

BOTULISM IN SOUTH AFRICA*

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The history of botulism in South Africa is the history of the identification of the disease among animals by Theiler and Robinson and of research on lamsiekte by them and other workers. This research was a model of painstaking and brilliant investigation covering a period of many years, for which Sir Arnold Theiler received a large monetary reward¹⁷ from the Government and which was recognized, together with his other research work, by the many honours conferred on him. There have been no identified human cases of botulism in the Republic, but a small outbreak was reported in Southern Rhodesia in August 1959.

The first reported cases of botulism were recorded as 'sausage poisoning', which was prevalent¹ in Germany in

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the early eighteenth century. Subsequently cases occurred elsewhere on the Continent. 'Fish' outbreaks were first reported in Russia in 1833 and have since occurred there and elsewhere. The number of cases and deaths reported stimulated research and it was soon recognized that poisoning occurred where preserved foods were prepared in an unsatisfactory manner¹ and that affected food might have a sour taste or bad odour. Warnings were issued accordingly.

Speculation and research into causes centred on metallic and other chemical poisons, putrefactive alkaloids, 'ptomaines', ferments, vegetable seeds, moulds, algae, etc., and it was not until 1895 that Van Ermengem isolated the responsible organism and proved its causal relationship.

American outbreaks have been recorded since 1900 and may have been prevalent earlier without having been

recognized or recorded. The incriminated foodstuffs have been mostly home-canned fruits and vegetables, but have also included meat products. No outbreaks caused by commercially canned products had occurred⁴ since 1925, but recently two women died^{18, 23} from botulism after eating canned tuna; *Clostridium botulinum* (type E) was isolated from the remains in the tin and from another tin in the same batch.

In Britain no authenticated cases were recorded¹ until 1922, when an outbreak occurred in Scotland due to potted wild-duck paste. Eight persons were affected and all died. No further cases were reported until 1935, and since then there have been a moderate number of small outbreaks.

BOTULISM IN MAN

Botulism is included among the food poisoning conditions although the classical gastro-intestinal symptoms are either little in evidence, transient, or absent altogether. They are always overshadowed by the characteristic nervous symptoms.

The *incubation period* averages 12 - 30 hours, with limits between 6 hours and 8 days.

The first *symptoms* may be nausea or vomiting; diarrhoea is uncommon—constipation is more likely and abdominal pain is usually absent.

Early symptoms are usually lassitude, fatigue, headache and dizziness, weakness of the arms and legs and muscular incoordination, and blurring of vision. Dizziness and hoarseness may be the first noticed, followed by diplopia and photophobia. The throat is dry, with difficulty in speaking and swallowing. Headache is unusual but insomnia is frequent. The mind remains clear but there may be drowsiness. In the later stages there is pronounced muscular weakness, progressing to paralysis of the diaphragm, dysphagia and aphonia.

Of the *physical signs* ptosis is the most constant and is often marked. There may be a divergent squint, midriasis, and a sluggish or absent eye reflex; accommodation is impaired but not usually completely lost. There is marked muscular weakness; reflexes are normal. In the early stages the temperature is normal or subnormal and the pulse very slow. The temperature remains low unless some complication supervenes, but the pulse rate increases. At this stage the combination of the low temperature and the rapid pulse is quite striking.

The *diagnosis* is made on the clinical picture, particularly the appearance of cranial-nerve paralysis in a group of persons who have consumed the same food within the incubation period of the disease.

Chickens that have eaten left-overs from the family meal may develop symptoms of limberneck, but only with type-A intoxications.

Toxin in the food may be demonstrated by mouse-protection tests made with saline suspensions or filtered broth cultures of the suspected food, heated to kill competing organisms.

In rare cases toxin has been demonstrated in the blood of patients and, after the 14th day from the ingestion of affected food, antitoxins are present.

Confusion may occur with the symptoms of acute encephalitis, encephalitis lethargica, bulbar poliomyelitis, belladonna and salanaceae poisoning, mushroom poisoning or of other types of food poisoning.⁵

The *prognosis* varies with the amount of toxin consumed and with the type of the organism responsible; type-A toxin causes most fatalities. A short incubation period usually means a more serious outcome. Death occurs most frequently 4 - 8 days after onset and is due to cardiac or respiratory failure. In patients who survive, convalescence is prolonged but recovery is ultimately complete. The case mortality varies widely, between 20 and 70%.

Treatment. A stomach wash-out and a high enema of liquid soap and olive oil may help to clear out toxin (and possibly to neutralize it) before it is absorbed.⁷ Antitoxin is of limited value in clinical cases; it should be given to neutralize any toxin not yet absorbed, but it will not do so once the toxin is absorbed and fixed in the tissues. A slow intravenous drip of 50 ml. of antiserum in a litre of 10% hypertonic glucose should be given daily until symptoms subside; 10 ml. of the serum may be given intramuscularly to those at risk who have not developed symptoms. The remaining treatment is good nursing and symptomatic relief. Preparations should be made in advance for artificial respiration in case it becomes necessary.

Outbreak in S. Rhodesia. Flemming⁶ reports a family outbreak in Gatooma affecting 2 adults and 3 children after eating cottage pie and home-preserved pears. An adult who ate pears only escaped. The mother became ill after 24 hours with nausea and vomiting followed by dysphagia and ocular signs and symptoms which reached a peak about the 4th day and subsided after 8 days. The son of 8 years had abdominal pain after 28 hours followed by blurred vision and other nervous symptoms. The symptoms in the other two children and the African cook set in later and were quite mild. The mother and son were severely ill, developing symptoms of impending respiratory failure, but both recovered. An organism, identified by cultural and agglutination tests as *Cl. botulinum*, type B, was isolated from the bones of the meat component of the cottage pie and from the faeces of all 5 patients.

BACTERIOLOGY

Clostridium botulinum occurs in 7 different types which, though they have a general similarity in morphology and cultural habits, show many variations between one type and another and in the same type from time to time—variations in the morphology, cultural characteristics, proteolytic properties, the readiness to form spores and the resistance of the spores to heat and other destructive agencies, the ability to form toxins and the potency of the toxins and their resistance to noxious agents, the immunological reactions, the effect on different animals, and the geographical distribution.

It is a large pleomorphic, Gram-positive, motile, sporulating rod, occurring singly and in chains, and with 4 - 8 peritrichal flagellae. The spores are oval and subterminal, and distend the bacillary outline; it is a strict anaerobe, emitting a butyric odour on certain media. Cultural characteristics are similar for all types up to a point, but they differ in certain sugar reactions and in proteolytic properties.

The *spores* are highly resistant to heat. They resist boiling for a period varying from 30 minutes to 22 hours,

and autoclaving at 120°C for 20 minutes. The spores with the most heat resistance are those of type A, type F, and the proteolytic strains of type B; those with the lowest resistance are types C₂ and F, and the non-proteolytic strains of type B; types C_β and D being intermediate in resistance.¹⁴ Resistance to heat is less in acid media (at pH 3.7 boiling for 10 minutes is lethal) and in strong salt solutions (8% and over). Spores are not killed by weak acids or by fairly strong brine solutions, but the resistance to heat is lowered. They survive freezing at -16°C for 14 months and are resistant to drying.

A soluble toxin is produced, but atoxic strains have been isolated from contaminated foods,⁹ being culturally, morphologically and serologically identical with *Cl. botulinum*. The toxin is moderately heat labile and is attenuated by heating, by certain chemicals (e.g. formalin), and by being kept at room temperature for a long time. It is destroyed by the prolonged action of direct sunlight, diffuse daylight, and air, but is relatively resistant to sunlight as well as to freezing or drying;² it retains its potency for long periods if kept sealed and in the dark. The toxin is resistant to acids and is not destroyed by the digestive enzymes. Virulence is unaffected by putrefaction of the culture medium under anaerobic conditions.¹ Dolman and Murakami¹⁴ (quoting Prevot and Brygoo) state that type-A toxin is almost completely destroyed after 2 minutes at 60°C, and types B and E in 2 minutes at 70°C. Type C is not completely destroyed after 2 minutes at 70°C, and type D is unaffected after 40 minutes at 70°C, but both toxins are destroyed in 2 minutes at 90°C. At 37°C the different types lose their toxicity gradually at varying rates. Type F toxin is heat labile, losing 99% of its toxicity in 6 days at 37°C.

The geographical distribution of *Cl. botulinum* is world-wide, but the distribution of the types varies. Type A and type B (proteolytic strain) predominate in different parts of America, while type B (non-proteolytic strain) outnumber type A in Europe. Type C is widely distributed, but type D has been reported only from Africa and type F only from an island in Denmark. Type E spores in sea mud and shoreline silt in Northern Japan, British Columbia, Alaska, and Labrador, but nowhere south of latitude 40°N.¹⁴

Soil and faeces. 'Topley and Wilson',³ quoting other authors, states that *Cl. botulinum* is naturally present in the soil and is probably commoner in virgin soils and pasture than in cultivated soils and manure. It is occasionally present in the faeces of animals, such as pigs, cattle and horses, which feed on soil produce, but there is no evidence that it is a natural intestinal inhabitant. It is rarely present in the faeces of healthy human beings. There have been no extensive surveys of the soil in Southern Africa, but type D is apparently the predominant type, with type C also present. Knock¹³ examined 102 soil samples from widely scattered areas and proved the presence of type B in 3 samples from Kirstenbosch and Ysterfontein (Cape Province) and Klein Windhoek (South West Africa). The chances of success are limited by the techniques available and by the hazards of sampling. On a 'lamsiekte' farm Scheuber¹² was able to prove the presence of the organism only in soil samples from the immediate

vicinity of a decomposing carcase, whereas 100 samples collected elsewhere on the same farm were negative. Meyer and Gunnison¹⁰ obtained type-A organisms from cultures of type D that had been isolated from carrion at Onderstepoort laboratory. The state of infectivity of our soils is largely unknown, but it would be surprising if types other than types C and D are not commoner than appears from our present knowledge.

Species affected. Under natural conditions each type of *Cl. botulinum* tends to be associated with botulism in a particular animal or group of animals. Lamanna⁷ suggests that this association is a function of the distribution of the organism and the feeding habits of the host rather than of susceptibility on the part of the host, but all these factors probably play a part. Some of the common associations are as follows: type A—man and chickens (limberneck); type B—man; type C₂—man (rare), chickens, ducks, equines, and birds; type C_β—cattle and man* (rare); type D—man (rare), cattle (lamsiekte), and equines; type E—man, mink (one case¹⁴); type F—man.

BOTULISM IN ANIMALS: LAMSIEKTE

The special interest in botulism in South Africa centres on the occurrence of lamsiekte. The symptoms of this disease of cattle were observed from pioneer times, but Hutcheon, in 1884, was the first to associate them with a depraved appetite and a deficiency of phosphorus in the diet. Various workers did extensive research before Theiler and Robinson¹⁹ demonstrated the full aetiology in the 1920s.

In extensive areas of Southern Africa the soil, and consequently the grazing, is deficient in phosphorus.¹⁵ This applies more particularly to grass veld, and the deficiency in the grass is more serious in summer than winter. The areas where the deficiency occurs include all the Provinces of the Republic (particularly Western and Northern Transvaal, North-West Cape and Griqualand West, Namaqualand, Eastern Cape and Border, Western Free State and Natal Highlands) and South West Africa and Bechuanaland.¹⁶ Thorold states¹⁶ that in areas north of South Africa lamsiekte is hardly known, and attributes this to scavengers and to 'safari' (soldier) ants.

Aphosphorosis in the animal, aggravated by deficiency of protein and other essential nutrients, leads to poor development and lack of condition, lowered fertility and milk yield, and may lead to serious deformities of the joints and hoofs and to so-called stiff-sickness. The cattle develop a depraved appetite (including osteophagia) that drives them to eat carcase debris and putrid bones, which they would normally avoid. If the material has been contaminated with *Cl. botulinum* (type C or D) and the toxins have had time to develop, lamsiekte results.

Cl. botulinum grows readily in carcase debris under natural conditions in the veld and the carcase is highly toxic after about 6 days. The remains of meerkats, birds, hares, and particularly tortoises, are more important than those of domestic animals. Tortoises may be responsible for 80% of lamsiekte in some areas, and 100% of tortoise carcasses may be toxic and retain toxicity for periods up to a year.

Another source of lamsiekte is swill from Bantu mine compounds, which is given to dairy cattle, with kaffir-beer waste, to increase the milk yield. Swill is mainly mealie meal, but contains small fragments of meat, which probably carry the toxin. Feeding with swill is not common, but 3 outbreaks on the Witwatersrand have been attributed to this practice.²² Attempts to isolate toxin from the milk of sick cows have failed, but nevertheless the milk should not be sold. Active steps are necessary to prevent meat-hungry Bantu labourers from eating the meat of cattle that have died of lamsiekte.

*Lamanna⁷ states that he knows of only two outbreaks in man due to type C and one to type D. No other human cases from types C or D have been traced in the literature.

These incidents pose some interesting questions. How did the swill become infected? Was the meat content at fault and, if so, is compound beef commonly contaminated? No cases of botulism have been reported among Bantu in mine compounds. How did they escape the disease if the food was contaminated? These aspects will be investigated as far as possible, although the results are more likely to be of scientific interest than of fundamental public-health importance. In any case the use of swill for this purpose should be discouraged.

The disease in cattle develops after an incubation period of 2-6 days (limits, 18 hours—16 days)—the shorter the incubation period the more likely the case is to be fatal. The symptoms are essentially paralysis of the muscles of locomotion, mastication, and deglutition, without any rise in temperature. The paralysis usually starts in the hindquarters and spreads progressively forwards to the forelimbs, the neck, and the head. The type of case varies from very acute, with death in a few hours, to chronic.

Preventive measures are to feed the cattle with bone meal to prevent osteophagia, to collect and destroy carcase debris that may be toxic, and to refrain from feeding dairy cattle with compound swill. In addition immunization with toxoid is necessary; 2 or 3 injections are required, with an annual booster. Three million doses of vaccine are issued from Onderstepoort annually.¹⁶ It produces a good immunity.

Other animals than bovines are also susceptible to the disease.²⁰ Sheep may develop osteophagia; goats may eat infected ruminal contents; equines may eat hay contaminated by the carcase of an infected rat; ostriches and poultry may pick up fragments of carcase. Water birds are also liable to the disease.²¹

THE TOXINS

Botulism is a toxæmia not associated with an infective process in the body. The toxin is preformed outside the body, and there is no invasion of the tissues. The organism is generally unable to grow within the body of warm-blooded animals, but cases of wound infection have been reported. In a group of 3 cases where contamination of a wound was proved there were no symptoms, but in another group, in which type A was isolated, symptoms were present and all 3 patients died.⁴

Botulin is the most potent of all known poisons. Only tetanus and shigella neurotoxins approach it in toxicity; other protein poisons, such as diphtheria toxin and animal venoms are many thousands of times less toxic. It is far more toxic than aconitine, one of the most poisonous non-protein substances known,¹⁴ and is ten thousand times more potent as a gastro-intestinal poison than cyanide.⁸

Experimental animals vary in their susceptibility to the toxin, and the lethal dose in a species probably does not depend on the body weight. The toxin acts at the terminal ends of autonomic nerves and at the neuromuscular junction and the number of these sites does not increase as an animal grows. The minimal lethal dose in man cannot be accurately assessed, but he appears to be very susceptible. Cases have been reported of severe illness or death that resulted from the mere tasting of contaminated food without swallowing an appreciable amount, and the attack rate of those eating contaminated food is very high.⁸ The small intestine is thought to be the major site of absorption in cases of natural poisoning, with some absorption from the stomach and colon. It is not known how the toxin, being a protein, escapes proteolytic digestion.

In isolated muscle-nerve preparations it has been demonstrated that the muscle will still respond normally to stimulation for half an hour after exposure to the toxin, but within a few minutes of its application no known treatment, including the use of antitoxin, will prevent paralysis after the lapse of the period of latency. Because of the irreversible nature of the toxin uptake and the short latent period, the use of antitoxin is probably without value except for a possible neutralization of toxin still in the process of escaping from the bowel.

Botulinal toxins are highly antigenic proteins, and small

quantities of toxin given under cover of antitoxin will induce rabbits to produce maximal quantities of antitoxin. Moreover, it is not difficult to detoxify toxins with formaldehyde, and the toxoids are non-toxic, strongly immunogenic, and quite stable. For use in human beings a preparation of highly purified polyvalent botulinal toxoid of great efficacy is now available; they contain haemagglutinins, which may be harmless, but more research is necessary to confirm this.

Much research has been done on the toxin by Lamanna⁷ and others, but much remains to be done. Research was first stimulated by the need to evolve safe canning methods and latterly by the threat of the use of these toxins as chemical warfare agents. The use of the toxins for this purpose has received some notice recently in the lay press, some of the accounts reading rather like science fiction, but reports have also been published in scientific journals. Defence authorities make substantial sums available for research; the Geneva convention prohibits the use of live germs in warfare but not the use of toxins. Rosebury and Kabat⁸ have extensively reviewed the use of these toxins as aggressive agents. Methods of distribution of the toxin would be the main difficulty to overcome. One possible method is the deliberate contamination of foods with the organisms or their spores during the process of canning or preservation. This implies sabotage and, if possible at all, could probably only be applied on a small scale. Another possible method is the dissemination of preformed desiccated toxin from the air to pollute drinking water supplies. The poisonous dose is very minute and toxin would not be oxidized for some 12 hours, after which the area would be safe for invasion troops. Large amounts of highly potent toxin can readily be produced. It has been calculated that about 350 lb. of purified toxin distributed evenly by plane in a 10 million gallon reservoir would result in a minimal lethal dose (estimated) in each 5 ml. Ordinary methods of purification and filtration of water will not neutralize the toxin, but heavy over-chlorination for 30-40 minutes is effective.¹¹ In the laboratory slow filtration through a 30 cm. depth of activated charcoal, or boiling for 10-15 minutes, will remove the toxin.

DISCUSSION

The almost complete absence in Southern Africa of botulism in the form of food poisoning in man is somewhat surprising in view of the occurrence of *Cl. botulinum* in our soils and of national traditions regarding home preservation of fruits, vegetables and meat. It is true that the prevailing types, so far as we know, are those associated with animal botulism (types C and D), but even these have been reported to cause human cases.⁷ Other types have been found, although there is no evidence on how prevalent they may be.

Man may have a high degree of natural immunity to types C and D, or there may be other factors that account for our freedom from cases. 'Topley and Wilson'³ expresses the opinion that heat treatment alone is not sufficient to explain why the majority of canned foods are innocuous, and that it is probable that a large number of factors determine the survival, germination, multiplication and toxin production in canned food. Unless these factors are favourably combined, which is more likely with home canning than commercial processes, the food remains innocuous. The most important factors are (1) the presence of spores in suitable numbers, (2) insufficient heating, (3) anaerobic conditions, (4) too slow cooking, and (5) the use of the food without final cooking.

Given very favourable conditions *Cl. botulinum* may produce detectable toxins within 12 hours, but usually it takes 2-14 days, according to the temperature at which the food is stored, and other factors.³ Little or no growth

occurs below 10°C or at 45°C. The optimum temperature appears to be 35-40°C.

Prevention and Control

Botulism may be associated with a wide variety of foods—canned and preserved meat and fish products, sausages, shellfish, pickled fish, cheese, and canned or bottled fruits and vegetables, including apricots, asparagus, string beans, beet, olives, onions, pears, peas, spinach, and sweetcorn.

Fresh food, either raw or cooked, does not cause botulism because, even if it is contaminated, toxins do not have time to form.

Commercially canned products can be virtually disregarded as a source of botulism. If modern methods are meticulously followed the high and prolonged temperatures will destroy any organisms or toxins present.

Home products are the chief danger. Facilities in the home kitchen fall short of those in a factory and the housewife is not so well trained or supervised as factory operatives. Moreover, sufficiently high and prolonged temperatures are not so easily attained except with a pressure cooker. Fractional sterilization is unreliable—only steam under pressure is really effective, especially with non-acid foods.

In most cases contaminated food shows some signs, more or less marked, of spoiling or decomposition, but these may be so slight as to escape notice and very occasionally contaminated food may seem normal. Non-proteolytic strains of the organism may be less liable to change the appearance of food. Home-preserved fruits and vegetables in jars, if contaminated, may have rising bubbles of gas, and the liquid contents may squirt out when opened. Cans may be 'blown'. In both cases the contents may be mushy in appearance, with a bitter taste and a rancid odour, which, however, may only appear on heating. The liquor or brine may be cloudy. Ham may be soft and pale with a rancid odour.

The first step in prevention is to avoid opportunities for foods to become contaminated with *Cl. botulinum*, or at least to prevent its growth and the elaboration of toxins. This involves the selection of fresh and sound foods and the paying of strict attention to hygiene in their preparation for canning to ensure, particularly, the exclusion of soil or other such material containing the organism. It also involves processing at a sufficiently high temperature for long enough to destroy any organisms present or their toxins, as is done in commercial canning. Home-canned products should be cooked in a domestic pressure cooker.

The containers of canned and bottled foods should be carefully examined before opening and the contents should be scrutinized and smelled (but not tasted). Tasting of food may be dangerous if it has been heavily contaminated. If any signs of deterioration are present the food should be discarded.

Boiling of home-canned products for 5 or 10 minutes before it is served is a worth-while safeguard, as this will destroy any toxins it may contain.

Dewberry¹ warns against allowing suspected food to come in contact with cuts, and quotes Geiger as the authority for the statement that the toxin may be absorbed

from broken skin areas, mucous surfaces, and fresh wounds.

The prevention of botulism from the consumption of made-up meat products is difficult. The use of strong brine in pickling hams may discourage the growth of the organism.

Specific immunization with toxoid is possible and effective, but the low incidence in man would hardly justify this procedure except possibly as a means of protecting the population in bacterial warfare. In man 3 doses of alum-precipitated toxoid are given at intervals of 2 months.

Polyvalent antitoxic serum (10 ml. intramuscularly) should be given to patients known to have eaten contaminated food and who have not yet developed symptoms; if it is given within a few hours the toxin may be neutralized before it is absorbed. Such conditions, however, will seldom be fulfilled.

SUMMARY

Botulism was first reported (as 'sausage poisoning') in Germany in 1834; in America in 1900; in Britain in 1922; in S. Rhodesia (Gatooma) in 1959. It is unknown in the Republic except in cattle.

Clostridium botulinum was identified in 1895. It occurs naturally in soil, occasionally in the faeces of pigs, cattle, and horses. Spores are heat resistant, toxins relatively heat labile. Seven types are known—all affect man, but type C_α, C_β and D only rarely.

Botulin is the most potent of all toxins. After exposure there is a latent period before onset of paralysis but the action is irreversible. The possible use of toxin in warfare has stimulated research. Antitoxins are readily produced.

In man a short incubation period precedes possible gastrointestinal symptoms with cranial nerve paralysis, prostration, muscular weakness, and respiratory failure. The case mortality varies between 20 and 70%.

In cattle, symptoms are essentially similar. It arises from phosphorus deficiency, osteophagia, and the ingestion of contaminated carcase debris. Prevention is by feeding with bone meal, collecting carcase debris, and protective inoculation.

Human botulism is prevented by selecting fresh and sound food, avoiding contamination of foods, preventing bacterial growth and toxin formation, and rejecting foods with any signs of spoilage. Process foods at high temperatures of sufficient duration (home products in pressure cookers). Examine containers and contents. Discard those with rising gas bubbles, blown cans, mushy or cloudy contents, rancid odour, etc. Give polyvalent antiserum to consumers of contaminated food, before symptoms develop.

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