Review

Eurasian Journal of Toxicology

Botulism: Clinical Features, Laboratory Insights And Management Options

Handan CIFTCI[1], Omer CANACIK[2]

[1]University of Health Sciences, Bursa Yuksek Ihtisas Training and Research Hospital, Department of Emergency Medicine, Bursa, Türkiye
[2]Dokuz Eylul University, School of Medicine Department of Emergency Medicine,Izmir, Turkey

Abstract

Botulism is a neuroparalytic disease caused by the neurotoxin produced by Clostridium botulinum (a gram-positive, anaerobic, endospore-forming bacillus). Botulinum neurotoxin (BoNT) is one of the most potent substances known, and seven toxin serotypes (serotypes A–G) have been identified. Type A serotype botulism is the most common cause of human botulism, and can occur in epidemic proportions. Botulism occurs after ingestion of food contaminated with BoNT, colonization of a wound by neurotoxin-producing Clostridium species, and exposure to botulinum neurotoxins by inhalation or injection. In all forms of botulism, progressive muscle weakness is usually seen, beginning in the cranial nerves and progressing in a proximal to distal manner to the extremities. This descending paralysis can lead to respiratory failure and death with involvement of the respiratory muscles. Treatment includes supportive care, intubation, and early administration of botulinum antitoxin.

Introduction

Botulism toxicity is caused by neurotoxins produced by *Clostridium botulinum* (*C. botulinum*) and occasionally by the closely related species *C. baratii* and *C. butyricum*[1]. *Clostridium botulinum* (*C. botulinum*) is a gram-positive, anaerobic, endospore-forming bacillus, and the causative agent is the botulinum neurotoxin (BoNT), one of the most potent biological substances known[2].

Clostridium botulinum is among the most resilient and efficient biological entities in nature and forms a spore that can withstand the harshest environmental conditions. In spore form, *C. botulinum* can be found ubiquitously in soil. It is estimated that spores can survive in liquid media for 30 years and under space conditions for many months[3]. When suitable environmental or laboratory conditions are provided (pH around 7.0, optimum growth at 35°C, and anaerobic conditions), *C. botulinum spores* can develop into toxin-producing bacilli[4]. *Clostridium botulinum* spores can withstand standard cooking and food processing measures, and for this reason, spores in foods preserved under appropriate conditions can germinate into the vegetative form and produce toxins. Consequently, the modern industrial canning technique was developed specifically to kill *C. botulinum* spores[3,4].

Botulism-related syndromes develop by exposure to botulinum neurotoxins through ingestion of preformed toxin (foodborne botulism), by colonization of a wound (wound botulism), or via the gastrointestinal tract (infant botulism and adult intestinal colonization botulism) by *Clostridium* species that produce neurotoxins, and via cosmetic or therapeutic injection (iatrogenic botulism)[1].

Epidemiology

Studies on botulism began in 1793 in Germany after an outbreak associated with blood sausages. *C. botulinum* was first isolated by Van Ermengem in 1897 from raw salted ham, which caused an epidemic that affected twenty-three people in Belgium and led to three deaths[5]. In 1949, Burgen and colleagues discovered that the botulinum neurotoxin inhibits neurotransmitter release. Later, a cellular mechanism of action was proposed that included three sequential steps, namely binding to neuronal acceptors, neuronal internalization, and intraneuronal action, and

Corresponding Author: Handan ÇİFTÇİ **e-mail:** drhandanc@hotmail.com
Received: 29.11.2024 • **Accepted:** 9.12.2024
Cite this article as: Çiftçi H, Canacık Ö. Botulism: Clinical Features, Laboratory Insights And Management Options. Eurasian J Tox. 2024;6(3): 49-54

©Copyright 2018 by Emergency Physicians Association of Turkey - Available online at https://dergipark.org.tr/ejtox

subsequently, the amino acid sequence of botulinum toxins was determined[6].

Outbreaks of foodborne botulism continue to be reported worldwide. The extent and duration of a botulism outbreak can vary depending on the type and source of exposure, as well as on whether the exposure is limited to a specific geographic location. The most common type of exposure to botulism is point source outbreaks, though intermittent common source outbreaks still account for a quarter of all outbreaks. Food preferences that vary by region indicate that the toxin type may also vary[7]. The use of biological agents to commit biocrime is a global problem. Due to its potential for use as a biological weapon, the storage and processing of BoNT requires enhanced safety measures and controls. BoNT is one of the most powerful and lethal biological agents known in nature, even with the lowest dose. In a person with an average body weight of 70 kg, 70 µg of ingested, 0.7-0.9 µg of inhaled, or 0.09-0.15 µg of intravenously administered concentrated BoNT/A toxin can be lethal[2]. It is not known whether exposure to purified toxin, such as might occur in the case of bioterrorism, would produce gastrointestinal signs and symptoms[1].

Pathophysiology

C. botulinum species are classified in four groups according to culture and serological characteristics[5]. Seven different botulinum toxin serotypes (BoNT A-G) have been identified through the study of botulism outbreaks in humans (BoNT/A, /B, /E and /F), birds (BoNT/C), cattle (BoNT/D), or isolated from soils (BoNT/G). The various BoNTs are produced by different *Clostridium botulinum* strains that exhibit heterogeneous bacteriological characteristics[8]. Most *Clostridium botulinum* strains produce a single antigenic type of toxin, but some strains produce two types of toxin, generally a large amount of one toxin and a small amount of the other (Table 1)[5]. BoNTs are single-chain polypeptides composed of a ≈50 kDa light chain and a ≈100 kDa heavy chain, linked by both disulfide bridges and non-covalent interactions functioning as a zinc-dependent endopeptidase[8]. Botulinum neurotoxin enters the bloodstream after ingestion, absorption from a colonized wound or intestine, inhalation, or injection. After entering the body, botulinum neurotoxins are transported by the circulatory system to the cholinergic synapses of the peripheral and cranial nervous system, where they affect the neuromuscular junctions. The toxin binds to high-affinity presynaptic receptors, and is then transported to the nerve cell by receptor-mediated endocytosis. The N-terminal heavy chain domain of BoNT facilitates the transport of the catalytic domain (light chain) from the endosomal membrane to the cytosol of the peripheral cholinergic nerve cell. After entering the cytosol, the light chain endoproteases prevent neurotransmitter (acetylcholine) release by specifically cleaving soluble N-ethylmaleimide-sensitive factor attachment protein receptor (SNARE) proteins in the postsynaptic nerve terminals. These proteins are the synaptosomal-associated protein 25 kDa (SNAP-25; cleaved by BoNT/A, BoNT/C, and BoNT/E), syntaxin (cleaved by BoNT/C), and synaptobrevin (also known as vesicle-associated membrane protein [VAMP], cleaved by BoNT/B, BoNT/D, BoNT/F, and BoNT/G)[2,3]. This causes descending symmetrical flaccid paralysis and cranial nerve palsy of varying severity[8]. In spite of their toxicity, they produce a prolonged but reversible effect at synapses[6,9].

In humans, botulism most often originates from strains producing toxin types A, B or E, and sometimes also from strains producing toxin type F[5]. Toxin type A in particular produces the most severe syndrome, and the proportion of patients requiring mechanical ventilation is high[9]. Toxin type B usually causes milder disease than type A and is the group with the lowest mortality. Cases of toxin types C, D, F and G are rare in humans, and this may be attributed to their lower presence in soil. Human cases caused by toxin type E are generally associated with the consumption of foods originating from water, producing a syndrome that includes gastrointestinal symptoms of varying severity[3]. Outbreaks involving toxin type E are the group with the shortest incubation period7. Moreover, although all toxin types can easily produce botulism in experimental animal models, conditions in the human intestine are not normally conducive to the germination of *C. botulinum* spores[9].

Clinical findings

Studies report that the symptoms of botulism are mostly food-related and most commonly develop due to toxin type

Table 1: Clostridium botulinum: Groups, Neurotoxins Formed, and Main Species Affected[2,5]

Group	Neurotoxin formed	Main species affected
Group I *Clostridium botulinum* (Proteolytic)	A	Humans, horses
	B	Humans, cattle, horses
	F	Humans
Group II *Clostridium botulinum* (Non-proteolytic)	B	Humans
	E	Humans, fish, birds
	F	Humans
Group III *Clostridium botulinum*	C	Birds, farmed chicken and pheasants, horses
	D	Cattle, sheep
Group IV *Clostridium argentinense*	G	No reported outbreaks, environmentally isolated
Group V *Clostridium baratii*	F	Humans
Group VI *Clostridium butyricum*	E	Humans

Table 2: Clinical criteria for early diagnosis of botulism[9,10]

1) Afebrile (≤37.8°C or ≤100.4°F)
2) At least one of the following symptoms: Blurred vision Double vision Difficulty speaking, including slurring Any change in voice, including hoarseness Dysphagia/accumulation of secretions/drooling Thick tongue
3) At least one of the following symptoms: Ptosis Extraocular palsy/decreased ability to track objects/tiredness caused by blocking light shining into the eyes Facial paresis/loss of facial expression/accumulation of secretions or milk/poor feeding/poor sucking when using a pacifier/tiredness while eating Fixed pupils Descending paralysis starting from the cranial nerves

A. Although the disease is most often associated with larger outbreaks, sporadic cases also exist. The reported incubation period ranges between 1-12 days, while the shortest reported incubation period is two hours. While the average time from the onset of the disease to hospitalization is two days, hospital admissions can vary up to a period of 14 days. The toxin dose exposed to is closely related to the incubation period and the severity of the disease[1].

The most common signs and symptoms of botulism are descending paralysis, dysphagia, weakness, diplopia, blurred vision, dysphonia, and dysarthria. Additionally, nausea, vomiting, ptosis, respiratory distress and respiratory failure may be observed in many patients[1,10]. There is a high need for mechanical ventilation in patients presenting with respiratory distress, especially in botulism due to toxin type A9. It is reported that most of these patients are admitted to hospital within the first 48 hours from the onset of the disease and are intubated1. Therefore, public health authorities need to take this situation into consideration, especially in cases where bioterrorism is suspected[4].

Less frequently, ocular palsy, dyspnea, dry mouth, abdominal pain, dizziness, abnormal pupillary reaction, and dilated pupils may also occur. Involvement of one or more cranial nerves may be seen accompanying the symptoms at the time of admission to hospital. These patients should be evaluated for weakness that progresses in a proximal to distal manner and for respiratory involvement[1,8]. Table 2 presents clinical criteria for early diagnosis of botulism[9,10].

The botulism patient is usually alert and has no fever provided there is no secondary infection. Fever is also rare in infants and young children, but may be more common than in adults10. When clouding of consciousness occurs in patients, respiratory failure, drug or alcohol use, pre-existing disease, or concurrent infection should be considered. Postural hypotension may occur[11].

Botulism causes prolonged flaccid paralysis which can last weeks or months. Aspiration of oral secretions and stomach contents may occur due to loss of protective airway reflexes. In the acute phase, death usually occurs due to early respiratory failure, while in the later stages of the disease, it is caused by long-term intensive care complications such as ventilator-associated pneumonia and deep vein thrombosis[9]. The mortality rate due to botulism increases with age. Mortality rates have decreased from 50% to less than 1% in recent years thanks to the developments in intensive care. Mortality rates are higher in wound botulism patients (15% to 17%) and lower in infant botulism patients (less than 1%). In recovered patients, it might not be possible for muscle strength and endurance to return to normal for up to 1 year, and long-term psychological problems may also occur[11].

Syndromes related to botulism

1. **Foodborne botulism:** Symptoms may appear within 12-36 hours or up to eight days after consuming food containing botulinum neurotoxin[5]. Foodborne botulism usually occurs due to inadequate storage conditions and exposure to undercooked home-canned foods or after consuming contaminated food from commercial sources[11]. The first symptoms may be nausea and vomiting. However, these symptoms may be caused by the toxin or may be due to other products of *C. botulinum* or other degradation products. Initially, the toxin causes symptoms such as double vision, inability to focus, ptosis, dry mouth, difficulty in speaking clearly (dysphonia), and dysphagia. Failure of the body muscles, especially the respiratory and cardiac muscles, can result in death. Some patients may be misdiagnosed with Guillain-Barré syndrome or myasthenia gravis instead of botulism[5]. Foodborne botulism outbreaks usually affect a small number of people. However, since large outbreaks are possible ("epidemic potential"), foodborne botulism is a public health emergency[9]. A botulism outbreak may escalate into a mass casualty incident when emergency medical services resources are inadequate in relation to the number and severity of cases. This can put great pressure on hospitals, as it requires long-term use of emergency services and frequently, intensive care facilities[12].
2. **Wound botulism (exposure to botulinum neurotoxin from a wound colonized by bacteria):** Wound botulism may develop in traumatic injuries and infected surgical wounds. The anaerobic conditions that occur in a wound abscess provide a suitable environment for bacteria to proliferate, and infection begins when BoNT passes into the bloodstream. Wound botulism is a risk for drug users who inject subcutaneously or intramuscularly and can cause outbreaks among drug users due to shared-use products. In some cases of wound botulism, nausea and vomiting may occur due to drug or alcohol use, botulism-induced ileus, diplopia, or coinfection (e.g., bacteremia, pneumonia)[1,3,5].

3. **Inhalation botulism:** This may occur in laboratory workers after inhalation of toxins and may develop after inhalation of spores in drugs[5]. It was first identified among German research laboratory workers in 1962. Clinical symptoms are similar to botulism caused by ingestion of the toxin, except for the absence of gastrointestinal symptoms (e.g., nausea, vomiting, abdominal cramps, diarrhea)[4].
4. **Iatrogenic botulism:** This may develop after exposure to botulinum neurotoxin through injection of high-concentration botulinum toxin for cosmetic or therapeutic purposes[13].
5. **Infant botulism:** This is seen in babies under one year old and has the highest incidence between 6 weeks and 6 months. Infant botulism occurs as a result of ingestion of spores that produce the toxin in vivo[11]. Following ingestion, the spores undergo germination and the organism can become established in the intestines of young infants. The most important reason for this is that the normal intestinal flora is not yet established enough to prevent this colonization. The infant experiences constipation, generalized weakness, and progressive paralysis and other neurological symptoms. Samples were taken from some patients diagnosed with sudden infant death syndrome and the presence of *C. botulinum* spores was detected[5]. Honey consumption is a risk factor for the disease, but this probably accounts for only 20% of cases. The clinical findings resemble adult forms of the disease, with common symptoms such as difficulty in sucking and swallowing, change in voice tone, ptosis, and drooping neck, and these symptoms may progress to general flaccid paralysis and respiratory failure[13].
6. **Adult intestinal colonization botulism:** This is associated with intestinal abnormality or surgery and/or antibiotic treatment. In most adults, the intestinal flora prevents the establishment of any ingested *C. botulinum* spores. Since it rarely occurs or is poorly recognized, few adult cases of this type of botulism have been reported[5,13].

Diagnostic methods for detecting botulism

We know that patients with suspected botulism receive a delayed diagnosis, as complete blood count and cerebrospinal fluid (CSF) examination and imaging methods are generally evaluated as normal. After 7 days of the disease, the amount of CSF protein may be found to have increased[14]. The Tensilon test (edrophonium test) can be used to rule out myasthenia gravis. Electrodiagnostic and neuromuscular conduction tests can be performed to reveal muscle weakness, and in the event of a large outbreak, electrodiagnostic studies can differentiate diseases such as Guillain-Barré syndrome and myasthenia gravis from botulism, thus contributing to the change of treatment. Repetitive nerve stimulation (RNS), electromyography (EMG), and nerve conduction studies (NCS) are recommended as electrodiagnostic studies. The distinctive classical findings seen in botulism include an increase in compound motor nerve action potential amplitude, repetitive nerve stimulation (RNS) rates of 30-50 Hz, fibrillation, decreased use of muscle units, decreased duration of muscle unit potentials in electromyography (EMG), and decreased motor-evoked amplitude on a nerve conduction study (NCS) when other findings are normal[15]. However, electrodiagnostic studies may be normal, and therefore unhelpful, in the early stages of the disease[15].

The diagnosis of symptomatic botulism is confirmed by identifying the neurotoxin:

1. Botulism neurotoxin in the stool, serum and gastric fluid,
2. Growth of *Clostridium* species in the stool or wound culture (those that produce neurotoxins),
3. The diagnosis is confirmed by detecting botulinum neurotoxin in food consumed by the symptomatic person[13.]

The gold standard for laboratory confirmation of botulism is the mouse bioassay[2,3]. Real-time polymerase chain reaction can detect *Clostridium* species growing in culture. The botulinum neurotoxin can be identified by distinguishing serotypes within hours using the mass spectrophotometry method. Sample collection, transportation and storage should be carried out in accordance with the laboratory's botulism recommendations[16,17].

Neurological observation of botulism

Patients initially experience nausea and vomiting, while cranial nerve palsy occurs in all patients over time. Respiratory failure and extremity weakness may occur due to involvement[18].

Respiratory observation of botulism

Around half of patients may require intubation due to involvement of the diaphragm or upper airway (preventing aspiration) muscles[1,18]. The need for intubation can be determined by spirometry, physical examination, respiratory rate, or nasal respiratory pressure measurement. Assessment with blood gas may not be reliable in the early period[19-20-21].

Treatment

Treatment includes supportive care, intubation and antitoxin administration. If botulism is suspected, it should be treated with botulinum antitoxin (BAT). If neurological findings continue to worsen after administering one vial of BAT, diagnoses other than botulism should be considered. BAT administration (ideally within the first 24 hours, preferably within 48 hours) leads to a decrease in mortality and morbidity. In BAT administration, skin sensitivity testing is

not required[22]. The antitoxin does not reverse paralysis, but halts the progression. BAT treatment may be reconsidered in children who continue to deteriorate neurologically despite dose-adjusted BAT treatment for foodborne botulism[23]. If botulism is suspected in pregnant women, BAT treatment should be performed[24]. Until she receives BAT, the mother should cease breastfeeding, and express and discard her milk in consultation with a breastfeeding specialist. Aminoglycosides, magnesium, clindamycin, tetracycline or calcium should be given to botulism patients only after careful assessment and appropriate monitoring[25,26]. Activated charcoal administered orally may cause morbidity in cases of ileus or aspiration[27,28]. The effectiveness of activated charcoal and plasmapheresis has not yet been fully proven[29]. Clinicians should be prepared for dry eye, ileus, pressure sores, urinary retention, and deep vein thrombosis[30-31]. Since toxin does not pass through the blood-brain barrier in botulism patients, it should be remembered that their cognitive functions are intact and psychological procedures should be applied[32,33].

Conclusion

- In crisis and emergency situations, early clinical diagnosis of botulism should be made by criteria.
- Botulism patients should be examined frequently.
- The airway should be protected.
- It should not be forgotten that the patients' cognitive functions are intact.
- BAT treatment should be given in case of doubt (regardless of the route of exposure).
- One should be prepared for an epidemic situation.

References

1. Chatham-Stephens K, Fleck-Derderian S, Johnson SD, Sobel J, Rao AK, Meaney-Delman D. Clinical features of foodborne and wound botulism: a systematic review of the literature, 1932–2015. Clinical Infectious Diseases. 2018; 66(suppl_1):11-6. https://doi.org/10.1093/cid/cix811
2. Rawson AM, Dempster AW, Humphreys C M, Minton NP. Pathogenicity and virulence of Clostridium botulinum. Virulence. 2023;14(1): 2205251. https://doi.org/10.1080/21505594.2023.2205251
3. Sobel J. Chapter 5 Botulism. In: Lutwick LI, Lutwick SM, editors. Beyond Anthrax, The Weaponization Of İnfectious Disease, Humana Press. 2009.p.85–105. https://doi.org/10.1007/978-1-59745-326-4_5
4. Dembek ZF, Smith LA, Rusnak JM. Botulism: cause, effects, diagnosis, clinical and laboratory identification, and treatment modalities. Disaster medicine and public health preparedness. 2007;1(2);122-34. https://doi.org/10.1097/DMP.0b013e318158c5fd
5. Lund BM, Peck MW. Clostridium botulinum. In: Guide to Foodborne Pathogens, 2nd ed.; Labbé RG, García S, editors. John Wiley & Sons Ltd.: Chichester, UK. 2013; pp. 91–111. https://doi.org/10.1002/9781118684856.ch6
6. Caleo M, Restani L. Exploiting botulinum neurotoxins for the study of brain physiology and pathology. Toxins. 2018; 10(5); 175. https://doi.org/10.3390/toxins10050175
7. Fleck-Derderian S, Shankar M, Rao AK, Chatham-Stephens K, Adjei S, Sobel J, Pillai S K. The epidemiology of foodborne botulism outbreaks: a systematic review. Clinical infectious diseases. 2018;66(suppl_1):73-S81. https://doi.org/10.1093/cid/cix846
8. Humeau Y, Doussau F, Grant NJ, Poulain B. How botulinum and tetanus neurotoxins block neurotransmitter release. Biochimie. 2000; 82(5); 427-446. https://doi.org/10.1016/S0300-9084(00)00216-9
9. Rao AK, Sobel J, Chatham-Stephens K, Luquez C. Clinical Guidelines for Diagnosis and Treatment of Botulism, 2021. MMWR Recommendations Rep. 2021;70(2):1-30.
10. Rao AK, Lin NH, Griese SE, Chatham-Stephens K, Badell ML, Sobel, J. Clinical criteria to trigger suspicion for botulism: an evidence-based tool to facilitate timely recognition of suspected cases during sporadic events and outbreaks. Clinical Infectious Diseases. 2018;66(suppl_1), 38-42. https://doi.org/10.1093/cid/cix814
11. Singh M, Fernández-Frackelton M. Section Twelve: Infectious Diseases, Chapter 118: Bacteria. In: Walls RM, Robert S, Hockberger R S, Gausche-Hill M, Erickson TB, Wilcox SR, editors. Rosen's, Emergency Medicine: Concepts and Clinical Practise, 10th Edition, Elsevier: 2023. p. 1586-1609.
12. Krebs W, Higgins T, Buckley M, Augustine JJ, Raetzke BD, Werman HA. Botulism outbreak in a regional community hospital: lessons learned in transfer and transport considerations. Prehospital emergency care. 2019;23(1);49-57. https://doi.org/10.1080/10903127.2018.1476636
13. Sobel J. The Organism And Its Toxins. Clinical Infectious Diseases. 2005 Oct 15;41(8):1167-73. https://doi.org/10.1086/444507.
14. Fokke C, Van Den Berg B, Drenthen J, Walgaard C, Van Doorn PA, Jacobs BC. Diagnosis of Guillain-Barré syndrome and validation of Brighton criteria. Brain. 2014 Jan;137(Pt 1):33-43. https://doi.org/10.1093/brain/awt285. Epub 2013 Oct 26. PMID: 24163275.
15. Maselli RA, Bakshi N. AAEM case report 16. Botulism. American Association of Electrodiagnostic Medicine. Muscle Nerve. 2000 Jul;23(7):1137-44. https://doi.org/10.1002/1097-4598(200007)23:7<1137::aid-mus21>3.0.co;2-7. PMID: 10883013.
16. Maslanka SE, Solomon HM, Sharma S, Johnson EA. Chapter 32. Clostridium botulinum and its toxins. In: Salfinger Y, Tortorello ML, editors. Compendium of methods for the microbiological examination of foods: Washington, DC: American Public Health Association Press; 2013.p.1-11.
17. Kalb SR, Baudys J, Wang D, Barr JR. Recommended mass spectrometry-based strategies to identify botulinum neurotoxin-containing samples. Toxins (Basel). 2015 May 19;7(5):1765-78. https://doi.org/10.3390/toxins7051765. PMID: 25996606; PMCID: PMC4448173.
18. Griese SE, Kisselburgh HM, Bartenfeld MT, Thomas E, Rao AK, Sobel J, Dziuban EJ. Pediatric Botulism and Use of

Equine Botulinum Antitoxin in Children: A Systematic Review. Clin Infect Dis. 2017 Dec 27;66(suppl_1):17-29. https://doi.org/10.1093/cid/cix812. PMID: 29293924.

19. Hutchinson D, Whyte K. Neuromuscular disease and respiratory failure. Pract Neurol. 2008 Aug;8(4):229-37. https://doi.org/10.1136/pn.2008.152611. PMID: 18644909.

20. Hughes RA, Bihari D. Acute neuromuscular respiratory paralysis. J Neurol Neurosurg Psychiatry. 1993 Apr;56(4):334-43. https://doi.org/10.1136/jnnp.56.4.334. PMID: 8482951; PMCID: PMC1014946.

21. Lawn ND, Fletcher DD, Henderson RD, Wolter TD, Wijdicks EF. Anticipating mechanical ventilation in Guillain-Barré syndrome. Arch Neurol. 2001 Jun;58(6):893-8. https://doi.org/10.1001/archneur.58.6.893. PMID: 11405803.

22. Yu PA, Lin NH, Mahon BE, Sobel J, Yu Y, Mody RK, Gu W, Clements J, Kim HJ, Rao AK. Safety and Improved Clinical Outcomes in Patients Treated With New Equine-Derived Heptavalent Botulinum Antitoxin. Clin Infect Dis. 2017 Dec 27;66(suppl_1):57-64. https://doi.org/10.1093/cid/cix816. PMID: 29293928; PMCID: PMC5866099.

23. Botulism antitoxin heptavalent. (A, B, C, D, E, F, G)—(Equine) [package insert]. Gaithersburg, MD: Cangene Corporation (as Emergent Biosolutions); 2017. https://www.fda.gov/media/85514/download

24. Badell ML, Rimawi BH, Rao AK, Jamieson DJ, Rasmussen S, Meaney-Delman D. Botulism During Pregnancy and the Postpartum Period: A Systematic Review. Clin Infect Dis. 2017 Dec 27;66(suppl_1):30-S37. https://doi.org/10.1093/cid/cix813. PMID: 29293925.

25. Robinson RF, Nahata MC. Management of botulism. Ann Pharmacother. 2003 Jan;37(1):127-31. https://doi.org/10.1345/aph.1C034. PMID: 12503947.

26. Lee JH, Lee SI, Chung CJ, Lee JH, Lee SC, Choi SR, Oh JN, Bae JY. The synergistic effect of gentamicin and clindamycin on rocuronium-induced neuromuscular blockade. Korean J Anesthesiol. 2013 Feb;64(2):143-51. https://doi.org/10.4097/kjae.2013.64.2.143.

27. Mazuet C, Dano J, Popoff MR, Créminon C, Volland H. Characterization of botulinum neurotoxin type A neutralizing monoclonal antibodies and influence of their half-lives on therapeutic activity. PLoS One. 2010 Aug 26;5(8):e12416. https://doi.org/10.1371/journal.pone.0012416.

28. Osterhoudt KC, Alpern ER, Durbin D, Nadel F, Henretig FM. Activated charcoal administration in a pediatric emergency department. Pediatr Emerg Care. 2004 Aug;20(8):493-8. https://doi.org/10.1097/01.pec.0000136064.14704.d1.

29. Paterson DL, King MA, Boyle RS, Pond SM, Whitby M, Wright M, Henderson A. Severe botulism after eating home-preserved asparagus. Med J Aust. 1992 Aug 17;157(4):269-70. https://doi.org/10.5694/j.1326-5377.1992.tb137135.x. PMID: 1435446.

30. Merz B, Bigalke H, Stoll G, Naumann M. Botulism type B presenting as pure autonomic dysfunction. Clin Auton Res. 2003 Oct;13(5):337-8. https://doi.org/10.1007/s10286-003-0118-2. PMID: 14564656.

31. Jenzer G, Mumenthaler M, Ludin HP, Robert F. Autonomic dysfunction in botulism B: a clinical report. Neurology. 1975 Feb;25(2):150-3. https://doi.org/10.1212/wnl.25.2.150.

32. Simpson LL. Identification of the major steps in botulinum toxin action. Annu Rev Pharmacol Toxicol. 2004;44:167-93. https://doi.org/10.1146/annurev.pharmtox.44.101802.121554.

33. Cohen RE, Anderson DL. Botulism: emotional impact on patient and family. J Psychosom Res. 1986;30(3):321-6. https://doi.org/10.1016/0022-3999(86)90009-7.