

Microbial update

Clostridia

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Clostridia are a class of bacteria within the Phylum Firmicutes (Latin: Firmus = strong; Cutis = skin; this referring to the strong cell wall structure). Classes within this Phylum include the Bacilli order Bacillales (bacillus, listeria and staphylococcus); the Bacilli order Lactobacillales (enterococcus, lactobacillus, lactococcus, leuconostoc, pediococcus, and streptococcus); the Clostridia (clostridium, acetobacterium, eubacterium, heliobacterium, megasphaera, pectinatus, selenomonas, zymophilus and sporomusa); and the Erysipelotrichi (erysipelothrix).

The Genus Clostridium consists of rod shaped Gram positive bacteria that are obligate anaerobes and form spores. They are a very diverse group being found in a large range of environments. In foods they can be the cause of food poisoning or spoilage incidents.

What do they cause?

Members of the genus clostridium can cause a range of human illnesses:

- C. botulinum – botulism.
- C. perfringens – food poisoning.
- C. difficile – colitis.
- C. teteni – tetanus.

In foods, we are usually concerned with C. botulinum and C. perfringens. There have been some reports which have suggested that recent problems with C. difficile infections in hospital environments may have a food related link.

The organism has been found in animals destined for food production and in various type of meat product in retail stores, but as yet there have been no instances where a case of C. difficile illness has been definitely traced to a food source, but we must remain aware of these concerns.

C. botulinum

This is the cause of botulism. Foodborne botulism (as distinct from wound botulism and infant botulism) is a severe type of food poisoning caused by the ingestion of foods containing a neurotoxin, formed during



growth of the organism within a food matrix. The toxin is heat labile and can be destroyed if heated at 80°C for 10 minutes or longer, but this is never used as a control option in food manufacturing.

The incidence of the disease is low, but the disease is of considerable concern because of its high mortality rate if not treated immediately and properly. The botulinum toxin interferes with nerve transmission in humans and if untreated will gradually cause paralysis. Death can occur due to paralysis of the breathing muscles.

Cases of botulism are very rare in the UK, but in other countries it is more frequently seen, with the USA reporting between 10-30 outbreaks annually. The organism and its spores are widely distributed in nature. They occur in both cultivated and forest soils, bottom sediments of streams, lakes, and coastal waters, and in the intestinal tracts of fish and mammals, and in the gills and viscera of crabs and other shellfish and these will be the original source of the organisms that give rise to food poisoning.

As the organism can produce spores and

these are heat resistant, normal cooking cannot be guaranteed to eliminate the organism from food products. The control of C. botulinum in foods is therefore dependent on a good knowledge of the organism and factors that can be used to prevent its growth. It is important to note that in standard botulism, it is the growth of the organism and subsequent production of toxin that is the hazard to health, not simply the presence of the organism.

In order to begin to construct control strategies for the organism, we have to know more about it. The organism can be split broadly into two groups; proteolytic (mesophilic) C. botulinum and non-proteolytic (psychrotrophic) C. botulinum.

These two have different sets of survival and growth characteristics (see Table 1).

Linked with this, the mesophilic types are very much more heat resistant than the psychrotrophic types.

Control of the organism in foods is therefore dependent on considering the intrinsic parameters of the food (for example pH, water activity and salt content), its packaging

Table 1. Survival and growth characteristics of C. botulinum.

	Mesophilic	Psychrotrophic
Minimum temperature (°C) for growth	10	3
Minimum pH for growth	4.6	5.0
Minimum Aw for growth	0.94	0.97
Maximum salt (%) allowing growth	10	5
Bot group	I	II
Toxin types	A,B,F	B,E,F
Proteolytic	yes	no

type, storage conditions, and process, and developing systems that will prevent the growth of, and toxin formation in the food itself. In recent years there have been concerns over the risks of *C. botulinum* growth within chilled, low oxygen packed MAP or vacuum packed foods. The conditions within such packs could allow the growth of psychrotrophic *C. botulinum* unless good controls are in place, and a series of control measures have now been suggested in the UK by the UK Food Standards Agency and industry guides.

This guidance suggests particular levels of pH, water activity or heat process that will prevent the growth of psychrotrophic *C. botulinum*. Any food that does not contain the suggested controls should have no more than a 10 day shelf life.

Infant botulism is a particular type of illness that tends only to affect infants under the age of 12 months old. It is caused by the ingestion of *C. botulinum* spores which can grow and produce toxin within the infant gut. This results in botulism. The illness tends to be limited to infants and is due to a limited gut microflora, which in adults will successfully compete with, and prevent *C. botulinum* spores from growing.

A number of infant botulism cases have been linked to the consumption of honey, in which there is believed to be a high concentration of botulinum spores. For this reason it is not recommended that infants under 12 months old, are given honey to eat in any form.

C. perfringens

Clostridium perfringens is widely distributed in the environment and frequently occurs in the intestines of humans and many domestic and wild animals. Spores of the organism persist in soil and sediments.

Over the past 100 year this organism has been most closely associated with clinical cases of gas gangrene. It was first linked with food poisoning about 50 years ago and became prominent after research papers were published by Hobbs in the 1950s.

If *C. perfringens* spores are present in raw foods, they are very likely to survive the cooking process as they are heat resistant. As a cooked food is cooled, the spores can germinate and grow. Under optimal growth conditions the organism has a generation time of 10-12 minutes, and can grow to very high numbers quickly. Food poisoning will follow, if high numbers of vegetative cells are consumed, these produce enterotoxin during sporulation within the small intestine.

This can occur if foods (particularly meat and meat based products such as gravy) are cooked then improperly cooled or stored under unchilled conditions, before being eaten. It is interesting to note that this is an unusual food toxin poisoning. In most cases (for example Staphylococcal enterotoxins poisoning, *Bacillus cereus* emetic toxin poisoning and *C. botulinum* poisoning) the



toxin is produced within the food and is consumed as a toxin. In the case of *C. perfringens*, it is viable bacterial cells that are consumed. These must survive the acidic nature of the stomach and pass into the small intestine where the toxin is formed in situ, causing illness.

C. perfringens food poisoning usually begins about 8-24 hours after eating contaminated foods and consists of abdominal cramps and diarrhoea. Recovery usually occurs within 24 hours without the need for medical intervention, although some residual cramping may persist for an additional 24-48 hours.

Control of *C. perfringens* is centred on the fast cooling of cooked food products after cooking. Normal cooking will not kill *C. perfringens* spores, but fast cooling will ensure that the potential germination and growth of spores is very limited, reducing the risks to the consumer considerably.

Spoilage clostridia

As well as being a known cause of food poisoning, there are a range of clostridia that will cause food spoilage. The spores of such organisms may survive cooking, and can grow in processed foods unless they are formulated or processed sufficiently to prevent clostridial growth.

Detection and enumeration

There are a number of standard reference methods that can be used to detect and enumerate *C. perfringens*. These are based on standard agars containing selective and differential agents and are described in European and International Methods. They tend to be easy to use.

For *C. botulinum* however, there are no standard methods available. There is an intention to produce an International Standard based on the use of a PCR approach, but this will not be easy for routine testing laboratories to use. Often labo-

ratories will assess the levels of sulphite reducing clostridia as an indication of potential presence of pathogenic clostridia, but this should never be used as a measure of safety. For *C. botulinum*, the only route to assure safety of foods is to use risk assessment and HACCP procedures to design and manufacture safety into the food product, by either eliminating the organism using a correctly designed process, or ensuring the food will not allow the growth of *C. botulinum*.

Conclusions

Clostridia a large group of bacteria that can be widely found in foods ingredients. Most are fairly innocuous, however some can be the cause of instances of food poisoning or spoilage.

In the case of food poisoning, clostridia, control should be achieved through correct formulation (to prevent growth), correct and validated processing (to eliminate the organism and its spores) and correct cooling after cooking. Such procedures should be identified through risk assessment and the HACCP process and will, if correctly applied, result in the production of safe and stable food products.

When using heat processing to eliminate these organisms, it is of great importance to ensure that the process used (the time and temperature are fully validated to achieve the required kill) and are applied consistently.

If intrinsic food conditions are used to control growth, then these to should be validated (and perhaps checked through predictive microbiology or challenge testing) to ensure that they are effective. ■

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