Knowledge of Botulism among Health Care Students

Kara Marie Schmidt

The University of Toledo

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Dedication

I am grateful for the ever present, and enormous amount of encouragement and support I received from my husband from the first time I ever mentioned my desire to become a Physician Assistant through the entire demanding program. I greatly appreciate the sacrifices made by my husband and children in giving me the gift of time and patience to achieve this professional goal. This goal would also not have been possible without my parents and in-laws who gave selflessly of their time and energy for my children and in support of me. Lastly, I am indebted to my classmates from whom I learned from and leaned on throughout the challenges faced these past two and a quarter years.

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Chapter 1/Introduction

The bacterial species *Clostridium botulinum* is capable of producing botulinum toxin, arguably the most potent toxin known. Symptoms can occur with just one microgram of toxin per kilogram of body weight, which is 275 times more poisonous than cyanide (Van Rynen, Rega, Budd, & Burkholder-Allen, 2009). Found in soil, this ubiquitous species is a gram-positive, rod-shaped, spore-forming, obligate anaerobic bacterium. The spores are heat resistant, killed only by exposing to high heat for several minutes (Josko, 2004). But, given the ideal environment the spores will germinate into toxin-producing bacteria. This toxin then causes botulism within its host, a rare but life-threatening illness presenting with symmetric, descending paralysis that can be fatal when it affects the body's ventilitory function.

Three forms of botulism poisoning were initially accepted until recent years when three more have been reported. They are differentiated by the means of acquisition: food-borne, infant (due to ingestion of spores via raw honey or dust), and wound. The additional types are adult intestinal (similar to infant botulism because toxin is produced within the gastrointestinal tract of an infected adult host), inadvertent injection-related, and inhalational botulism (of which there are 3 lab cases reported, and the form that would occur if aerosolized toxin was released in an act of bioterrorism) (Horowitz, 2005). Bioterrorism is the use of biological agents (both infectious agents and the toxins they produce) to intimidate governments or societies on behalf of an ideological cause. It is estimated that one gram of aerosolized botulinum toxin could kill at least one million people (Arnon et al., 2001).

C. botulinum toxin is too large to cross the blood-brain barrier and therefore it targets cells in the peripheral nervous system where acetylcholine acts to activate muscles. The toxin is a polypeptide made up of a heavy and a light chain joined together by a single disulfide bond. The light chain, an endopeptidase, breaks down the protein complex required for the fusion of acetylcholine-containing vesicles at the nerve terminal. By blocking this step, the toxin irreversibly acts to prevent release of acetylcholine at the neuromuscular junction, thus inhibiting muscle contraction and causing paralysis. Depending upon the amount of toxin absorbed into the blood, botulism may manifest mildly or rather severely, causing such paralysis that some patients may require months of ventilator support. It is important to recognize that botulism presents as symmetric, descending flaccid paralysis that always begins in the bulbar musculature as multiple cranial nerve palsies. Common symptoms are sometimes referred to as the "4 D's": diplopia, dysarthria, dsyphonia, and dysphagia. Additionally, botulism (except wound botulism) is an afebrile illness that does not decrease the patient's mental status despite ensuing weakness and paralysis until later in the process when carbon dioxide narcosis occurs due to hypoventilation (Arnon et al., 2001).

Treatment includes close monitoring for respiratory failure, as this is the most common cause of death due to botulism poisoning. Supportive therapy may include mechanical ventilation, and nutritional support via tube feeding or hyperalimentation due to dysphagia and other complications. Other than supportive therapy, the only treatment available is antitoxin therapy. And if given early, will minimize further disruption of the release of acetylcholine in other nerves, but not reverse present paralysis from nerves

already affected (Arnon et al., 2001). There are two types of medications available in the United States: antitoxin and an immune globulin. First is equine serum botulism antitoxin, which used to treat children over one year of age and adults. Secondly, there is human-derived botulinum immune globulin (BIG-IV or BabyBIG), which is used for infants less than one year of age. The antitoxin is a form of artificial acquired passive immunity (i.e. antibodies transferred from horse or human) that attaches and neutralizes circulating toxin that has not entered any cell or receptor site. Therapy available for infants less than one year of age is human-derived botulinum immune globulin (Baby-BIG) (Horowitz, 2005). At any age, recovery occurs when the body sprouts new motor axon threads for reinnervation of paralyzed muscle fibers, which takes up to six months (Arnon et al., 2001). According to Horowitz (2005), "Delay to administering the antitoxin is the most important factor that affects clinical course and outcome" (p. 836). Delay must be minimized by prompt identification of symptoms, which is essential for a favorable prognosis and effective treatment of botulism. Ultimately this all begins with awareness and knowledge.

According to the Centers for Disease Control and Prevention (CDC), approximately 145 cases of botulism are reported yearly in the United States. The majority of cases are infant (65%), and less frequent is food-borne (15%), and wound botulism (20%). Seldom is adult enteric infection reported (U.S. Centers for Disease Control and Prevention, 2008).

Due to its extreme potency, lethality, ease of production, and stability in environment, botulinum toxin is a major threat as a naturally occurring illness and a biological weapon (Shukla & Sharma, 2005). Thus, it is listed along with anthrax,

plague, smallpox, tularemia, and viral hemorrhagic fevers as Category A agents on the CDCs list of organisms that pose the highest risk/highest priority to national security. Consider that botulinum toxin is the most poisonous substance known and is 275 times more poisonous than cyanide and 100,000 times more toxic than sarin (Van Rynen et al., 2008).

According to Arnon et al. (2001), only the tiniest amount of toxin, 0.001 micrograms per kilogram will cause symptoms in the human being. Additionally the same researchers noted, "A single gram of crystalline toxin, evenly dispersed and inhaled, would kill more than 1 million people. Even if practical details might make this type of dissemination difficult, it is a nonetheless an accurate statement. While means of bioterrorism via inhalation of botulinum toxin has been studied, so has its ingestion via intentionally contaminated food. Specifically, a mathematical model was calculated by Wein and Liu in 2005 which predicted that 1 gram of the toxin in commercially-distributed milk consumed by more than 550,000 urban dwellers would result in 100,000 casualties. Consider 10 grams of toxin in the previous scenario and the casualties could rise to 500,000. Therefore, unlike a hypothetical smallpox epidemic, the threat of a mass casualty incident from botulism should be considered a definite and credible potential threat.

With this unfortunate distinction, the effects of botulinum toxin should be well known by at least those in the health care community. Currently, it is unclear how knowledgeable students in the health care professions are about botulism. Great efforts should be taken to ensure that at a minimum, those entering into and practicing in

healthcare are prepared to recognize and respond to this illness in the time necessary to avoid a fatal outcome.

Chapter 2/Literature Review

According to the CDC (2008), rates of food-borne and infant botulism have remained steady in recent years, while wound botulism has seen an increase due to black tar heroin use. In addition to *C. botulinum* causing these naturally occurring forms of disease, the CDC has placed this species among the most worrisome for their use as a bioweapon. Numerous studies demonstrate the seriousness of botulism regardless of its route to producing disease. Due to the number of people who could be sickened with so little toxin makes the threat of intentional release in an act of terrorism very concerning and very real.

The interest in botulinum toxin as a biologic agent began at least 65 years ago. In fact, during the Japanese's occupation in Manchuria in the 1930's, their biological warfare group (Unit 731) fed cultures of *C. botulinum* to prisoners and produced lethal outcome. In addition, during WWII, several countries investigated the use of botulinum toxin as a biologic agent. And, even after 1972, when the offensive research and production of biological weapons was prohibited by the Biological and Toxin Weapons Convention, Iraq and the Soviet Union still produced the toxin for use as a weapon (Arnon et al., 2001).

Indeed, terrorists continued its use as demonstrated in the early 1990's when the Japanese cult, Aum Shinrikyo released an ineffective *C. botulinum* aerosol preparation against a U.S. military facility (Horowitz, 2005). Additionally, United Nations inspections of Iraq after the first Gulf War, revealed approximately 19,000 liters of concentrated botulinum toxin were produced, more than any other biological agent. Approximately half of that was loaded into military weapons, but much remains unaccounted for (Arnon

et al., 2001). Additionally, countries that have developed or are believed to be developing botulinum toxin for use as a weapon currently and are listed by the US government as "state sponsors of terrorism" include Iran, Iraq, North Korea, and Syria (Arnon et al., 2001). While some researchers may question the potential for terrorist use of aerosol dissemination of botulinum toxin due to difficulty in stabilization and concentration, inhalational botulism has actually occurred in humans accidentally. Moreover, there is concern of deliberate contamination of the food or water supply.

Of the bioterror attacks not involving genetic engineering, one of the possible scenarios that pose the greatest threat to humans is a release of botulinum toxin in cold drinks. For example, the milk supply has been studied as a potential bioterrorist target because of the multiple and rapid steps in transportation and processing, wide distribution, and for the fact that it is a highly consumed food. In 2005, Wein and Liu described a hypothetical bioterrorism scenario that analyzed the outcome if 1 gram of botulinum neurotoxin was released into a holding tank of approximately 50,000 gallons of raw milk. It was determined that if all of this milk was distributed and consumed without detection, there would be a mean of 100,000 casualties based on 68.4% inactivation of the toxin by heat treatment (170^0 F/77^0C for 15 min.). Of great importance is noting that early symptomatic detection in this model could avoid about 67% of the casualties.

In response to this mathematical model, Weingart and Schreiber et al., (2010) analyzed milk contamination with botulinum toxin via a laboratory-scale pasteurization process. They used heat treatment commonly used by the dairy industry called high-temperature short-time (HTST) pasteurization similar to industry standards (161^0F/72^0C

for 15 sec.), and showed it would inactivate at least 99.5% of botulinum toxin in their milk samples. But, researchers also admit that "other complex food matrices or slight changes to the heating parameters might have a different effect on the stability of BoNT" (botulinum neurotoxin).

The possibility of contamination of water supplies is an additional threat, as botulinum toxin has been shown to retain up to 50% of its activity for 5 to 70 days in various types of untreated water and beverages. There are many barriers to be noted such as dilution, and inactivation from chlorine, other disinfectants, hydrolysis, sunlight, microbes, and filtration to make this contamination less likely. But, personal-use water purification devices without reverse osmosis fail to remove the toxin. Most bottled water in the United States does utilize the process of reverse osmosis, but not all international bottled water sources do. Still, toxin introduction to municipal water supplies or to bottled water or other beverages is a threat that should be considered (Villar, Elliott, Davenport, & Davenport, 2006).

With the reality that this toxin can cause disease via intentional use or otherwise, the health care community must be able to clinically recognize its symptoms and diagnose it efficiently. Indeed, in the United States, the detection of such events is based on clinician reporting to the Centers for Disease Control and Prevention in order to maintain their well-established surveillance system.

To illustrate the importance of the clinician's ability to recognize botulism poisoning, the following is a discussion of a case that went undetected for a prolonged period of time. In Vancouver between July and September of 1985 one bottle of garlic intermittently used in a restaurant caused a botulism outbreak affecting 36 people over

2 distinct clusters of illnesses separated by about a month. "More than 2 months after the beginning of the outbreak and several days after the last patient ate at the restaurant, botulism was first diagnosed", even when the median incubation period from ingestion to neurologic symptoms was only 2.3 days and the median period from ingestion to hospitalization was 7 days (St. Louis et al., 1988). In this outbreak, the patients did present with typical symptoms of botulism poisoning including muscle weakness (100%), dysphagia (91%), blurred vision (88%), dry mouth (85%), dysphonia (82%), dizziness (73%), and diplopia (67%). Most patients were hospitalized, some required mechanical ventilation, and there was no mortality associated with this outbreak. The most common working diagnoses in these patients before correct identification of botulism was: myasthenia gravis, stroke, the atypical or descending variant of Guillain-Barre syndrome, psychiatric disorders and viral syndromes.

Since the events in September 2001, studies have been done to assess physician preparedness for bioterrorism. One national survey of hundreds of randomly selected family physicians, revealed that about 75% of them "did not feel prepared to respond to a bioterrorist attack" (Chen, Hickner, Fink, Galliher, & Burstin, 2001). And, only 24% of those surveyed thought they "could recognize signs and symptoms of an illness in their patients due to bioterrorism" and 38% rated their current knowledge of the diagnosis and management of bioterrorism related illness as "poor". Lastly, in this study, only 18% of physicians reported previous training in bioterrorism preparedness.

A similar study in 2003 was conducted, but compared the preparedness of emergency and primary care physicians via a random sample, national survey. More emergency physicians reported that they were "well prepared to play a role in

responding to possible bioterror attack" than primary care physicians (43% vs. 21%) (Alexander, Larkin, & Wynia, 2006).

Another study in 2005 assessed the ability of physicians to diagnose and manage illness caused by four of the category A bioterrorism agents (smallpox, anthrax, botulism, and plague). Six hundred thirty one internal medicine physicians (including residents and some attending physicians) were included in the research which showed 49.6% were able to correctly diagnose botulism poisoning on the pretest and about 90%, after the didactic education, on the post test. Results of the study "demonstrated that physicians are undertrained in the diagnosis and management of infection caused by 4 pathogens identified as likely to be used in a potential bioterrorism attack" (Cosgrove, Perl, Song, & Sisson, 2005).

Since the 2001 terrorist attack with anthrax, research regarding botulism poisoning has shifted from various cases of food borne ingestion, to the threat of its use as a weapon and our preparedness to recognize such an event. With botulinum toxin as one of the top agents feared for use in such an attack, it places healthcare providers at the forefront in its initial detection. Therefore, with this group holding responsibility to recognize and report botulism, it's worth not only asking them how ready they feel to take on such a task, but to actually determine how much they know on the subject. In addition, most studies focus their attention on physicians' ability to diagnose botulism poisoning, but in reality, health care is a multidisciplinary team effort that involves nurses in triage to any and all type of clinical provider. Today there are physician assistants and nurse practitioners also work in emergency departments and urgent care centers, diagnosing and treating patients. And some patients may present at their

pharmacy and question the pharmacist on which over the counter medication may help their symptoms instead of, or before presenting in to an emergency department.

Essentially, potential providers in the healthcare system have expanded over the years and so too, this study attempts to analyze the knowledge of botulism in such a group as a means to identify preparedness to handle this disease whether it is of natural or intentional cause.

Chapter 3/Methods

A survey questionnaire was developed for use as a tool to assess the current knowledge of botulism in health care students. The study was reviewed and approved by the UTHSC Institutional Review Board.

The sample for the study included students of the UTHSC in the progressions of nursing, pharmacology and physician assistant studies, who were required to complete the AMA BDLS training course. The survey questionnaire and a cover letter were included in the packets for the BDLS course. Participants were asked to read the cover letter, which stated that participation was voluntary and anonymity would be ensured.

Participants were asked to complete a pretest before the training course which they subsequently turned into a collection box during the break. The training course was conducted, which included content on botulism poisoning and upon conclusion, the participants were asked to complete and return the post test. A number was included on these pre and post test questionnaires for the purpose of matching them pre course and post course, while still maintaining anonymity. All of the completed surveys were kept in a locked office until coded and entered into a Statistical Package for the Social Sciences (SPSS)[1] data file for analysis.

This survey was used to assess the current knowledge of botulism in healthcare students, and included demographic items and questions about botulism. The first questions on the survey asked the participants which profession they are studying and if they have had classes on infectious diseases. The next eleven questions addressed knowledge of botulism in regard to its diagnosis and treatment. The survey instructed participants not to guess if unsure of an answer.

Chapter 4/Results

A total of 301 pre-tests were completed and received for analysis. Demographic results showed that 108 (36%) participants were nursing students, 156 (52%) were pharmacy students and 37 (12%) were physician assistant students (Table 1). Ninety three percent of students reported they had had classes on infectious diseases during the course of their studies. Consider the statistical hypotheses for this study, which are as follows:

Ho1: There will be no statistically significant differences in the pretest scores among the student groups (α = 0.05).

Ho2: None of the student groups will achieve a mean score of 60% or greater on the pretest (α = 0.05).

Ho3: There will be no statistically significant differences among the posttest scores among the student groups.

Ho4: There will be no statistically significant difference in the pretest versus posttest score for nursing students.

Ho5: There will be no statistically significant difference in the pretest versus posttest score for pharmacy students.

Ho6: There will be no statistically significant difference in the pretest versus posttest score for physician assistant students.

After analyzing the results, the mean percentage of correct answers over all groups on the pre-test was very poor, 25.55% (sd ± 19.40%, n=301). Specific group pre-test results are as follows: nursing students scored 17.33% (sd ± 15.14%, n=108), pharmacy students 25.23% (sd 17.26%, n=156), and physician assistant students

50.86% (sd 17.60%, n=37) (Table 2). A 95% confidence interval was used to show that the lower and upper boundaries for the means are the following: overall (23.5 – 27.7), nursing students (14.4 - 20.2), pharmacy students (22.5 – 27.9), physician assistant students (44.9 – 56.7).

Although none of the professions achieved a passing mean score of 60% there were significant differences among the professions. Analysis of variance indicated there were significant differences among the professions ($F_{2,298}$ = 56.388, p < 0.001). Levene's test for equality of variance indicated variances were equal ($F_{2,298}$ = 1.938, p = ns) and a Tukey HSD was used for post hoc tests. Because the group sizes were unequal the harmonic mean was used for comparisons. Table 3 indicates the mean score for the pretest for each of the professions were significantly different from the others.

Following the BDLS course the students were retested on their knowledge of botulism. The post-test showed improvement from the pre-test in all three groups of health care students. The mean post-test score for all students was 73.3% (sd ±15.32%, n=149). There were a total of 149 post-tests returned representing 52 nursing students, 78 pharmacy students, and 19 physician assistant students. The means for percentage of questions answered correctly are as follows: 72.30% (sd ± 15.32%, n=149) overall, and among groups of students 68.88% (sd ± 16.23%, n=52) for nursing students, 73.89% (sd ± 14.08%, n=78) for pharmacy students, and 75.11% (sd ± 16.82) for physician assistant students (Table 4).

All of the professions achieved a passing mean score of 60% or greater, and there were no significant differences among the professions. Analysis of variance

indicated there were no significant differences among the professions ($F_{2,146}$ = 2.066, p

= ns). When the pre-test and post-test mean score were compared, paired t tests

revealed a significant increase in the mean score for each the three professions. For

the nursing students, the pre-test mean was 18.00 (sd ±15.53, n=52,) while the post-test

mean was increased to 68.88 (sd ±16.23, n=52) and t_{51} =16.736, p<0.001. The

pharmacy students pre-test mean was 25.28 (sd±17.06, n=78) and for the post-test the

mean raised to 73.89 (sd ±14.08, n=78) and t_{77} =22.095, p<0.001. Lastly, the physician

assistant students pre-test mean was 47.84 (sd ±19.36, n=19) and post-test mean of

75.11 (sd ±16.82, n=19) and t_{18} =6.829, p<0.001.

Chapter 5/Discussion

The results of this study plainly show that students in health care do not have adequate knowledge regarding botulism. This is evidenced by the fact that the overall mean on a pre-test with 11 questions on botulism was 25.5% correct answers. While none of the three groups of students had a mean with a passing score (60% or greater), the physician assistant students answered the most questions correctly, followed by pharmacy students and lastly, nursing. This is likely a reflection of curriculum content differences related to infectious disease among the areas the students were studying.

The post-test clearly shows the improvement of knowledge base after teaching on botulism was included in the BDLS course. The overall mean on the post-test was 71.8% correct answers, which is a passing score and shows over 45% improvement. For each group of students, the post-test mean was improved and all were passing percentages.

Furthermore, the hypotheses presented earlier in the study can now be assessed:

Ho1: There will be no statistically significant differences in the pretest scores among the student groups (α = 0.05) - *rejected*.

Ho2: None of the student groups will achieve a mean score of 60% or greater on the pretest (α = 0.05) – *fail to reject*.

Ho3: There will be no statistically significant differences among the posttest scores among the student groups – *fail to reject*.

Ho4: There will be no statistically significant difference in the pretest versus posttest score for nursing students - *rejected*.

Ho5: There will be no statistically significant difference in the pretest versus posttest score for pharmacy students - *rejected*.

Ho6: There will be no statistically significant difference in the pretest versus posttest score for physician assistant students - *rejected*.

While the study included just over 300 students, there were some limitations in the fact that the sample analyzed was taken from one Midwestern university. This may make it harder to generalize our results across the country and among healthcare students studying at other universities, but it is consistent with the lack of knowledge of botulism seen in health care professionals as cited previously.

Less than half of the students who completed the pretest completed the posttest. This decreases the number of subjects available to analyze in our posttest sample. In addition, posttest knowledge was acquired immediately after the BDLS course was completed, and thus it is uncertain how much information on botulism was retained.

This study also did not utilize a true control group. Even with these limitations, there was no other reason for such improvement in test scores than the education on botulism that participants received during the course.

Conclusion

Clostridium botulinum and the toxin it produces have been extensively studied and its effects well documented since first investigated in the 1820's. Yet it is often misdiagnosed when it occurs from natural or accidental causes, and it is questionable at what cost it would be recognized if used intentionally. Despite the facts that have been so carefully laid out about botulinum toxin and potential for misuse by terrorists, it is not well know by students in health care professions.

Like any other infectious disease or category A agent, education on botulism poisoning must be included in the curriculum of health care students. Whether it is included as part of a program didactic requirement or via a BDLS training course required by the institution, as it was for the UTHSC students, it must be taught. Those in health care professions can only respond efficiently and effectively to this disease if there is education and preparedness.

It cannot be acceptable that a toxin as potent as botulinum that discriminates against no age, race, or gender, is found naturally across the world, has been produced and released by terrorists, has no cure, and requires early detection to save the victim's life, could be so unknown to students in health care professions.

Recommendations for further research on this topic are to use a larger sample size which includes health care students from around the country and to utilize a Quasi-experimental design which would include a separate sample for the pretest and posttest.

References

Alexander, G. C., Larkin, G. L., & Wynia, M. K. (2006). Physicians' preparedness for

bioterrorism and other public health priorities. *Academic Emergency Medicine, 13*(11),

1238-41.

Arnon, S. S., Schechter, R., Inglesby, T. V., Henderson, D. A., Bartlett, J. G., Ascher, M.

S.,…& Tonat, K. (2001). Botulinum toxin as a biological weapon: Medical and public

health management. *JAMA, 285*(8), 1059-70.

Chen, F. M., Hickner, J., Fink, K. S., Galliher, J.M., & Burstin, H. (2002). On the front lines:

family physicians' preparedness for bioterrorism. *Journal of Family Practice, 51*(9), 745-

50.

Cosgrove, S. E., Perl, T. M., Song, X., & Sisson, S. D. (2005). Ability of physicians to

diagnose and manage illness due to category A bioterrorism agents. *Archives of*

Internal Medicine, 165, 2002-6.

Council of State and Territorial Epidemiologists. Botulism Surveillance Summary. (2008).

Retrieved from http://www.cdc.gov/nationalsurveillance/botulism_surveillance.html

Horowitz, B. Z. (2005). Botulinum toxin. *Critical Care Clinics, 21*(4), 825-39.

Josko, D. (2004). Botulin toxin: A weapon in terrorism. Clinical Laboratory Science, 17(1),

30-4.

Shukla, H. D., & Sharma, S. K. (2005). Clostridium botulinum: a bug with beauty and

weapon. *Critical Reviews in Microbiology, 31*(1), 11-8.

St. Louis, M. E., Peck, H. S., Bowering, D., Morgan, G. B., Blatherwick, J., Banerjee, S.,…&

Blake, P. A. (1988). Botulism from chopped garlic: Delayed recognition of a major

outbreak. *Annals of Internal Medicine, 108*(3), 363-8.

Van Rynen, J. L., Rega, P. P., Budd, C., & Burkholder-Allen, K. J. (2009). The use of negative inspiratory force by ED personnel to monitor respiratory deterioration in the event of a botulism-induced MCI. *Journal of Emergency Nursing, 35*(2), 114-7.

Villar, R. G., Elliot, S. P., Davenport, K. M., & Davenport, K. M. (2006). Botulism: the many faces of botulinum toxin and its potential for bioterrorism. *Infectious Disease Clinics of North America, 20*, 313-27.

Wein, L. M., & Liu, Y. (2005). Analyzing a bioterror attack on the food supply: The case of botulinum toxin in milk. *Proceedings of the National Academy of Sciences, 102*(28), 9984-89.

Weingart, O. G., Schreiber, T., Mascher C., Pauly, D., Dorner, M. B., Berger, T. F. H.,…& Dorner, B. G. (2010). The case of botulinum toxin in milk: Experimental data. *Applied and Environmental Microbiology, 76*(10), 3293-3300.

Tables

Table 1 Demographics

Profession studying	Number	Percentage
Nursing	108	36%
Pharmacy	156	52%
Physician Assistant	37	12%
Total	301	

Table 2 Pre-test scores by profession:

Profession studying	Number	Mean	Standard Deviation
Nursing	108	17.3394	15.14746
Pharmacy	156	25.2322	17.26625
Physician Assistant	37	50.8602	17.60014
Total	301	25.5505	19.40019

Table 3 Significant differences in the mean pre-test scores.

Tukey HSD[a,]

What profession are you studying?	N	Subset for alpha = 0.05		
		1	2	3
Nursing	108	17.3394		
Pharmacy	156		25.2322	
Physician Assistant	37			50.8602
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 70.263.

Table 4 Post-test scores by profession:

Profession studying	Number	Mean	Deviation Standard
Nursing	52	68.8811	16.23700
Pharmacy	78	73.8928	14.08045
Physician Assistant	19	75.1196	16.82899
Total	149	72.3002	15.32667

Table 5 Pre-test and post-test mean score comparisons with paired t tests:

Nursing students

	Mean	Number	Standard Deviation
Pre-test	18.0056	52	15.53720
Post-test	68.8811	52	16.23700

$t_{51} = 16.736$, $p < 0.001$

Pharmacy students

	Mean	Number	Standard Deviation
Pre-test	25.2895	78	17.06355
Post-test	73.8928	78	14.08045

$t_{77} = 22.095$, $p < 0.001$

Physician Assistant students

	Mean	Number	Standard Deviation
Pre-test	47.8474	19	19.36782
Post-test	75.1196	19	16.82899

$t_{18} = 6.829$, $p < 0.001$

Appendices

Appendix A: Cover Letter

Christopher E. Bork, Ph.D., PT, EMT-B
Paul P. Rega, MD, FACEP
Department of Public Health and Homeland Security
Health Science Campus
Glendale Medical Building
3355 Glendale Avenue
Toledo, Ohio 43614
Phone: 419-383-6301

Dear Colleague,

Botulism is a condition that occurs regularly across this nation and the world. Although botulism typically involves, at most, a handful of cases, it must be recognized as a potential weapon for terrorists. Therefore we feel it is our responsibility to prepare professionals to recognize and treat botulism. Regretfully little is known about what practicing clinicians (and students who are preparing to become clinicians) know about botulism. The purpose of this letter and included questionnaire is to enlist your help in assisting us to assess your knowledge about botulism. The results will help provide information to develop education.

The accompanying questionnaire consists of two sections: demographic information and knowledge of botulism (including signs, symptoms and treatment). The questionnaire should take less than ten minutes to complete. Please answer the questions to the best of your ability, but if you are uncertain of the correct answer, please do not guess. Participation in this study is completely voluntary, and you have the right to discontinue the survey at any time and to leave any questions blank.

All returned questionnaires will be compiled and will have no specific identifiers for you. This is to maintain your anonymity. There will be no monetary or gift compensation for your participation in this study, but your participation is greatly appreciated.

If you should have any questions, please feel free to contact, Dr. Christopher Bork at 419-383-6301 or Dr. Paul P. Rega at 419-383-6722. We can also be reached via e-mail at Christopher.Bork@utoledo.edu or Paul.Rega@utoledo.edu.

Thank you for your time and helping to further research in emergency preparedness and disaster medicine.

Sincerely,

Christopher E. Bork, Ph.D., P.T., EMT-B   Paul P. Rega, MD, FACEP
College of Medicine   College of Medicine
The University of Toledo   The University of Toledo

IRB #106390
Appendix B: Survey

Demographic Information   **IRB Approval #106390**

1. What is your professional background?
    ☐ Nurse   ☐ Physician   ☐ Physician Assistant   ☐ EMT
    ☐ Paramedic (EMT-P) ☐ Other _____
    ☐ **Student** (please skip to question 5)

2. What is your specialty/area of practice? _____

3. How many years have you worked in your profession? _____

4. Are you currently practicing your profession?   ☐ Yes ☐ No

5. **Students**: what profession are you studying?
    ☐ Medicine
    ☐ Nursing
    ☐ Pharmacy
    ☐ Physician Assistant
    ☐ Other _____

6. **Students:** Have you had classes on infectious diseases?   ☐ Yes ☐ No

The next eleven (11) items address knowledge about botulism and its diagnosis and treatment. Please do not guess if you are unsure of an answer.

7. The appropriate personal protective equipment to wear when caring for a patient with suspected botulism includes
    a. A surgical mask within three feet of the patient.
    b. An N-95 respirator within three feet of the patient.
    c. Does not require any respiratory protection.
    d. Don't know

8. Botulism, in its early stages, principally affects what system?
    a. Cardiovascular
    b. Immunological
    c. Neurological
    d. Don't know

9. Common forms of botulism include food-borne, infant, and
    a. Wound.
    b. Naso-pharyngeal
    c. Bubonic

d. Don't know

10. Baby-BIG (Botulism Immune Globulin) IV is medication reserved for babies with infant botulism up to the age of
    a. Six months.
    b. One year.
    c. Five years.
    d. Don't know

Please complete the items on the next side

11. One of the earliest signs of infant botulism is
    a. Fever
    b. Cough
    c. Constipation
    d. Don't know

12. Which poison is more toxic than botulinum toxin?
    a. Cyanide
    b. Sarin
    c. Neither
    d. Don't know

13. One of the keys to the diagnosis of botulism is
    a. A widened mediastinum on a chest x-ray.
    b. Swollen lymph nodes in the groin and axillae.
    c. Neither
    d. Don't know

13. In the Centers for Disease Control and Prevention (CDC) categories of biological weapons of mass destruction, botulism resides in
    a. Category A
    b. Category B
    c. Category C
    d. Don't know

14. If botulism antitoxin is administered within one or two days of the onset of disease, symptoms will typically resolve in
    a. Less than 48 hours.
    b. Less than one week.
    c. Neither.
    d. Don't know

15. One manifestation typically found in a patient with botulism is
    a. Ptosis
    b. Pruritus
    c. Pancytopenia

       d.   Don't know

16. A syndrome that is typically seen early with food-borne botulism is
       a.   Altered sensorium.
       b.   Nausea, vomiting and diarrhea.
       c.   Paresthesia
       d.   Don't know

Thank you for participating in this survey.

Abstract

Objective: To assess the current knowledge of botulism in health care students.

Methods: A pre-test survey on botulism was given to students in the progressions of nursing, pharmacology and physician assistant studies at the University of Toledo Health Science Campus, before they began an AMA Basic Disaster Life Support (BDLS) training course. After the course, which included information on botulism, the students completed a post-test survey.

Results: A total of 301 pre-test surveys were collected with a mean of 25% correct. Ninety three percent reported having had classes on infectious diseases. After the BDLS course with teaching on botulism, the post-test scores were significantly improved among all student groups with a mean score of 72%.

Conclusions: Students in health care have inadequate knowledge of botulism and there are statistically significant differences between groups of students based on area of study. Additionally, BDLS serves as a reasonable platform for botulism education.