Scholars LITERATURE

Journal of Food and Nutritional Health

Review Article

DOI: https://doi. org/10.47275/2692-5222-125
Volume 5 Issue 1

Review on Foodborne Botulism: Historical Notes, Diagnosis and Treatment

G. Kavyasri[1*], Anusha Manchukonda[2*], Anil Kumar Gowda[3] and Soumitha Mondal[4]

[1]Government Medical College, Siddipet, Telangana, India
[2]Mamata Academy of Medical Sciences, Hyderabad, Telangana, India
[3]Kempegowda Institute of Medical Science, Bengaluru, Karnataka, India
[4]Dr. BR. Ambedkar Medical College and Hospital, Bangalore, Karnataka, India

Abstract

It is likely that mankind has been plagued by food-borne botulism since the beginning of time. Prior to the 19th century, food poisoning was only documented in a few historical sources. There is evidence that some ancient dietary laws and taboos were based on a knowledge of the life-threatening effects of poisoned food. Botulism was first described in 1735. However, in 1793, another intoxication occurred in Wildbad, in Baden-Württemberg, Germany, when six persons over 13 died. The cause of the intoxication was a popular blood sausage (black pudding). Blood sausages were prohibited by the edict of Emperor Leo VI of Byzantium in the 10th century. Atropine intoxication cannot result in dilated pupils combined with fatal muscle paralysis as described in some ancient case reports on Atropa belladonna intoxications. The discovery of botulinum toxin in Southern Germany at the end of the 18th century was spurred by well-documented outbreaks of "sausage poisoning" in Württemberg. Between 1817 and 1822, Justinus Kerner (1786-1862) published the first accurate and complete description of the symptoms of food-borne botulism. Clostridia species produce botulinum neurotoxins (BoNTs), which are the most potent natural toxins known. An acute symmetric descending flaccid paralysis is a classic symptom of toxic neurological syndrome (afebrile). In most cases, botulism occurs as a result of food poisoning. A toxin-induced neuromuscular paralysis causes all forms of the disease to exhibit the same symptoms. The diagnosis of botulism and the choice of antidote are essentially clinical decisions. Labs are mandatory in confirming clinical suspicions to regulatory agencies, identifying the BoNTs involved, and identifying the source of intoxication. Detection of BoNTs in clinical specimens/food samples and isolation of BoNTs from stool are the two steps in the diagnosis of foodborne botulism in the laboratory. The initial symptoms of foodborne botulism can be confused with more common clinical conditions (stroke, myasthenia gravis, Guillain-Barré syndrome-Miller-Fisher variant, Eaton-Lambert syndrome, tick paralysis, and shellfish or tetrodotoxin poisoning). As part of the treatment, decontamination procedures, antidotes are administered, and, when necessary, respiratory support is provided; little difference is related to the way that the patient was exposed to the substance.

Keywords: Botulism, Diagnosis, Treatment, Food, Toxicity, Poison center, Poisoning, Intoxication, Rehabilitation

[*]***Correspondence to:*** G. Kavyasri and Anusha Manchukonda[1], Government Medical College, Siddipet, Telangana, India and Mamata Academy of Medical Sciences, Hyderabad, Telangana, India.

Citation: Kavyasri G, Manchukonda A, Gowda AK, Mondal S (2024) Review on Foodborne Botulism: Historical Notes, Diagnosis and Treatment. J Food Nutr Health, Volume 5:1. 125. DOI: https://doi. org/10.47275/2692-5222-125

Received: June 05, 2024; **Accepted:** August 30, 2024; **Published:** September 02, 2024

Introduction

BoNT is one of the strongest natural toxins, mainly found in species. Known as botulism, these BoNTs can cause a life-threatening neuroparalytic syndrome. Acute, afebrile, symmetric descending flaccid paralysis is the classic clinical presentation of human botulism. Clinically, this severe intoxication could be an emergency requiring an early diagnosis and source identification. Moreover, every case of botulism may also constitute a public health emergency if it is suspected that a commercial product has been consumed, and the clinician should report the suspected case to the ministry of health or the national reference agencies as soon as possible [1].

As botulism has several different routes of exposure, there are many different types: foodborne, infantile or adult intestinal, wound, iatrogenic, and inhalation. Some authors mention botulism from an unknown source as well. Toxin-induced neuromuscular paralysis is the common denominator in all these forms. Preformed BoNT complexes in foods result in foodborne botulism, which is the most common form of botulism in the EU. BoNTs are xenobiotics that have effects on specific cells in all different forms. In the literature, human cases of botulism have been described for types A, B, and E (rarely F) neurotoxins [2]. Type C and D primarily cause illness in animals, however, there have been cases of human botulism reported. In the case of wound botulism, type G toxin was suspected. It is always advisable to take standard precautions when evaluating and treating a patient. It is impossible to absorb botulinum toxin through intact skin. The toxins can be absorbed through mucosal surfaces, the eyes, and non-intact skin. Person-to-person transmission of botulinum has never been reported, even in patient care settings. Even so, people exposed to bodily fluids or stool from patients with botulism should be informed of the early signs of botulism and should seek medical attention. Botulism is an "old" and well described disease, however, from a clinical standpoint, it remains a rare intoxication with difficulty in detecting. In the differential diagnosis process, medical professionals should be aware of this intoxication [3]. However, an early clinical diagnosis is crucial for managing the intoxicated patient appropriately with supportive and antidote treatment.

History

Botulism was first described in 1735. In 1793, another intoxication

occurred in Wildbad, in Baden-Württemberg, Germany, when six persons over 13 died. The cause of the intoxication was a popular blood sausage (black pudding). There have been 230 cases of sausage intoxication recorded after this first well documented outbreak, and Kerner (a German physician) has described the characteristic clinical syndrome associated with them. A syndrome with analogous clinical characteristics, called "fish poisoning," has been described by some Russian doctors as well [4].

In 1870, the German physician Muller coined the term "botulism" from the Latin botulus, meaning sausage. Emilie Pierre Van Ermengem, a microbiologist in the 19th century, investigated an outbreak of botulism in Ellezelles (Belgium) in December 1895, and isolated the clostridial organism that caused the disease. There were 23 of 34 participating musicians who manifested neuromuscular paralysis over the next two days (mydriasis, diplopia, dysphagia, dysarthria and progressive muscle paralysis) after a funeral meal which mainly contained raw-salted ham. Three of them died. Therefore, Van Ermengem defined botulism as an intoxication, not an infection, and determined that the toxin was produced by a spore-forming anaerobic bacterium, *Bacillus botulinus*. Its name was later changed to *Clostridium botulinum* to better express both its spindle shape and its anaerobic metabolism [2, 5].

Pathophysiology of Botulism

The cause of botulism is a bacterial toxin produced by the anaerobic gram-positive bacterium *C. botulinum*, and rarely by closely related species (*Clostridium butyricum* and *Clostridium baratii*). In addition to being ubiquitous in the environment, these organisms form spores that can survive most naturally occurring conditions or routine cooking practices indefinitely. When humans consume spores, they do not normally germinate in their intestines. The toxins are produced only when the spores germinate, and this occurs under a rare combination of conditions, including anaerobic conditions, low acidity (pH > 4.5), low salt and sugar content, and temperatures between 37 ˚F and 99 ˚F (3 ˚C and 37 ˚C). Among the most powerful biologic toxins are botulinum toxins [6]. Studies on primates have been extrapolated to humans, although the precise lethal dose has not been determined. Purified crystalline botulinum toxin type A has been estimated to be lethal at 70 g when taken orally and at 0.80 to 0.90 g when inhaled for a 154 lb (70 kg) man. Older studies suggested lower doses.

A, B, C, D, E, F, and G are the seven antigenically distinct botulinum toxins identified between 1919 and 1970. Most strains of *C. botulinum* release only one type of toxin, although there are strains that produce two types. Additionally, two new botulinum-toxin-like proteins were identified and assembled from gene sequences: one from a *Enterococcus faecalis* isolate and one from a *C. botulinum* isolate. Botulinum toxins all contain zinc-endopeptidase proteins composed of an approximately 100,000 Dalton heavy chain and a 50,000 Dalton light chain. A botulinum neurotoxin is transported to peripheral cholinergic nerve terminals, including neuromuscular junctions, postganglionic parasympathetic nerve endings, and peripheral ganglia, through ingestion, absorption from colonized wounds or intestines, inhalation, or injection [7]. By similar pharmacological mechanisms at the neuromuscular junction, all toxin types produce similar clinical symptoms of cranial nerve palsies followed by descending symmetric flaccid paralysis.

During botulinum neurotoxin activity at the neuromuscular junction, the heavy chain binds to a neuronal cell, internalizes via receptor-mediated endocytosis, travels to the cytosol, and cleaves the proteins responsible for releasing the neurotransmitter acetylcholine (specific to each serotype) [8]. By inhibiting acetylcholine release at the presynaptic motor neuron terminal, acetylcholine transmission across the neuromuscular junction is blocked. It is unlikely that botulinum toxin can pass through the blood-brain barrier to reach the central nervous system due to its large molecular structure. Although direct effects on the central nervous system have not been documented in humans, botulinum toxin might be transported axonally, like tetanus toxin. After sprouting of new nerve terminals, recovery takes weeks to months [9].

Human disease is caused by toxins serotypes A, B, E, and (more rarely) F. Patients who are affected by toxins serotype A require mechanical ventilation at a higher rate than patients who are affected by toxins serotype B. The disease caused by toxins serotype B tends to be milder than the disease caused by toxins serotype A. Toxins of type C and D have caused only two cases of illness in humans since the 1950s. Human gastrointestinal tracts might not absorb toxin type C even though it blocks neuromuscular transmission in laboratory experiments. Neuromuscular transmission is not blocked by toxin type D in studies of human tissue. Human cases of toxin type G have not been reported [10]. A syndrome of variable severity, accompanied by gastrointestinal symptoms, is normally caused by toxins type E, found in aquatic foods. Cases of type F are rare and tend to progress rapidly, cause extensive paralysis, and cause respiratory failure, but they recover sooner. Animal models expressing all types of toxins are readily susceptible to botulism (Figure 1) [11].

Botulism in Humans: Toxic Mechanisms

Symptoms of foodborne botulism in humans include a neurological toxidrome due to the blocking of voluntary motor and autonomic cholinergic junctions induced by the toxin. BoNT is ingested with food in this form. BoNT types and quantities ingested do not affect the toxic mechanism, which is quite similar, but instead the onset (time) of clinical manifestations and severity of the symptoms dominate the toxic mechanism. *C. botulinum* and other BoNT producing clostridia grow and produce toxin only in an anaerobic environment, with a pH > 4.6, low salt and sugar levels, and a temperature range of 4 - 45 ˚C. The main source of intoxication is home-canned foods or traditional local foods [12].

Bont-producing clostridia produce a polypeptide toxin (150,000 Daltons) that acts specifically on neuromuscular junctions and cholinergic sites in the autonomic nervous system (ganglionic synapses

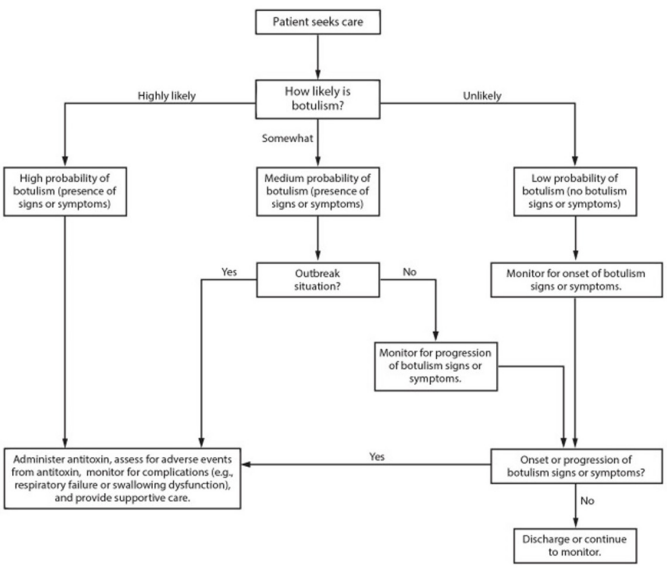

Figure 1: Approaches to analyze tumor images. Pathologists currently use artificial intelligence (AI) and machine learning techniques [3].

and parasympathetic synapses) by binding to presynaptic receptors [13]. The toxin then blocks the normal calcium-induced quantitative release of acetylcholine from presynaptic nerve terminals by a complex process (e.g., translocation of light chain (Lc) into the cytosol, cleavage of soluble N-ethylmaleimide-sensitive factor attachment protein receptors (SNAREs), due to its metalloproteinase activity). This process is irreversible. The first neurological stage of intoxication primarily involves cranial nerves and muscles. Ophthalmoparesis is also regarded as a good indicator of overall intoxication severity and progression. It is probably due to three main reasons that cranial nerves are involved first: the neurons that innervate cranial muscles are short, resulting in poor tertiary deposits. In addition to the presence of phasic fibers in facial muscles, high turnover of neurotransmitters and fast internalization of toxins, facial muscles are constantly active. Postsynaptic receptors in the facial muscular membrane are more quickly affected by acetylcholine deficiency [14].

Axon regeneration (sprouting) may occur only by forming new axon terminals at the original synaptic sites [2, 15]. Neuromuscular block associated with neurogenic atrophy (chemo-denervation) and regeneration speed of nerve terminals and presynaptic membranes determine the length of time it takes to recover. Affected central nervous system and placenta are not affected by the toxin.

Botulism in Humans: Clinical Syndrome

Since foodborne botulism is historically the most common form, its typical toxidrome is best known and described. Even so, clinicians may have difficulty detecting this poison because of their unfamiliarity with it [16]. Botulism is easier to diagnose when there are several cases with similar symptoms and signs. Based on the type and quantity of toxin ingested, the onset time of botulism may vary by patient, resulting in symptoms within 12 to 36 - 48 h of ingestion of contaminated food, delayed in some cases up to 10 - 15 days. Diagnosis can be complicated by these differences in presentation time.

With regard to the different types of BoNT, it is well known that BoNT/A is more severe and requires intubation most likely [2, 17]. However, type-E botulism has the shortest incubation period, while type-B botulism has the longest incubation period. All these characteristics make the specific treatment aimed at preventing the gastrointestinal absorption of BoNTs and the binding of BoNTs to neuromuscular junctions. The in-situ production of toxins in other forms than foodborne botulism (e.g., wounds and intestinal tracts) can also be prevented or limited through the addition of other treatments. In spite of the prevalence of gastrointestinal disturbances, some 30% of patients present with no signs or symptoms of abdominal/gastrointestinal distress [18]. It is often believed that these early gastrointestinal symptoms are caused by the accidental ingestion of other bacteria or their toxins in improperly preserved food, which are associated with nausea, vomiting, abdominal discomfort, pain, and diarrhea when present. After these first gastrointestinal symptoms are resolved, about 70% of patients experience constipation once they have resolved the neurological symptoms. Dry mouths and throats are often misinterpreted as pharyngitis, despite being symptoms of intoxication [19]. A disturbance of the autonomic nervous system can cause postural hypotension.

Afebrile, symmetric descending flaccid paralysis is the classic manifestation of foodborne botulism. Sensory and intellectual functions are not involved. Symptoms always emerge in the bulbar muscles, densely innervated by the cranial nerves IX, X, XI, and XII. Botulism cannot be diagnosed at the first stage because cranial nerves are not involved. The main symptoms are dysarthria (frequent), rhinolalia, dysphonia and dysphagia (as IX nerve involvement) preceding gastrointestinal discomfort (such as diarrhea followed by constipation) and often associated with autonomic dysfunction (i.e., dry or sore mouth and throat) in patients with normal mental status and reflexes, without sensitivity disorders [20]. There is a tendency to underestimate these complaints. Typical toxidromes are characterized by diplopia, blurred vision, mydriasis (often fixed), lateral rectus palsy, external ophthalmoplegia and bilateral ptosis (Figure 2) [3].

Despite supportive and specific therapy, severe cases may quickly worsen into respiratory failure (e.g., without gasping or agitation due to muscle paralysis), requiring mechanical ventilation. The sixth cranial nerve palsy may be the first neurological manifestation of type B botulism, and patients with a third cranial nerve palsy eventually developed respiratory failure [21]. According to oculography studies, the toxins limit saccadic burst innervation to the extraocular muscles by causing multiple hypometric saccades. During the first clinical assessment, pupillary reactions may be present, and may persist for months after the patient has recovered from motor function [22]. There are only a few instances of nystagmus. The four most specific neurological symptoms are visual disturbances, dysarthria, dysphagia and dry or sore mouth and throat, which are all caused by cholinergic blockade. When botulism is mild, the patient's symptoms end after the first few days with a gradual clinical improvement [23, 24]. The most severe cases of prolonged extensive flaccid paralysis may result in life-threatening complications: respiratory dysfunction due to weakened glottis or diaphragmatic weakness (the weakened glottis tends to close during inspiration). It is common for airways obstruction or aspiration pneumonia to occur during the first phase of botulism [25]. A patient can remain ventilator-dependent for seven to eight months when mechanical ventilation is necessary between one and eight weeks. According to the BoNT type, ventilator support is typically provided for 58 days for type-A and 26 days for type-B. When the ventilator is used for an extended period of time, complications like nosocomial infections may occur. After one year, many patients report residual symptoms such as easy fatigability, exertional dyspnea, hypothermia, changes in resting heart rate, and urinary retention. Regeneration occurs from new motor axons that sprout and reinnervate paralyzed muscles over several months [26].

Differential Diagnosis

Clinical suspicion is key to diagnosing botulism, as well as applying the appropriate antidote. It is crucial for the laboratory to confirm the clinical diagnosis, especially in the context of regulatory agencies, as well as to determine the source of intoxication and the different BoNTs involved [2].

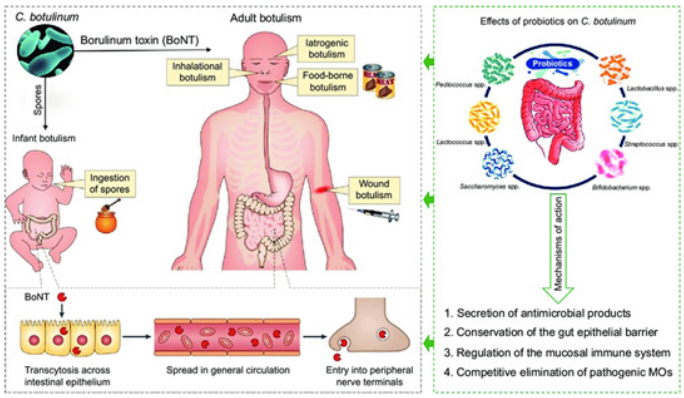

Figure 2: The mechanism of action of probiotics bacteria in inhibiting *C. botulinum* [5].

Due to the rarity of foodborne botulism, it is often underdiagnosed or misdiagnosed, and the initial symptoms do not seem pathognomonic and may be confused with common clinical conditions such as stroke, myasthenia gravis, Guillain-Barré syndrome, Eaton–Lambert syndrome, tick paralysis, shellfish or tetrodotoxin poisoning, and the autoimmune acute demyelinating polyneuropathy known as Miller–Fisher variant, Guillain–Barré syndrome (Miller–Fisher variant), Eaton–Lambert syndrome. It is more difficult to suspect botulism when the gastrointestinal syndrome is present, and some cases remain totally unrecognized, primarily in cases where the neurological symptoms remain vague [27, 28]. A large outbreak makes diagnosis easier for physicians. In spite of this, it is not always easy to recognize an outbreak and many times the first cases are mistakenly diagnosed. Sometimes, an outbreak is recognized only after a food vehicle is linked to a second cluster of cases. Patients with botulism intoxication are frequently referred to different specialists first (for example, an oculist, an otolaryngologist, or a neurologist) and only after these evaluations, due to the worsening clinical picture, are they taken to the emergency room.

There are cases where the diagnosis of botulism was only made after death because the existence of a cluster of cases finally led public health authorities to diagnose an outbreak. The correct diagnosis of the first case of botulism is therefore crucial to ensuring the correct detection and treatment of subsequent cases and particularly to avoid treatment delays. A patient presenting with any kind of weakness should be considered for botulism, including generalized, ocular or oropharyngeal weakness, as well as acute gastrointestinal dysfunction. When both cranial nerves are abnormal and descending paralysis progresses, botulism should be suspected [29]. Regarding the confirmed diagnosis of botulism suspected by the CDC, approximately 10.5% of patients get Guillain-Barré syndrome, 6.2% get food poisoning, and 3% get carbon monoxide poisoning.

In the absence of other complications, routine laboratory tests cannot confirm clinical suspicion. Tests can be used to differentiate botulism from similar diseases: (1) Normal cerebrospinal fluid (CSF) may distinguish botulism from Guillain–Barré syndrome (although it is rare for a slightly elevated CSF protein level to be observed in botulism, whereas in Guillain–Barré syndrome, the protein level may be initially normal; (2) Tension testing allows to differentiate intoxication from myasthenia gravis; (3) Neuroradiological studies can rule out strokes or ischemic. In 1979, a test called the "ice pack test" was developed for the diagnosis of ptosis and diplopia by applying ice to closed eyelids. Myasthenic ptosis was found to have a maximum specificity of 100%, and a maximum sensitivity of 89% when the test was used to diagnose ocular myasthenia gravis. In diseases with presynaptic failure of neuromuscular transmission, ptosis can occur, but no evidence has been reported in the medical literature on the response to this test. It is reported that one case of Miller-Fisher syndrome had a positive result [30, 31]. It is possible to hypothesize that presynaptic mechanisms of neuromuscular transmission are facilitated by cooling, resulting in a positive ice pack test. Using ice pack tests for botulism diagnosis is risky, as a case of botulism intoxication was reported with a positive result.

Laboratory investigations

Detection methods in biologic samples must be rapid and reliable in order to assist clinicians in making rapid diagnoses and assist surveillance systems in identifying the source of contamination and analyzing epidemiologically the cluster as a potential public health emergency [32]. BoNTs can be detected in clinical specimens or food samples, and BoNT producing clostridia can be isolated from stool specimens to confirm foodborne botulism. The most direct way to confirm the diagnosis is to inject serum or stool from the patient into mice and look for signs of botulism. Due to the possibility that other Clostridium species produce botulism, the isolation of other clostridium species (e.g., *C. butyricum* and *C. baratii*) should be considered in the laboratory diagnosis criteria. As a "gold standard" for confirming the presence of BoNTs in clinical specimens, food and/or environment samples eventually linked to botulism outbreaks, in vivo mouse lethality bioassays (intraperitoneal injections of mice with and without antitoxin) are routinely performed [33]. A type-specific botulinum antitoxin is used to identify the type (typing) of toxins produced by different strains of the bacteria. The mouse bioassay suffers from several disadvantages (costs, time, animal facilities, dedicated personnel, long turnaround time).

In vitro methods for detection of BoNTs and neurotoxigenic clostridia have been proposed and validated. To date, immunoassays for detecting BoNTs, assays for detecting catalytic activity of BoNTs, cell-based assays for detecting biologic activity of BoNTs and nucleic acid-based methods have been developed, and some of these methods will replace the *in vivo* mouse lethality bioassay in the future [2, 34]. The likelihood of obtaining case confirmation may be greater if samples are collected before antitoxin administration (stool and/or gastric contents). In cases of foodborne botulism reported in the USA (1975 to 1988), the toxin was isolated in 37% of sera (126/240 cases), 23% of stool (65/288 cases), and 5 % of gastric aspirates (3/63 cases). A specimen's collection time influences its results, for example, 50% of sera and 60% of stool samples are positive if samples are collected within 1-2 days [35].

An epidemiological study conducted in Italy (1986–2015) identified 285 laboratory-confirmed incidents with a total of 421 cases. Twenty-four percent (56/275) of confirmed food-borne cases were tested for serum and 65.3% resulted in positive results [36]. Other cases were confirmed by direct detection of toxins in feces (52 patients) or foods (159 patients). Additional 154 foodborne cases presenting with botulism symptoms were laboratory confirmed by the isolation of BoNT-producing Clostridia from feces. To obtain laboratory confirmation of botulism, different days (up to four) are usually required [37]. Botulinum antitoxin has an increased effect when administered early, so it is crucial to emphasize that treatment must be administered before lab confirmation. The rapid, reliable and sensitive assay for detecting BoNTs will be of great benefit to poisoned patients, especially those with special needs, such as children.

The endopep-mass spectrometry assay, electrochemiluminescence (ECL) immunoassay, immuno-PCR, and enhanced chemiluminescence-based ELISA all exhibit high sensitivity, compared with a mouse lethality bioassay, and are capable of rapidly detecting active BoNTs in sera or other clinical samples [38, 39].

Treatment

Key points in the treatment of botulism are (1) The decontamination, (2) The administration of the specific antidote and (3) The support of respiratory function if necessary. There are only a few differences related to the way of exposure.

Gastrointestinal decontamination

After excluding eventual contraindications, gastrointestinal decontamination should be performed in all cases of foodborne botulism to remove spores and toxin. In general, patients manifest their first clinical symptoms a few days after consuming contaminated food; therefore, gastric lavage (or induced emesis) should be considered only in cases of recent consumption of possible contaminated food. If the constipation is caused by an anticholinergic effect, then gastrointestinal decontamination must be performed. If there are no clear evidence of

efficacy for upper or lower decontamination via oro-gastric tube and cathartics/whole bowel irrigation when a suspected or confirmed case of botulism intoxication exists, then upper and lower decontamination could be performed in these cases [40]. Sorbitol is better than magnesium salts when it comes to catharsis because magnesium salts may exacerbate neuromuscular blockade. There may be challenges when it comes to irrigation of the whole bowel due to the ileus toxin-induced ileus; in some cases, neostigmine is useful for reversing ileus, inhibiting the enzymatic breakdown of acetylcholine. As a treatment option for intoxicated patients, activated charcoal absorbs BoNT/A in vitro [41].

Antidotes

Toxins that remain unbound at presynaptic level of nerve endings are neutralized by antitoxin treatment. A second point is that antitoxin reduces nerve ending involvement; usually the clinical symptoms may not appear until 12 hours after antitoxin administration. Any antigen can't be countered by type-specific antitoxins. It is generally recommended to administer antitoxin as soon as botulism is suspected. Besides botulism, the antitoxin is effective against all other types of botulism. According to animal study results, early administration of botulinum toxin after an aerosolized release of the toxins (lethal concentration) may be effective in treating inhalation botulism [42].

The only antidote for equine-derived antitoxins has been available since 1970. Animal experiments have demonstrated the effectiveness of antidotes to date. To evaluate antitoxin therapy in humans, no randomized controlled studies have been conducted (not ethical) [43]. Only case reports, retrospective studies, and clinical experiences have been used to evaluate the efficacy of antidotes. Because of the rarity of intoxication and the late diagnosis, it is difficult to perform morbidity and mortality studies. Antidotes are incapable of reversing the endo cellular mechanism of the toxin once the BoNT has entered and fixed at presynaptic level in the nervous system. According to clinical experience, the early administration of antitoxins (within 24 h) reduces the duration of mechanical ventilation and intensive care stays in patients with neurological syndromes. In an analysis of 132 cases of BoNT/A foodborne botulism (1973 - 1980), Tacket and co-authors examined the impact of antitoxin therapy on patient outcomes. In patients treated with antitoxin within 24 h after onset of symptoms, the fatality rate was lower (10% vs 15%). Patients who did not receive antitoxin had a very high mortality rate (46%) [44]. The median hospital stays for those who received early antitoxin was 10 days compared to 41 days for those who received antitoxin > 24 h and 56 days for those who were not treated with antitoxin. In some cases, however, patients may require respiratory support for longer periods of time, typically 2 - 12 weeks, whereas for botulism due to BoNT/A and BoNT/B, the duration may be 58 and 26 days, respectively. For some patients who have been severely intoxicated, a prolonged rehabilitation program is also necessary [45].

It was previously studied by the US army medical research institute of infectious diseases (USAMRIID) before the US Food and drug administration approved heptavalent botulism antitoxin (HBAT). As the Fc portion of the equine IgG antibodies has been removed, leaving the F(ab')2 portion, HBAT is derived from "despeciated" equine IgG antibodies. Currently, this is the most common formulation in the world. One vial (20 - 50 ml) of HBAT is recommended as an intravenous infusion for adults (diluted with 0.9% sodium chloride in a 1:10 ratio before use). Testing for skin sensitivity is optional. The dose may need to be adjusted when pediatric patients are involved (weight-based correction). According to the antitoxin serotype, one vial of HBAT has a half-life of 7.51 h to 34.20 h. It is unclear how effective HBAT is against the many subtypes and mosaics [46].

During the Investigational New Drug ("compassionate use" IND) study period (2010 - 2013), BAT was evaluated for safety and improved clinical outcomes. 249 patients were treated with BAT, ranging in age from 10 days to 88 years (median age 46 years) [1]. The median age of the children was 6 years; the range was 10 days - 17 years. There were no pregnant or nursing patients among the 249 patients treated. 104 (42%) patients had botulism confirmed by laboratory or epidemiological testing. All 104 patients treated within 24 h of symptom onset (early treatment) survived, while 90% (64/71) of those treated later survived (not statistically significant). In contrast, early BAT treatment was associated with statistically significant shorter hospital stays (median, 15 vs 25 days; $p = 0.04$) and ICU stays (10 vs 17 days; $p = 0.04$) stays. Out of 249 patients, 9% experienced at least one adverse effect related to BAT: fever (3%), rash (2%), chills (1%) and agitation, edema, slight hypertension, nausea (1%). The adult treated group reported bronchospasm, chest pressure, diaphoresis, erythema, increased respiratory rate, jitteriness, leukocytosis, mild hypotension, tachycardia, urinary retention, and vomiting [47]. One case of fever, agitation/anxiety and a feeling of "hurting all over" was described in the pediatric group ($n = 17$). One severe adverse reaction occurred in a 10-year-old boy (29 kg body weight) who developed bradycardia leading to asystole 90 min after BAT infusion and resolved rapidly after epinephrine therapy. After 30 min, a second episode of severe bradycardia occurred. At this point, BAT infusion was halted (an estimated 73% of the recommended dose was administered) [48, 49].

In a 64-year-old man, serum sickness occurred 11 days after BAT administration and was described by his physician as mild, self-limiting serum sickness characterized by arthralgia and myalgia treated with ibuprofen; the principal investigator also considered it to be not serious. The history of adverse effects between 1967 and 1977 reported a 9 - 17% risk of hypersensitivity, a 1 percent risk of serum sickness, and a 1.9% risk of anaphylaxis [50]. In the past, however, the recommended dose was 2 - 4 times higher than the current dose. In addition to a reduced rate of hypersensitivity, there is a 1 - 4% risk of serum sickness associated with a single vial administration. Botulism antitoxin heptavalent (HBAT) is currently available and used in the EU as well [51].

Antibiotic therapy, experimental treatments and supportive airway treatment

Toxin mechanism cannot be interfered with by antibiotic therapy. Antibiotic therapy alone is not sufficient for wound botulism; secondary infections should be treated as well [52]. The neuromuscular blockade may be exacerbated by aminoglycoside antibiotics and clindamycin in all forms.

In the last two decades, several attempts have been made to develop new drugs (e.g., monoclonal antibodies) that block the catalytic activity of BoNTs [53]. In addition to designing small molecules, peptidic inhibitors, aptamers and testing some natural substances for their anti-botulinum activity, research has been conducted on small molecules, peptidic inhibitors, aptamers and natural substances. The development of specific inhibitors that prevent the neuroparalytic action of BoNTs (regardless of their serotypes and subtypes) could be useful without knowing the particular type of BoNT present in poisoned patients is ongoing and appears promising. Research efforts have mainly focused on BoNT/A and light chain proteolytic activity [54-57]. There appears to be promise in the development of pan-BoNT inhibitors that act independently of BoNT immunological properties and target a common step of intoxication. Experimental studies have been conducted on different chemicals or molecules that can interfere

with BoNTs mechanisms at different stages. Toxin binding may be inhibited, toxin internalization and trafficking may be inhibited, toxin translocation may be inhibited, toxin disulfide bonds may be reduced, SNARE cleavage by L-chain may be inhibited, and BoNT paralysis may be reversed [58-63].

During intoxication, BoNTs bind to receptors located at the presynaptic membrane. There are some antagonists of ganglioside receptors (e.g., quinic, lectins from Limax Flavus and tricum vulgaris, thearubigin). There are two main limitations of these treatments: serotype specificity and a short therapeutic window [64]. In addition to Dyngo-4a, methylamine hydrochloride, bafilomycin A, nigericin, and quinolinol, other drugs interfere with internalization (mainly mediated by dynamin-dependent endocytic pathways) [65-67].

The other option is to interfere with a specific intracellular toxic mechanism (e.g., adamantanes, lomofungin, chicoric acid botulin, benzimidazole acrylonitrile). Although L-chain metalloprotease inhibitors have been studied, none of these molecules has demonstrated sufficient efficacy to be considered a drug. Intoxicated nerve terminals are functionally recovered by a final group of molecules [68]. As a potassium channel-blocking agent, 3,4 diaminopyridine (3,4-DAP) and analogs increase presynaptic action potential duration, causing an increase in Ca^{2+} influx and acetylcholine release. Neither the efficacy nor the cross-over of 3,4-diaminopyridine has been confirmed [69]. There has only been an improvement in ocular and limb muscle strength, but there has been no improvement in respiratory paralysis. Guanidine has been used in the past to enhance acetylcholine release, but its application, evaluated in placebo-controlled studies, failed to improve clinical outcomes. The benefits of steroids, immunoglobulins, chloroquine, and plasmapheresis have been debated in individual cases [70].

Botulism intoxication is treated with prompt and supportive care. It is necessary to monitor respiration closely due to the high risk of rapid respiratory failure and respiratory compromise [3, 71-73]. In mild cases and when suspicions of botulism intoxication are present, Arnon and colleagues suggest placing patients in a reverse Trendelenburg position at 20 - 25° with cervical support; traditionally, this position promotes diaphragmatic function by reducing the pressure on abdominal viscera and reducing aspiration risk.

Botulism and Pregnancy

Experimental and clinical data indicate that the large molecular weight of the toxin (150 kDa) prevents passive diffusion through the placenta; no cases of botulism were observed in infants born to poisoned mothers [74]. According to a recent review, 16 cases of botulism occurred during pregnancy (11 during the third trimester) and one during the postpartum period. The source of ten cases was confirmed or likely foodborne, two cases were ultimately diagnosed as wound botulism due to heroin use, and five cases remain unknown. A total of eleven women (65%) required mechanical ventilation and intensive care [75]. There were two deaths and two persistent vegetative states. There were no maternal or neonatal adverse reactions reported in the 11 women who received antitoxin. Contraindications to the administration of antidotes do not include pregnancy. An antitoxin has been administered with positive results. A total of six premature infants were born, but no cases of congenital botulism were reported. Although BoNT-A has not yet been studied in pregnant women, it appears to be relatively safe for both mother and fetus. The use of BoNT type A during lactation in humans has not been reported. Consequently, breast milk would not be able to excrete the toxin, since it would not be detectable in the systemic circulation. As a result, an infant is probably not at risk. There is no definitive data on foodborne botulism [76].

Conclusion

Symptoms of botulism include acute, afebrile, symmetric descending flaccid paralysis. The identification of the source may result in a medical emergency in some cases. In these circumstances, every case of botulism is also a public health emergency and must be reported immediately to the ministry of health or national agencies. In Europe, foodborne botulism, the most common form, is caused by ingestion of preformed toxins mainly from improperly preserved home canned vegetables, fish or meat. The unfamiliarity of clinicians may complicate the initial clinical suspicion, however. As long as standard precautions are taken when evaluating and treating patients with foodborne botulism, the disease is not contagious. Clinical manifestations are nonspecific at first, including nausea, vomiting, and xerostomia, followed by neurological signs. Even though the laboratory plays a role in confirming the clinical diagnosis, identifying the BoNTs involved and eventually identifying the source of botulism, its diagnosis is essentially clinical. This process may take months to complete, but it involves sprouting and re-innerving paralyzed muscles through motor axon twigs. In order to treat botulism, patients must undergo gastrointestinal decontamination, antidotes (antitoxins), and ultimately respiratory support. It is only antitoxins that neutralize unbound toxins that are circulating in the blood (at presynaptic level). Clinical suspicions may lead to the start of treatment as soon as possible. It is currently available in the EU as a safe and effective antitoxin against botulism heptavalent (HBAT). Several efforts have been made to design new drugs over the last two decades, primarily to block BoNT catalysis. BoNT inhibitor studies are underway and appear promising in preventing the neuroparalytic effect of BoNTs in the future.

Acknowledgements

None.

Conflict of Interest

None.

References

1. Arnon SS, Schechter R, Inglesby TV, Henderson DA, Bartlett JG, et al. (2001) Botulinum toxin as a biological weapon: medical and public health management. J Am Med Assoc 285: 1059-1070. https://doi.org/10.1001/jama.285.8.1059
2. Lonati D, Schicchi A, Crevani M, Buscaglia E, Scaravaggi G, et al. (2020) Foodborne botulism: clinical diagnosis and medical treatment. Toxins 12: 1-17. https://doi.org/10.3390/toxins12080509
3. Rao AK, Sobel J, Chatham-Stephens K, Luquez C (2021) Clinical guidelines for diagnosis and treatment of botulism, 2021. MMWR Recommen Rep 70: 1-30. http://dx.doi.org/10.15585/mmwr.rr7002a1
4. Smith LD (1977) Botulism. The organism, its toxins, the disease. Springfield, IL.
5. Alizadeh AM, Hashempour-Baltork F, Alizadeh-Sani M, Maleki M, Azizi-Lalabadi M, et al. (2020) Inhibition of *Clostridium botulinum* and its toxins by probiotic bacteria and their metabolites: an update review. Qual Assur Saf Crop Foods 12: 59-68. https://doi.org/10.15586/qas.v12iSP1.823
6. Zhang S, Masuyer G, Zhang J, Shen Y, Lundin D, et al. (2017) Identification and characterization of a novel botulinum neurotoxin. Nat Commun 8: 1-10. https://doi.org/10.1038/ncomms14130
7. Lonati D, Schicchi A, Auricchio B, Petrolini VM, Anniballi F, et al. (2017) Nosocomial transmission of *Clostridium butyricum* type E responsible for two cases (one outbreak) of infant botulism. Clin Toxicol 55: 400.
8. Kerner J (1817) Poisoning from spoiled sausages. Tub Pap Nat Sci Pharm 3: 1-25.
9. Young JH (1976) Botulism and the ripe olive scare of 1919-1920. Bull Hist Med 50: 372-391.

10. Torrens JK (1998) *Clostridium botulinum* was named because of association with "sausage poisoning". Br Med J 316: 151. https://doi.org/10.1136/bmj.316.7125.151c

11. Van Ermengem E (1897) On a new anaerobic bacillus and its relationship to Botulism. Veit 26: 1–56.

12. Erbguth FJ (2008) From poison to remedy: the chequered history of botulinum toxin. J Neural Transm 115: 559-565. https://doi.org/10.1007/s00702-007-0728-2

13. Anniballi F, Lonati D, Fiore A, Auricchio B, Medici D (2014) New targets in the search for preventive and therapeutic agents for botulism. Expert Rev Anti Infect 12: 1075-1086. https://doi.org/10.1586/14787210.2014.945917

14. Rossetto O, Pirazzini M, Lista F, Montecucco C (2019) The role of the single interchains disulfide bond in tetanus and botulinum neurotoxins and the development of antitetanus and antibotulism drugs. Cell Microbiol 21: 1-10. https://doi.org/10.1111/cmi.13037

15. Howard L, Geyer HL (2019) Botulism. In Nelson LS, Howland MA, Lewin NA, Smith SW, Goldfrank LR, et al. Goldfrank's Toxicologic Emergencies. McGraw-Hill Professional, New York, USA, pp 574–591.

16. Woodruff BA, Griffin PM, McCroskey LM, Smart JF, Wainwright RB, et al. (1992) Clinical and laboratory comparisons of botulism from toxin types A. B and E in the United States, 1975–1988. J Infect Dis 166: 1281-1286. https://doi.org/10.1093/infdis/166.6.1281

17. Sobel J (2005) Botulism. Clin Infect Dis 41: 1167–1173. https://doi.org/10.1086/444507

18. Hughes JM, Blumenthal JR, Merson MH, Lombard GL, Dowell VR, et al. (1981) Clinical features of types A and B food-borne botulism. Ann Intern Med 95: 442-445. https://doi.org/10.7326/0003-4819-95-4-442

19. Centers for Disease Control and Prevention (CDC) (1998). Botulism in the United States 1899–1996: Handbook for Epidemiologists, Clinicians, and Laboratory Workers. Atlanta, USA.

20. Terranova W, Palumbo JN, Breman JG (1979) Ocular findings in botulism type B. J Am Med Assoc 241: 475-477. https://doi.org/10.1001/jama.1979.03290310015004

21. Simcock PR, Kelleher S, Dunne JA (1994) Neuro-ophthalmic findings in botulism type B. Eye 8: 646-648. https://doi.org/10.1038/eye.1994.160

22. Friedman DI, Fortanasce VN, Sadun AA (1990) Tonic pupils as a result of botulism. Am J Ophthalmol 109: 236-237. https://doi.org/10.1016/s0002-9394(14)76000-3

23. Schmidt-Nowara WW, Samet JM, Rosario PA (1983) Early and late pulmonary complications of botulism. Arch Intern Med 14: 451-456. https://doi.org/10.1001/archinte.1983.00350030061012

24. Vita G, Girlanda P, Puglisi RM, Marabello L, Messina C (1987) Cardiovascular-reflex testing and single-fiber electromyography in botulism: a longitudinal study. Arch Neurol 44: 202-206. https://doi.org/10.1001/archneur.1987.00520140064019

25. Chatham-Stephens K, Fleck-Derderian S, Johnson SD, Sobel J, Rao AK, et al. (2017) Clinical features of foodborne and wound botulism: a systematic review of the literature, 1932–2015. Clin Infect Dis Off Publ Infect Dis Soc Am 6: 11-16. https://doi.org/10.1093/cid/cix811

26. Shapiro RL, Hatheway C, Swerdlow DL (1998) Botulism in the United States: a clinical and epidemiologic review. Ann Intern Med 129: 221-228. https://doi.org/10.7326/0003-4819-129-3-199808010-00011

27. St. Louis ME, Peck SH, Bowering D, Morgan GB, Blatherwick J, et al. (1988) Botulism from chopped garlic: delayed recognition of a major outbreak. Ann Intern Med 108: 363-368. https://doi.org/10.7326/0003-4819-108-3-363

28. Saavedra J, Femminini R, Kochen S, de Zarate JC (1979) A cold test for myasthenia gravis. Neurology 29: 1075. https://doi.org/10.1212/WNL.29.7.1075

29. Golnik KC, Pena R, Lee AG, Eggenberger ER (1999) An ice test for the diagnosis of myasthenia gravis. Ophthalmology 106: 1282-1286. https://doi.org/10.1016/S0161-6420(99)00709-5

30. Lertchavanakul A, Gamnerdsiri P, Hirunwiwatkul P (2001) Ice test for ocular myasthenia gravis. J Med Assoc Thai 84: 131-136.

31. Reid JM, Morrison I, Gorrie G, Metcalfe R (2008) Positive "ice-on-eyes" test in miller fisher syndrome. Pract Neurol 8: 193-204. https://doi.org/10.1136/jnnp.2008.148072

32. Ricker K, Hertel G, Stodieck S (1977) Influence of temperature on neuromuscular transmission in myasthenia gravis. J Neurol 216: 273-282. https://doi.org/10.1007/BF00314051

33. Molgò J., Thesleff S (1984) Studies on the mode of action of botulinum toxin type A at the frog neuromuscular junction. Brain Res 297: 309-316. https://doi.org/10.1016/0006-8993(84)90572-9

34. Larner AJ, Thomas DJ (2000) Can myasthenia gravis be diagnosed with the "ice pack test"? a cautionary note. Postgrad Med J 76: 162-163. https://doi.org/10.1136/pmj.76.893.162

35. Cosentino G, Alfonsi E, Lonati D, Locatelli CA, Valentino F, et al. (20120 Positive ice pack test in a case of food-borne botulism: a clinical note. J Neurol 259: 2486-2487. https://doi.org/10.1007/s00415-012-6591-6

36. Thirunavukkarasu N, Johnson E, Pillai S, Hodge D, Stanker L, et al. (2018) Botulinum neurotoxin detection methods for public health response and surveillance. Front Bioeng Biotechnol 6: 1-12. https://doi.org/10.3389/fbioe.2018.00080

37. Fenicia L, Anniballi F, De Medici D, Delibato E, Aureli P (2007) SYBR green real-time PCR method to detect Clostridium botulinum type-A. Appl Environ Microbiol 73: 2891-2896. https://doi.org/10.1128/AEM.02234-06

38. Singh AK, Stanker LH, Sharma SK (2013) Botulinum neurotoxin: where are we with detection technologies? Crit Rev Microbiol 39: 43-56. https://doi.org/10.3109/1040841X.2012.691457

39. Koh CY, Schaff UY, Piccini ME, Stanker LH, Cheng LW, et al. (2015) Centrifugal microfluidic platform for ultrasensitive detection of botulinum toxin. Anal Chem 87: 922-928. https://doi.org/10.1021/ac504054u

40. Babrak L, Lin A, Stanker LH, McGarvey J, Hnasko R (2016) Rapid microfluidic assay for the detection of botulinum neurotoxin in animal sera. Toxins 8: 1-9. https://doi.org/10.3390/toxins8010013

41. Chao HY, Wang YC, Tang SS, Liu HW (2004) A highly sensitive immuno-polymerase chain reaction assay for *Clostridium botulinum* neurotoxin type-A. Toxicon 43: 27-34. https://doi.org/10.1016/j.toxicon.2003.10.013

42. Guglielmo-Viret V, Attree O, Blanco-Gros V, Thullier P (2005) Comparison of electrochemiluminescence assay and ELISA for the detection of *Clostridium botulinum* type B neurotoxin. J Immunol Methods 301: 164-172. https://doi.org/10.1016/j.jim.2005.04.003

43. Cheng LW, Stanker LH (2013) Detection of botulinum neurotoxin serotypes A and B using a chemiluminescent versus electrochemiluminescent immunoassay in food and serum. J Agric Food Chem 61: 755-760. https://doi.org/10.1021/jf3041963

44. Isbister GK, Oakley P, Whyte I, Dawson A (2001) Treatment of anticholinergic-induced ileus with neostigmine. Ann Emerg Med 38: 689-693. https://doi.org/10.1067/mem.2001.119458

45. Gomez HF, Johnson R, Guven H, McKinney P, Phillips S, et al. (1995) Adsorption of botulinum toxin to activated charcoal with a mouse bioassay. Ann Emerg Med 25: 818-822. https://doi.org/10.1016/S0196-0644(95)70214-8

46. Dembek ZF, Smith LA, Rusnak JM (2007) Botulism: cause, effects, diagnosis, clinical and laboratory identification, and treatment modalities. Disaster Med Public Health Prep 1: 122-134. https://doi.org/10.1097/DMP.0b013e318158c5fd

47. Tacket CO, Shandera WX, Mann JM, Hargrett NT, Blake PA (1984) Equine antitoxin use and other factors that predict outcome in type A foodborne botulism. Am J Med 76: 794-798. https://doi.org/10.1016/0002-9343(84)90988-4

48. Chang GY, Ganguly G (2003) Early antitoxin treatment in wound botulism results in better outcome. Eur Neurol 49: 151-153. https://doi.org/10.1159/000069073

49. Lonati D, Flore L, Vecchio S, Giampreti A, Petrolini VM, et al. (2015) Clinical management of foodborne botulism poisoning in emergency setting: an Italian case series. Clin Toxicol 53: 338.

50. Hatheway CH, Snyder JD, Searl JE, Edell TA, Lewis GE (1984) Antitoxin levels in botulism patients treated with trivalent equine botulism antitoxin to toxin types A., B., and E. J Infect Dis 150: 407-412. https://doi.org/10.1093/infdis/150.3.407

51. Black RE, Gunn RA (1980) Hypersensitivity reactions associated with botulinal antitoxin. Am J Med 69: 567-570. https://doi.org/10.1016/0002-9343(80)90469-6

52. Pirazzini M, Rossetto O (2017) Challenges in searching for therapeutics against botulinum neurotoxins. Expert Opin Drug Discov 12: 497-510. https://doi.org/10.1080/17460441.2017.1303476

53. Yu PA, Lin NH, Mahon BE, Sobel J, Yu Y, et al. (2017) Safety and improved clinical outcomes in patients treated with new equine-derived heptavalent botulinum antitoxin. Clin Infect Dis 66: 57-64. https://doi.org/10.1093/cid/cix816

54. Lonati D, Grassi GC, Lindal Baldvinsdottir L, Neou P, Moens J, et al. (2019) Poison centre data on botulism: results from an EAPCCT survey. Clin Toxicol 57: 453-454. https://doi.org/10.1080/15563650.2019.1598646

55. Santos JI, Swensen P, Glasgow LA (1981) Potentiation of *Clostridium botulinum* toxin by aminoglycoside antibiotics: clinical and laboratory observations. Pediatr 68: 50-54. https://doi.org/10.1542/peds.68.1.50

56. Azarnia Tehran D, Zanetti G, Leka O, Lista F, Fillo S, et al. (2015) Novel inhibitor prevents the peripheral neuroparalysis of botulinum neurotoxins. Sci Rep 5: 17513. https://doi.org/10.1038/srep17513

57. Duregotti E, Zanetti G, Scorzeto M, Megighian A, Montecucco C, et al. (2015) Snake and spider toxins induce a rapid recovery of function of botulinum neurotoxin paralysed neuromuscular junction. Toxins 7: 5322-5336. https://doi.org/10.3390/toxins7124887

58. Zanetti G, Azarnia Tehran D, Pirazzini M, Binz T, et al. (2015) Inhibition of botulinum neurotoxins interchain disulfide bond reduction prevents the peripheral neuroparalysis of botulism. Biochem Pharm 98: 522-530. https://doi.org/10.1016/j.bcp.2015.09.023

59. Qiao J, Hayes KC, Hsieh JTC, Potter PJ, Delaney GA (1997) Effects of 4-aminopyridine on motor evoked potentials in patients with spinal cord injury. J Neurotrauma 14: 135-149. https://doi.org/10.1089/neu.1997.14.135

60. Dock M, Ali AB, Karras A, Misset B, Garrouste-Orgeas M, et al. (2002) Treatment of severe botulism with 3,4-diaminopiridine. La Presse Med 31: 601-602.

61. Neal KR, Dunbar EM (1990) Improvement in bulbar weakness with guanoxan in type-B botulism. Lancet 335: 1286-1287. https://doi.org/10.1016/0140-6736(90)91360-M

62. Roblot P, Roblot F, Fauchere JL, Devilleger A, Marechaud R, et al. (1994) Retrospective study of 108 cases of botulism in Poitiers, France. J Med Microbiol 40: 379-384. https://doi.org/10.1099/00222615-40-6-379

63. Polo JM, Martin J, Berciano J (1996) Botulism and pregnancy. Lancet 348: 195. https://doi.org/10.1016/S0140-6736(05)66139-9

64. Magri K, Bresson V, Barbier C (2006) Botulism and pregnancy. J Gynecol Obstet Biol Reprod 35: 624-626. https://doi.org/10.1016/s0368-2315(06)76453-5

65. Badell ML, Rimawi BH, Rao AK, Jamieson DJ, Rasmussen S, et al. (2017) Botulism during pregnancy and the postpartum period: a systematic review. Clin Infect Dis 66: 30-37. https://doi.org/10.1093/cid/cix813

66. Hughes RA, Bihari D (1993) Acute neuromuscular respiratory paralysis. J Neurol Neurosurg Psychiatry 56: 334-343. https://doi.org/10.1136%2Fjnnp.56.4.334

67. Dressler D, Benecke R (2003) Autonomic side effects of botulinum toxin type B treatment of cervical dystonia and hyperhidrosis. Eur Neurol 49: 34-38. https://doi.org/10.1159/000067023

68. Dolly JO, Aoki KR (2006) The structure and mode of action of different botulinum toxins. Eur J Neurol 13: 1-9. https://doi.org/10.1111/j.1468-1331.2006.01648.x

69. Jenzer G, Mumenthaler M, Ludin HP, Robert F (1975) Autonomic dysfunction in botulism B: a clinical report. Neurology 25: 150-153. https://doi.org/10.1212/WNL.25.2.150

70. Koenig MG, Drutz DJ, Mushlin AI, Schaffner W, Rogers DE (1967) Type B botulism in man. Am J Med 42: 208-219. https://doi.org/10.1016/0002-9343(67)90020-4

71. Merz B, Bigalke H, Stoll G, Naumann M (2003) Botulism type B presenting as pure autonomic dysfunction. Clin Auton Res 13: 337-348. https://doi.org/10.1007/s10286-003-0118-2

72. Potulska-Chromik A, Zakrzewska-Pniewska B, Szmidt-Sałkowska E, Lewandowski J, Siński M, et al. (2013) Long lasting dysautonomia due to botulinum toxin B poisoning: clinical-laboratory follow up and difficulties in initial diagnosis. BMC Res Notes 6: 1-3. https://doi.org/10.1186/1756-0500-6-438

73. Truax BT (1984) Autonomic Disturbances in the Guillain-Barré Syndrome. Seminars in Neurology. Thieme Medical Publishers.

74. Dalos NP, Borel C, Hanley DF (1988) Cardiovascular autonomic dysfunction in Guillain-Barré syndrome. Therapeutic implications of swan-ganz monitoring. Arch Neurol 45: 115-117. https://doi.org/10.1001/archneur.1988.00520250125034

75. Hund EF, Borel CO, Cornblath DR, Hanley DF, McKhann GM (1993) Intensive management and treatment of severe Guillain-Barré syndrome. Crit Care Med 21: 433-446.

76. Hughes RA, Wijdicks EF, Benson E, Cornblath DR, Hahn AF, et al. (2005) Supportive care for patients with Guillain-Barre syndrome. Arch Neurol 62: 1194-1198. https://doi.org/10.1001/archneur.62.8.1194