoxin types produced	A, B, and F	B, E, and F	E
Minimum growth temperature (°C)	10-12	3.0	10-15
Minimum growth pH	4.6	5.0	4.0-5.2
Minimum water activity (a_w)	0.935	0.97	-
Inhibitory salt (NaCl) concentration (%)	10	5-6	-
Thermal D values for endospores (Min)	$D_{100°C}$ = >15	$D_{100°C}$ = <0.1	$D_{100°C}$ = <1-5
Foods involved in outbreaks	Home canned, faulty commercial processed foods	Fermented marine products, dried fish, vacuum packaged fish	Vegetable-based foods in Asia

[1)]Compiled from references, Hauschild (2); Rhodehamel *et al.* (3); Peck (8).

bottom deposits of streams, lakes, and coastal waters, in the gills and viscera of crabs and other shellfish, and in the intestinal tracts of fish and animals (3, 5, 7, 15). Comprehensive surveys on the distribution and occurrence of *C. botulinum* spore types in the environments (in both land and marine environments) of many parts of the world have been published (2, 4). The types of *C. botulinum* spores present and their numbers and distribution in the environment vary depending on the location. Type A is found more frequently in soils of the western United States, China, Brazil, and Argentina. Type B is found more frequently in the soils of the eastern United States, Great Britain, and Denmark, and to a lesser extent in the Netherlands. The majority of type B strains from North America are of the proteolytic type (group I), whereas the type B strains from Europe generally belong to the nonproteolytic type (group II). Type E is especially common in freshwater and marine sediments, and is found in fish, shellfish, and fishery products. Huss (16) published an excellent review on the incidence of *C. botulinum* spores in fishery products. Nonproteolytic types B and F generally exist in a habitat similar to that of type E (3). *C. botulinum* spores have also been detected in honey and corn syrup (17).

Botulinum neurotoxin (BoNT)

All *C. botulinum* types produce a neurotoxin (BoNT), also known as 'Botox', which is a simple dichain polypeptide that consists of a 100-kDa 'heavy' chain joined by a single disulfide bond to a 50-kDa 'light' chain, corresponding to between 1,251 and 1,297 amino acids. In this configuration, the polypeptide has a low biological activity. The three-dimensional structure of type A BoNT has been resolved to 3.3 Å resolution using data collected from multiple crystals at 4°C (18). The light chain of BoNT binds to the receptors on nerve endings and blocks the release of the neurotransmitter acetylcholine at the neuromuscular junction, causing a flaccid paralytic action that requires regeneration of nerve endings for recovery. BoNT is the most poisonous known substance and is extremely toxic to humans and animals. Group I strains (proteolytic types) of *C. botulinum* possess endogenous proteolytic enzymes that cleave the BoNT polypeptide at a site between the disulfide links into a dichain molecule with subunits of 100-kDa and 50-kDa (19). In this state, the 'nicked' protein expresses its full potency. The BoNTs from nonproteolytic strains of *C. botulinum* can be activated by exogenous proteolytic enzymes or by adding trypsin to a pure culture or extract. Type A BoNT is more lethal than types B and E (20). BoNTs are the most lethal substance known to man, being 15,000 to 100,000 times more lethal than sarin (a potent organophosphate nerve agent) (20). The exact lethal dose of BoNT for humans is not known. However, by extrapolation, Arnon *et al.* (21) estimated the lethal dose of crystalline type A BoNT for humans from primate data. The lethal dose of crystalline type A BoNT for a 70 kg (154 lb) human would be approximately 0.09-0.15 μg via intravenous or intramuscular injection, 0.70-0.90 μg via inhalation, and 70 μg via oral ingestion (1 μg/kg).

BoNTs are heat-labile and their thermal inactivation is non-linear. The inactivation times of BoNTs depend on their initial concentrations. Reported thermal inactivation times for BoNT types A, B, E, and F with an initial concentration of 10^4 mouse lethal doses (MLD) per g at 78-80°C range from <1 to 2 min (22, 23). Minimum heat treatments of 20 min at 79°C or 5 min at 85°C are recommended to inactivate a BoNT concentration of 10^5 MLD in foods (24). BoNT in solution is colorless and odorless, and can be absorbed into the bloodstream through the respiratory mucous membranes as well as through the walls of the stomach and intestine (25). Type A BoNT is the first biological toxin licensed for treatment of human diseases, namely for cervical torticollis, strabismus, and blepharospasm associated with dystonia, and to reduce facial wrinkles and hemifacial spasm; type A BoNT is also used for several unlicensed treatments for other human diseases (21, 26, 27).

***C. botulinum* growth and toxin production**

The proteolytic (group I) and nonproteolytic (group II) types differ in their tolerance to salt, water activity (a_w), minimum growth temperature, and heat resistance of their spores (Table 1). The optimum temperature for growth and toxin production by the proteolytic types is approximately 35°C, with very slow growth at both 10-12°C and 50°C. However, at 50°C, the toxin of proteolytic types may be inactivated slowly. The nonproteolytic types can grow

between 3 or 3.3°C and 45°C, with optimal growth and toxin production at approximately 30°C. Refrigeration may not provide a complete safeguard against botulism resulting from foods containing nonproteolytic type strains. The pH values that limit the growth of *C. botulinum* range from 4.6-5.0 depending on the group, type, strain, and medium. In general, proteolytic types are slightly more tolerant to acidic conditions than are the nonproteolytic types (Table 1). The minimum pH for toxin production in canned foods varies from 4.88 to 5.44, depending on the type of food (28). Occasional outbreaks of botulism may occur from the ingestion of high-acid foods such as tomato products as a result of growth of other contaminating microorganisms such as molds, which raise the pH of the product and thus permit *C. botulinum* growth and toxin production (29).

Proteolytic strains (group I) of types A, B, and F produce heat-resistance spores that are of major concern in the processing of low-acid canned foods. Nonproteolytic strains (group II) of types B, E, and F produce spores of relatively low heat resistance and are of concern primarily in pasteurized or unheated foods (refrigerated foods consumed without prior heating), including seafoods and fermented seafood products (3, 28). Proteinaceous foods such as meats and meat-based products and non-proteinaceous foods such as vegetables and vegetable-based products can provide sufficient nutrients for the growth of *C. botulinum* and the production of toxin. The proteolytic types digest complex proteins in foods; their growth is generally accompanied by mild to strong putrid odors that may warn consumers of a potential botulism hazard. However, nonproteolytic types may not sufficiently alter the odor and appearance of the food during their growth to warn consumers of possible risk of botulism.

Botulinum types

Botulism is currently classified into four categories: food-borne, infant, wound, and undetermined classification (child or adult botulism from intestinal colonization). The Centers for Disease Control and Prevention (CDC) investigates botulism outbreaks and identifies the carrier source and strain involved in the outbreak. It publishes the details of outbreak investigations in its Morbidity and Mortality Weekly Report (MMWR). The MMWR report also publishes outbreaks related to other pathogens. One can access to these reports online using the website, http://www.cdc.gov/mmwr/.

Food-borne botulism is a form of intoxication that results from the consumption of food in which *C. botulinum* has grown and produced toxin. Signs and symptoms of botulism develop as early as 2 hr or as long as 8 days after consumption of the toxin-containing food. The usual incubation period is 18 to 72 hr. The symptoms and signs of botulism are nausea, vomiting, fatigue, dizziness, headache, constipation, paralysis of muscles, double vision, difficulty in breathing, and dryness of the skin, mouth, and throat. The duration of illness ranges anywhere from 1 to 10 days or more depending upon the host resistance, type, and amount of toxin ingested, as well as the type of food. Treatment includes administration of botulinal antitoxin and appropriate supportive care, particularly respiratory assistance lasting from 2 to 8 weeks (20). Recovery involves the regeneration of nerve endings and may take several weeks or months. Over the years, the mortality rate has decreased from >60 to <10 % in the USA as a result of improvements in treatment such as supportive and respiratory intensive care and prompt administration of antitoxin (3).

Many types of food product [dairy products, vegetables, jarred peanuts, fishery products, meat products (beef, pork, and poultry), and condiments (chili sauce, chili peppers, tomato relish, and salad dressing)] have been implicated in food-borne botulism outbreaks (3, 5, 7, 8, 21, 30-32). Outbreaks occurred from these foods as a result of their preparation at home, commercial establishments, or restaurant establishments where proper control measures were not used during food manufacturing and subsequent handling and storage. The spores in these products survive inadequate cooking and canning processes, and then germinate, reproduce, and produce toxin in the anaerobic environment of the food. Commercially processed foods are less often involved in outbreaks. Recent major food-borne outbreaks involving various foods that resulted from under-processing or temperature abuse are presented in Table 2 (3, 7, 8, 18, 30, 31, 33, 34). Sobel *et al.* (35) reported that 263 cases of food-borne botulism from 160 food-borne events occurred between 1990 and 2000 in the USA; these events involved home-canned vegetables, fermented marine products, and other food products. This amounts to an average of about 26 cases per year. Over the past 20 years, food-borne botulism was recorded for approximately 35 cases per year in Italy (e.g., home-prepared vegetables in oil), 35 cases per year in Germany (e.g., salted hams), 25 cases per year in France (e.g., salted hams), and 10 cases per year in Spain (e.g., home-canned vegetables) (8).

The first food-borne outbreak of *C. butyricum* (produces type E neurotoxin) was reported in China in 1994; the food implicated in this outbreak was homemade salted, fermented paste of soybeans and wax gourds (36). Six individuals became ill during this outbreak; three of these six died (Table 2). After this outbreak, several previously reported *C. botulinum* type E food-borne botulism cases involving soybean products in China in 1973 and 1983 were re-examined and recognized to be associated with neurotoxigenic *C. butyricum*. Another major outbreak of neurotoxigenic *C. butyricum* associated with sevu (a crisp made with gram flour) was reported from India in 1996 (34). This outbreak involved 34 young school children who became ill (3 of the 34 died) after eating suspected sevu at a school cafeteria. The outbreak-associated sevu had been improperly stored. During the improper storage, the sevu was contaminated with *C. butyricum* spores, which subsequently germinated and produced toxin.

Control of *C. botulinum*

The presence of *C. botulinum* in foods can be controlled by preventing the conditions that support its growth and toxin production during storage. The conditions that favor *C. botulinum* growth and toxin production in foods include relatively high moisture (a_w >0.94), low-salt, low-acid (pH >4.6) food that is devoid of oxygen and stored without refrigeration (>3.0 or 3.3°C). Food processors use a variety

Table 2. Examples of major outbreaks of food-borne botulism resulted from foods produced at commercial establishments[1)]

Outbreak year/location	Food product involved	Number of cases/deaths	Factors contributing to outbreak	*C. botulinum* type involved
1985/British Columbia, Canada	Chopped garlic-in-oil (pH >4.6, Bottled, no preservatives)	36/0	Temperature abuse and consumption of unheated product	Proteolytic type B
1989/Lancashire and North Wales, UK	Hazelnut Yogurt	27/1	Under processing of canned hazelnut puree ingredient containing aspartame (a_w=0.99)	Proteolytic type B
1991/Cairo, Egypt	Faseikh (uneviscerated fermented and salted fish)	92/20	Toxin production during fermentation and before salting	Type E
1994/USA	Potato dip "Skordalla and Aubergine Dip"	30/0	Foil-wrapped potatoes left at room temperature before use in dip	Type A
1996/Italy	Commercially prepared "Mascarpone Cheese"	8/1	Improper processing, handling and storage	Type A
1997/Iran	Traditional cheese preserved in oil	27/1	Unsafe process	Type A
1997/Argentina	Home-cured ham	6/0	Unsafe process and temperature abuse	Type E
1998/UK	Bottled mushrooms	2/1	Unsafe process and temperature abuse	Proteolytic type B
1999/Morocco	Meat and chicken dish	80/15	Temperature abuse	Type B[2)]
2001/USA	Chilli made from frozen chilli	15/0	Improper handling and storage of frozen chilli	Type A
1994/China	Home-made salted and fermented paste of soybeans and wax gourds	6/3	Unsafe process and storage	*C. butyricum* type E
1996/India	Sevu (crisp snack made of gram flour)	34/3	Unsafe process and storage	*C. butyricum* type E

[1)]Compiled from references, O'Mahony *et al.* (33); Rhodehamel *et al.* (3); Aureli *et al.* (30); Peck (8); Kalluri *et al.* (31); Chaudhry *et al.* (34); Lund and Peck (7).
[2)]Only toxin identified - unclear whether it was proteolytic or nonproteolytic type B.

of physical and chemical treatments to either destroy *C. botulinum* spores in the product or control its growth and subsequent toxin production in the product during extended storage. Physical treatments, namely dehydration, low temperature storage, pasteurization, thermal sterilization, irradiation, and nonthermal technologies such as high pressure processing, can be used to control *C. botulinum* in foods. Some specific constraints may restrict application of physical treatments to certain types of food products. For example, product stability and identity may be altered substantially by increased thermal processing or dehydration (3). Chemical treatments used to inhibit or delay growth and toxin production by *C. botulinum* consist of ingredients that are added to food products during their preparation. Ingredients such as salt (NaCl), acids for acidification, nitrite, ascorbate or isoascorbate, polyphosphates, sugars or syrups, antioxidants, smoke and its components, seasonings, spices, extenders, and binders can be used to control *C. botulinum* in foods. These ingredients are added to food products during their manufacturing, either by themselves at high levels or in combination with others at lower levels (hurdle concept) depending on the type of food product, processing, and storage conditions. Details on each of these physical and chemical treatments used to control *C. botulinum* in a variety of food products have been reviewed and published previously (2, 3, 7, 37).

Spores of *C. botulinum* types A, B, and F are the most heat-resistant of the *C. botulinum* types, and are of particular concern in the sterilization of low-acid, shelf-stable canned foods (pH >4.6). Low-acid, shelf-stable foods (Table 3) (2) packaged in hermetically sealed containers (i.e., canned foods) are commercially processed by thermal sterilization at temperatures around 121°C in order to destroy heat-resistant spores of *C. botulinum* and to achieve 'commercial sterility', which is defined as "a process that renders a product free of pathogens and spoilage organisms under normal conditions of storage and distribution" (Code of Federal Regulations, 21 CFR 113). Commercial sterility is usually expressed in min at 121°C, the sterilization process equivalent time (F_o) given to a particular type of product. Food processors employ F_o values that exceed the critical F_o value (3.0 min) necessary for a 12-log reduction of heat-resistant spores of *C. botulinum* (7). Additional sterilization process equivalent time (F_o value) is given to some products, and results in the complete destruction of spores of various mesophilic and thermophilic bacteria that possess higher heat resistance than *C. botulinum* spores (2). For example, sterilizing F_o values for various commercially processed low-acid canned vegetable and marine products are

Table 3. Treatments used to control *C. botulinum* in shelf-stable canned foods

Processes	Spores killed	Spores inhibited	Foods
Thermal process	Yes	-	Low-acid canned foods (pH >4.6)
Thermal process plus salt and nitrite	Yes[1)]	Yes	Low-acid canned meats (e.g. canned ham)
Pasteurization and pH (acidification)	No	Yes	Canned acidic foods (pH <4.6) (e.g. pickles, mayonnaise, canned vegetable and fruit products)

[1)]Partial inactivation, i.e., 6-7 log units.

presented in Table 4 (38). Each food processor generates its own F_o values for its low-acid canned food products based on parameters such as product type, ingredients in the particular type of product, can size and type, type of process, and packaging used.

Many canned cured meats become organoleptically unacceptable after thermal sterilization. Therefore, these commercially-processed meat products receive milder thermal treatment (far below the F_o that low-acid canned foods traditionally receive), and nitrite and salt (in a brine solution) are normally added to control the growth of *C. botulinum* (Table 3). This less severe thermal treatment results in the destruction of vegetative microflora and partial inactivation (i.e., 6-7 logs) of bacterial spores. The surviving spores are inhibited by the added nitrite and salt. The shelf-stable canned meats that fall into this category include canned deviled ham, potted meat, Vienna sausage, canned corned beef, and corned beef hash (39). Acidification is used to control *C. botulinum* in acidic canned food products. It is generally known that the growth and toxin production of *C. botulinum* is completely inhibited in acidic foods with an equilibrium pH lower than 4.6. Acidified products such as pickled pigs feet, pickled vegetables, and relishes are acidified through the process of pickling in vinegar solutions. Canned fruit and vegetable products are acidified by the addition of acids.

Table 4. Examples of approximate sterilizing values (F_o) for some commercially-processed canned food products

Product type	Approximate F_o (min)
Asparagus, in brine	3.0
Green beans, brine packed	3.5
Corn, whole kernel, brine packed	9.0
Corn, cream style	5.0
Peas, brine packed	7.0
Mackerel in brine	2.9-3.6
Sausage, Vienna, in brine	5.0
Chili con carne	6.0

These acidic products receive heat treatment less than that needed to control *C. botulinum*, but sufficient to inactivate microorganisms that are able to grow at acidic pH.

Many extended shelf-life, chilled canned foods such as pasteurized canned crab meat have a pH >5.0, low salt content, and a_w >0.97. A combination of heat treatment (i.e., pasteurization) and maintenance of storage temperature <10°C is used on these products to ensure safety from *C. botulinum* growth and toxin production (7). For example, pasteurized canned blue crab meat in the USA receives a heat treatment in which each can is heated in a water bath at 88°C until the cold-point temperature in the can reaches 85°C, and remains at that temperature for 1 min (Table 5). This treatment gives the canned product a shelf-life of 6 to 18 months when stored at ≤2.2°C. The low heat treatment used for these products results in several log-units destruction of *C. botulinum* type E spores and sub-lethal damage of many other spores. Storage of these products at temperatures ≤2.2°C plays an integral role, providing protection against *C. botulinum* growth and toxin production. Failure to maintain low storage temperature may result in germination, growth, and toxin formation by the sub-lethally damaged spores.

Non-thermal technologies such as high pressure processing have received a great deal of attention recently because of their ability to destroy various vegetative pathogens and viruses. Currently, high pressure processing is being evaluated at the National Center for Food Safety and Technology (NCFST), Summit-Argo, IL, USA for its ability to inactivate *C. botulinum* spores. Studies at the NCFST (40-42) have evaluated the effects of high pressure (120,000 psi) in combination with moderate to elevated process temperatures (i.e., 35 to 75°C) on spores of *C. botulinum* type A strains (BS-A and 62-A), type E strains (Alaska and Beluga), and nonproteolytic type B strains (2-B, 17-B, KAP8-B, and KAP9-B). Spores of type A strains, BS-A and 62-A, cannot be completely inactivated at high pressure and moderate to elevated temperature (120,000 psi and 60-75°C). Type A spores were found to be more resistant to high pressure processing; only partial inactivation (≤3-log units) of these spores was obtained when a combination of high pressure and temperature (120,000 psi and 75°C) was applied (Fig.

Table 5. Treatments to control *C. botulinum* in perishable canned foods[1)]

Process	Botulinum spores killed	Botulinum spores inhibited	Foods
Pasteurization and refrigeration	No	Yes	Hermetically sealed and canned seafood products[2)]

[1)]Compiled from Hauschild (2).
[2)]Blue crab meat pasteurized to 85°C internal temperature.

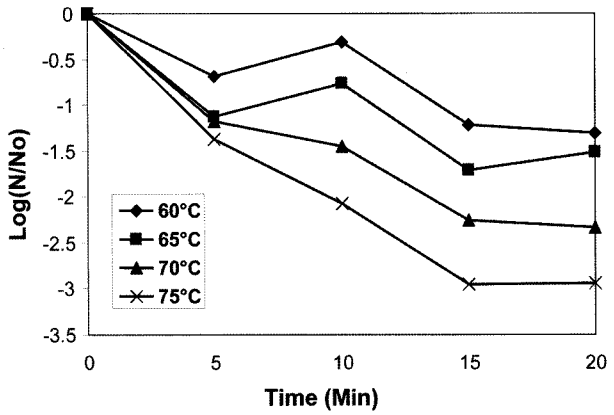

Fig. 1. Reduction of *C. botulinum* strain, 62-A spores in phosphate buffer (0.067M, pH 7.0) at high temperatures with a pressure of 827 MPa (120,000 psi).

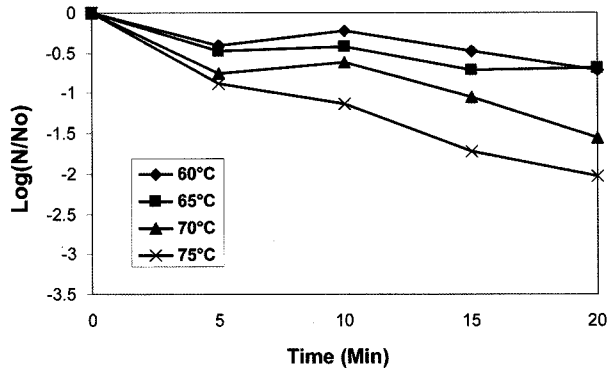

Fig. 2. Reduction of *C. botulinum* strain, BS-A spores in phosphate buffer (0.067M, pH 7.0) at high temperatures with a pressure of 827 MPa (120,000 psi).

1 and 2). Reductions of over 5.5 log-units of spores of nonproteolytic type B strains, 2-B, 17-B, and KAP9-B, were obtained at this pressure and temperature combination (Fig. 3). Based on the limited number of strains tested, resistance of *C. botulinum* spore types to high pressure processing appears to be in the order of type E < nonproteolytic type B < type A, with type E being the least resistant. High pressure processing has the potential to destroy *C. botulinum* type A spores in low-acid, shelf-stable canned food products (i.e., commercially sterile process) when the process temperature is above 110°C and in type E spores in refrigerated canned products when the process temperature is above 50°C. High pressure processing as a method of commercial sterilization for low-acid canned foods must be designed to yield an equivalent protection factor (i.e., F_o) to that used for thermal treatment of low-acid canned foods.

Conclusions

The use of thermal sterilization alone or in combination with added inhibitory ingredients or compounds are the common commercial methods used for preventing growth and toxin production by *C. botulinum* types A and B in low-acid canned foods. Most of the reported botulism outbreaks result from improper processing, handling, and storage of foods in the home and in commercial food service establishments. Although outbreaks of botulism from commercially-processed canned foods are rare, they do occur occasionally as a result of improper processing and storage temperature or post-processing contamination (i.e., container damage or leakage). The rarity of the occurrence of outbreaks of commercially-processed canned foods can be attributed to greater awareness of food processors towards the control of key factors during manufacturing, including the use of an HACCP system that inhibits *C. botulinum* growth and toxin production.

For further reading on *C. botulinum*, the reader can visit the website created by the National Center for Food Safety and Technology at Summit-Argo, IL, USA. This website maintains an online publication database on *C. botulinum*. It can be accessed by logging on to: http://www.ncfst.iit.edu/CBOT/cbotbibl.html.

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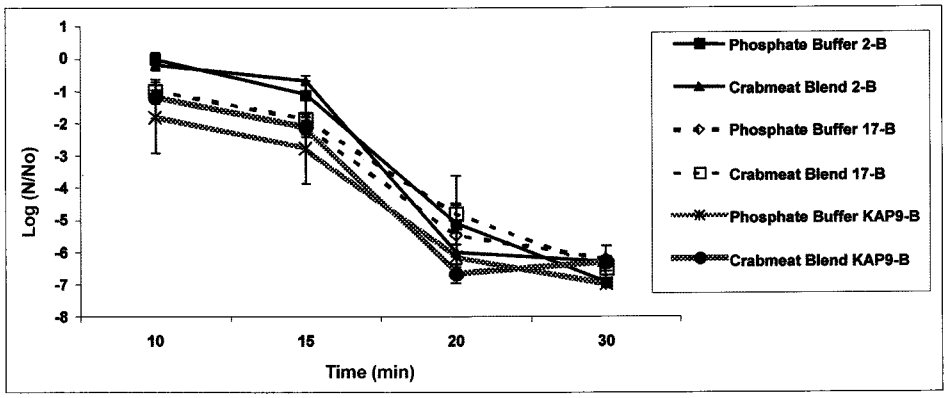

Fig. 3. Effect of time at 827 MPa (120,000 psi) and 75°C combination on the reduction of spores of nonproteolytic type B *C. botulinum* strains, 2-B, 17-B, and KAP9-B, in a phosphate buffer (0.067M, pH 7.0) and crabmeat blend.

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