

Clostridium perfringens Disease

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Abstract: *C. perfringens* (Clostridium perfringens) is a pathogen whose infects human and animals. In spite of its potential danger as an infectious agent, avirulent forms of bacillus can be found in many different habitats, such as the normal flora of human gastrointestinal (GI) tract, and environment, such as sewage and soil. Several common diseases associated with *C. perfringens* were recorded such as food-poisoning, gas gangrene, necrotic enteritis and many veterinary diseases. The trouble starts when the balance of bacteria in the gut is disrupted, giving *C. perfringens* a chance to proliferate unchecked. It may contaminate soil, animal feed and litter, or be transmitted directly from infected to healthy animals. *C. perfringens* related livestock infections have been reported in most parts of the world. Once an animal contracts a disease caused by *C. perfringens* it's often too late to do anything about it, Death comes quickly and violently. The most practical way to handle perfringens-related illnesses in animals is to prevent them in the first place. The present literature review of the diagnosis, type determination of major lethal toxins and diseases caused by *C. perfringens*.

[Nagwa, Ata, Eman A. Khairy, Sohad M. Dorgham and Mona S, Zaki. **Clostridium perfringens Disease**. *Life Sci J* 2013;10(1):1599-1602] (ISSN: 1097-8135). <http://www.lifesciencesite.com>. 235

Keywords: Key words: *C. perfringens*; *C. perfringens* toxins; food poisoning; necrotic enteritis.

Introduction

C. perfringens is a Gram-positive, rod-shaped, anaerobic, spore-forming bacterium of the genus *Clostridium* (1), widely distributed in nature and usually forms a part of the normal intestinal flora of man and animals and can be found in soil. The pathogenic importance of this organism as the causative agent of gas gangrene, food poisoning in man, various forms of acute enteritis and fatal enterotoxemias in animals. The pathogenicity of *C. perfringens* is associated with several toxins. The alpha, beta, epsilon and iota toxins are the major lethal poisonous substances produced by the organism and are closely related to its virulence, even though they produce several minor extracellular toxins. Usually, *C. perfringens* has been classified into five toxigenic types (A through E) on the basis of their ability to produce the major lethal toxins (2, 3). Two other major toxins (i.e. enterotoxin and beta-2) can also be produced by all types of *C. perfringens*, although they are not used for its typing (4). *C. perfringens* enterotoxin (CPE) is the main virulent factor that initiates many critical GI diseases. When food contaminated with *C. perfringens* is consumed, CPE begins its membrane action in a unique four-step mechanism. The first step of the mechanism is the binding CPE to the target receptor on plasma membrane protein or claudin proteins, which leads to the formation of a small complex. The complex then undergoes physical change when it binds to other membrane proteins and forms a larger complex in the membranes, which results in the disruption of the

membrane's permeability. This usually leads to cell death, because the osmotic equilibrium is not maintained due to the breakdown of the membrane's permeability (5). *C. perfringens* type A is consistently recovered both from the intestinal tracts of animals and from the environment, while others (types B, C, D and E) are less common in animal intestinal tracts (6). The main toxins that are produced by *C. perfringens* are alpha, beta, epsilon and iota. Indeed, these toxins are linked to the virulence and *C. perfringens* type, which depends on the various toxin combinations that are elaborated by the bacteria. Based on these major lethal toxins, *C. perfringens* is divided into types A, B, C, D and E, which all commonly produce the alpha toxin (7) (Table I). As these toxins are antigenic, typing is achieved by neutralization of the lethal toxins with type-specific antisera using mice or guinea pigs as test animals.

Table I: Major Lethal Toxins Of C.Perfringens For Type Determination

Toxins				
Type	Alpha	Beta	Epsilon	Iota
A	++	-	-	-
B	+	++	+	-
C	+	++	-	-
D	+	-	++	-
E	+	-	-	++

++ = Produced as a predominant toxic fraction.

+ = Produced in smaller quantities.

- Not produced.

Diagnosis

C. perfringens can be diagnosed by Nagler's Reaction where the suspect organism is cultured on an egg yolk media plate. One side of the plate contains anti-alpha-toxin, while the other side does not. A streak of suspect organism is placed through both sides. An area of turbidity will form around the side that does not have the anti-alpha-toxin, indicating uninhibited lecithinase activity. *C. perfringens* can be isolated and cultured in Cooked Meat Medium (CMM) at 37°C for 24 to 48 hours and streaked in a plate containing neomycin sulphate sheep blood agar, incubated under anaerobic conditions in anaerobic jars at 37°C for 24 to 48 hours. After incubation, colonies were analyzed according to the shape, color, and type of hemolysis. Bacterial morphology was microscopically assessed in gram stained smears. Biochemical tests for species identification: production of catalase, lecithinase and gelatinase, fermentation of glucose and lactose, and skim milk coagulation. Interpretation was performed according to (8,9)

Type A

This is the most common of all the *C. perfringens* types, the most variable in toxigenic properties and also the most confusing organism in respect to its pathogenicity. In spite of continued intensive study, its role in the pathogenesis of diseases is still not fully understood. In its toxigenic behavior type A can be subdivided into two varieties. The "classical" variety, characterized mainly by alpha toxin production, is associated with gas gangrene, traumatic infections, avian necrotic enteritis and the normal intestinal tract. The enterotoxigenic or human food poisoning variety, characterized by enterotoxin production, is capable of causing human enteritis. Alpha-toxin production by strains isolated from normal intestinal contents or feces may be so little on laboratory media that it is impossible to type the organism by the toxin neutralization method. Alpha-toxin is an enzyme, chemically known as phospholipase-C (lecithinase-C), which hydrolyzes lecithin into phosphorylcholine and a diglyceride (10). As the membranes of most cells consist of lipoprotein complexes containing lecithin, alpha-toxin leads to their destruction. The resultant biological effect is either hemolysis, necrosis or death, depending on tissues accessible to the toxin. Type A has also been reported as causing enterotoxemia in sheep and calves, sometimes referred to as "sudden death" (11, 12). In poultry, type A has been shown to be capable of causing necrotic enteritis (13).

Type B

Clostridium perfringens type B produces alpha, beta and epsilon toxins and has been associated with enterotoxaemia in sheep and goats (14).

Infection with *Clostridium perfringens* types B causes severe enteritis, dysentery, toxemia, and high mortality in young lambs, calves, pigs, and foals. Types B produce the highly necrotizing and lethal β toxin that is responsible for severe intestinal damage. This toxin is sensitive to proteolytic enzymes, and disease is associated with inhibition of proteolysis in the intestine. The diseases are Lamb dysentery in lambs up to 3 wk of age, Calf enterotoxemia in well-fed calves up to 1 mo. and Foal enterotoxemia in foals in the first week of life.

Lamb dysentery is an acute disease of lambs up to 3 wk old. Many may die before signs are seen, but some newborn lambs stop nursing, become listless, and remain recumbent. A fetid, blood-tinged diarrhea is common, and death usually occurs within a few days.

In calves, there is acute diarrhea, dysentery, abdominal pain, convulsions, and opisthotonos. Death may occur in a few hours, but less severe cases survive for a few days, and recovery over a period of several days is possible.

In foals, there is acute dysentery, toxemia, and rapid death. Struck in adult sheep is characterized by death without premonitory signs.

Hemorrhagic enteritis with ulceration of the mucosa is the major lesion in all species. Grossly, the affected portion of the intestine is deep blue-purple and appears at first glance to be an infarction associated with mesenteric torsion.

Treatment is usually ineffective because of the severity of the disease, but if available, specific hyperimmune serum is indicated, and oral administration of antibiotics may be helpful. The disease is best controlled by vaccination of the pregnant dam during the last third of pregnancy: initially, 2 vaccinations 1 month apart, and annually thereafter. When outbreaks occur in newborn animals from unvaccinated dams, antiserum should be administered immediately after birth.

Type C

Type C of *C. perfringens* produces alpha and beta toxins. Beta toxin is crucial for pathogenesis of necrotic enteritis in animals and man (15).

Beta-toxin is more sensitive to enzymes, elevated temperatures and other environmental factors than the other major lethal toxins of *C. perfringens*; it is readily inactivated by trypsin and other proteolytic enzymes (16, 17). It causes Struck disease in adult sheep, sudden death in goats and feedlot cattle. Necrotic enteritis in chickens, Hemorrhagic enteritis in neonatal piglets. Different variants of

disease caused by *C. perfringens* of toxin types A and C have been reported as important in commercial poultry especially in broiler production (18).

Type D

C. perfringens type D is the etiological agent of enterotoxaemia (pulpy kidney disease) of several animal species (19). According to current knowledge, the disease is caused by epsilon toxin, a major exotoxin produced by this microorganism (20). This classic enterotoxemia of sheep is seen less frequently in goats and rarely in cattle. It has a worldwide distribution affecting animals of any age. It is most common in lambs that are either less 2 wk old or weaned in feedlots and on a high-carbohydrate diet or, less often, on lush green pastures. The disease has been suspected in well-nourished beef calves nursing high-producing cows grazing lush pasture and in sudden death syndrome in feedlot cattle; however, supportive laboratory evidence in the latter is lacking. Predisposing factors are essential, the most common being the ingestion of excessive amounts of feed or milk in the very young and of grain in feedlot lambs. In young lambs, the disease usually is restricted to ewes with single lambs, because ewes with twins seldom give enough milk to allow enterotoxemia to develop. In the feedlot, the disease usually is seen in lambs switched rapidly to high-grain diets. As starch intake increases, it provides a suitable medium for overgrowth of *C. perfringens*, producing epsilon toxin. The toxin causes vascular damage, particularly in capillaries of the brain. Many adult sheep carry strains of *C. perfringens* type D as part of their normal intestinal microflora, which is the source of organisms that infect the newborn. Most such carriers have nonvaccinal antitoxin serum titers. Usually, sudden deaths in the best-conditioned lambs are the first indication of enterotoxemia. In some cases excitement, incoordination, and convulsions occur before death. Opisthotonos, circling, and pushing the head against fixed objects are common neurologic signs; frequently, hyperglycemia or glycosuria is present. Diarrhea may or may not develop. Occasionally, adult sheep are affected too, showing weakness, incoordination, convulsions, and death within 24 hr. In goats, the course of disease ranges from peracute to chronic, with signs that vary from watery diarrhea with or without blood to sudden death. Affected calves not found dead show mania, convulsions, blindness, and death within a few hours. Subacutely affected calves are stuporous for a few days and may recover. In goats, diarrhea and nervous signs are seen, and death may occur over several weeks. Type D enterotoxemia occasionally is seen in young horses that have overeaten. The method of control depends on the age of the lambs, the frequency with which the disease appears on a

particular property, and the method of husbandry. If the disease is seen consistently in young lambs, ewe immunization probably is the most satisfactory method of control. Breeding ewes should be given 2 injections of type D toxoid in their first year, a booster injection 4–6 wk before lambing, and each year thereafter. Enterotoxemia in feedlot lambs can be controlled by reducing the amount of concentrate in the diet. However, this may not be economical, and immunization of all lambs with toxoid upon entering the feedlot likely will reduce losses to an acceptable level. Two injections, 2 wk apart, will protect lambs through the feeding period. When alum-precipitated toxoids or bacterins are used, the injection should be given at such a site that the local cold abscesses, which commonly develop, can be removed easily during normal dressing and will not blemish the carcass.

Type E:

Type E of *C. perfringens* produces alpha and iota toxins (14). The principal toxin is iota-toxin. The characteristics of this toxin are somewhat similar to epsilon-toxin of type D, but they are not antigenically related. The toxin is produced by intact cells and fully activated by proteolytic enzymes. Biological effects of this toxin include necrotizing action and marked increase in capillary permeability (21). It causes hemorrhagic enteritis in calves and enteritis in rabbits.

Conclusion

The present study was concluded that the *C. perfringens* species is divided into five types, from A to E, based on their ability to produce any of the four major lethal toxins.

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1/22/2013