

Development of botulinum toxin therapy

Alan B. Scott, MD

Smith-Kettlewell Eye Research Institute, 2318 Fillmore Street, San Francisco, CA 94115, USA

Botulism occurs mostly from eating improperly preserved food. In the eighteenth and nineteenth centuries in Bavaria, botulism was caused by sausages that were preserved with inadequate boiling, smoking, and salting. Justinus Kerner collected data on 230 cases of botulism and published two important monographs in 1820 and 1822. Kerner gave a remarkably complete and accurate description of clinical botulism: its symptoms, time course, and physical findings, especially that the tear fluid disappears, the pupil dilates, the eye muscles are paralyzed, mucus and saliva secretion is suppressed, the skin is dry, the skeletal muscles and gut are paralyzed, and until the last, cognition is preserved. Finally, Kerner suggested the potential therapeutic use of toxin to block abnormal motor movements, such as chorea, and speculated on its use in other disorders with hypersecretion, for example. However, he stopped there. Kerner was an important romantic poet and a busy medical officer; lacking a university appointment, he went off in these directions at age 37, after remarkably insightful and creative research on botulism. His work is summarized in further detail by Smith [1] and more recently by Erbguth and Naumann [2].

Seventy-five years later, van Ermengem, professor of bacteriology and a student of Koch, correctly described the bacterial basis of botulism. Of 34 individuals who had attended a funeral and ate some raw, partially salted ham, 23 were paralyzed and three died. van Ermengem found extracts of the ham to be toxic to laboratory animals, producing a paralytic disease akin to botulism. The toxicity in the animals, as well as in those who had eaten the ham, was related to the amount consumed, with only a small amount needed.

Animals vary in their susceptibility—in particular, carrion eaters, such as dogs, are resistant. From the ham and from the spleen of a man who had died, van Ermengem isolated the anaerobic bacterium, grew it, named it, characterized its culture requirements, and described its toxin.

Over the next three decades, food canning and botulism grew together. K.F. Myer, a Swiss veterinarian, developed a major focus of botulinum investigation at the Hooper Foundation in San Francisco. New strains of the organism and toxin were characterized. Techniques for reliably killing the spores in the canning process and knowledge of the correct pH (<4.0) and salt concentration to prevent organism growth and toxin production were defined. The requirements for toxin inactivation by heating were also defined. The California canning industry was saved, and knowledge of how to grow the organism and extract the toxin was developed. Type F toxin was recognized later in 1960 and type G in 1970.

Swords into plowshares

The potential for botulinum toxin as a warfare agent was a second focus of investigation from the 1930s onward. The bacteria are remarkably easy to grow in culture, with tremendous toxin yields in a 5-gallon container after just 3 or 4 days. It is easy to concentrate (although not so easy to crystallize), and quantities to paralyze whole cities or armies have been made by several nations—the United States, the United Kingdom, Russia, Iraq, and perhaps many others. But distribution for ingestion is not easy, as the toxin loses potency with time in dilute without protein-buffering solutions, as might occur in a water supply. Inhalation of the dried toxin as an aerosol is frightening to contemplate but was never developed.

E-mail address: abs@ski.org

Development of concentration and crystallization techniques at Fort Detrick by Lamanna and Duff in 1946–7, using acid precipitation techniques, became the basis of the clinical product. The breakup of the Army's Chemical Corps at Fort Detrick led to the move by Edward Schantz to the Food Research Institute in Wisconsin. There, he continued to manufacture toxin in concentrated form for experimental use and gave it out generously to the academic community.

Among these experimenters was Drachman [3], at Johns Hopkins, who used small doses of toxin to paralyze the hind limb in chicks. At that time, techniques had been developed to accurately inject extraocular muscles with local anesthetics to assess their contribution to the eye movement performance. Because strabismus surgery had high reoperative rates in many categories, other alternatives to strabismus surgical treatment were being sought systematically by injecting various anesthetics, alcohol, enzymes, enzyme blockers, snake neurotoxins, and finally, motivated by Drachman's work, botulinum toxin. The effect was remarkable. An injection of a few picograms would induce paralysis confined to the target muscle, long in duration, and with no side effects whatsoever. The results of these animal experiments on strabismus were published in 1973, and an application was sent to the Food and Drug Administration (FDA) for human use after working out techniques for freeze-drying, buffering with albumin, and assuring sterility, potency, and safety. Human experimentation began first in strabismus in 1977. By 1982, the eye muscles were injected for strabismus and nystagmus, the lid muscles for retraction, hemifacial spasm, and blepharospasm, and the limbs and neck for dystonia, all as predicted in the 1973 study. It was astonishing that none of the local neurology, orthopedic, and rehabilitation physicians at the Children's Hospital, University of California at San Francisco, Stanford, and the Shriner's Hospital would try toxin for muscle contractures with stroke, dystonia, torticollis, or cerebral palsy. This strong aversion to the idea of toxin use makes it possible to grasp why no one in the century and a half since the time of Kerner had tried this. Without local support, I had no possibility of trying out the wide spectrum of toxin uses and, therefore, enlisted others. My cases of torticollis, the first three injected, were later published by Dr. Tsui of Vancouver [4], and many others undertook to expand the use in dystonia and muscular disorders. It was only in the late 1980s that Dr. Koman of Wake Forest University would pioneer the use of toxin in pediatric treatment of leg spasm and cerebral palsy. The use gradually expanded, because there was no adequate alternative treatment for many motility disorders. With active patient groups

and Internet access, blepharospasm patients rapidly disseminated information of good results. Torticollis patients likewise came soon to know that pain was markedly decreased by toxin injection, motility increased, head position improved a little, and tremor was not changed much. And so on through the range of motor and muscle disorders such as spasmodic dysphonia, spasm in various gastroenteric and urinary sphincters, muscle spasm in stroke, and pretty well every muscle, including those producing low back pain. With data from thousands of patients and 240 investigators, the FDA approved use in adult strabismus and blepharospasm in December 1989.

The use to reduce hyperhidrosis came from the original and creative application of botulinum toxin by Drobnic and Laskawi in 1994 [5] to treat Frey's Syndrome, gustatory sweating usually occurring after parotid gland surgery with subsequent anomalous connection of nerves in that region. It was an easy transfer of this concept of blocking cholinergic innervation to sweat glands as a treatment for hyperhidrosis in the axilla, hands, and elsewhere. Remarkable in this application in Frey's Syndrome is the extraordinary duration of toxin effect, sometimes more than 1 year, much longer than the typical 3 to 4 months seen after injection of muscles. An extension of this approach is the use to diminish salivation by parotid gland injection to ameliorate the poorly handled secretions in amyotrophic lateral sclerosis and to decrease excessive lacrimal gland secretion effects, which harkens directly back to clinical findings of dryness so prominent in Kerner's patients of 1820.

Cosmetic use of botulinum toxin, probably its greatest single application, is the creation of Alistair and Jean Carruthers. For many years, a few blepharospasm patients injected at intervals of 3 or 4 months around the eyes and upper face would mention as a joke upon return that they were "back to get the wrinkles out." But only somebody working in aesthetic dermatology and ophthalmology could grasp the potential for this application of botulinum toxin. The Carruthers' thoughtful and rational application of toxin to selective agonist-antagonistic muscle groups in the face, to lift the brow, flatten folds, is probably overtaken now by less discriminate use. The idea that toxin use in young people may prevent skin wrinkle development is an intriguing prospect. It is from widespread use around the face that the beneficial effect of toxin on headache has emerged. Direct release of muscle tension by the paralytic effect at neuromuscular junctions is the principal mechanism. Intriguing theories on toxin operating by effect on proprioceptors, thus altering centrally controlled muscle tonus in migraine, are speculative.

An important historical event is the appearance of toxin type B, Myobloc. In choosing the toxin type to concentrate on in our original studies, we looked at epidemics and found that type A typically produced extensive muscle paralysis, and type B was often associated with autonomic disorders. This has been proved by the high degree of autonomic side effects of type B toxin, where use for torticollis brought about good paralytic effects, but in a sizable percentage also dryness of the mouth, difficulty in accommodation, reduced sweating, constipation, and so on [6]. If limited for muscle paralysis in higher doses because of side effects, type B toxin would seem superior for injection into lacrimal or salivary glands and in the GI tract. Type B is essential in those patients for whom type A toxin is impossible because of antibody development, although the development of resistance to type B toxin in five of 21 patients resistant to type A toxin is a high and troubling occurrence rate [7]. A drawback for cosmetic use of type B is the stinging of the current low pH solution when injected subcutaneously. This can be ameliorated by dilution with bicarbonate buffer, but then higher volume injection at each site is needed. Type C is under development in Italy, and types D, E, and G will certainly be future alternatives. Type F toxin developed in Japan and used in clinical experimental series had a shorter duration of effect than type A toxin and is not available.

Antibodies to botulinum toxin should become increasingly less of a problem. The initial lot of type A (79-11) licensed to me by the FDA and later used by Allergan to formulate Botox in the United States had a relatively low potency, and thus a higher

antigenic protein content. This resulted in antibody production in doses >200 U or even lesser doses at injection intervals of <30 days. The lot used in Europe for Botox (88-4), had a much higher potency and probably fewer antibodies developed. Both of these have now been replaced by still higher potencies for the Allergan product, Botox. In the future, we might hope for cheaper toxin as competition develops, but high regulatory costs in the United States will prevent this.

References

- [1] Smith Louis DS, Thomas Charles C. Botulism. Springfield (IL): Charles C. Thomas; 1977.
- [2] Erbguth FJ, Naumann M. Historical aspects of botulinum toxin: Justinus Kerner (1786–1862) and the “sausage poison”. *Neurology* 1999;53:1850–3.
- [3] Drachman DB. Neuropoisons. In: Simpson LL, editor. New York: Plenum Press; 1971. p. 315–39.
- [4] Tsui JK, Eisen A, Mak E, Carruthers J, Scott A, Calne DB. A pilot study on the use of botulinum toxin in spasmodic torticollis. *Can J Neurol Sci* 1985;12(4):314–6.
- [5] Drobik C, Laskawi R. Frey-Syndrom: Behandlung mit Botulinum-Toxin. *HNO Aktuell* 1994;2:142–4.
- [6] Dressler D, Benecke R. Initial experiences with clinical use of botulinum toxin type B. *Nervenarzt* 2002;73(2):194–8.
- [7] Kumar R, Seeberger L. Long-term safety, efficacy, and dosing of botulinum toxin B (Myobloc) in cervical dystonia (CD) and other movement disorders. *Mov Disord* 2002;17(Suppl 5):S292–3.