IN THE UNITED STATES DISTRICT COURT
FOR THE DISTRICT OF COLUMBIA

ABBVIE INC., *Plaintiff*, v. U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES, *et al.*, *Defendants*.	**Case No. 1:26-cv-00431 (CJN)**

DECLARATION OF MITCHELL F. BRIN, MD

I, Mitchell F. Brin, pursuant to 28 U.S.C. § 1746, declare as follows:

1. I am Senior Vice President and Chief Scientific Officer for BOTOX and Neurotoxins at AbbVie Inc. and Allergan Aesthetics (an AbbVie Company). I provide scientific, technical, and strategic leadership for AbbVie's development, commercialization, and manufacturing of biological products.

2. I am a Clinical Professor of Neurology at University of California Irvine (UCI) and serve on UCI's School of Biological Sciences Dean's Leadership Council. I am also a Fellow of the American Academy of Neurology, the American Neurological Association, and the American Headache Society. I have authored more than 550 scientific publications and continue to treat patients in clinical practice. I hold a biology degree from the University of Pennsylvania and a medical degree from Columbia College of Physicians and Surgeons.

3. Over the course of my career, I have played key roles in the research and development of BOTOX and have been involved in developing nearly every condition for which BOTOX is a licensed treatment, from inception through licensure. I am also part of the team responsible for overseeing the advancement of our pipeline of products. I have personal

knowledge of the facts stated herein through my work and my review of AbbVie's products and correspondence with regulatory agencies. I submit this declaration in support of AbbVie's Motion for Summary Judgment.

4. AbbVie works to discover and manufacture innovative medicines that solve the most pressing medical needs of today and tomorrow. AbbVie's products have therapeutic uses across numerous fields, including immunology, oncology, neuroscience, and eyecare. AbbVie annually invests billions of dollars into the research and development of new medicines, and its products treat 16 million Americans annually.

5. AbbVie manufactures BOTOX®, a biological product that has twelve FDA therapeutic indications across eight chronic and debilitating diseases spanning adult and pediatric populations, including chronic migraine, spasticity, overactive bladder, cervical dystonia, and neurogenic detrusor overactivity. Today, BOTOX is the only FDA-licensed biological product providing localized treatment of chronic migraine, bladder disorders, involuntary eye dysfunctions, and hyperhidrosis, and AbbVie continues to research BOTOX's potential efficacy as to other severe and difficult-to-treat medical conditions.

6. BOTOX's current FDA-licensed therapeutic indications and their licensure dates are as follows:

- **Blepharospasm** (1989): a condition involving involuntary, repeated blinking or spasm of eyelid muscles, resulting in uncontrollable blinking or eyelid closure;
- **Strabismus** (1989): a condition involving eye misalignment due to poor control of eye muscles resulting in double vision, reduced depth perception, eye strain, and potential vision loss;
- **Cervical dystonia** (2000): a painful neurological condition due to involuntary neck muscle contractions, causing the head to twist, turn, or tilt uncontrollably in various directions;
- **Hyperhidrosis** (2004): a skin disorder resulting in excessive pathologic sweating due to overactive sweat glands;

- **Spasticity** (2010 for adult upper limb; 2016 for adult lower limb; 2019 for pediatric upper and lower limb): a debilitating neurological condition causing muscle stiffness and impaired movement of limbs that can interfere with movement and function, often seen after stroke, spinal cord injury, multiple sclerosis, traumatic brain injury, or cerebral palsy;
- **Chronic migraine** (2010): headaches on 15 or more days per month, often severely impacting daily life due to pain and other symptoms;
- **Neurogenic detrusor overactivity** (2011 for adults; 2021 for children): uncontrolled contraction of bladder muscles causing frequent, urgent urination or leakage, resulting from nerve damage due to neurological conditions in adults and children; and
- **Overactive bladder** (2013): a condition involving the urge to urinate caused by uncontrolled contraction of bladder muscles without underlying neurological cause.

7. While administering BOTOX to treat overactive facial muscles, researchers noticed that BOTOX reduces wrinkles near injection sites. FDA has since licensed BOTOX (under the brand name BOTOX Cosmetic®) for four cosmetic indications.

8. I was involved in and have reviewed the regulatory history of AbbVie's licensed biological product BOTOX and am familiar with AbbVie's operations more generally.

9. BOTOX is licensed and marketed under Biologics License Application (BLA) 103000. AbbVie is the holder of that BLA. Attached hereto as **Exhibit 1** is a true and correct copy of the U.S. Food & Drug Administration License Revocation and Product Transfer Letter dated November 15, 2023, which provides on page 1 that AbbVie is "authorized to introduce or deliver for introduction into interstate commerce" BOTOX.

10. In 1989, FDA licensed the product known today as BOTOX under the name Oculinum for the treatment of strabismus and blepharospasm, two eye disorders caused by muscle dysfunction. Attached hereto as **Exhibit 2** is a true and correct copy of Alan B. Scott et al., *Treatment of strabismus and blepharospasm with Botox (onabotulinumtoxinA): Development, insights, and impact*, 102 Medicine S23 (2023), which describes the development of Oculinum as a treatment for strabismus and blepharospasm on pages S24 through S26.

11. In 1991, Allergan Inc. acquired Oculinum, Inc., and thereby also acquired the product Oculinum, which Allergan renamed BOTOX a year later. AbbVie acquired Allergan in 2020.

12. FDA has only licensed BOTOX in its finished dosage forms (50 Unit, 100 Unit, and 200 Unit single-dose vials), which have always consisted of three ingredients: onabotulinumtoxinA (onabotA), human serum albumin (HSA), and sodium chloride. Attached hereto as **Exhibit 3** is a true and correct copy of the 1989 United States Prescribing Information for Oculinum (now known as BOTOX), which notes on page 1 that "[e]ach vial of Oculinum® contains 100 units (U) of *Clostridium botulinum* toxin type A, 0.5 milligrams of albumin (human), and 0.9 milligrams of sodium chloride in a sterile, lyophilized form without a preservative." To this day, the prescribing information for BOTOX states that the product "contains albumin, a derivative of human blood." Attached hereto as **Exhibit 4** is a true and correct copy of the 2023 United States Prescribing Information for BOTOX, which confirms the presence of HSA in section 5.14. AbbVie's ability to market BOTOX under its BLA depends on the presence of all three ingredients, and each of these ingredients plays an important role.

13. OnabotA is a natural, biological protein toxin produced by the growth of the bacterium *Clostridium botulinum*. After growth of the bacteria under controlled conditions, the toxin is purified to yield the onabotA drug substance, a complex consisting of several naturally occurring proteins. Attached hereto as **Exhibit 5** is a true and correct copy of Mitchell F. Brin et al., *Update on Non-Interchangeability of Botulinum Neurotoxin Products*, 16(6) Toxins (Basel) 266 (June 2024), which describes, on page 6, properties of botulinum neurotoxins. Since onabotA is an extremely potent protein, it must be dosed in minute quantities. For example, the mass of

onabotA contained in a 100 U BOTOX vial (~5 nanograms) is only millionths the mass of a single grain of sand.

14. The HSA contained in BOTOX is a protein obtained from human blood plasma. That plasma is collected from qualified human donors, who are tested for infectious agents. The plasma is then fractionated as part of a months-long process to isolate therapeutic proteins like HSA. HSA constitutes the vast majority of the protein content in BOTOX. Each BOTOX vial contains approximately 100,000 times more HSA by mass than onabotA.

15. Sodium chloride produces an isotonic solution with the same osmotic balance as fluids in the human body to protect cells from osmotic damage and minimize patient discomfort at the site of injection.

16. Since it was first licensed, BOTOX has always been formulated with HSA to protect onabotA from environmental factors affecting the quality, stability, and performance of BOTOX. Given the minute quantities of onabotA in each BOTOX vial, any loss of biological activity or toxin protein could have disproportionate impacts, as even a small absolute loss may represent a substantial percentage of the total dose. HSA plays an essential role in preventing the breakdown of onabotA into other compounds (*i.e.*, oxidation), minimizing protein "clumping" (*i.e.*, protein aggregation) that could reduce efficacy or trigger immune responses, and stopping onabotA from sticking to surfaces or syringes (*i.e.*, adsorption).

17. Since BOTOX was first licensed, researchers have found that HSA plays a more important role in influencing BOTOX's therapeutic effect than initially understood. By 2000, several peer-reviewed studies reported that the concentration of HSA affects toxin activity and enables the effective administration of lower amounts of toxin to achieve the same therapeutic effect. Attached hereto as **Exhibit 6** is a true and correct copy of Bahram Mohammadi et al.,

Experience with long-term treatment of albumin-supplemented botulinum toxin type A, 116 J. Neural Transm. 437 (2009), which cites on page 440 two studies published in 2000 and 2001 that evaluated reducing toxin dose by increasing HSA concentration in patients with cervical dystonia, blepharospasm, and hemifacial spasm. Generally, lowering the effective dose of a product can reduce the risk of side effects and adverse immune responses. By 2009, researchers demonstrated that increasing the concentration of HSA allowed for the reduction of a toxin dose by more than half while maintaining the same level of efficacy during long-term use over 5-10 years. *Id.* (observing that the average dose of 184 units with albumin-supplemented treatment compared to other long-term observations of the same botulinum toxin product with doses ranging from 500 to 1,072 units).

18. As AbbVie recounted in a scientific addendum to an October 16, 2025 letter submitted to CMS, recent *in vitro* and *in vivo* studies conducted by AbbVie have further identified the mechanism by which HSA enhances the pharmaceutical effects of onabotA and thus influences BOTOX's efficacy. Attached as **Exhibit 7** is a true and correct copy of AbbVie's October 16, 2025 letter to CMS, which incorporates the scientific addendum on pages 12 through 16. I have reviewed this document and am familiar with its contents, including the scientific addendum. I led the team that designed, executed, and evaluated the studies that were summarized in the scientific addendum.

19. [redacted]

[redacted]

20. On December 5, 2025, AbbVie submitted to CMS a declaration about the role of HSA from Dr. Andrew Pickett, an independent consultant and scientist with 40 years of experience working with biological products and 85 peer-reviewed publications. Attached hereto as **Exhibit 8** is a true and correct copy of the declaration of Dr. Pickett. I have reviewed the document and am familiar with its contents. Dr. Pickett's declaration summarizes the literature reporting the safety and long-term effectiveness of a lower dose of toxin administered with a higher concentration of HSA. Ex. 8 § 2. Based on these reported observations and AbbVie's recent studies, Dr. Pickett agrees that HSA plays an integral role in BOTOX by influencing key therapeutic parameters of onabotA. *Id.* at p. 6.

21. On December 5, 2025, AbbVie also submitted to CMS a declaration from Dr. Martin Gastens, the Vice President of Parenteral Product Development, Science & Technology at AbbVie, describing how BOTOX supply is heavily reliant on qualified HSA supply. Attached hereto as **Exhibit 9** is a true and correct copy of the declaration of Dr. Gastens. As discussed therein, AbbVie maintains two qualified HSA suppliers under its BLA and invests significant time and resources to qualifying, validating, and periodically re-validating suppliers and incoming materials for blood and blood components. *See* Ex. 9 ¶¶ 10-13.

22. [redacted]

23. [redacted]

24. Even modest changes to how HSA is introduced into the BOTOX manufacturing process require extensive development, validation, and FDA review because of the sensitivity of onabotA to its manufacturing environment. *See* Ex. 9 ¶ 7; *see also* 21 U.S.C. § 356a; 21 C.F.R.

§ 601.12 (setting forth FDA regulations governing changes to licensed biological products). [redacted]

[redacted] In August 2025, for example, FDA approved a prior-approval supplemental BLA [redacted]

I declare under penalty of perjury that the foregoing is true and correct.

Dated: April 26, 2026

Mitchell F. Brin, MD

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ABBVIE INC., *Plaintiff*, v. U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES, *et al.*, *Defendants*.	**Case No. 1:26-cv-00431 (CJN)**

INDEX OF EXHIBITS TO DECLARATION OF MITCHELL F. BRIN IN SUPPORT OF PLAINTIFF'S MOTION FOR SUMMARY JUDGMENT

Exhibit 1	Letter from Dr. Sarah Yim, Director Office of Therapeutic Biologics and Biosimilars, FDA, to Dr. Claire Whitley, AbbVie Inc., License Revocation and Product Transfer (Nov. 15, 2023).
Exhibit 2	Alan B. Scott et al., *Treatment of strabismus and blepharospasm with Botox (onabotulinumtoxinA): Development, insights, and impact*, 102 Medicine S23 (2023).
Exhibit 3	United States Prescribing Information, Oculinum® (1989).
Exhibit 4	United States Prescribing Information, BOTOX® (2023).
Exhibit 5	Mitchell F. Brin et al., *Update on Non-Interchangeability of Botulinum Neurotoxin Products*, 16(6) Toxins (Basel) 266 (Jun 2024).
Exhibit 6	Bahram Mohammadi et al., *Experience with long-term treatment of albumin-supplemented botulinum toxin type A*, 116 J. Neural Transm. 437 (2009).
Exhibit 7 (REDACTED)	Letter from Johanna Corbin, Senior Vice President, Chief Patent and Innovation Counsel, AbbVie Inc., to Mike Stuart, General Counsel, Department of Health and Human Services, and to Beth Kelley, Acting Deputy

	General Counsel, Department of Health and Human Services, Inflation Reduction Act: Exclusion of BOTOX® (onabotulinumtoxinA), A Human Plasma-Derived Biological Product, from Medicare Drug Price Negotiation Program. (Oct. 16, 2025).
Exhibit 8 (REDACTED)	Letter from Dr. Andrew Pickett, Founder, Toxin Science Limited, to Office of the General Counsel, Department of Health and Human Services, The Role of Human Serum Albumin on the Biological Activity of OnabotulinumtoxinA (Dec. 5, 2025).
Exhibit 9 (REDACTED)	Declaration of Dr. Martin Gastens, Vice President of Parenteral Product Development, AbbVie Inc., to Office of the General Counsel, Department of Health and Human Services (Dec. 5, 2025).

EXHIBIT 1 to Brin Declaration

U.S. FOOD & DRUG ADMINISTRATION

BLA 103000

LICENSE REVOCATION AND PRODUCT TRANSFER

AbbVie Inc.
Attention: Claire Whitley, BSc (Hons) Int.
1 North Waukegan Road
North Chicago, IL 60064

Dear Dr. Whitley:

Please refer to your biologics license application (BLA) under section 351(a) of the Public Health Service Act for Botox and Botox Cosmetic (onabotulinumtoxinA) for injection.

We have received your letters dated February 2, 2023, and March 3, 2023, that your company, formerly identified as Allergan, Inc., U.S. License No. 1145, has been acquired by AbbVie Inc. It is our understanding that AbbVie Inc. will continue to manufacture Botox and Botox Cosmetic (onabotulinumtoxinA) in the same manner as Allergan, Inc., using the same equipment, manufacturing procedures and methods, and responsible personnel.

It is our understanding that you will remain the authorized official for AbbVie Inc.. Final draft labeling submitted on October 11, 2023, and other information required for the change in licensure have been reviewed and found to be in compliance with the required standards.

Therefore, under the provisions of 21 CFR 601.5(a), the U.S. License No. 1145 under Allergan, Inc. is hereby revoked, effective this date.

In accordance with the provisions of section 351(a) of the Public Health Service Act, you are hereby authorized to introduce or deliver for introduction into interstate commerce, Botox and Botox Cosmetic (onabotulinumtoxinA) under your existing U.S. License No. 1889 effective this date.

As soon as possible, but no later than 14 days from the date of this letter, submit content of labeling [21 CFR 601.14(b) in structured product labeling (SPL) format via the FDA automated drug registration and listing system (eLIST), as described at FDA.gov.[1] Information on submitting SPL files using eLIST may be found in the guidance for industry *SPL Standard for Content of Labeling Technical Qs and As*.[2]

[1] http://www.fda.gov/ForIndustry/DataStandards/StructuredProductLabeling/default.htm
[2] We update guidances periodically. For the most recent version of a guidance, check the FDA Guidance Documents Database https://www.fda.gov/RegulatoryInformation/Guidances/default.htm.

The SPL will be accessible via publicly available labeling repositories.

Submit final printed carton and container labels with U.S. License No. 1889 no more than 30 days after they are printed. Submit these labels electronically according to the guidance for industry *Providing Regulatory Submissions in Electronic Format — Certain Human Pharmaceutical Product Applications and Related Submissions Using the eCTD Specifications*. For administrative purposes, designate this submission "**Final Printed Carton and Container Labels for approved BLA 103000**." Approval of this submission by FDA is not required before the labeling is used.

Please note that this letter supersedes any previously issued license certificates (see October 20, 1999, FR Doc No: 99-27159). You may place these certificates in your historical files. However, we recommend that you keep a copy of this letter available for review at the time of FDA inspections.

Cite the BLA number listed above at the top of the first page of all submissions to this application.

If you have any questions, please contact Susan Daugherty, Regulatory Project Manager, at (301) 796-0878.

Sincerely,

{See appended electronic signature page}

Sarah Yim, MD
Director
Office of Therapeutic Biologics and Biosimilars
Office of New Drugs
Center for Drug Evaluation and Research

This is a representation of an electronic record that was signed electronically. Following this are manifestations of any and all electronic signatures for this electronic record.

/s/

SARAH K YIM
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EXHIBIT 2 to Brin Declaration

Supplement Article

Medicine

OPEN

Treatment of strabismus and blepharospasm with Botox (onabotulinumtoxinA)

Development, insights, and impact

Alan B. Scott, MD[a], Stanley Fahn, MD[b], Mitchell F. Brin, MD[c,d,*]

Abstract

Strabismus, deviation of the ocular alignment, can adversely affect quality of life and activities of daily living. Surgery was the prior standard of care for strabismus, but up to 40% of patients required additional surgeries. This need for more effective and less invasive treatment, along with the convergence of other events such as the development of electromyography, purification of botulinum toxin A, and the finding that injection of botulinum toxin type A could paralyze the hind limbs of chicks, led Dr. Alan Scott to investigate injection of his formulation for strabismus. The positive results of initial trials in monkeys segued to human trials with observations of alignment improvements and few adverse events. The success of botulinum toxin type A in the treatment of strabismus led to interest in its use to treat other skeletal muscles, particularly in blepharospasm, a type of focal dystonia involving eyelid spasms and involuntary eye closure that lacked an effective pharmacological treatment. Patient groups helped to increase awareness of this novel treatment, and results from clinical trials confirmed its effectiveness. Dr. Scott's formulation, then known as Oculinum, received its first Food and Drug Administration approvals in 1989 for strabismus and blepharospasm. Allergan acquired Oculinum in 1991, renaming it Botox. These initial uses led to its application in a myriad of other indications as outlined in other articles of this supplement.

Abbreviations: AEs = adverse events, EMG = electromyography, FDA = Food and Drug Administration, IRB = institutional review board, QOL = quality of life.

Keywords: botulinum toxin, neuromuscular agents

1. Overview of strabismus and blepharospasm

Strabismus is a deviation of ocular alignment in which the primary lines of sight deviate by at least 1 prism diopter.[1] It can be further categorized by direction of the deviation: inwards (esotropia), outwards (exotropia), upwards (hypertropia), downwards (hypotropia), and rotary (cyclotropia).[2] Prevalence is estimated to be 1.1–5% in the general population (from studies spanning 1992–2018) and is >40% in patients with cerebral palsy or Down syndrome.[1,3,4] Strabismus can range in severity, and can be corrected in some patients with lenses, prisms, and/or vision therapy.[1] Strabismus can be congenital or due to a brain motor control problem or an injury.[2] It usually develops in childhood but can develop at any age.[1] Ocular disturbances associated with strabismus include blurred vision, diplopia (double vision), and impaired depth perception.[2] Strabismus can also affect activities of daily living, quality of life (QOL), and work productivity.[1]

Blepharospasm is a form of focal dystonia characterized by spasms of the orbicularis oculi muscles of the eyelid, involuntary eye closures, and enhanced blinking, sometimes to a severity that renders a patient functionally blind.[5,6] Crude estimates of the prevalence of blepharospasm range from 16–133 per million.[7] Onset generally occurs in the fifth or sixth decade of life, and older age and female sex may be risk factors for blepharospasm development.[7] Though the etiology of primary blepharospasm is unclear, patients with blepharospasm had a higher frequency of prior eye diseases such blepharitis and keratoconjunctivitis than those without blepharospasm.[6,8] About one-quarter of patients with blepharospasm have at least one family member with dystonia.[6] Secondary blepharospasm, which is less common than idiopathic or primary blepharospasm, can develop after focal lesions in the brain or can develop in patients with parkinsonism, tardive dyskinesia, or conditions associated with lid weakness.

As in patients with other forms of dystonia, those with blepharospasm often employ a geste antagoniste, or sensory trick, that temporarily reduces dystonia. In one study, the frequency of gestes antagonistes in patients with blepharospasm was over 70%, which were most commonly stretching or rubbing the

This manuscript was funded by AbbVie. AbbVie was involved in the manuscript concept and participated in writing, reviewing, and approval of the final version. No honoraria or payments were made for authorship.

AB Scott and S Fahn have nothing to disclose. MF Brin is a full-time employee of AbbVie and holds stock in the company.

[a] Strabismus Research Foundation, San Francisco, CA, USA, [b] Columbia University, New York, NY, USA, [c] Allergan/AbbVie, Irvine, CA, USA, [d] University of California, Irvine, CA, USA.

** Correspondence: Mitchell F. Brin, Senior Vice President, Chief Scientific Officer, Botox & Neurotoxins, Allergan, an AbbVie Company, 2525 Dupont Drive; T2-3, Irvine, CA 92623-9534, USA (e-mail: mitchell.brin@abbvie.com).*

How to cite this article: Scott AB, Fahn S, Brin MF. Treatment of strabismus and blepharospasm with Botox (onabotulinumtoxinA): Development, insights, and impact. Medicine 2022;102:S1(e32374).

Received: 31 August 2022 / Accepted: 1 December 2022

http://dx.doi.org/10.1097/MD.0000000000032374

eyebrows or eyelids.[9] Blepharospasm can also have adverse effects on patient QOL.[5] Depression is more frequent in patients with blepharospasm compared with healthy controls. Many patients find difficulties in activities of daily living due to their condition, including social activities and work, and begin to withdraw from them.[10] Photophobia is common in blepharospasm,[11] and patients may have problems with driving, particularly night driving, and watching cinema, because the lights of the oncoming car and from the movie screen worsen the blepharospasm.

The following historical narrative was compiled based on review of the literature and interviews with the authors, and the quoted portions reflect the personal observations and reflections of the individuals who were interviewed. In some instances, this article describes uses for which Allergan has not sought and/or received regulatory approval in individual countries and are mentioned for historical context or background only.

2. Unmet need for the treatment of strabismus and blepharospasm

Prior to the use of botulinum toxin for strabismus, the standard of care was surgical treatment. However, many cases were not successfully treated.

*Dr. Scott (*Fig. 1*), who was the director of the Smith-Kettlewell Eye Research Institute, where he performed the initial injections of botulinum toxin in animals and humans: In some categories [of strabismus] as many as 40% of patients needed re-operations, so we were looking for something new and different.*

There was no effective pharmacological treatment for blepharospasm prior to botulinum toxin.[12,13] Medications to decrease muscle contraction were mainly ineffective and some conferred intolerable side effects.[6] Benztropine mesylate, clonazepam, pimozide, baclofen, lorazepam, amantadine hydrochloride, tridihexethyl hydrochloride, and trihexyphenidyl hydrochloride were used without much success.[14–16] In one study, two-thirds of patients with blepharospasm experienced improvement with clonazepam, but only 11% had a marked and lasting effect.[17]

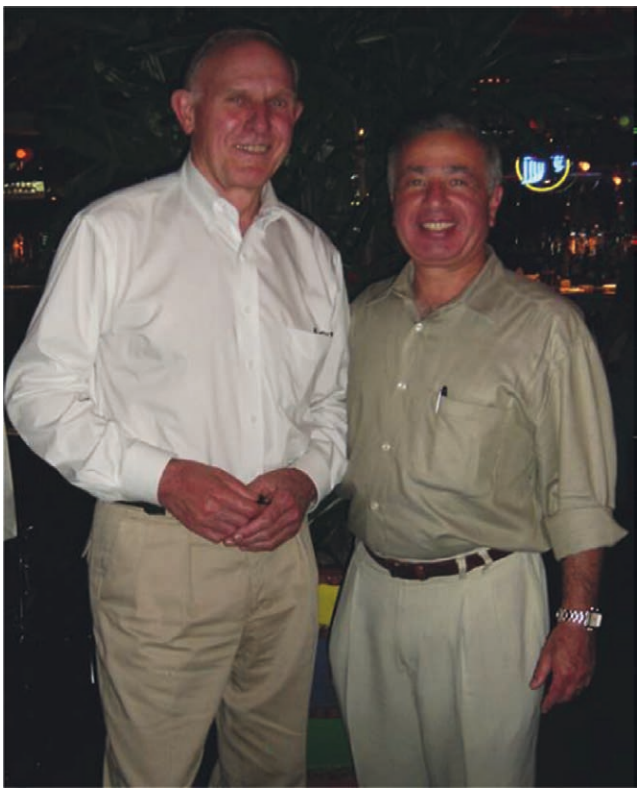

Figure 1 . Drs. Alan Scott and Joseph Jankovic. Photo provided by Dr. Joseph Jankovic.

Surgery to remove branches of the cranial nerve VII or part of the eyelid or brow muscles could result in long-term improvements in blepharospasm.[13,18] However, as with many other types of surgeries, general anesthesia, hospitalization, and weeks of convalescence are required.[14] In addition, scarring and other complications could occur.[13,14] The surgical treatment often left eye weakness, and as a consequence, sometimes people could not close their eyes adequately.

*Dr. Brin, who was a fellow and faculty member at Columbia University when he first used botulinum toxin A to treat blepharospasm under the mentorship of Dr. Fahn: In mild cases [of blepharospasm] we also tried "lid lifters." These were made by fashioning wire on eyeglasses, which would press against the eyelids to help keep them open [*Fig. 2*]. The mechanism of lid lifters is unknown, but it may be related to sensory feedback, such as occurs when blepharospasm patients reduce muscle spasm by touching their eyelids—a sensory trick.*[19] *The physical effect of lifting the lids may also be involved. But, in general, there was no good treatment for blepharospasm before botulinum toxin.*

3. Early use of botulinum toxin A as a treatment for strabismus and blepharospasm and its development program

Historically, the German physician Justinus Kerner gave the first detailed clinical description of "sausage poisoning," now known as botulism, including weakness, dry mouth, and reduced body secretions.[20] Kerner was also the first to suggest a therapeutic use of botulinum toxin, particularly for conditions involving hyperexcitability of the nervous system such as hypersecretion of sweat or mucus and motor hyperkinesias.

As referenced in the first chapter of this supplement, a combination of ideas and events converged for the modern-day use of botulinum toxin A for therapeutic purposes.

Dr. Scott: There were 2 or 3 things that came together in the early 1970s. Another researcher, Dr. Carter Collins, and I had been working on muscle physiology and one of the techniques which we developed was electromyography to find the muscle. We developed the needle electrodes that allowed us to inject and localize in the muscle itself with substantial accuracy. The second thing that was going on at that time was that the outcomes from strabismus surgery were far from perfect. And then another thing happened about that time: Lance Simpson wrote a book on neuropoisons.[21] *We'd been trying various things, injecting alcohol and various enzymes in the muscle to try to influence it but none of these had had a great effect. Simpson's book had a chapter wherein Dr. Daniel Drachman described his experiments injecting the hind limb of chicks and showed that selectively with a small enough amount of botulinum toxin, you could interfere with the muscle.*[22] *Clearly, it seemed possible then that we could use very small amounts of toxin. And so we did what Drachman did; he got his toxin from Ed Schantz. I wrote to Ed and he sent us some toxin. [Dr. Edward Schantz at the University of Wisconsin developed a bulk purification method for botulinum toxin A.*[23]*] He supplied the scientific world with toxin, or the few people who were interested in it at the time, and sent it out freely and we started to use it in monkeys. First in some other animals, but then we rapidly moved to monkeys because we could very easily see the effects of tiny doses and work up until we found an appropriate dose. The miracle moment for me was injecting it in the monkey. I could immediately see that we really had something quite special,*

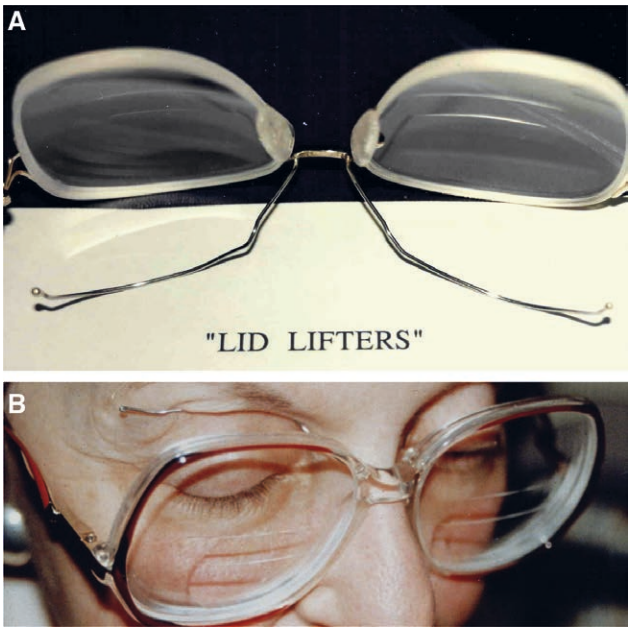

Figure 2. Lid lifters designed for blepharospasm patients. (A) Photograph showing wire lifters fashioned on eyeglasses by the patient's husband, who was a dentist. (B) Blepharospasm patient wearing lid lifters. Photos provided by Dr. Mitchell Brin.

quite specific, not toxic to anything else, long-lasting. From that experience, you could tell that this was going to be very, very valuable.[24]

In the first published article on botulinum toxin A for injection, Dr. Scott and colleagues used electromyography (EMG) for guidance to inject botulinum toxin A into either the medial or lateral ocular rectus muscle in rhesus monkeys and observed a dose-dependent transient weakness from 2 weeks to at least 8 months without serious local or systemic adverse events (AEs).[25] The success of these experiments led to its testing in humans.

Dr. Scott, on the very first patient he injected with botulinum toxin, a man with strabismus: The first injection patient was a man who had a retinal detachment operation that had scarred his muscles and pulled his eye out of line. He did not have good functional visual return from his retinal detachment. Our chance of creating a serious problem for him was pretty low. He wasn't really seeing double because his vision was poor. Though I'd done many injections behind the eye, here we were doing something new and different, always an exciting experience in clinical research.

A case series of botulinum toxin A injections using EMG guidance to correct strabismus documented that 19 patients had been treated with 67 doses, with no systemic effects.[26] A maximum correction of 40 diopters and duration of effect of at least 6 months were observed. Example photos before and after botulinum toxin A treatment are shown in Fig. 3, with resultant decreases in deviation in Fig. 4.

The success of botulinum toxin A in the treatment of strabismus led to its use in other disorders such as blepharospasm and those involving skeletal muscles, as predicted in the early paper on results of its use in animals.[25] Patient support and education groups also began forming, which assisted in communicating new therapies to patients.

Dr. Scott: As an ophthalmologist-clinician looking at the problem of blepharospasm, it appeared to be an extraordinarily rare disorder described in books but for which you almost never saw a case. There really aren't a lot of cases around—there's a few thousand—but many of those people were off the streets. They were confined, they couldn't get out. They'd been told by a number of doctors that there was no effective treatment really for it and we didn't see the patients. So, when I first started to see blepharospasm patients, it was new to me. It only took studying 2 to 3 patients to develop a reasonable protocol for how we should inject: multiple injections around the eye in the affected muscles. By that time, 1982, Mattie Lou [Koster], had made herself known. She was featured in a Wall Street Journal *article, and she developed a patient help foundation [the Benign Essential Blepharospasm Research Foundation]. The foundation provided a place for patients to find out information, to get moral support for their disorder, and receive guidance on treatment. Once Oculinum or the idea of Oculinum (later called Botox), got out there, it spread like wildfire through that community and those patients knew quite a lot more than most of the doctors very, very quickly and came asking for it.*

Dr. Fahn, who is the H. Houston Merritt Professor of Neurology and Director Emeritus of the Center for Parkinson's Disease and Other Movement Disorders at Columbia University, had been studying focal and generalized dystonia at Columbia University Medical Center, evaluating medications to alleviate the excessive muscle contractions. After he became aware of the *Wall Street Journal* article, he contacted Mattie Lou Koster to learn more about her blepharospasm foundation. In their subsequent correspondence, he encouraged her to utilize the foundation to raise money for blepharospasm research, and she informed him of the early results of botulinum toxin A injections by Dr. Scott. Dr. Fahn flew to Iowa City, Iowa, to meet Dr. Scott, who was invited by Dr. Richard Anderson at the University of Iowa to demonstrate how to inject botulinum toxin into the orbicularis oculi muscles to treat blepharospasm. When Dr. Fahn returned to New York, he prepared a protocol for a clinical trial to study botulinum toxin injections for the treatment of blepharospasm and other forms of focal dystonia. However, a hurdle at Columbia University was obtaining institutional review board (IRB) approval of the research protocol.

Dr. Fahn: One of the stumbling blocks was [that] the IRB thought it was too dangerous. Basically, the review board members insisted that the study investigators be inoculated with botulinum toxoid to protect ourselves in case we stuck ourselves with a needle and got botulinum toxin into us. It seemed a little strange to me, but I went and talked to our group… and we agreed that this was too much for us to accept. We felt that if we eventually got blepharospasm or some other focal dystonia and that botulinum toxin would be a good treatment for it, we might be immune to it and would not get the clinical benefit from this approach. So, it ended up that we agreed not to be inoculated with botulinum toxoid, but we offered to sign a waiver to Columbia University and the medical center that we would not hold them liable if we accidentally got botulism symptoms. And that was satisfactory to the IRB, giving us the green light to go ahead with our study.

Once work could begin in this area at Columbia University, open-label trials were begun. With that success, the first double-blind trial testing botulinum toxin A for blepharospasm was conducted.[27] Eight subjects were injected; 5 were injected unilaterally, with the other eye receiving saline as a control. Despite the double-blind protocol, the 5 patients and the investigators were able to detect the eye that received botulinum toxin owing to the less forceful contractions and shorter times of eyelid closure. Evoked motor potentials, contractions at rest, and maximally forced contractions were all significantly reduced.

An additional Columbia study tested injections of botulinum toxin type A in patients with a variety of focal dystonias, including 49 with blepharospasm.[28] Motor symptom relief was observed in 69% of blepharospasm patients, with a mean of 2.7 days to onset and an 11.4 week duration of effect. Ptosis (19%), conjunctivitis (7%), and entropion (2%) were reported. Similarly, in another open-label study, 39 patients with blepharospasm were injected with a mean of 21.3U botulinum toxin

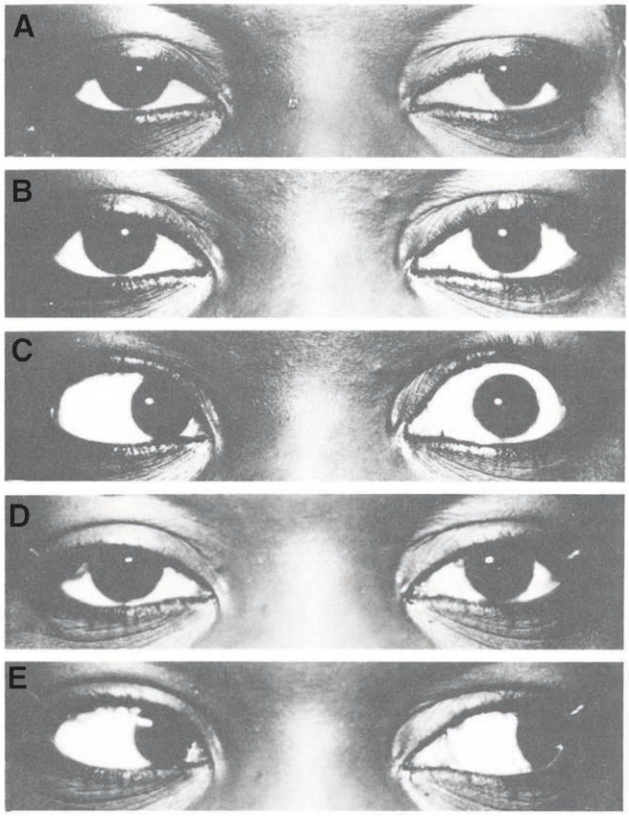

Figure 3 . Before and after photos of botulinum toxin A treatment for strabismus. (A) Prior to injection with botulinum toxin A. (B) Primary position gaze 2 days after injection with 1.56 × 10-3 µg botulinum toxin A. (C) Left gaze 2 days after injection, with absence of abduction due to lateral rectus paralysis. (D) Primary position gaze at 3 months after injection, with reduction of exotropia. (E) Left gaze 3 months after injection, with full return of abduction. Reprinted from Ophthalmology, Vol 87, Scott AB. Botulinum toxin injection into extraocular muscles as an alternative to strabismus surgery. Pages 1044–9, Copyright 1980, with permission from Elsevier.

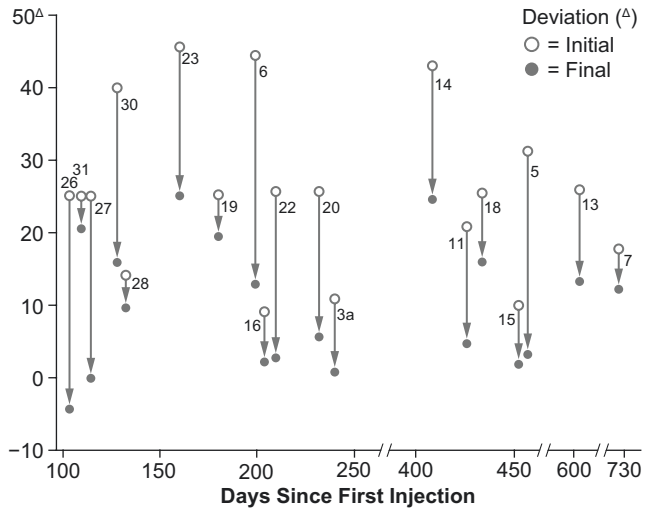

Figure 4. Decreases in deviation of ocular alignment by patients following injection of botulinum toxin A for the treatment of strabismus. Data are included for patients (numbered) who received at least the therapeutic dose threshold of 3.12 x 10-4 µg botulinum toxin A, did not receive eyelid injections, and were followed for at least 100 days after injection. Patients with paralytic strabismus or neuromyotonia were excluded. Reprinted with permission of the American Ophthalmological Society from Scott AB. Botulinum toxin injection of eye muscles to correct strabismus. Trans Am Ophthalmol Soc. 1981;79:734–70.

A per eye over 124 treatment visits with a mean between-treatment interval of 9.9 weeks.[29] Eyelid closure force decreased following treatment (Fig. 5). In the patients who had not had previous surgery, an 8.5% rate of AEs was seen, with ptosis the most common AE.

Early research supply was disrupted in 1985, when the insurance carried by Smith-Kettlewell for the work on botulinum toxin A was canceled. As a result of this loss of insurance, Dr. Scott established a company, Oculinum, so that work could continue.

Dr. Scott: I was personally not very worried about being sued, whereas the directors of the lab had this concern for the laboratory facility more generally. If I were to be sued, and the laboratory had no insurance, the magnitude of impact on the laboratory was unknown, and that might take the institution down. So, they had a responsibility to say, "We can't be doing this without insurance." This is when I established Oculinum Inc. That was a kind of a construct word. "Oculo": eyeball and "linum": line-them-up with the Latinate ending "-um," which was euphonic. So "Oculinum."

In both strabismus and blepharospasm, injection of botulinum toxin into the muscles around the affected eye blocks presynaptic release of acetylcholine at the neuromuscular junction and reduces muscle contraction. In the case of blepharospasm, injections of botulinum toxin into the eyelid and brow muscles block neural signals to the orbicularis oculi and associated periocular muscles, thus reducing eye spasm. For strabismus, injections into the extraocular muscle inhibit contractions in the deviated eye, allowing the opposing muscles of the same eye to assume a greater movement force and reposition the eye into a straighter alignment.[2]

4. Efficacy and safety highlights

The first indications for which any botulinum toxin (at the time, Oculinum) received Food and Drug Administration (FDA) approval were for the treatment of strabismus and blepharospasm associated with dystonia, both for patients aged ≥12 years, on December 29, 1989.[30]

The package insert consolidated much of the early research performed for both strabismus and blepharospasm. The dataset supporting the strabismus approval included Dr. Scott's multicenter open-label trial in which patients (N=677) received one or more injections of Oculinum.[30] Improvements in alignment of ≤10 prism diopters were seen in 55% of patients at evaluations ≥6 months following treatment. A subsequent study that reported health-related QOL assessments showed improved reading function following treatment with onabotulinumtoxinA.[31]

AEs from the treatment with botulinum toxin A for strabismus were local to the injection area. Those affecting extraocular muscles occurred at a rate of 17% in a population of 2058 adults who received 3650 injections. Ptosis varied by the location of the injected muscles, with incidences of 38% following injections into the superior rectus, 16% in the horizontal rectus, and 1% in the inferior rectus. Retrobulbar hemorrhage occurred at a rate of 0.3% in a series of 5587 injections.[30]

Oculinum was the first botulinum toxin approved for blepharospasm, and botulinum toxin type A is now considered to be the first-line treatment for this condition.[32] Due to the dramatic results from the open-label trials, few randomized, controlled trials have been undertaken for blepharospasm.[12,27,32] In one of the trials submitted to the FDA for approval, 27 patients with persistent moderate to severe blepharospasm, of which 26 had not responded to previous drug treatments, were treated with 2U of Oculinum in 6 sites per side (total of 12U per eye).[14] Of these, 25 patients reported an improvement in eyelid force and eyelid spasm within 48 hours, with a mean duration of effect of 8.1 weeks (range: 2–17 weeks). Ptosis was the most frequent AE, occurring in 22.2% (6/27) patients; all cases resolved. No systemic effects related to treatment were identified.

In a small (N=11), double-blind, placebo-controlled study, patients with blepharospasm received 6.25U injected

subcutaneously, medially, and laterally (25U per eye).[33] All patients significantly improved after treatment with Oculinum. Of 68 injections (performed in controlled and open-label phases), AEs observed in the Oculinum-treated patients included blurred vision (n=6), tearing (n=5), ecchymosis (n=3), ptosis (n=2), and diplopia (n=1).

Dr. Scott's large multicenter (N=1684), open-label trial conducted with multiple investigators showed that patients with blepharospasm experienced clinical improvements in eyelid force and lid spasm intensity that lasted for a mean of 12 weeks before needing retreatment.[30] As noted in the package insert, there have been no reports of definitive serious AEs associated with distant spread of onabotulinumtoxinA at the recommended dose for blepharospasm (≤30U).[30] The most frequent AEs with blepharospasm are ptosis (21%), superficial punctate keratitis (6%), and eye dryness (6%).[30] As physicians have refined the injection techniques over time, ptosis rates have decreased, with a 2007 review reporting a mean ptosis rate of 13%[34] and several studies reporting significantly lower ptosis rates for blepharospasm and hemifacial spasm following injection into the pretarsal rather than preseptal orbicularis oculi (eg, 13% pretarsal vs 16% preseptal[35] and 0% pretarsal vs 7.5% preseptal).[36] Importantly, unit doses are not interchangeable among different botulinum toxin products, each of which has its own dosing guidelines and clinical profile.[30,37]

Dr. Brin: With our first patient, the remarkable thing is when she came for a follow-up at Columbia University Medical Center in upper Manhattan, she mentioned that after one of her visits to us, she walked all the way down to the bottom of Manhattan, to Bowery and Canal Street. When I asked her why, she said, "Because I now could."

Subsequent studies captured QOL improvements relative to baseline. These were observed with repeat treatments of onabotulinumtoxinA, particularly in patients who were previously naïve to treatment.[38] A retrospective analysis found that 92% (33/36) and 90% (18/20) of patients with blepharospasm experienced substantial benefit at 2 and 5 years of treatment, respectively, with AEs occurring in 10.1% of patients over 398 treatment cycles, with ptosis and dry eye the most common AEs.[39] Another retrospective analysis of 73 patients with blepharospasm with at least 10 years of follow-up after a mean of 8.7 onabotulinumtoxinA treatments found a mean duration of effect of 18.2 weeks and increases in doses after the third or fourth treatments.[40] Ptosis, ecchymosis, and diplopia occurred in 19.2%, 8.2%, and 5.4% of patients, respectively. Region-specific information regarding safety and efficacy can be found in local labeling.

5. Impact of onabotulinumtoxinA on patients and the broader biomedical community

The development of botulinum toxin A (Oculinum) for therapeutic use has had a profound effect on patients with a variety of conditions as well as on the biomedical community. In 1991, Allergan purchased the manufacturing and licensing rights to Oculinum and changed the name to Botox (non-proprietary name: onabotulinumtoxinA). Since that time, it has received FDA approval for multiple additional indications.

Dr. Brin: Patients rallied around the treatment for which they saw hope and they felt that there was a light at the end of the tunnel in terms of making progress in their disease. Encouraged by their patients, movement disorder clinicians explored this treatment off-label, and professional societies moved quickly to develop guidelines. Stanley van der Noort was the chair of the American Academy of Neurology's Therapeutics and Technology Assessment (TTA) subcommittee. He asked me to convene [a] team of experts to assess botulinum toxin therapy, and the TTA subcommittee issued the report on the clinical use of botulinum toxin type A in neurology in 1990.[41] *In addition, Mark Hallett at the National Institutes of Health chaired the Consensus Development Conference in November 1990, followed by a published report in 1991.*[42] *The TTA subcommittee published the AAN Training Guidelines in 1994.*[43]

Dr. Scott: After the word got out [about Botox], there was a rapid expansion in use because a large number of these conditions didn't have any adequate treatment at this time. We mentioned in the very first paper that we wrote in 1973[25] *that we expected its use would go beyond just the ocular and extraocular muscles and indeed, that's where it went.*

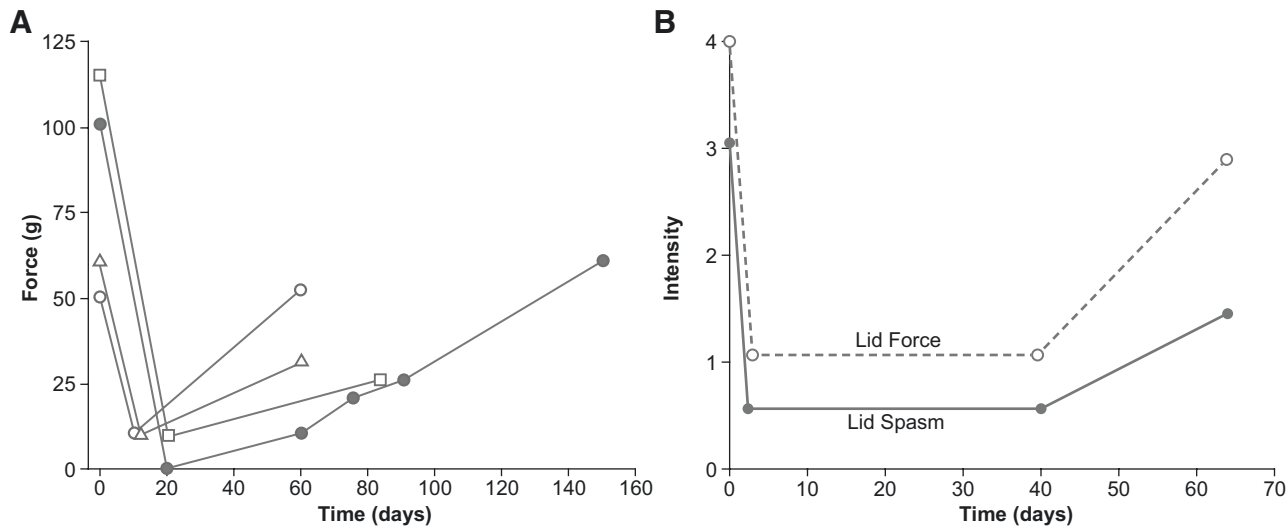

Figure 5 . Eyelid closure force before and after botulinum toxin A treatment for blepharospasm (A) measured with a modified calibrated spring-loaded speculum in four patients, and (B) mean eyelid closure force and eyelid spasm scores from 27 patients, each measured on scales ranging from 0 to 4. For force, 0 indicated minimal effort for eyelid separation and 4 an inability to separate the eyelids with the thumbs. For spasm, 0 indicated no spasm, with 4 indicating severe, incapacitating spasm. Panel A: Reproduced with permission from Arch Ophthalmol. 1985;103(3):347–50. Copyright (1985) American Medical Association. All rights reserved. Panel B: Figure 2 from "Treatment of blepharospasm with medication, surgery and type A botulinum toxin" by Arthurs B, Flanders M, Codere F, et al published in the Canadian Journal of Ophthalmology. 1987;22(1):24–8 is used under a CC BY-ND 4.0 license.

Dr. Fahn: It's rather shocking that a potent food poison can actually be something good. It's interesting that if you take the dose down, it's a helpful agent for the right condition. It's been remarkable to see how well this compound has done in helping so many people, and it's not just our specialty of movement disorders. So much of the rest of medicine has been helped. I was always amazed when I heard it was helpful in migraine, for example, so you're always learning something new.

In addition, training physicians who went on to bring the new therapy to their home countries had an especially important impact on the medical community.

Dr. Fahn: We were getting more and more patients referred to us, and eventually I stopped doing the injections myself because everybody else in our team wanted to do them, and it was very gratifying to see so many patients getting better and everybody learning how to do it. I remember some of the foreign fellows going back to their home countries and being the first ones to do botulinum toxin injections. For example, Andrzej Friedman had injected with us, and he went back and started injecting in Poland. We trained Nir Giladi from Israel who went back and started injecting patients there; he was the first in Israel to use botulinum toxin. Nearly all my trainees, from both the US and other countries, learned about botulinum toxin treatments. Training people and spreading the word was very, very important to us.

Physicians who trained with Dr. Fahn within the first 10 years of testing experimental botulinum toxin: US: Susan Bressman, Mitchell Brin, Robert Burke, Arif Dalvi, Rolando Diaz-Olivo (Puerto Rico), Enrico Fazzini, Blair Ford, Paul E. Greene, Ann Hunt, Un Jung Kang, Elan Louis, Giselle Petzinger, Seth Pullman, John Rogers, Miran Salgado, Rachel Saunders-Pullman, Heidi Shale, Marie-Helene St. Hilaire, Daniel Togasaki, Richard Trosch, Daniel Truong; Outside the US: Sylvain Chouinard (Montreal, Canada), Oren Cohen (Tel Aviv, Israel), Andrej Friedman (Warsaw, Poland), Nir Giladi (Tel Aviv, Israel), Zygmunt Jamrozic (Warsaw, Poland), Beom Jeon (Seoul, South Korea), Vladimir Kostic (Belgrade, Serbia), Timothy Lynch (Dublin, Ireland), Uday Muthane (Bangalore, India), Gianni Pezzoli (Milan, Italy), Samer Tabbal (Beirut, Lebanon), Dominic Thyagarajan (Melbourne, Australia).

Acknowledgments

Writing and editorial assistance was provided to the authors by Jennifer L. Giel, PhD, on behalf of Evidence Scientific Solutions, Inc, and was funded by Allergan, an AbbVie company. All authors met the ICMJE authorship criteria. Neither honoraria nor other form of payments were made for authorship.

Author contributions

Conceptualization: Mitchell F. Brin.
Funding acquisition: Mitchell F. Brin.
Supervision: Mitchell F. Brin.
Writing – review & editing: Alan B. Scott, Stanley Fahn, Mitchell F. Brin.

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EXHIBIT 3 to Brin Declaration

Oculinum® (Botulinum Toxin Type A)

Manufacturer: Oculinum, Inc.
Distributor: Allergan Pharmaceuticals
A Division of Allergan, Inc.

Description: Oculinum® (Botulinum Toxin Type A) is a sterile, lyophilized form of purified botulinum toxin type A, produced from a culture of the Hall strain of *Clostridium botulinum* grown in a medium containing N-Z amine and yeast extract. It is purified from the culture solution by a series of acid precipitations to a crystalline complex consisting of the active high molecular weight toxin protein and an associated hemagglutinin protein. The crystalline complex is re-dissolved in a solution containing saline and albumin and sterile filtered (0.2 microns) prior to lyophilization. **Oculinum®** is to be reconstituted with sterile non-preserved saline prior to intramuscular injection.

Each vial of **Oculinum®** contains 100 units (U) of *Clostridium botulinum* toxin type A, 0.5 milligrams of albumin (human), and 0.9 milligrams of sodium chloride in a sterile, lyophilized form without a preservative. One unit (U) corresponds to the calculated median lethal intraperitoneal dose (LD/50) in mice of the reconstituted **Oculinum®** injected.

Clinical Pharmacology: Oculinum® blocks neuromuscular conduction by binding to receptor sites on motor nerve terminals, entering the nerve terminals, and inhibiting the release of acetylcholine. When injected intramuscularly at therapeutic doses, **Oculinum®** produces a localized chemical denervation muscle paralysis. When the muscle is chemically denervated, it atrophies and may develop extrajunctional acetylcholine receptors. There is evidence that the nerve can sprout and reinnervate the muscle, with the weakness thus being reversible.

The paralytic effect on muscles injected with **Oculinum®** is useful in reducing the excessive, abnormal contractions associated with blepharospasm. When used for the treatment of strabismus, it is postulated that the administration of **Oculinum®** affects muscle pairs by inducing an atrophic lengthening of the injected muscle and a corresponding shortening of the muscle's antagonist. Following peri-ocular injection of **Oculinum®**, distant muscles show electrophysiologic changes but no clinical weakness or other clinical change for a period of several weeks or months, parallel to the duration of local clinical paralysis.[1]

In one study, botulinum toxin was evaluated in 27 patients with essential blepharospasm. Twenty-six of the patients had previously undergone drug treatment utilizing benztropine mesylate, clonazepam and/or baclofen without adequate clinical results. Three of these patients then underwent muscle stripping surgery still without an adequate outcome. One patient of the 27 was previously untreated. Upon using botulinum toxin, 25 of the 27 patients reported improvement within 48 hours. One of the other patients was later controlled with a higher dosage. The remaining patient reported only mild improvement but remained functionally impaired.[2]

In another study, twelve patients with blepharospasm were evaluated in a double-blind, placebo-controlled study. All patients receiving botulinum toxin (n = 8) were improved compared with no improvements in the placebo group (n = 4). The mean dystonia score improved by 72%, the self-assessment score rating improved by 61%, and a videotape evaluation rating improved by 39%. The effects of the treatment lasted a mean of 12.5 weeks.[3]

One thousand six hundred eighty-four patients with blepharospasm evaluated in an open trial showed clinical improvement lasting an average of 12.5 weeks prior to the need for re-treatment.[4]

Six hundred seventy-seven patients with strabismus treated with one or more injections of **Oculinum®** were evaluated in an open trial. Fifty-five percent of these patients were improved to an alignment of 10 prism diopters or less when evaluated 6 months or more following injection.[5] These results are consistent with results from additional open label trials which were conducted for this indication.[4]

Indications and Usage: Oculinum® is indicated for the treatment of strabismus and blepharospasm associated with dystonia, including benign essential blepharospasm or VII nerve disorders in patients 12 years of age and above.

The efficacy of **Oculinum®** in deviations over 50 prism diopters, in restrictive strabismus, in Duane's syndrome with lateral rectus weakness, and in secondary strabismus caused by prior surgical over-recession of the antagonist is doubtful, or multiple injections over time may be required. **Oculinum®** is ineffective in chronic paralytic strabismus except to reduce antagonist contracture in conjunction with surgical repair.

Presence of antibodies to botulinum toxin type A may reduce the effectiveness of **Oculinum®** therapy. In clinical studies, reduction in effectiveness due to antibody production has occurred in one patient with blepharospasm receiving 3 doses of **Oculinum®** over a 6 week period totalling 92 U, and in several patients with torticollis who received multiple doses experimentally, totalling over 300 U in a one month period. For this reason, the dose of **Oculinum®** for strabismus and blepharospasm should be kept as low as possible, in any case below 200 U in a one month period.

Contraindications: Oculinum® is contraindicated in individuals with known hypersensitivity to any ingredient in the formulation.

Warnings: The recommended dosages and frequencies of administration for **Oculinum®** should not be exceeded. There have not been any reported instances of systemic toxicity resulting from accidental injection or oral ingestion of **Oculinum®**. Should accidental injection or oral ingestion occur, the person should be medically supervised for several days on an office or outpatient basis for signs or symptoms of systemic weakness or muscle paralysis. The entire contents of a vial is below the estimated dose for systemic toxicity in humans weighing 6 kg. or greater.

In the event of overdosage or injection into the wrong muscle, additional information may be obtained by contacting Allergan Pharmaceuticals at (800) 347-5063 from 8:00 a.m. to 4:00 p.m. Pacific Time, or at (714) 724-5954 for a recorded message at other times.

The effect of botulinum toxin may be potentiated by aminoglycoside antibiotics or any other drugs that interfere with neuromuscular transmission. Caution should be exercised when **Oculinum®** is used in patients taking any of these drugs.[6]

Precautions: *General:* The safe and effective use of **Oculinum®** depends upon proper storage of the product, selection of the correct dose, and proper reconstitution and administration techniques. Physicians administering **Oculinum®** must understand the relevant neuromuscular and orbital anatomy and any alterations to the anatomy due to prior surgical procedures, and standard electromyographic techniques.

As with all biologic products, epinephrine and other precautions as necessary should be available should an anaphylactic reaction occur.

During the administration of **Oculinum®** for the treatment of strabismus, retrobulbar hemorrhages sufficient to compromise retinal circulation have occurred from needle penetrations into the orbit. It is recommended that appropriate instruments to decompress the orbit be accessible. Ocular (globe) penetrations by needles have also occurred. An ophthalmoscope to diagnose this condition should be available.

Reduced blinking from Oculinum injection of the orbicularis muscle can lead to corneal exposure, persistent epithelial defect and corneal ulceration, especially in patients with VII nerve disorders. One case of corneal perforation in an aphakic eye requiring corneal grafting has occurred because of this effect. Careful testing of corneal sensation in eyes previously operated upon, avoidance of injection into the lower lid area to avoid ectropion, and vigorous treatment of any epithelial defect should be employed. This may require protective drops, ointment, therapeutic soft contact lenses, or closure of the eye by patching or other means.

Information for Patients: Patients with blepharospasm may have been extremely sedentary for a long time. Sedentary patients should be cautioned to resume activity slowly and carefully following the administration of **Oculinum®**.

Drug Interactions: The effect of botulinum toxin may be potentiated by aminoglycoside antibiotics or any other drugs that interfere with neuromuscular transmission. Caution should be exercised when **Oculinum®** is used in patients taking any of these drugs.[6] (See Warnings).

Pregnancy: ***Pregnancy Category C:*** Animal reproduction studies have not been conducted with **Oculinum®**. It is also not known whether **Oculinum®** can cause fetal harm when administered to a pregnant woman or can affect reproduction capacity. **Oculinum®** should be administered to pregnant women only if clearly needed.

Carcinogenesis, Mutagenesis, Impairment of Fertility: Long term studies in animals have not been performed to evaluate carcinogenic potential of **Oculinum®**.

Nursing Mothers: It is not known whether this drug is excreted in human milk. Because many drugs are excreted in human milk, caution should be exercised when **Oculinum®** is administered to a nursing woman.

Pediatric Use: Safety and effectiveness in children below the age of 12 have not been established.

Adverse Reactions:[4] There have been reports of seven cases of diffuse skin rash and two cases of local swelling of the eyelid skin lasting for several days following eyelid injection.

Strabismus: Inducing paralysis in one or more extraocular muscles may produce spatial disorientation, double vision, or past-pointing. Covering the affected eye may alleviate these symptoms. Extraocular muscles adjacent to the injection site are often affected, causing ptosis or vertical deviation, especially with higher doses of **Oculinum®**. The incidence rates of these side effects in 2058 adults who received 3650 injections for horizontal strabismus are listed below:

Ptosis	15.7%
Vertical deviation	16.9%

The incidence of ptosis was much less after inferior rectus injection (0.9%) and much greater after superior rectus injection (37.7%).

The incidence rates of these side effects persisting for over 6 months in an enlarged series of 5587 injections of horizontal muscles in 3104 patients are listed below:

Ptosis lasting over 180 days	0.3%
Vertical deviation greater than 2 prism diopters lasting over 180 days	2.1%

In these patients, the injection procedure itself caused 9 scleral perforations. A vitreous hemorrhage occurred and later cleared in one case. No retinal detachment or visual loss occurred in any case. Sixteen retrobulbar hemorrhages occurred. Decompression of the orbit after 5 minutes was done to restore retinal circulation in one case. No eye lost vision from retrobulbar hemorrhage. Five eyes had pupillary change consistent with ciliary ganglion damage (Adies pupil).

Blepharospasm: In 1684 patients who received 4258 treatments (involving multiple injections) for blepharospasm, the incidence rates of adverse reactions per treated eye are listed below:

Ptosis	11.0%
Irritation/Tearing (includes dry eye, lagophthalmos, and photophobia)	10.0%

Ectropion, keratitis, diplopia and entropion were reported rarely (incidence less than 1%)

Ecchymosis occurs easily in the soft eyelid tissues. This can be prevented by applying pressure at the injection site immediately after the injection.

In two cases of VII nerve disorder (one case of an aphakic eye) reduced blinking from **Oculinum®** injection of the orbicularis muscle led to serious corneal exposure, persistent epithelial defect and corneal ulceration. Perforation requiring corneal grafting occurred in one case, an aphakic eye. Avoidance of injection into the lower lid area to avoid ectropion may reduce this hazard. Vigorous treatment of any corneal epithelial defect should be employed. This may require protective drops, ointment, therapeutic soft contact lenses, or closure of the eye by patching or other means.

Two patients previously incapacitated by blepharospasm experienced cardiac collapse attributed to over-exertion within three weeks following **Oculinum®** therapy. Sedentary patients should be cautioned to resume activity slowly and carefully following the administration of **Oculinum®**.

Overdosage: In the event of overdosage or injection into the wrong muscle, additional information may be obtained by contacting Allergan Pharmaceuticals at (800) 347-5063 from 8:00 a.m. to 4:00 p.m. Pacific Time, or at (714) 724-5954 for a recorded message at other times.

Dosage and Administration: *Strabismus:* **Oculinum®** is intended for injection into extraocular muscles utilizing the electrical activity recorded from the tip of the injection needle as a guide to placement within the target muscle. Injection without surgical exposure or electromyographic guidance should not be attempted. Physicians should be familiar with electromyographic technique.

An injection of **Oculinum®** is prepared by drawing into a sterile 1.0 mL tuberculin syringe an amount of the properly diluted toxin (see Dilution Table) slightly greater than the intended dose. Air bubbles in the syringe barrel are expelled and the syringe is attached to the electromyographic injection needle, preferably a 1½", 27 gauge needle. Injection volume in excess of the intended dose is expelled through the needle into an appropriate waste container to assure patency of the needle and to confirm that there is no syringe-needle leakage. A new, sterile needle and syringe should be used to enter the vial on each occasion for dilution or removal of **Oculinum®**.

To prepare the eye for Oculinum injection, it is recommended that several drops of a local anesthetic and an ocular decongestant be given several minutes prior to injection.

Note: The volume of **Oculinum®** injected for treatment of strabismus should be between 0.05 mL to 0.15 mL per muscle.

Strabismus dosage: The initial listed doses of the diluted **Oculinum®** (see Dilution Table below) typically create paralysis of injected muscles beginning one to two days after injection and increasing in intensity during the first week. The paralysis lasts for 2-6 weeks and gradually resolves over a similar time period. Overcorrections lasting over 6 months have been rare. About one half of patients will require subsequent doses because of inadequate paralytic response of the muscle to the initial dose, or because of mechanical factors such as large deviations or restrictions, or because of the lack of binocular motor fusion to stabilize the alignment.

I. Initial doses in units (abbreviated as U). Use the lower listed doses for treatment of small deviations. Use the larger doses only for large deviations.
   A. For vertical muscles, and for horizontal strabismus of less than 20 prism diopters: 1.25 U to 2.5 U in any one muscle.
   B. For horizontal strabismus of 20 prism diopters to 50 prism diopters: 2.5 U to 5.0 U in any one muscle.
   C. For persistent VI nerve palsy of one month or longer duration: 1.25 U to 2.5 U in the medial rectus muscle.

II. Subsequent doses for residual or recurrent strabismus.
   A. It is recommended that patients be re-examined 7-14 days after each injection to assess the effect of that dose.
   B. Patients experiencing adequate paralysis of the target muscle that require subsequent injections should receive a dose comparable to the initial dose.
   C. Subsequent doses for patients experiencing incomplete paralysis of the target muscle may be increased up to twice the size of the previously administered dose.
   D. Subsequent injections should not be administered until the effects of the previous dose have dissipated as evidenced by substantial function in the injected and adjacent muscles.
   E. The maximum recommended dose as a single injection for any one muscle is 25 U.

Blepharospasm: For blepharospasm, diluted **Oculinum®** (see Dilution Table) is injected using a sterile, 27-30 gauge needle without electromyographic guidance. 1.25 U to 2.5 U (0.05 mL to 0.1 mL volume at each site) injected into the medial and lateral pre-tarsal orbicularis oculi of the upper lid and into the lateral pre-tarsal orbicularis oculi of the lower lid is the initial recommended dose. In general, the initial effect of the injections is seen within three days and reaches a peak at one to two weeks post-treatment. Each treatment lasts approximately three months, following which the procedure can be repeated indefinitely. At repeat treatment sessions, the dose may be increased up to two-fold if the response from the initial treatment is considered insufficient—usually defined as an effect that does not last longer than two months. However there appears to be little benefit obtainable from injecting more than 5.0 Units per site. Some tolerance may be found when Oculinum is used in treating blepharospasm if treatments are given any more frequently than every three months, and it is rare to have the effect be permanent.

The cumulative dose of **Oculinum®** in a 30-day period should not exceed 200 U.

Dilution Technique: To reconstitute lyophilized **Oculinum®**, use sterile normal saline **without** a preservative; 0.9% Sodium Chloride Injection is the recommended diluent. Draw up the proper amount of diluent in the appropriate size syringe. Since **Oculinum®** is denatured by bubbling or similar violent agitation, inject the diluent into the vial gently. Discard the vial if a vacuum does not pull the diluent into the vial. Record the date and time of reconstitution on the space on the label. **Oculinum®** should be administered within 4 hours after reconstitution.

During this time period, reconstituted **Oculinum®** should be stored in a refrigerator (2° to 8°C). Reconstituted **Oculinum®** should be clear, colorless and free of particulate matter. Parenteral drug products should be inspected visually for particulate matter and discoloration prior to administration and whenever the solution and the container permit. The use of one vial for more than one patient is not recommended because the product and diluent do not contain a preservative.

Dilution Table

Diluent Added (0.9% Sodium Chloride Injection)	Resulting dose in Units per 0.1 mL
1.0 mL	10.0 U
2.0 mL	5.0 U
4.0 mL	2.5 U
8.0 mL	1.25 U

Note: These dilutions are calculated for an injection volume of 0.1 mL. A decrease or increase in the **Oculinum®** dose is also possible by administering a smaller or larger injection volume — from 0.05 mL (50% decrease in dose) to 0.15 mL (50% increase in dose).

How supplied: Each vial contains 100 U of lyophilized *Clostridium botulinum* Toxin type A, NDC 0023-0504-01.

Caution: Federal law prohibits dispensing without a prescription.

Storage: Store the lyophilized product in a freezer at or below -5°C. Administer **Oculinum®** within 4 hours after the vial is removed from the freezer and reconstituted. During these four hours, reconstituted **Oculinum®** should be stored in a refrigerator (2° to 8°C). Reconstituted **Oculinum®** should be clear, colorless and free of particulate matter.

All vials, including expired vials, or equipment used with the drug should be disposed of carefully as is done with all medical waste.

December 1989

Manufactured by: Oculinum, Inc., Berkeley, CA 94710 Distributed by: Allergan Pharmaceuticals, A Division of Allergan, Inc., Irvine, CA 927

1. Sanders D, Massey W, Buckley E. Botulinum toxin for blepharospasm: Single-fiber EMG studies. Neurology 1986;36:545-547.
2. Arthurs B, Flanders M, Codere F, Gauthier S, Dresner S, Stone L. Treatment of blepharospasm with medication, surgery and type A botulinum toxin. Can J Ophthalmol 1987;22:24-28.
3. Jankovic J, Orman J. Botulinum A toxin for cranial-cervical dystonia: A double-blind, placebo-controlled study. Neurology 1987;37: 616-623.
4. Data on file, Oculinum, Inc.
5. Scott A B. Botulinum toxin treatment of strabismus. American Academy of Ophthalmology, Focal Points 1989: Clinical Modules for Ophthalmologists Vol VII Module 12.
6. Wang Y C, Burr D H, Korthals G J, Sugiyama H. Acute toxicity of aminoglycoside antibiotics as an aid in detecting botulism. Appl Environ Microbiol 1984; 48:951-955.

EXHIBIT 4 to Brin Declaration

HIGHLIGHTS OF PRESCRIBING INFORMATION
These highlights do not include all the information needed to use BOTOX® safely and effectively. See full prescribing information for BOTOX.

BOTOX® (onabotulinumtoxinA) for injection, for intramuscular, intradetrusor, or intradermal use
Initial U.S. Approval: 1989

WARNING: DISTANT SPREAD OF TOXIN EFFECT
See full prescribing information for complete boxed warning.
The effects of BOTOX and all botulinum toxin products may spread from the area of injection to produce symptoms consistent with botulinum toxin effects. These symptoms have been reported hours to weeks after injection. Swallowing and breathing difficulties can be life threatening and there have been reports of death. The risk of symptoms is probably greatest in children treated for spasticity but symptoms can also occur in adults, particularly in those patients who have an underlying condition that would predispose them to these symptoms. (5.1)

INDICATIONS AND USAGE

BOTOX is an acetylcholine release inhibitor and a neuromuscular blocking agent indicated for:

- Treatment of overactive bladder (OAB) with symptoms of urge urinary incontinence, urgency, and frequency, in adults who have an inadequate response to or are intolerant of an anticholinergic medication (1.1)
- Treatment of urinary incontinence due to detrusor overactivity associated with a neurologic condition [e.g., spinal cord injury (SCI), multiple sclerosis (MS)] in adults who have an inadequate response to or are intolerant of an anticholinergic medication (1.1)
- Treatment of neurogenic detrusor overactivity (NDO) in pediatric patients 5 years of age and older who have an inadequate response to or are intolerant of anticholinergic medication. (1.2)
- Prophylaxis of headaches in adult patients with chronic migraine (≥15 days per month with headache lasting 4 hours a day or longer) (1.3)
- Treatment of spasticity in patients 2 years of age and older (1.4)
- Treatment of cervical dystonia in adult patients, to reduce the severity of abnormal head position and neck pain (1.5)
- Treatment of severe axillary hyperhidrosis that is inadequately managed by topical agents in adult patients (1.6)
- Treatment of blepharospasm associated with dystonia in patients 12 years of age and older (1.7)
- Treatment of strabismus in patients 12 years of age and older (1.7)

Limitations of Use

Safety and effectiveness of BOTOX have not been established for:

- Prophylaxis of episodic migraine (14 headache days or fewer per month) (1.3)
- Treatment of hyperhidrosis in body areas other than axillary (1.6)

DOSAGE AND ADMINISTRATION

- Follow indication-specific dosage and administration recommendations. In a 3 month interval, do not exceed a total dose of:
  - Adults: 400 Units
  - Pediatrics: the lesser of 10 Units/kg or 340 Units (2.1)
- See Preparation and Dilution Technique for instructions on BOTOX reconstitution, storage, and preparation before injection (2.2)
- Overactive Bladder: Recommended total dose 100 Units, as 0.5 mL (5 Units) injections across 20 sites into the detrusor (2.3)
- Adult Detrusor Overactivity associated with a Neurologic Condition: Recommended total dose 200 Units, as 1 mL (~6.7 Units) injections across 30 sites into the detrusor (2.3)
- Pediatric Detrusor Overactivity associated with a Neurologic Condition: 0.5 mL injections across 20 sites into the detrusor (2.4)
  - Greater than or equal to 34 kg: Recommended total dose is 200 Units
  - Less than 34 kg: Recommended total dose is 6 Units/kg
- Chronic Migraine: Recommended total dose 155 Units, as 0.1 mL (5 Units) injections per each site divided across 7 head/neck muscles (2.5)
- Adult Upper Limb Spasticity: Recommended total dose up to 400 Units divided among affected muscles (2.6)
- Adult Lower Limb Spasticity: Recommended total dose 300 Units to 400 Units divided across ankle and toe muscles (2.6)
- Pediatric Upper Limb Spasticity: Recommended total dose 3 Units/kg to 6 Units/kg (maximum 200 Units) divided among affected muscles (2.7)
- Pediatric Lower Limb Spasticity: Recommended total dose 4 Units/kg to 8 Units/kg (maximum 300 Units) divided among affected muscles (2.7)
- Cervical Dystonia: Base dosing on the patient's head and neck position, localization of pain, muscle hypertrophy, patient response, and adverse event history; use lower initial dose in botulinum toxin naïve patients (2.8)
- Axillary Hyperhidrosis: 50 Units per axilla (2.9)
- Blepharospasm: 1.25 Units-2.5 Units into each of 3 sites per affected eye (2.10)
- Strabismus: The dose is based on prism diopter correction or previous response to treatment with BOTOX (2.11)

DOSAGE FORMS AND STRENGTHS

For Injection: 100 Units or 200 Units vacuum-dried powder in a single-dose vial (3)

CONTRAINDICATIONS

- Hypersensitivity to any botulinum toxin preparation or to any of the components in the formulation (4, 5.4, 6)
- Infection at the proposed injection site (4)
- Intradetrusor Injections: Urinary tract infection or urinary retention (4)

WARNINGS AND PRECAUTIONS

- Spread of toxin effects; swallowing and breathing difficulties can lead to death. Seek immediate medical attention if respiratory, speech or swallowing difficulties occur (5.1, 5.6)
- Potency Units of BOTOX are not interchangeable with other preparations of botulinum toxin products (5.2, 11)
- Potential serious adverse reactions after BOTOX injections for unapproved uses (5.3)
- Concomitant neuromuscular disorder may exacerbate clinical effects of treatment (5.5)
- Use with caution in patients with compromised respiratory function (5.6, 5.7, 5.10)
- Corneal exposure and ulceration due to reduced blinking may occur with BOTOX treatment of blepharospasm (5.8)
- Retrobulbar hemorrhages and compromised retinal circulation may occur with BOTOX treatment of strabismus (5.9)
- Bronchitis and upper respiratory tract infections in patients treated for spasticity (5.10)
- Urinary tract infections in patients treated for OAB (5.12)
- Urinary retention: Post-void residual urine volume should be monitored in patients treated for OAB or adult detrusor overactivity associated with a neurologic condition who do not catheterize routinely, particularly patients with multiple sclerosis or diabetes mellitus. (5.13)

ADVERSE REACTIONS

The most common adverse reactions (≥5% and >placebo, if applicable) are (6.1):

- OAB: urinary tract infection, dysuria, urinary retention
- Adult Detrusor Overactivity associated with a neurologic condition: urinary tract infection, urinary retention
- Pediatric Detrusor Overactivity associated with a neurologic condition: urinary tract infection, leukocyturia, bacteriuria
- Chronic Migraine: neck pain, headache
- Adult Spasticity: pain in extremity
- Pediatric Spasticity: upper respiratory tract infection
- Cervical Dystonia: dysphagia, upper respiratory infection, neck pain, headache, increased cough, flu syndrome, back pain, rhinitis
- Axillary Hyperhidrosis: injection site pain and hemorrhage, non-axillary sweating, pharyngitis, flu syndrome

To report SUSPECTED ADVERSE REACTIONS, contact AbbVie at 1-800-678-1605 or FDA at 1-800-FDA-1088 or www.fda.gov/medwatch.

DRUG INTERACTIONS

Patients receiving concomitant treatment of BOTOX and aminoglycosides or other agents interfering with neuromuscular transmission (e.g., curare-like agents), or muscle relaxants, should be observed closely because the effect of BOTOX may be potentiated (7.1, 7.4)

USE IN SPECIFIC POPULATIONS

- Pregnancy: Based on animal data, may cause fetal harm. (8.1)

See 17 **for PATIENT COUNSELING INFORMATION and Medication Guide.**

Revised: 11/2023

FULL PRESCRIBING INFORMATION: CONTENTS*

* Sections or subsections omitted from the full prescribing information are not listed.

FULL PRESCRIBING INFORMATION

WARNING: DISTANT SPREAD OF TOXIN EFFECT

Postmarketing reports indicate that the effects of BOTOX and all botulinum toxin products may spread from the area of injection to produce symptoms consistent with botulinum toxin effects. These may include asthenia, generalized muscle weakness, diplopia, ptosis, dysphagia, dysphonia, dysarthria, urinary incontinence and breathing difficulties. These symptoms have been reported hours to weeks after injection. Swallowing and breathing difficulties can be life threatening and there have been reports of death. The risk of symptoms is probably greatest in children treated for spasticity but symptoms can also occur in adults treated for spasticity and other conditions, particularly in those patients who have an underlying condition that would predispose them to these symptoms. In unapproved uses and in approved indications, cases of spread of effect have been reported at doses comparable to those used to treat cervical dystonia and spasticity and at lower doses *[see Warnings and Precautions (5.1)]*.

1 INDICATIONS AND USAGE

1.1 Adult Bladder Dysfunction

Overactive Bladder

BOTOX for injection is indicated for the treatment of overactive bladder with symptoms of urge urinary incontinence, urgency, and frequency, in adults who have an inadequate response to or are intolerant of an anticholinergic medication.

Detrusor Overactivity associated with a Neurologic Condition

BOTOX is indicated for the treatment of urinary incontinence due to detrusor overactivity associated with a neurologic condition (e.g., SCI, MS) in adults who have an inadequate response to or are intolerant of an anticholinergic medication.

1.2 Pediatric Detrusor Overactivity Associated with a Neurologic Condition

BOTOX is indicated for the treatment of neurogenic detrusor overactivity (NDO) in pediatric patients 5 years of age and older who have an inadequate response to or are intolerant of anticholinergic medication.

1.3 Chronic Migraine

BOTOX is indicated for the prophylaxis of headaches in adult patients with chronic migraine (≥15 days per month with headache lasting 4 hours a day or longer).

Limitations of Use

Safety and effectiveness have not been established for the prophylaxis of episodic migraine (14 headache days or fewer per month) in seven placebo-controlled studies.

1.4 Spasticity

BOTOX is indicated for the treatment of spasticity in patients 2 years of age and older.

Limitations of Use

BOTOX has not been shown to improve upper extremity functional abilities, or range of motion at a joint affected by a fixed contracture.

1.5 Cervical Dystonia

BOTOX is indicated for the treatment of adults with cervical dystonia, to reduce the severity of abnormal head position and neck pain associated with cervical dystonia.

1.6 Primary Axillary Hyperhidrosis

BOTOX is indicated for the treatment of severe primary axillary hyperhidrosis that is inadequately managed with topical agents.

Limitations of Use

The safety and effectiveness of BOTOX for hyperhidrosis in other body areas have not been established. Weakness of hand muscles and blepharoptosis may occur in patients who receive BOTOX for palmar hyperhidrosis and facial hyperhidrosis, respectively. Patients should be evaluated for potential causes of secondary hyperhidrosis (e.g., hyperthyroidism) to avoid symptomatic treatment of hyperhidrosis without the diagnosis and/or treatment of the underlying disease.

Safety and effectiveness of BOTOX have not been established for the treatment of axillary hyperhidrosis in pediatric patients under age 18.

1.7 Blepharospasm and Strabismus

BOTOX is indicated for the treatment of strabismus and blepharospasm associated with dystonia, including benign essential blepharospasm or VII nerve disorders in patients 12 years of age and older.

2 DOSAGE AND ADMINISTRATION

2.1 Instructions for Safe Use

The potency Units of BOTOX (onabotulinumtoxinA) for injection are specific to the preparation and assay method utilized. They are not interchangeable with other preparations of botulinum toxin products and, therefore, units of biological activity of BOTOX cannot be compared to nor converted into units of any other botulinum toxin products assessed with any other specific assay method *[see Warnings and Precautions (5.2) and Description (11)]*.

Indication specific dosage and administration recommendations should be followed. When initiating treatment, the lowest recommended dose should be used. In treating adult patients for one or more indications, the maximum cumulative dose should not exceed 400 Units, in a 3-month interval. In pediatric patients, the total dose should not exceed the lower of 10 Units/kg body weight or 340 Units, in a 3-month interval *[see Dosage and Administration (2.7)]*.

The safe and effective use of BOTOX depends upon proper storage of the product, selection of the correct dose, and proper reconstitution and administration techniques. An understanding of standard electromyographic techniques is also required for treatment of strabismus, upper or lower limb spasticity, and may be useful for the treatment of cervical dystonia. Physicians administering BOTOX must understand the relevant neuromuscular and structural anatomy of the area involved and any alterations to the anatomy due to prior surgical procedures and disease, especially when injecting near the lungs.

Do not use BOTOX and contact AbbVie (1-800-678-1605) if:

- the tamper evident features on the carton appear to be broken or compromised, or
- the U.S. License number 1889 is not present on the vial label and carton labeling *[see How Supplied/Storage and Handling (16)]*.

2.2 Preparation and Dilution Technique

Prior to injection, reconstitute each vacuum-dried vial of BOTOX with only sterile, preservative-free 0.9% Sodium Chloride Injection, USP. Draw up the proper amount of diluent in the appropriate size syringe (see Table 1, or for specific instructions for detrusor overactivity associated with a neurologic condition, see Section *2.3*), and slowly inject the diluent into the vial. Discard the vial if a vacuum does not pull the diluent into the vial. Gently mix BOTOX with the diluent by rotating the vial. Record the date and time of reconstitution on the space on the label. BOTOX should be administered within 24 hours after reconstitution. During this time period, unused reconstituted BOTOX should be stored in a refrigerator (2° to 8°C) for up to 24 hours until time of use. BOTOX vials are for single-dose only. Discard any unused portion.

Table 1: Dilution Instructions for BOTOX Vials (100 Units and 200 Units)**

Diluent* Added to 100 Unit Vial	Resulting Dose Units per 0.1 mL	Diluent* Added to 200 Unit Vial	Resulting Dose Units per 0.1 mL
1 mL	10 Units	1 mL	20 Units
2 mL	5 Units	2 mL	10 Units
4 mL	2.5 Units	4 mL	5 Units
8 mL	1.25 Units	8 mL	2.5 Units
10 mL	1 Unit	10 mL	2 Units

*Preservative-free 0.9% Sodium Chloride Injection, USP Only
**For Detrusor Overactivity associated with a Neurologic Condition Dilution, see Section *2.3*

Note: These dilutions are calculated for an injection volume of 0.1 mL. A decrease or increase in the BOTOX dose is also possible by administering a smaller or larger injection volume - from 0.05 mL (50% decrease in dose) to 0.15 mL (50% increase in dose).

An injection of BOTOX is prepared by drawing into an appropriately sized sterile syringe an amount of the properly reconstituted toxin slightly greater than the intended dose. Air bubbles in the syringe barrel are expelled and the syringe is attached to an appropriate injection needle. Patency of the needle should be confirmed. A new, sterile needle and syringe should be used to enter the vial on each occasion for removal of BOTOX.

Reconstituted BOTOX should be clear, colorless, and free of particulate matter. Parenteral drug products should be inspected visually for particulate matter and discoloration prior to administration and whenever the solution and the container permit.

2.3 Adult Bladder Dysfunction

General

Patients must not have a urinary tract infection (UTI) at the time of treatment. Prophylactic antibiotics, except aminoglycosides, *[see Drug Interactions (7.1)]* should be administered 1-3 days pre-treatment, on the treatment day, and 1-3 days post-treatment to reduce the likelihood of procedure-related UTI.

Patients should discontinue anti-platelet therapy at least 3 days before the injection procedure. Patients on anti-coagulant therapy need to be managed appropriately to decrease the risk of bleeding.

Appropriate caution should be exercised when performing a cystoscopy.

Overactive Bladder

An intravesical instillation of diluted local anesthetic with or without sedation may be used prior to injection, per local site practice. If a local anesthetic instillation is performed, the bladder should be drained and irrigated with sterile saline before injection.

The recommended dose is 100 Units of BOTOX, and is the maximum recommended dose. The recommended dilution is 100 Units/10 mL with preservative-free 0.9% Sodium Chloride Injection, USP (see Table 1). Dispose of any unused saline.

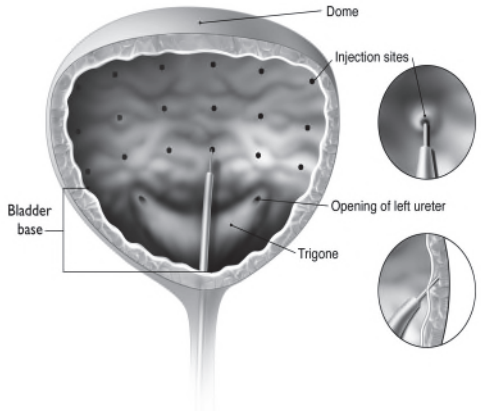
Reconstituted BOTOX (100 Units/10 mL) is injected into the detrusor muscle via a flexible or rigid cystoscope, avoiding the trigone. The bladder should be instilled with enough saline to achieve adequate visualization for the injections, but over-distension should be avoided.

The injection needle should be filled (primed) with approximately 1 mL of reconstituted BOTOX prior to the start of injections (depending on the needle length) to remove any air.

The needle should be inserted approximately 2 mm into the detrusor, and 20 injections of 0.5 mL each (total volume of 10 mL) should be spaced approximately 1 cm apart (see Figure 1). For the final injection, approximately 1 mL of sterile normal saline should be injected so that the remaining BOTOX in the needle is delivered to the bladder. After the injections are given, patients should demonstrate their ability to void prior to leaving the clinic. The patient should be observed for at least 30 minutes post-injection and until a spontaneous void has occurred.

Patients should be considered for reinjection when the clinical effect of the previous injection has diminished (median time until patients qualified for the second treatment of BOTOX in double-blind, placebo-controlled clinical studies was 169 days [~24 weeks]), but no sooner than 12 weeks from the prior bladder injection.

Figure 1: Injection Pattern for Intradetrusor Injections for Treatment of Overactive Bladder and Detrusor Overactivity Associated with a Neurologic Condition



Detrusor Overactivity associated with a Neurologic Condition

An intravesical instillation of diluted local anesthetic with or without sedation, or general anesthesia may be used prior to injection, per local site practice. If a local anesthetic instillation is performed, the bladder should be drained and irrigated with sterile saline before injection.

The recommended dose is 200 Units of BOTOX per treatment, and should not be exceeded.

200 Unit Vial of BOTOX
- Reconstitute a 200 Unit vial of BOTOX with 6 mL of preservative-free 0.9% Sodium Chloride Injection, USP and mix the vial gently.
- Draw 2 mL from the vial into each of three 10 mL syringes.
- Complete the reconstitution by adding 8 mL of preservative-free 0.9% Sodium Chloride Injection, USP into each of the 10 mL syringes, and mix gently. This will result in three 10 mL syringes each containing 10 mL (~67 Units in each), for a total of 200 Units of reconstituted BOTOX.
- Use immediately after reconstitution in the syringe. Dispose of any unused saline.

100 Unit Vial of BOTOX
- Reconstitute two 100 Unit vials of BOTOX, each with 6 mL of preservative-free 0.9% Sodium Chloride Injection, USP and mix the vials gently.
- Draw 4 mL from each vial into each of two 10 mL syringes. Draw the remaining 2 mL from each vial into a third 10 mL syringe for a total of 4 mL in each syringe.
- Complete the reconstitution by adding 6 mL of preservative-free 0.9% Sodium Chloride Injection, USP into each of the 10 mL syringes, and mix gently. This will result in three 10 mL syringes each containing 10 mL (~67 Units in each), for a total of 200 Units of reconstituted BOTOX.
- Use immediately after reconstitution in the syringe. Dispose of any unused saline.

Reconstituted BOTOX (200 Units/30 mL) is injected into the detrusor muscle via a flexible or rigid cystoscope, avoiding the trigone. The bladder should be instilled with enough saline to achieve adequate visualization for the injections, but over-distension should be avoided.

The injection needle should be filled (primed) with approximately 1 mL of reconstituted BOTOX prior to the start of injections (depending on the needle length) to remove any air.

The needle should be inserted approximately 2 mm into the detrusor, and 30 injections of 1 mL (~6.7 Units) each (total volume of 30 mL) should be spaced approximately 1 cm apart (see Figure 1). For the final injection, approximately 1 mL of sterile normal saline should be injected so that the remaining BOTOX in the needle is delivered to the bladder. After the injections are given, the saline used for bladder wall visualization should be drained. The patient should be observed for at least 30 minutes post-injection.

Patients should be considered for re-injection when the clinical effect of the previous injection diminishes (median time to qualification for re-treatment in the double-blind, placebo-controlled clinical studies was 295-337 days [42-48 weeks] for BOTOX 200 Units), but no sooner than 12 weeks from the prior bladder injection.

2.4 Pediatric Detrusor Overactivity Associated with a Neurologic Condition

Patients must not have a urinary tract infection (UTI) at the time of treatment. Oral prophylactic antibiotics, except aminoglycosides, *[see Drug Interactions (7.1)]* should be administered 1-3 days pre-treatment, on the treatment day, and 1-3 days post-treatment to reduce the likelihood of procedure-related UTI. Alternatively, for patients receiving general anesthesia (or conscious sedation) for the treatment of detrusor overactivity associated with a neurologic condition, one dose of IV prophylactic antibiotics, except aminoglycosides, *[see Drug Interactions (7.1)]* may be administered prior to treatment administration on the day of treatment.

Patients should discontinue anti-platelet therapy at least 3 days before the injection procedure. Patients on anti-coagulant therapy need to be managed appropriately to decrease the risk of bleeding.

Appropriate caution should be exercised when performing a cystoscopy.

- In patients 5 years to less than 12 years of age: Consider general anesthesia (or conscious sedation) prior to injection, per local site practice.
- In patients 12 years of age or older: Consider an intravesical instillation of diluted local anesthetic with or without sedation, or general anesthesia prior to injection, per local site practice.

At a minimum, consider a diluted instillation of local anesthetic for all age groups. If a local anesthetic instillation is performed, drain and irrigate the bladder with sterile saline before injection.

If patient's body weight is greater than or equal to 34 kg, the recommended dosage is 200 Units of BOTOX per treatment administered as an intradetrusor injection after dilution:
- Reconstitute BOTOX to result in 20 Units BOTOX/mL in the vial(s):
  - BOTOX 200 Unit vial: add 10 mL of preservative-free 0.9% Sodium Chloride Injection, USP and mix the vial gently.

  - BOTOX 100 Unit vials: add 5 mL of preservative-free 0.9% Sodium Chloride Injection, USP to each of two 100 Unit vials of BOTOX and mix the vials gently.
- Draw 10 mL from the vial(s) into one 10 mL dosing syringe.
- Use immediately after reconstitution in the syringe. Dispose of any unused saline.

If patient's body weight is less than 34 kg, the recommended dosage is 6 Units/kg body weight administered as a bladder injection after dilution (refer to Table 2):

- Reconstitute BOTOX to result in 20 Units BOTOX/mL in the vial(s):
  - BOTOX 200 Unit vial: add 10 mL of preservative-free 0.9% Sodium Chloride Injection, USP and mix the vial gently.
  - BOTOX 100 Unit vial(s): add 5 mL of preservative-free 0.9% Sodium Chloride Injection, USP to one 100 Unit vial of BOTOX (if final dose is less than or equal to 100 U) or to each of two 100 Unit vials of BOTOX (if final dose is greater than 100 U) and mix the vial(s) gently.
- Refer to Table 2 for dilution instructions (i.e., the amount of reconstituted BOTOX and additional diluent to draw into one 10 mL dosing syringe).
- Use BOTOX immediately after reconstitution in the syringe. Dispose of any unused preservative-free 0.9% Sodium Chloride Injection, USP.

Table 2: BOTOX Dilution Instructions and Final Dosing for Patients with Body Weight < 34 kg

Body Weight (kg)	Volume of reconstituted BOTOX and Diluent* (mL) to draw into dosing syringe to achieve a final volume of 10 mL		Final dose of BOTOX in dosing syringe
	BOTOX (mL)	Diluent* (mL)	
12 to less than 14	3.6	6.4	72 Units
14 to less than 16	4.2	5.8	84 Units
16 to less than 18	4.8	5.2	96 Units
18 to less than 20	5.4	4.6	108 Units
20 to less than 22	6	4	120 Units
22 to less than 24	6.6	3.4	132 Units
24 to less than 26	7.2	2.8	144 Units
26 to less than 28	7.8	2.2	156 Units
28 to less than 30	8.4	1.6	168 Units
30 to less than 32	9	1	180 Units
32 to less than 34	9.6	0.4	192 Units

*Preservative-free 0.9% Sodium Chloride Injection, USP Only

Reconstituted BOTOX is injected into the detrusor muscle via a flexible or rigid cystoscope, avoiding the trigone. The bladder should be instilled with enough saline to achieve adequate visualization for the injections, but over-distension should be avoided.

The injection needle should be filled (primed) with approximately 1 mL of reconstituted BOTOX prior to the start of injections (depending on the needle length) to remove any air.

The needle should be inserted approximately 2 mm into the detrusor, and 20 injections of 0.5 mL each (total volume of 10 mL) should be spaced approximately 1 cm apart (see Figure 1). For the final injection, approximately 1 mL of sterile normal saline should be injected so that the remaining BOTOX in the needle is delivered to the bladder. After the injections are given, the saline used for bladder wall visualization should be drained. The patient should be observed for at least 30 minutes post-injection.

Patients should be considered for re-injection when the clinical effect of the previous injection diminishes (median time to qualification for re-treatment in the double-blind, parallel group clinical study was 207 days [30 weeks] for BOTOX 200 Units), but no sooner than 12 weeks from the prior bladder injection.

2.5 Chronic Migraine

The recommended dilution is 200 Units/4 mL or 100 Units/2 mL, with a final concentration of 5 Units per 0.1 mL (see Table 1). The recommended dose for treating chronic migraine is 155 Units administered intramuscularly using a sterile 30-gauge, 0.5 inch needle as 0.1 mL (5 Units) injections per each site. Injections should be divided across 7 specific head/neck muscle areas as specified in the diagrams and Table 3 below. A one inch needle may be needed in the neck region for patients with thick neck muscles. With the exception of the procerus muscle, which should be injected at one site (midline), all muscles should be injected bilaterally with half the number of injection sites administered to the left, and half to the right side of the head and neck. The recommended re-treatment schedule is every 12 weeks.

Diagrams 1-4: Recommended Injection Sites (A through G) for Chronic Migraine

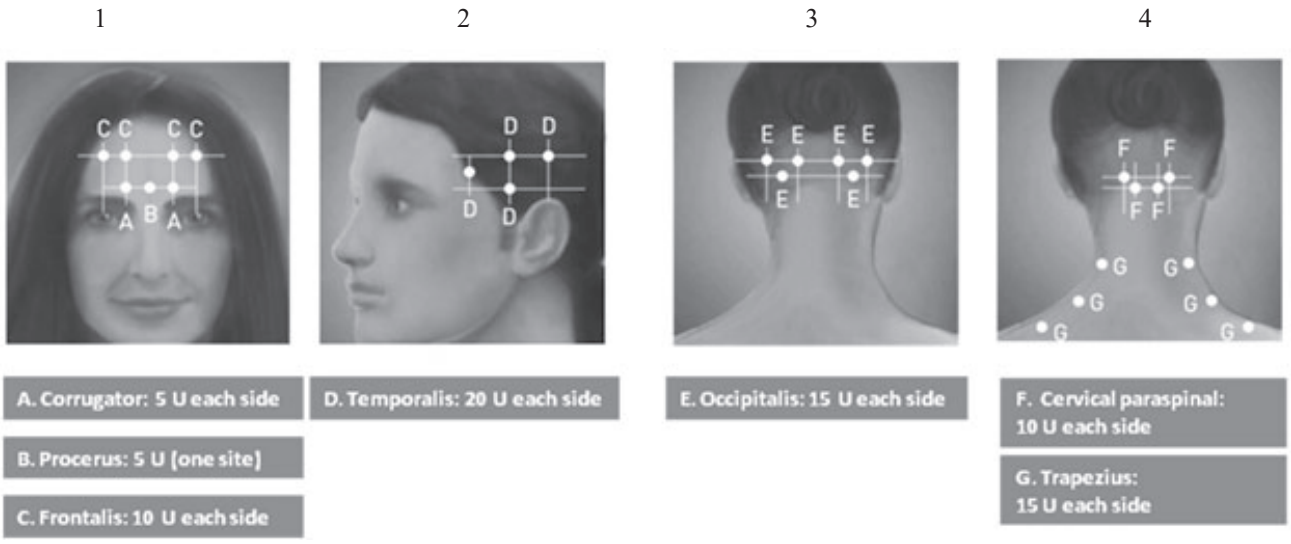


Table 3: BOTOX Dosing by Muscle for Chronic Migraine

Head/Neck Area	Recommended Dose (Number of Sites[a])
Frontalis[b]	20 Units divided in 4 sites
Corrugator[b]	10 Units divided in 2 sites
Procerus	5 Units in 1 site
Occipitalis[b]	30 Units divided in 6 sites
Temporalis[b]	40 Units divided in 8 sites
Trapezius[b]	30 Units divided in 6 sites
Cervical Paraspinal Muscle Group[b]	20 Units divided in 4 sites
Total Dose:	**155 Units** divided **in 31 sites**

[a] Each IM injection site = 0.1 mL = 5 Units BOTOX
[b] Dose distributed bilaterally

2.6 Adult Spasticity

General

Dosing in initial and sequential treatment sessions should be tailored to the individual based on the size, number and location of muscles involved, severity of spasticity, the presence of local muscle weakness, the patient's response to previous treatment, or adverse event history with BOTOX.

The recommended dilution is 200 Units/4 mL or 100 Units/2 mL with preservative-free 0.9% Sodium Chloride Injection, USP (see Table 1). The lowest recommended starting dose should be used, and no more than 50 Units per site should generally be administered. An appropriately sized needle (e.g., 25-30 gauge) may be used for superficial muscles, and a longer 22 gauge needle may be used for deeper musculature. Localization of the involved muscles with techniques such as needle electromyographic guidance, nerve stimulation, or ultrasound is recommended.

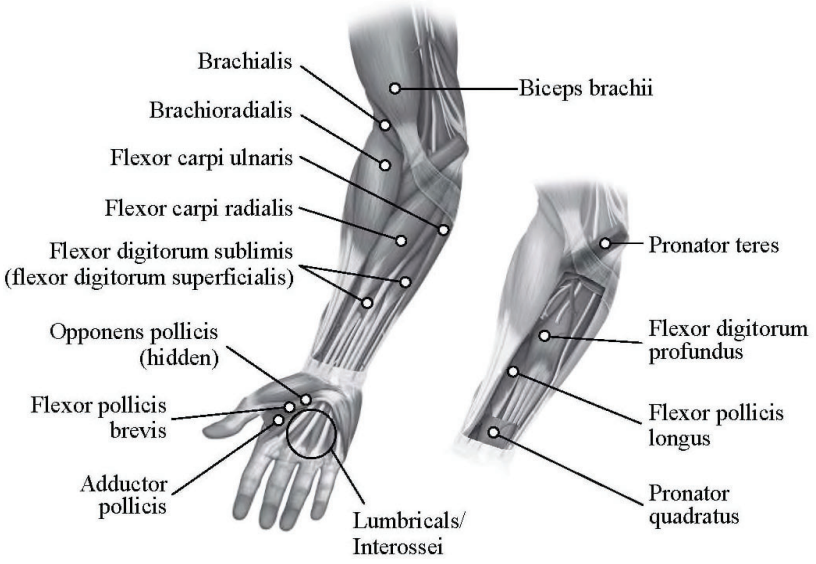
Repeat BOTOX treatment may be administered when the effect of a previous injection has diminished, but generally no sooner than 12 weeks after the previous injection. The degree and pattern of muscle spasticity at the time of re-injection may necessitate alterations in the dose of BOTOX and muscles to be injected.

Adult Upper Limb Spasticity
In clinical trials, doses ranging from 75 Units to 400 Units were divided among selected muscles (see Table 4 and Figure 2) at a given treatment session.

Table 4: BOTOX Dosing by Muscle for Adult Upper Limb Spasticity

Muscle	Recommended Dose Total Dosage (Number of Sites)
Biceps Brachii	60 Units to 200 Units divided in 2 to 4 sites
Brachioradialis	45 Units to 75 Units divided in 1 to 2 sites
Brachialis	30 Units to 50 Units divided in 1 to 2 sites
Pronator Teres	15 Units to 25 Units in 1 site
Pronator Quadratus	10 Units to 50 Units in 1 site
Flexor Carpi Radialis	12.5 Units to 50 Units in 1 site
Flexor Carpi Ulnaris	12.5 Units to 50 Units in 1 site
Flexor Digitorum Profundus	30 Units to 50 Units in 1 site
Flexor Digitorum Sublimis	30 Units to 50 Units in 1 site
Lumbricals/Interossei	5 Units to 10 Units in 1 site
Adductor Pollicis	20 Units in 1 site
Flexor Pollicis Longus	20 Units in 1 site
Flexor pollicis brevis/ Opponens pollicis	5 Units to 25 Units in 1 site

Figure 2: Injection Sites for Adult Upper Limb Spasticity

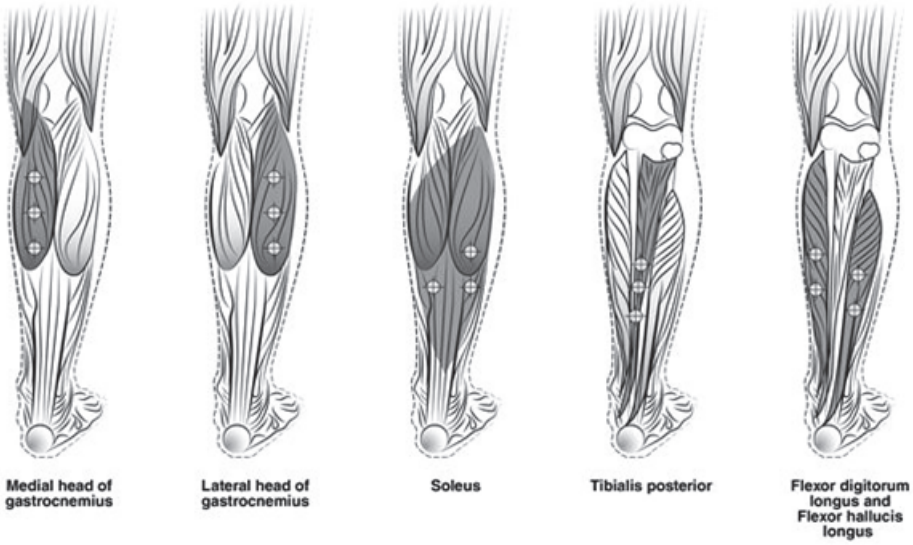


Adult Lower Limb Spasticity
The recommended dose for treating adult lower limb spasticity is 300 Units to 400 Units divided among 5 muscles (gastrocnemius, soleus, tibialis posterior, flexor hallucis longus and flexor digitorum longus) (see Table 5 and Figure 3).

Table 5: BOTOX Dosing by Muscle for Adult Lower Limb Spasticity

Muscle	Recommended Dose Total Dosage (Number of Sites)
Gastrocnemius medial head	75 Units divided in 3 sites
Gastrocnemius lateral head	75 Units divided in 3 sites
Soleus	75 Units divided in 3 sites
Tibialis Posterior	75 Units divided in 3 sites
Flexor hallucis longus	50 Units divided in 2 sites
Flexor digitorum longus	50 Units divided in 2 sites

Figure 3: Injection Sites for Adult Lower Limb Spasticity



2.7 Pediatric Spasticity

General

Localization of the involved muscles with techniques such as needle electromyographic guidance, nerve stimulation, or ultrasound is recommended. When treating both lower limbs or the upper and lower limbs in combination, the total dose should not exceed the lower of 10 Units/kg body weight or 340 Units, in a 3-month interval *[see Boxed Warning and Warnings and Precautions (5.1, 5.6)]*. Additional general adult spasticity dosing information is also applicable to pediatric spasticity patients *[see Dosage and Administration (2.6)]*.

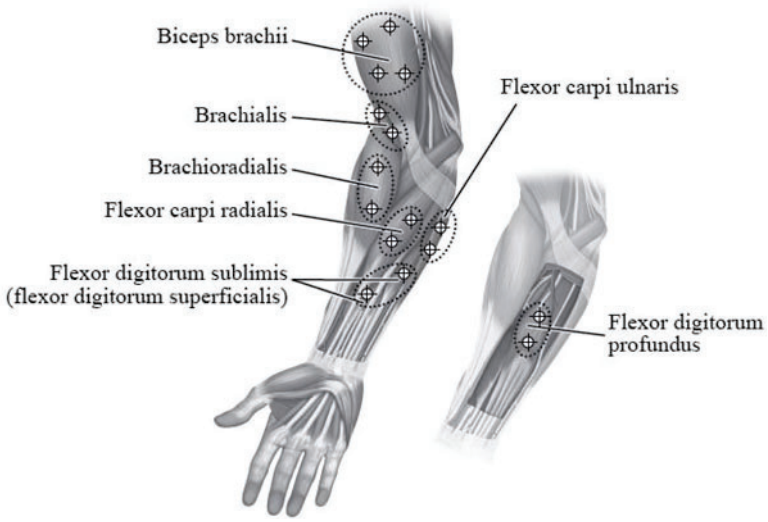
Pediatric Upper Limb Spasticity

The recommended dose for treating pediatric upper limb spasticity is 3 Units/kg to 6 Units/kg divided among the affected muscles (see Table 6 and Figure 4). The total dose of BOTOX administered per treatment session in the upper limb should not exceed 6 Units/kg or 200 Units, whichever is lower.

Table 6: BOTOX Dosing by Muscle for Pediatric Upper Limb Spasticity

Muscle	Recommended Dose and Number of Sites
Biceps Brachii	1.5 Units/kg to 3 Units/kg divided in 4 sites
Brachialis	1 Unit/kg to 2 Units/kg divided in 2 sites
Brachioradialis	0.5 Units/kg to 1 Unit/kg divided in 2 sites
Flexor Carpi Radialis	1 Unit/kg to 2 Units/kg divided in 2 sites
Flexor Carpi Ulnaris	1 Unit/kg to 2 Units/kg divided in 2 sites
Flexor Digitorum Profundus	0.5 Units/kg to 1 Unit/kg divided in 2 sites
Flexor Digitorum Sublimis	0.5 Units/kg to 1 Unit/kg divided in 2 sites

Figure 4: Injection Sites for Pediatric Upper Limb Spasticity



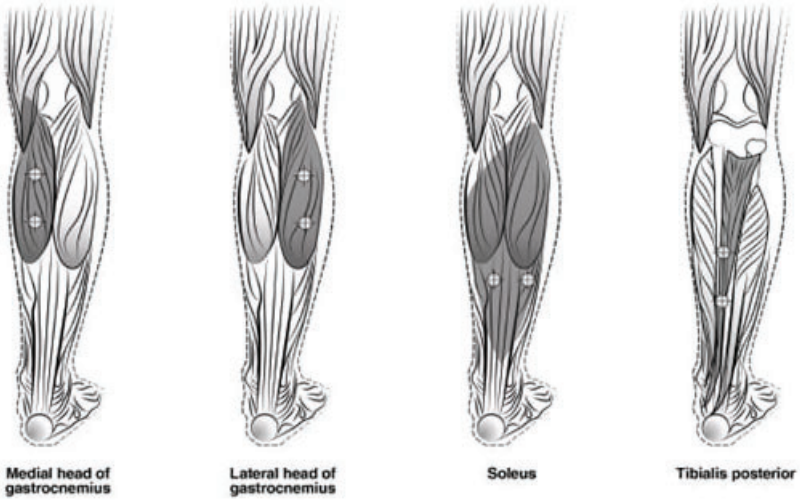
Pediatric Lower Limb Spasticity

The recommended dose for treating pediatric lower limb spasticity is 4 Units/kg to 8 Units/kg divided among the affected muscles (see Table 7 and Figure 5). The total dose of BOTOX administered per treatment session in the lower limb should not exceed 8 Units/kg or 300 Units, whichever is lower.

Table 7: BOTOX Dosing by Muscle for Pediatric Lower Limb Spasticity

Muscle	Recommended Dose Total Dosage (Number of Sites)
Gastrocnemius medial head	1 Unit/kg to 2 Units/kg divided in 2 sites
Gastrocnemius lateral head	1 Unit/kg to 2 Units/kg divided in 2 sites
Soleus	1 Unit/kg to 2 Units/kg divided in 2 sites
Tibialis Posterior	1 Unit/kg to 2 Units/kg divided in 2 sites

Figure 5: Injection Sites for Pediatric Lower Limb Spasticity



2.8 Cervical Dystonia

A double-blind, placebo-controlled study enrolled patients who had extended histories of receiving and tolerating BOTOX injections, with prior individualized adjustment of dose. The mean BOTOX dose administered to patients in this study was 236 Units (25th to 75th percentile range of 198 Units to 300 Units). The BOTOX dose was divided among the affected muscles *[see Clinical Studies (14.7)]*.

Dosing in initial and sequential treatment sessions should be tailored to the individual patient based on the patient's head and neck position, localization of pain, muscle hypertrophy, patient response, and adverse event history. The initial dose for a patient without prior use of BOTOX should be at a lower dose, with subsequent dosing adjusted based on individual response. Limiting the total dose injected into the sternocleidomastoid muscle to 100 Units or less may decrease the occurrence of dysphagia *[see Warnings and Precautions (5.1, 5.5, 5.6)]*.

The recommended dilution is 200 Units/2 mL, 200 Units/4 mL, 100 Units/1 mL, or 100 Units/2 mL with preservative-free 0.9% Sodium Chloride Injection, USP, depending on volume and number of injection sites desired to achieve treatment objectives (see Table 1). In general, no more than 50 Units per site should be administered using a sterile needle (e.g., 25-30 gauge) of an appropriate length. Localization of the involved muscles with electromyographic guidance may be useful.

Clinical improvement generally begins within the first two weeks after injection with maximum clinical benefit at approximately six weeks post-injection. In the double-blind, placebo-controlled study most subjects were observed to have returned to pre-treatment status by 3 months post-treatment.

2.9 Primary Axillary Hyperhidrosis

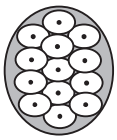
The recommended dose is 50 Units per axilla. The hyperhidrotic area to be injected should be defined using standard staining techniques, e.g., Minor's Iodine-Starch Test. The recommended dilution is 100 Units/4 mL with preservative-free 0.9% Sodium Chloride Injection, USP (see Table 1). Using a sterile 30 gauge needle, 50 Units of BOTOX (2 mL) is injected intradermally in 0.1 to 0.2 mL aliquots to each axilla evenly distributed in multiple sites (10-15) approximately 1-2 cm apart.

Repeat injections for hyperhidrosis should be administered when the clinical effect of a previous injection diminishes.

Instructions for the Minor's Iodine-Starch Test Procedure:
Patients should shave underarms and abstain from use of over-the-counter deodorants or antiperspirants for 24 hours prior to the test. Patient should be resting comfortably without exercise or hot drinks for approximately 30 minutes prior to the test. Dry the underarm area and then immediately paint it with iodine solution. Allow the area to dry, then lightly sprinkle the area with starch powder. Gently blow off any excess starch powder. The hyperhidrotic area will develop a deep blue-black color over approximately 10 minutes.

Each injection site has a ring of effect of up to approximately 2 cm in diameter. To minimize the area of no effect, the injection sites should be evenly spaced as shown in Figure 6.

Figure 6: Injection Pattern for Primary Axillary Hyperhidrosis



Each dose is injected to a depth of approximately 2 mm and at a 45° angle to the skin surface, with the bevel side up to minimize leakage and to ensure the injections remain intradermal. If injection sites are marked in ink, do not inject BOTOX directly through the ink mark to avoid a permanent tattoo effect.

2.10 Blepharospasm

For blepharospasm, reconstituted BOTOX is injected using a sterile, 27-30 gauge needle without electromyographic guidance. The initial recommended dose is 1.25 Units-2.5 Units (0.05 mL to 0.1 mL volume at each site) injected into the medial and lateral pre-tarsal orbicularis oculi of the upper lid and into the lateral pre-tarsal orbicularis oculi of the lower lid. Avoiding injection near the levator palpebrae superioris may reduce the complication of ptosis. Avoiding medial lower lid injections, and thereby reducing diffusion into the inferior oblique, may reduce the complication of diplopia. Ecchymosis occurs easily in the soft eyelid tissues. This can be prevented by applying pressure at the injection site immediately after the injection.

The recommended dilution to achieve 1.25 Units is 100 Units/8 mL; for 2.5 Units it is 100 Units/4 mL (see Table 1).

In general, the initial effect of the injections is seen within three days and reaches a peak at one to two weeks post-treatment. Each treatment lasts approximately three months, following which the procedure can be repeated. At repeat treatment sessions, the dose may be increased up to two-fold if the response from the initial treatment is considered insufficient, usually defined as an effect that does not last longer than two months. However, there appears to be little benefit obtainable from injecting more than 5 Units per site. Some tolerance may be found when BOTOX is used in treating blepharospasm if treatments are given any more frequently than every three months, and is rare to have the effect be permanent.

The cumulative dose of BOTOX treatment for blepharospasm in a 30-day period should not exceed 200 Units.

2.11 Strabismus

BOTOX is intended for injection into extraocular muscles utilizing the electrical activity recorded from the tip of the injection needle as a guide to placement within the target muscle. Injection without surgical exposure or electromyographic guidance should not be attempted. Physicians should be familiar with electromyographic technique.

To prepare the eye for BOTOX injection, it is recommended that several drops of a local anesthetic and an ocular decongestant be given several minutes prior to injection.

The volume of BOTOX injected for treatment of strabismus should be between 0.05-0.15 mL per muscle.

The initial listed doses of the reconstituted BOTOX *[see Dosage and Administration (2.2)]* typically create paralysis of the injected muscles beginning one to two days after injection and increasing in intensity during the first week. The paralysis lasts for 2-6 weeks and gradually resolves over a similar time period. Overcorrections lasting over six months have been rare. About one half of patients will require subsequent doses because of inadequate paralytic response of the muscle to the initial dose, or because of mechanical factors such as large deviations or restrictions, or because of the lack of binocular motor fusion to stabilize the alignment.

Initial Doses in Units

Use the lower listed doses for treatment of small deviations. Use the larger doses only for large deviations.

- For vertical muscles, and for horizontal strabismus of less than 20 prism diopters: 1.25 Units-2.5 Units in any one muscle.
- For horizontal strabismus of 20 prism diopters to 50 prism diopters: 2.5 Units-5 Units in any one muscle.
- For persistent VI nerve palsy of one month or longer duration: 1.25 Units-2.5 Units in the medial rectus muscle.

Subsequent Doses for Residual or Recurrent Strabismus

- It is recommended that patients be re-examined 7-14 days after each injection to assess the effect of that dose.
- Patients experiencing adequate paralysis of the target muscle that require subsequent injections should receive a dose comparable to the initial dose.
- Subsequent doses for patients experiencing incomplete paralysis of the target muscle may be increased up to two-fold compared to the previously administered dose.
- Subsequent injections should not be administered until the effects of the previous dose have dissipated as evidenced by substantial function in the injected and adjacent muscles.
- The maximum recommended dose as a single injection for any one muscle is 25 Units.

The recommended dilution to achieve 1.25 Units is 100 Units/8 mL; for 2.5 Units it is 100 Units/4 mL (see Table 1).

3 DOSAGE FORMS AND STRENGTHS

For Injection: sterile 100 Units or 200 Units vacuum-dried powder in single-dose vials for reconstitution only with sterile, preservative-free 0.9% Sodium Chloride Injection, USP prior to injection.

4 CONTRAINDICATIONS

BOTOX is contraindicated:

- In patients who are hypersensitive to any botulinum toxin product or to any of the components in the formulation *[see Warnings and Precautions (5.4)]*.
- In the presence of infection at the proposed injection site(s).
- For intradetrusor injection in patients with a urinary tract infection; or in patients with urinary retention or post-void residual (PVR) urine volume >200 mL who are not routinely performing clean intermittent self-catheterization (CIC) *[see Warnings and Precautions (5.12, 5.13)]*.

5 WARNINGS AND PRECAUTIONS

5.1 Spread of Toxin Effect

Postmarketing safety data from BOTOX and other approved botulinum toxins suggest that botulinum toxin effects may, in some cases, be observed beyond the site of local injection. The symptoms are consistent with the mechanism of action of botulinum toxin and may include asthenia, generalized muscle weakness, diplopia, ptosis, dysphagia, dysphonia, dysarthria, urinary incontinence, and breathing difficulties. These symptoms have been reported hours to weeks after injection. Swallowing and breathing difficulties can be life threatening and there have been reports of death related to spread of toxin effects. The risk of symptoms is probably greatest in children treated for spasticity but symptoms can also occur in adults treated for spasticity and other conditions, and particularly in those patients who have an underlying condition that would predispose them to these symptoms. In unapproved uses and in approved indications, symptoms consistent with spread of toxin effect have been reported at doses comparable to or lower than doses used to treat cervical dystonia and spasticity. Patients or caregivers should be advised to seek immediate medical care if swallowing, speech or respiratory disorders occur.

No definitive serious adverse event reports of distant spread of toxin effect associated with BOTOX for blepharospasm at the recommended dose (30 Units and below), severe primary axillary hyperhidrosis at the recommended dose (100 Units), strabismus, or for chronic migraine at the labeled doses have been reported.

5.2 Lack of Interchangeability between Botulinum Toxin Products

The potency Units of BOTOX are specific to the preparation and assay method utilized. They are not interchangeable with other preparations of botulinum toxin products and, therefore, units of biological activity of BOTOX cannot be compared to nor converted into units of any other botulinum toxin products assessed with any other specific assay method *[see Description (11)]*.

5.3 Serious Adverse Reactions with Unapproved Use

Serious adverse reactions, including excessive weakness, dysphagia, and aspiration pneumonia, with some adverse reactions associated with fatal outcomes, have been reported in patients who received BOTOX injections for unapproved uses. In these cases, the adverse reactions were not necessarily related to distant spread of toxin, but may have resulted from the administration of BOTOX to the site of injection and/or adjacent structures. In several of the cases, patients had pre-existing dysphagia or other significant disabilities. There is insufficient information to identify factors associated with an increased risk for adverse reactions associated with the unapproved uses of BOTOX. The safety and effectiveness of BOTOX for unapproved uses have not been established.

5.4 Hypersensitivity Reactions

Serious and/or immediate hypersensitivity reactions have been reported. These reactions include anaphylaxis, serum sickness, urticaria, soft tissue edema, and dyspnea. If such a reaction occurs, further injection of BOTOX should be discontinued and appropriate medical therapy immediately instituted. One fatal case of anaphylaxis has been reported in which lidocaine was used as the diluent, and consequently the causal agent cannot be reliably determined.

5.5 Increased Risk of Clinically Significant Effects with Pre-Existing Neuromuscular Disorders

Individuals with peripheral motor neuropathic diseases, amyotrophic lateral sclerosis or neuromuscular junction disorders (e.g., myasthenia gravis or Lambert-Eaton syndrome) should be monitored when given botulinum toxin. Patients with known or unrecognized neuromuscular disorders or neuromuscular junction disorders may be at increased risk of clinically significant effects

including generalized muscle weakness, diplopia, ptosis, dysphonia, dysarthria, severe dysphagia and respiratory compromise from therapeutic doses of BOTOX *[see Warnings and Precautions (5.1, 5.6)]*.

5.6 Dysphagia and Breathing Difficulties

Treatment with BOTOX and other botulinum toxin products can result in swallowing or breathing difficulties. Patients with pre-existing swallowing or breathing difficulties may be more susceptible to these complications. In most cases, this is a consequence of weakening of muscles in the area of injection that are involved in breathing or oropharyngeal muscles that control swallowing or breathing *[see Warnings and Precautions (5.1)]*.

Deaths as a complication of severe dysphagia have been reported after treatment with botulinum toxin. Dysphagia may persist for several months, and require use of a feeding tube to maintain adequate nutrition and hydration. Aspiration may result from severe dysphagia and is a particular risk when treating patients in whom swallowing or respiratory function is already compromised.

Treatment with botulinum toxins may weaken neck muscles that serve as accessory muscles of ventilation. This may result in a critical loss of breathing capacity in patients with respiratory disorders who may have become dependent upon these accessory muscles. There have been postmarketing reports of serious breathing difficulties, including respiratory failure.

Patients with smaller neck muscle mass and patients who require bilateral injections into the sternocleidomastoid muscle for the treatment of cervical dystonia have been reported to be at greater risk for dysphagia. Limiting the dose injected into the sternocleidomastoid muscle may reduce the occurrence of dysphagia. Injections into the levator scapulae may be associated with an increased risk of upper respiratory infection and dysphagia.

Patients treated with botulinum toxin may require immediate medical attention should they develop problems with swallowing, speech or respiratory disorders. These reactions can occur within hours to weeks after injection with botulinum toxin *[see Warnings and Precautions (5.1)]*.

5.7 Pulmonary Effects of BOTOX in Patients with Compromised Respiratory Status Treated for Spasticity or for Detrusor Overactivity Associated with a Neurologic Condition

Patients with compromised respiratory status treated with BOTOX for spasticity should be monitored closely. In a double-blind, placebo-controlled, parallel group study in adult patients treated for upper limb spasticity with stable reduced pulmonary function (defined as FEV_1 40-80% of predicted value and $FEV_1/FVC \leq 0.75$), the event rate in change of Forced Vital Capacity (FVC) ≥15% or ≥20% was generally greater in patients treated with BOTOX than in patients treated with placebo (see Table 8).

Table 8: Event Rate Per Patient Treatment Cycle Among Adult Upper Limb Spasticity Patients with Reduced Lung Function Who Experienced at Least a 15% or 20% Decrease in FVC From Baseline at Week 1, 6, 12 Post-injection with Up to Two Treatment Cycles with BOTOX or Placebo

	BOTOX 360 Units		BOTOX 240 Units		Placebo	
	≥15%	≥20%	≥15%	≥20%	≥15%	≥20%
Week 1	4%	0%	3%	0%	7%	3%
Week 6	7%	4%	4%	2%	2%	2%
Week 12	10%	5%	2%	1%	4%	1%

Differences from placebo were not statistically significant

In adult spasticity patients with reduced lung function, upper respiratory tract infections were also reported more frequently as adverse reactions in patients treated with BOTOX than in patients treated with placebo *[see Warnings and Precautions (5.10)]*.

In a double-blind, placebo-controlled, parallel group study in adult patients with detrusor overactivity associated with a neurologic condition and restrictive lung disease of neuromuscular etiology [defined as FVC 50-80% of predicted value in patients with spinal cord injury between C5 and C8, or MS] the event rate in change of Forced Vital Capacity ≥15% or ≥20% was generally greater in patients treated with BOTOX than in patients treated with placebo (see Table 9).

Table 9: Number and Percent of Patients Experiencing at Least a 15% or 20% Decrease in FVC From Baseline at Week 2, 6, 12 Post-Injection with BOTOX or Placebo

	BOTOX 200 Units		Placebo	
	≥15%	≥20%	≥15%	≥20%
Week 2	0/15 (0%)	0/15 (0%)	1/11 (9%)	0/11 (0%)
Week 6	2/13 (15%)	1/13 (8%)	0/12 (0%)	0/12 (0%)
Week 12	0/12(0%)	0/12 (0%)	0/7 (0%)	0/7 (0%)

5.8 Corneal Exposure and Ulceration in Patients Treated with BOTOX for Blepharospasm

Reduced blinking from BOTOX injection of the orbicularis muscle can lead to corneal exposure, persistent epithelial defect, and corneal ulceration, especially in patients with VII nerve disorders. Vigorous treatment of any epithelial defect should be employed. This may require protective drops, ointment, therapeutic soft contact lenses, or closure of the eye by patching or other means.

5.9 Retrobulbar Hemorrhages in Patients Treated with BOTOX for Strabismus

During the administration of BOTOX for the treatment of strabismus, retrobulbar hemorrhages sufficient to compromise retinal circulation have occurred. It is recommended that appropriate instruments to decompress the orbit be accessible.

5.10 Bronchitis and Upper Respiratory Tract Infections in Patients Treated for Spasticity

Bronchitis was reported more frequently as an adverse reaction in adult patients treated for upper limb spasticity with BOTOX (3% at 251 Units-360 Units total dose), compared to placebo (1%). In adult patients with reduced lung function treated for upper limb spasticity, upper respiratory tract infections were also reported more frequently as adverse reactions in patients treated with BOTOX (11% at 360 Units total dose; 8% at 240 Units total dose) compared to placebo (6%). In adult patients treated for lower limb spasticity, upper respiratory tract infections were reported more frequently as an adverse reaction in patients treated with BOTOX (2% at 300 Units to 400 Units total dose) compared to placebo (1%). In pediatric patients treated for upper limb spasticity, upper respiratory tract infections were reported more frequently as an adverse reaction in patients treated with BOTOX (17% at 6 Units/kg and 10% at 3 Units/kg) compared to placebo (9%). In pediatric patients treated for lower limb spasticity, upper respiratory tract infection was not reported with an incidence greater than placebo.

5.11 Autonomic Dysreflexia in Patients Treated for Detrusor Overactivity Associated with a Neurologic Condition

Autonomic dysreflexia associated with intradetrusor injections of BOTOX could occur in patients treated for detrusor overactivity associated with a neurologic condition and may require prompt medical therapy. In clinical trials, the incidence of autonomic dysreflexia was greater in adult patients treated with BOTOX 200 Units compared with placebo (1.5% versus 0.4%, respectively).

5.12 Urinary Tract Infections in Patients with Overactive Bladder

BOTOX increases the incidence of urinary tract infection *[see Adverse Reactions (6.1)]*. Clinical trials for overactive bladder excluded patients with more than 2 UTIs in the past 6 months and those taking antibiotics chronically due to recurrent UTIs. Use of BOTOX for the treatment of overactive bladder in such patients and in patients with multiple recurrent UTIs during treatment should only be considered when the benefit is likely to outweigh the potential risk.

5.13 Urinary Retention in Adults Treated for Bladder Dysfunction

Due to the risk of urinary retention, treat only patients who are willing and able to initiate catheterization post-treatment, if required, for urinary retention.

In patients who are not catheterizing, post-void residual (PVR) urine volume should be assessed within 2 weeks post-treatment and periodically as medically appropriate up to 12 weeks, particularly in patients with multiple sclerosis or diabetes mellitus. Depending on patient symptoms, institute catheterization if PVR urine volume exceeds 200 mL and continue until PVR falls below 200 mL. Instruct patients to contact their physician if they experience difficulty in voiding as catheterization may be required.

The incidence and duration of urinary retention is described below for adult patients with overactive bladder and detrusor overactivity associated with a neurologic condition who received BOTOX or placebo injections.

Overactive Bladder

In double-blind, placebo-controlled trials in patients with OAB, the proportion of subjects who initiated clean intermittent catheterization (CIC) for urinary retention following treatment with BOTOX or placebo is shown in Table 10. The duration of post-injection catheterization for those who developed urinary retention is also shown.

Table 10: Proportion of Patients Catheterizing for Urinary Retention and Duration of Catheterization Following an Injection in Double-Blind, Placebo-Controlled Clinical Trials in OAB

Timepoint	BOTOX 100 Units (N=552)	Placebo (N=542)
Proportion of Patients Catheterizing for Urinary Retention		
At any time during complete treatment cycle	6.5% (n=36)	0.4% (n=2)
Duration of Catheterization for Urinary Retention (Days)		
Median	63	11
Min, Max	1, 214	3, 18

Patients with diabetes mellitus treated with BOTOX were more likely to develop urinary retention than those without diabetes, as shown in Table 11.

Table 11: Proportion of Patients Experiencing Urinary Retention Following an Injection in Double-Blind, Placebo-Controlled Clinical Trials in OAB According to History of Diabetes Mellitus

	Patients with Diabetes		Patients without Diabetes	
	BOTOX 100 Units (N=81)	Placebo (N=69)	BOTOX 100 Units (N=526)	Placebo (N=516)
Urinary retention	12.3% (n=10)	0	6.3% (n=33)	0.6% (n=3)

Adult Detrusor Overactivity associated with a Neurologic Condition

In two double-blind, placebo-controlled trials in adult patients with detrusor overactivity associated with a neurologic condition (NDO-1 and NDO-2), the proportion of subjects who were not using clean intermittent catheterization (CIC) prior to injection and who subsequently required catheterization for urinary retention following treatment with BOTOX 200 Units or placebo is shown in Table 12. The duration of post-injection catheterization for those who developed urinary retention is also shown.

Table 12: Proportion of Adult Patients Not Using CIC at Baseline and then Catheterizing for Urinary Retention and Duration of Catheterization Following an Injection in Double-Blind, Placebo-Controlled Clinical Trials

Timepoint	BOTOX 200 Units (N=108)	Placebo (N=104)
Proportion of Patients Catheterizing for Urinary Retention		
At any time during complete treatment cycle	30.6% (n=33)	6.7% (n=7)
Duration of Catheterization for Urinary Retention (Days)		
Median	289	358
Min, Max	1, 530	2, 379

Among adult patients not using CIC at baseline, those with Multiple Sclerosis (MS) were more likely to require CIC post-injection than those with Spinal Cord Injury (SCI) (see Table 13).

Table 13: Proportion of Adult Patients by Etiology (MS and SCI) Not Using CIC at Baseline and then Catheterizing for Urinary Retention Following an Injection in Double-Blind, Placebo-Controlled Clinical Trials

Timepoint	MS		SCI	
	BOTOX 200 Units (N=86)	Placebo (N=88)	BOTOX 200 Units (N=22)	Placebo (N=16)
At any time during complete treatment cycle	31% (n=27)	5% (n=4)	27% (n=6)	19% (n=3)

A placebo-controlled, double-blind post-approval 52 week study with BOTOX 100 Units (Study NDO-3) was conducted in non-catheterizing adult MS patients with urinary incontinence due to detrusor overactivity associated with a neurologic condition. Catheterization for urinary retention was initiated in 15.2% (10/66) of patients following treatment with BOTOX 100 Units versus 2.6% (2/78) on placebo at any time during the complete treatment cycle. The median duration of post-injection catheterization for those who developed urinary retention was 64 days for BOTOX 100 Units and 2 days for placebo.

5.14 Human Albumin and Transmission of Viral Diseases

This product contains albumin, a derivative of human blood. Based on effective donor screening and product manufacturing processes, it carries an extremely remote risk for transmission of viral diseases and variant Creutzfeldt-Jakob disease (vCJD). There is a theoretical risk for transmission of Creutzfeldt-Jakob disease (CJD), but if that risk actually exists, the risk of transmission would also be considered extremely remote. No cases of transmission of viral diseases, CJD or vCJD have ever been identified for licensed albumin or albumin contained in other licensed products.

6 ADVERSE REACTIONS

The following adverse reactions to BOTOX (onabotulinumtoxinA) for injection are discussed in greater detail in other sections of the labeling:

- Spread of Toxin Effects *[see Warnings and Precautions (5.1)]*
- Serious Adverse Reactions with Unapproved Use *[see Warnings and Precautions (5.3)]*
- Hypersensitivity Reactions *[see Contraindications (4) and Warnings and Precautions (5.4)]*
- Increased Risk of Clinically Significant Effects with Pre-Existing Neuromuscular Disorders *[see Warnings and Precautions (5.5)]*
- Dysphagia and Breathing Difficulties *[see Warnings and Precautions (5.6)]*
- Pulmonary Effects of BOTOX in Patients with Compromised Respiratory Status Treated for Spasticity or for Detrusor Overactivity Associated with a Neurologic Condition *[see Warnings and Precautions (5.7)]*
- Corneal Exposure and Ulceration in Patients Treated with BOTOX for Blepharospasm *[see Warnings and Precautions (5.8)]*
- Retrobulbar Hemorrhages in Patients Treated with BOTOX for Strabismus *[see Warnings and Precautions (5.9)]*
- Bronchitis and Upper Respiratory Tract Infections in Patients Treated for Spasticity *[see Warnings and Precautions (5.10)]*
- Autonomic Dysreflexia in Patients Treated for Detrusor Overactivity Associated with a Neurologic Condition *[see Warnings and Precautions (5.11)]*
- Urinary Tract Infections in Patients with Overactive Bladder *[see Warnings and Precautions (5.12)]*
- Urinary Retention in Patients Treated for Bladder Dysfunction *[see Warnings and Precautions (5.13)]*

6.1 Clinical Trials Experience

Because clinical trials are conducted under widely varying conditions, the adverse reaction rates observed in the clinical trials of a drug cannot be directly compared to rates in the clinical trials of another drug and may not reflect the rates observed in clinical practice.

BOTOX and BOTOX Cosmetic contain the same active ingredient in the same formulation, but with different labeled Indications and Usage. Therefore, adverse reactions observed with the use of BOTOX Cosmetic also have the potential to be observed with the use of BOTOX.

In general, adverse reactions occur within the first week following injection of BOTOX and, while generally transient, may have a duration of several months or longer. Localized pain, infection, inflammation, tenderness, swelling, erythema, and/or bleeding/bruising may be associated with the injection. Symptoms associated with flu-like symptoms (e.g., nausea, fever, myalgia) have been reported after treatment. Needle-related pain and/or anxiety may result in vasovagal responses (including syncope, hypotension), which may require appropriate medical therapy.

Local weakness of the injected muscle(s) represents the expected pharmacological action of botulinum toxin. However, weakness of nearby muscles may also occur due to spread of toxin *[see Warnings and Precautions (5.1)]*.

Overactive Bladder

Table 14 presents the most frequently reported adverse reactions in double-blind, placebo-controlled clinical trials for overactive bladder occurring within 12 weeks of the first BOTOX treatment.

Table 14: Adverse Reactions Reported by ≥2% of BOTOX Treated Patients and More Often than in Placebo-Treated Patients Within the First 12 Weeks after Intradetrusor Injection, in Double-Blind, Placebo-Controlled Clinical Trials in Patients with OAB

Adverse Reactions	BOTOX 100 Units (N=552) %	Placebo (N=542) %
Urinary tract infection	18	6
Dysuria	9	7
Urinary retention	6	0
Bacteriuria	4	2
Residual urine volume*	3	0

*Elevated PVR not requiring catheterization. Catheterization was required for PVR ≥350 mL regardless of symptoms, and for PVR ≥200 mL to <350 mL with symptoms (e.g., voiding difficulty).

A higher incidence of urinary tract infection was observed in patients with diabetes mellitus treated with BOTOX 100 Units and placebo than in patients without diabetes, as shown in Table 15.

Table 15: Proportion of Patients Experiencing Urinary Tract Infection Following an Injection in Double-Blind, Placebo-Controlled Clinical Trials in OAB According to History of Diabetes Mellitus

	Patients with Diabetes		Patients without Diabetes	
	BOTOX 100 Units (N=81) %	Placebo (N=69) %	BOTOX 100 Units (N=526) %	Placebo (N=516) %
Urinary tract infection (UTI)	31	12	26	10

The incidence of UTI increased in patients who experienced a maximum post-void residual (PVR) urine volume ≥200 mL following BOTOX injection compared to those with a maximum PVR <200 mL following BOTOX injection, 44% versus 23%, respectively. No change was observed in the overall safety profile with repeat dosing during an open-label, uncontrolled extension trial.

Adult Detrusor Overactivity associated with a Neurologic Condition
Table 16 presents the most frequently reported adverse reactions in the double-blind, placebo-controlled studies within 12 weeks of injection for patients with detrusor overactivity associated with a neurologic condition treated with BOTOX 200 Units.

Table 16: Adverse Reactions Reported by ≥2% of BOTOX-Treated Patients and More Frequent than in Placebo-Treated Patients Within the First 12 Weeks after Intradetrusor Injection in Double-Blind, Placebo-Controlled Clinical Trials

Adverse Reactions	BOTOX 200 Units (N=262) %	Placebo (N=272) %
Urinary tract infection	24	17
Urinary retention	17	3
Hematuria	4	3

The following adverse reactions with BOTOX 200 Units were reported at any time following initial injection and prior to re-injection or study exit (median duration of exposure was 44 weeks): urinary tract infections (49%), urinary retention (17%), constipation (4%), muscular weakness (4%), dysuria (4%), fall (3%), gait disturbance (3%), and muscle spasm (2%).

In the Multiple Sclerosis (MS) patients enrolled in the double-blind, placebo-controlled trials, the MS exacerbation annualized rate (i.e., number of MS exacerbation events per patient-year) was 0.23 for BOTOX and 0.20 for placebo.

No change was observed in the overall safety profile with repeat dosing.

Table 17 presents the most frequently reported adverse reactions in a placebo-controlled, double-blind post-approval 52 week study with BOTOX 100 Units (Study NDO-3) conducted in MS patients with urinary incontinence due to detrusor overactivity associated with a neurologic condition. These patients were not adequately managed with at least one anticholinergic agent and not catheterized at baseline. The table below presents the most frequently reported adverse reactions within 12 weeks of injection.

Table 17: Adverse Reactions Reported in a Post Approval Study (NDO-3) by >2% of BOTOX Treated Patients and More Frequent than in Placebo-Treated Patients Within the First 12 Weeks after Intradetrusor Injection

Adverse Reactions	BOTOX 100 Units (N=66) %	Placebo (N=78) %
Urinary tract infection	26	6
Bacteriuria	9	5
Urinary retention	15	1
Dysuria	5	1
Residual urine volume*	17	1

* Elevated PVR not requiring catheterization. Catheterization was required for PVR ≥350 mL regardless of symptoms, and for PVR ≥200 mL to <350 mL with symptoms (e.g., voiding difficulty).

The following adverse events with BOTOX 100 Units were reported at any time following initial injection and prior to re-injection or study exit (median duration of exposure was 51 weeks): urinary tract infections (39%), bacteriuria (18%), urinary retention (17%), residual urine volume* (17%), dysuria (9%), and hematuria (5%).

No difference in the MS exacerbation annualized rate (i.e., number of MS exacerbating events per patient-year) was observed (BOTOX =0, placebo =0.07).

Pediatric Detrusor Overactivity associated with a Neurologic Condition
Table 18 presents the most frequently reported adverse reactions in Study 191622-120, a double-blind, parallel-group study conducted in pediatric patients with detrusor overactivity associated with a neurologic condition. These patients were not adequately managed with at least one anticholinergic agent and were using clean intermittent catheterization at baseline *[see Clinical Studies (14.3)]*. The table below presents the most frequently reported adverse reactions during the 12 weeks following intradetrusor administration of BOTOX 200 Units.

Table 18: Adverse Reactions Reported by ≥ 3% of BOTOX Treated Pediatric Patients within the First 12 Weeks after Intradetrusor Injection, Study 191622-120

Adverse Reactions	BOTOX 200 Unit (N=30)
Urinary tract infection	2 (7%)
Bacteriuria	6 (20%)
Leukocyturia	2 (7%)
Hematuria	1 (3%)

No change was observed in the overall safety profile with repeat dosing.

The most common adverse reactions in patients who received BOTOX 6 U/kg and less than a total dose of 200 U in Study 191622-120 were urinary tract infection (UTI), bacteriuria and hematuria.

Chronic Migraine
In double-blind, placebo-controlled chronic migraine efficacy trials (Study 1 and Study 2), the discontinuation rate was 12% in the BOTOX treated group and 10% in the placebo-treated group. Discontinuations due to an adverse event were 4% in the BOTOX group and 1% in the placebo group. The most frequent adverse events leading to discontinuation in the BOTOX group were neck pain, headache, worsening migraine, muscular weakness and eyelid ptosis.

The most frequently reported adverse reactions following injection of BOTOX for chronic migraine appear in Table 19.

Table 19: Adverse Reactions Reported by ≥2% of BOTOX Treated Patients and More Frequent than in Placebo-Treated Patients in Two Chronic Migraine Double-Blind, Placebo-Controlled Clinical Trials

Adverse Reactions	BOTOX 155 Units-195 Units (N=687) %	Placebo (N=692) %
Nervous system disorders		
Headache	5	3
Migraine	4	3
Facial paresis	2	0
Eye disorders		
Eyelid ptosis	4	<1
Infections and Infestations		
Bronchitis	3	2
Musculoskeletal and connective tissue disorders		
Neck pain	9	3
Musculoskeletal stiffness	4	1
Muscular weakness	4	<1
Myalgia	3	1
Musculoskeletal pain	3	1
Muscle spasms	2	1
General disorders and administration site conditions		
Injection site pain	3	2
Vascular Disorders		
Hypertension	2	1

Other adverse reactions that occurred more frequently in the BOTOX group compared to the placebo group at a frequency less than 1% and potentially BOTOX related include: vertigo, dry eye, eyelid edema, dysphagia, eye infection, and jaw pain. Severe worsening of migraine requiring hospitalization occurred in approximately 1% of BOTOX treated patients in Study 1 and Study 2, usually within the first week after treatment, compared to 0.3% of placebo-treated patients.

Adult Upper Limb Spasticity

The most frequently reported adverse reactions following injection of BOTOX for adult upper limb spasticity appear in Table 20.

Table 20: Adverse Reactions Reported by ≥2% of BOTOX Treated Patients and More Frequent than in Placebo-Treated Patients in Adult Upper Limb Spasticity Double-Blind, Placebo-Controlled Clinical Trials

Adverse Reactions	BOTOX 251 Units - 360 Units (N=115) %	BOTOX 150 Units - 250 Units (N=188) %	BOTOX <150 Units (N=54) %	Placebo (N=182) %
Gastrointestinal disorder				
Nausea	3	2	2	1
General disorders and administration site conditions				
Fatigue	3	2	2	0
Infections and infestations				
Bronchitis	3	2	0	1
Musculoskeletal and connective tissue disorders				
Pain in extremity	6	5	9	4
Muscular weakness	0	4	2	1

Twenty-two adult patients, enrolled in double-blind placebo controlled studies, received 400 Units or higher of BOTOX for treatment of upper limb spasticity. In addition, 44 adults received 400 Units of BOTOX or higher for four consecutive treatments over approximately one year for treatment of upper limb spasticity. The type and frequency of adverse reactions observed in patients treated with 400 Units of BOTOX were similar to those reported in patients treated for upper limb spasticity with 360 Units of BOTOX.

Adult Lower Limb Spasticity

The most frequently reported adverse reactions following injection of BOTOX for adult lower limb spasticity appear in Table 21. Two hundred thirty-one patients enrolled in a double-blind placebo controlled study (Study 7) received 300 Units to 400 Units of BOTOX, and were compared with 233 patients who received placebo. Patients were followed for an average of 91 days after injection.

Table 21: Adverse Reactions Reported by ≥2% of BOTOX Treated Patients and More Frequent than in Placebo-Treated Patients in Adult Lower Limb Spasticity Double-Blind, Placebo-Controlled Clinical Trial (Study 7)

Adverse Reactions	BOTOX (N=231) %	Placebo (N=233) %
Musculoskeletal and connective tissue disorders		
Arthralgia	3	1
Back pain	3	2
Myalgia	2	1
Infections and infestations		
Upper respiratory tract infection	2	1
General disorders and administration site conditions		
Injection site pain	2	1

Pediatric Upper Limb Spasticity

The most frequently reported adverse reactions following injection of BOTOX in pediatric patients 2 to 17 years of age with upper limb spasticity appear in Table 22. In a double-blind, placebo-controlled trial (Study 1), 78 patients were treated with 3 Units/kg of BOTOX, and 77 patients received 6 Units/kg to a maximum dose of 200 Units of BOTOX, and were compared to 79 patients who received placebo *[see Clinical Studies (14.6)]*. Patients were followed for an average of 91 days after injection.

Table 22: Adverse Reactions Reported by ≥2% of BOTOX 6 Units/kg Treated Patients and More Frequent than in Placebo-Treated Patients in Pediatric Upper Limb Spasticity Double-Blind, Placebo-Controlled Clinical Trial (Study 1)

Adverse Reactions	BOTOX 6 Units/kg (N=77) %	BOTOX 3 Units/kg (N=78) %	Placebo (N=79) %
Infections and infestations			
Upper respiratory tract infection*	17	10	9
General disorders and administration site conditions			
Injection site pain	4	3	1
Gastrointestinal disorders			
Nausea	4	0	0
Constipation	3	0	1
Respiratory, thoracic and mediastinal disorders			
Rhinorrhea	4	0	1
Nasal congestion	3	0	1
Nervous system disorders			
Seizure**	5	1	0

*Includes upper respiratory tract infection and viral upper respiratory tract infection

**Includes seizure and partial seizure

Pediatric Lower Limb Spasticity

The most frequently reported adverse reactions following injection of BOTOX in pediatric patients 2 to 17 years of age with lower limb spasticity appear in Table 23. In a double-blind, placebo-controlled trial (Study 2), 126 patients were treated with 4 Units/kg of BOTOX, and 128 patients received 8 Units/kg to a maximum dose of 300 Units of BOTOX, and were compared to 128 patients who received placebo *[see Clinical Studies (14.6)]*. Patients were followed for an average of 89 days after injection.

Table 23: Adverse Reactions Reported by ≥2% of BOTOX 8 Units/kg Treated Patients and More Frequent than in Placebo-Treated Patients in Pediatric Lower Limb Spasticity Double-Blind, Placebo-Controlled Clinical Trial (Study 2)

Adverse Reactions	BOTOX 8 Units/kg (N=128) %	BOTOX 4 Units/kg (N=126) %	Placebo (N=128) %
General disorders and administration site conditions			
Injection site erythema	2	0	0
Injection site pain	2	2	0
Respiratory, thoracic and mediastinal disorders			
Oropharyngeal pain	2	0	1
Injury, poisoning and procedural complications			
Ligament sprain	2	1	0
Skin abrasion	2	0	0
Metabolism and nutrition disorders			
Decreased appetite	2	0	0

Cervical Dystonia
In cervical dystonia patients evaluated for safety in double-blind and open-label studies following injection of BOTOX, the most frequently reported adverse reactions were dysphagia (19%), upper respiratory infection (12%), neck pain (11%), and headache (11%).

Other events reported in 2-10% of patients in any one study in decreasing order of incidence include: increased cough, flu syndrome, back pain, rhinitis, dizziness, hypertonia, soreness at injection site, asthenia, oral dryness, speech disorder, fever, nausea, and drowsiness. Stiffness, numbness, diplopia, ptosis, and dyspnea have been reported.

Dysphagia and symptomatic general weakness may be attributable to an extension of the pharmacology of BOTOX resulting from the spread of the toxin outside the injected muscles *[see Warnings and Precautions (5.1, 5.6)]*.

The most common severe adverse reaction associated with the use of BOTOX injection in patients with cervical dystonia is dysphagia with about 20% of these cases also reporting dyspnea *[see Warnings and Precautions (5.1, 5.6)]*. Most dysphagia is reported as mild or moderate in severity. However, it may be associated with more severe signs and symptoms *[see Warnings and Precautions (5.6)]*.

Additionally, reports in the literature include a case of a female patient who developed brachial plexopathy two days after injection of 120 Units of BOTOX for the treatment of cervical dystonia, and reports of dysphonia in patients who have been treated for cervical dystonia.

Primary Axillary Hyperhidrosis
The most frequently reported adverse reactions (3-10% of adult patients) following injection of BOTOX in double-blind studies included injection site pain and hemorrhage, non-axillary sweating, infection, pharyngitis, flu syndrome, headache, fever, neck or back pain, pruritus, and anxiety.

The data reflect 346 patients exposed to BOTOX 50 Units and 110 patients exposed to BOTOX 75 Units in each axilla.

Blepharospasm
In a study of blepharospasm patients who received an average dose per eye of 33 Units (injected at 3 to 5 sites) of the currently manufactured BOTOX, the most frequently reported adverse reactions were ptosis (21%), superficial punctate keratitis (6%), and eye dryness (6%).

Other events reported in prior clinical studies in decreasing order of incidence include: irritation, tearing, lagophthalmos, photophobia, ectropion, keratitis, diplopia, entropion, diffuse skin rash, and local swelling of the eyelid skin lasting for several days following eyelid injection.

In two cases of VII nerve disorder, reduced blinking from BOTOX injection of the orbicularis muscle led to serious corneal exposure, persistent epithelial defect, corneal ulceration and a case of corneal perforation. Focal facial paralysis, syncope, and exacerbation of myasthenia gravis have also been reported after treatment of blepharospasm.

Strabismus
Extraocular muscles adjacent to the injection site can be affected, causing vertical deviation, especially with higher doses of BOTOX. The incidence rates of these adverse effects in 2058 adults who received a total of 3650 injections for horizontal strabismus was 17%.

The incidence of ptosis has been reported to be dependent on the location of the injected muscles, 1% after inferior rectus injections, 16% after horizontal rectus injections and 38% after superior rectus injections.

In a series of 5587 injections, retrobulbar hemorrhage occurred in 0.3% of cases.

6.2 Immunogenicity
As with all therapeutic proteins, there is a potential for immunogenicity. The detection of antibody formation is highly dependent on the sensitivity and specificity of the assay. Additionally, the observed incidence of antibody (including neutralizing antibody) positivity in an assay may be influenced by several factors including assay methodology, sample handling, timing of sample collection, concomitant medications, and underlying disease. For these reasons, comparison of the incidence of antibodies to onabotulinumtoxinA in the studies described below with the incidence of antibodies in other studies or to other products may be misleading.

In a long term, open-label study evaluating 326 cervical dystonia patients treated for an average of 9 treatment sessions with the current formulation of BOTOX, 4 (1.2%) patients had positive antibody tests. All 4 of these patients responded to BOTOX therapy at the time of the positive antibody test. However, 3 of these patients developed clinical resistance after subsequent treatment, while the fourth patient continued to respond to BOTOX therapy for the remainder of the study.

One patient among the 445 hyperhidrosis patients (0.2%), two patients among the 380 adult upper limb spasticity patients (0.5%), and no patients among 406 migraine patients with analyzed specimens developed the presence of neutralizing antibodies.

In one Phase 3 study and the open-label extension study in patients with pediatric lower limb spasticity, neutralizing antibodies developed in 2 of 264 patients (0.8%) treated with BOTOX for up to 5 treatment cycles. Both patients continued to experience clinical benefit following subsequent BOTOX treatments.

In overactive bladder patients with analyzed specimens from the two phase 3 studies and the open-label extension study, neutralizing antibodies developed in 0 of 954 patients (0.0%) while receiving BOTOX 100 Unit doses and 3 of 260 patients (1.2%) after subsequently receiving at least one 150 Unit dose. Response to subsequent BOTOX treatment was not different following seroconversion in these three patients.

In detrusor overactivity associated with neurologic condition patients with analyzable specimens in the adult drug development program (including the open-label extension study), neutralizing antibodies developed in 3 of 300 patients (1.0%) after receiving only BOTOX 200 Unit doses and 5 of 258 patients (1.9%) after receiving at least one 300 Unit dose. Following development of neutralizing antibodies in these 8 patients, 4 continued to experience clinical benefit, 2 did not experience clinical benefit, and the effect on the response to BOTOX in the remaining 2 patients is not known. In 99 pediatric patients who had a negative baseline result for binding antibodies or neutralizing antibodies and had at least one evaluable post-baseline value from one randomized double-blind study and one double-blind extension study, no patients developed neutralizing antibodies after receiving 50 Units to 200 Units of BOTOX.

The data reflect the patients whose test results were considered positive for neutralizing activity to BOTOX in a mouse protection assay or negative based on a screening ELISA assay or mouse protection assay.

Formation of neutralizing antibodies to botulinum toxin type A may reduce the effectiveness of BOTOX treatment by inactivating the biological activity of the toxin. The critical factors for neutralizing antibody formation have not been well characterized. The results from some studies suggest that BOTOX injections at more frequent intervals or at higher doses may lead to greater incidence of antibody formation. The potential for antibody formation may be minimized by injecting with the lowest effective dose given at the longest feasible intervals between injections.

6.3 Postmarketing Experience
The following adverse reactions have been identified during post-approval use of BOTOX. Because these reactions are reported voluntarily from a population of uncertain size, it is not always possible to reliably estimate their frequency or establish a causal relationship to drug exposure. These reactions include: abdominal pain; alopecia, including madarosis; anorexia; brachial plexopathy; denervation/muscle atrophy; diarrhea; dry eye; eyelid edema (following periocular injection); hyperhidrosis; hypoacusis; hypoaesthesia; localized muscle twitching; malaise; Mephisto sign; paresthesia; peripheral neuropathy; radiculopathy; erythema multiforme, dermatitis psoriasiform, and psoriasiform eruption; strabismus; tinnitus; and visual disturbances.

There have been spontaneous reports of death, sometimes associated with dysphagia, pneumonia, and/or other significant debility or anaphylaxis, after treatment with botulinum toxin *[see Warnings and Precautions (5.4, 5.6)]*.

There have also been reports of adverse events involving the cardiovascular system, including arrhythmia and myocardial infarction, some with fatal outcomes. Some of these patients had risk factors including cardiovascular disease. The exact relationship of these events to the botulinum toxin injection has not been established.

New onset or recurrent seizures have also been reported, typically in patients who are predisposed to experiencing these events. The exact relationship of these events to the botulinum toxin injection has not been established.

7 DRUG INTERACTIONS

7.1 Aminoglycosides and Other Agents Interfering with Neuromuscular Transmission

Co-administration of BOTOX and aminoglycosides or other agents interfering with neuromuscular transmission (e.g., curare-like compounds) should only be performed with caution as the effect of the toxin may be potentiated.

7.2 Anticholinergic Drugs

Use of anticholinergic drugs after administration of BOTOX may potentiate systemic anticholinergic effects.

7.3 Other Botulinum Neurotoxin Products

The effect of administering different botulinum neurotoxin products at the same time or within several months of each other is unknown. Excessive neuromuscular weakness may be exacerbated by administration of another botulinum toxin prior to the resolution of the effects of a previously administered botulinum toxin.

7.4 Muscle Relaxants

Excessive weakness may also be exaggerated by administration of a muscle relaxant before or after administration of BOTOX.

8 USE IN SPECIFIC POPULATIONS

8.1 Pregnancy

Risk Summary

There are no studies or adequate data from postmarketing surveillance on the developmental risk associated with use of BOTOX in pregnant women. In animal studies, administration of BOTOX during pregnancy resulted in adverse effects on fetal growth (decreased fetal weight and skeletal ossification) at clinically relevant doses, which were associated with maternal toxicity *[see Data]*.

In the U.S. general population, the estimated background risk of major birth defects and miscarriages in clinically recognized pregnancies is 2-4% and 15-20%, respectively. The background risk of major birth defects and miscarriage for the indicated populations is unknown.

Data

Animal Data

When BOTOX (4, 8, or 16 Units/kg) was administered intramuscularly to pregnant mice or rats two times during the period of organogenesis (on gestation days 5 and 13), reductions in fetal body weight and decreased fetal skeletal ossification were observed at the two highest doses. The no-effect dose for developmental toxicity in these studies (4 Units/kg) is approximately equal to the human dose of 400 Units, on a body weight basis (Units/kg).

When BOTOX was administered intramuscularly to pregnant rats (0.125, 0.25, 0.5, 1, 4, or 8 Units/kg) or rabbits (0.063, 0.125, 0.25, or 0.5 Units/kg) daily during the period of organogenesis (total of 12 doses in rats, 13 doses in rabbits), reduced fetal body weights and decreased fetal skeletal ossification were observed at the two highest doses in rats and at the highest dose in rabbits. These doses were also associated with significant maternal toxicity, including abortions, early deliveries, and maternal death. The developmental no-effect doses in these studies of 1 Unit/kg in rats and 0.25 Units/kg in rabbits are less than the human dose of 400 Units, based on Units/kg.

When pregnant rats received single intramuscular injections (1, 4, or 16 Units/kg) at three different periods of development (prior to implantation, implantation, or organogenesis), no adverse effects on fetal development were observed. The developmental no-effect level for a single maternal dose in rats (16 Units/kg) is approximately 2 times the human dose of 400 Units, based on Units/kg.

8.2 Lactation

Risk Summary

There are no data on the presence of BOTOX in human or animal milk, the effects on the breastfed infant, or the effects on milk production. The developmental and health benefits of breastfeeding should be considered along with the mother's clinical need for BOTOX and any potential adverse effects on the breastfed infant from BOTOX or from the underlying maternal conditions.

8.4 Pediatric Use

Detrusor Overactivity associated with a Neurologic Condition

The safety and effectiveness of BOTOX for detrusor overactivity associated with a neurologic condition have been established in pediatric patients 5 years of age and older who have an inadequate response to or are intolerant of anticholinergic medication. Use of BOTOX in this patient population is based on the results of a randomized, double-blind, parallel group trial in 113 pediatric patients 5 to 17 years of age (inclusive) with detrusor overactivity associated with a neurologic condition (Study 191622-120) and a long-term, multicenter, double-blind, long-term extension trial (Study 191622-121) *[see Clinical Studies (14.3)].* The most common adverse reactions in this population were urinary tract infection, bacteriuria, hematuria, and leukocyturia *[see Adverse Reactions (6.1)]*.
The safety and effectiveness of BOTOX have not been established in patients with NDO younger than 5 years of age.

Overactive Bladder

The safety and effectiveness of BOTOX for the treatment of overactive bladder have not been established in pediatric patients.

Efficacy was not demonstrated in a multicenter, randomized, double-blind, parallel-group, multiple-dose clinical study which was conducted to evaluate the efficacy and safety of BOTOX in pediatric patients aged 12 to 17 years with overactive bladder. Fifty-five patients who had an inadequate response to or were intolerant of at least one anticholinergic medication were treated with BOTOX. There was not a statistically significant difference in the mean change from baseline in the daily average frequency of daytime urinary incontinence episodes (primary efficacy endpoint) at week 12 post-treatment when a medium and high dose were each compared to a low dose of BOTOX. The adverse reactions in pediatric patients treated with BOTOX were comparable with the known safety profile in adults with overactive bladder.

Prophylaxis of Headaches in Chronic Migraine

Safety and effectiveness in patients below the age of 18 years have not been established.

In a 12-week, multicenter, double-blind, placebo-controlled clinical trial, 123 adolescent patients (ages 12 to below 18 years) with chronic migraine were randomized to receive BOTOX 74 Units, BOTOX 155 Units, or placebo, for one injection cycle. This trial did not establish the efficacy of BOTOX, compared with placebo, for the prophylaxis of headaches in adolescents with chronic migraine.

Spasticity

Safety and effectiveness have been established in pediatric patients 2 to 17 years of age *[see Warnings and Precautions (5.1), Adverse Reactions (6.1), and Clinical Studies (14.6)]*. The safety and effectiveness of BOTOX have been established by evidence from adequate and well-controlled studies of BOTOX in patients 2 to 17 years of age with upper and lower limb spasticity.

Safety and effectiveness in pediatric patients below the age of 2 years have not been established *[see Boxed Warning and Warnings and Precautions (5.1)].*

Axillary Hyperhidrosis

Safety and effectiveness in patients below the age of 18 years have not been established.

Cervical Dystonia

Safety and effectiveness in pediatric patients below the age of 16 years have not been established.

Blepharospasm and Strabismus

Safety and effectiveness in pediatric patients below the age of 12 years have not been established.

Juvenile Animal Data

In a study in which juvenile rats received intramuscular injection of BOTOX (0, 8, 16, or 24 Units/kg) every other week from postnatal day 21 for 12 weeks, changes in bone size/geometry associated with decreased bone density and bone mass were observed at all doses, in association with limb disuse, decreased muscle contraction, and decreased body weight gain. Impairment of fertility and male reproductive organ histopathology (degeneration of seminiferous tubules of the testis) were observed at the mid and high doses. Bone and male reproductive organ effects showed evidence of reversibility after dosing cessation. The no-effect dose for adverse developmental effects in juvenile animals (8 Units/kg) is similar to the human dose (400 Units) on a body weight (kg) basis.

8.5 Geriatric Use

Of the 2145 adult patients in placebo-controlled clinical studies of BOTOX for the treatment of spasticity, 33.5% were 65 or older, and 7.7% were 75 years of age or older. No overall differences in safety were observed between elderly patients and adult patients younger than 65 years of age.

In clinical studies of BOTOX across other indications, no overall differences in safety were observed between elderly patients and younger adult patients, with the exception of Overactive Bladder (see below). Other reported clinical experience has not identified differences in responses between the elderly and younger adult patients, but greater sensitivity of some older individuals cannot be ruled out.

Overactive Bladder

Of 1242 overactive bladder patients in placebo-controlled clinical studies of BOTOX, 41.4% were 65 years of age or older, and 14.7% were 75 years of age or older. Adverse reactions of UTI and urinary retention were more common in patients 65 years of age or older in both placebo and BOTOX groups compared to younger patients (see Table 24). Otherwise, there were no overall differences in the safety profile following BOTOX treatment between patients aged 65 years and older compared to adult patients younger than 65 years of age in these studies.

Table 24: Incidence of Urinary Tract Infection and Urinary Retention according to Age Group during First Placebo-Controlled Treatment, Placebo-Controlled Clinical Trials in Patients with OAB

	<65 Years		65 to 74 Years		≧75 Years	
Adverse Reactions	**BOTOX 100 Units (N=344) %**	**Placebo (N=348) %**	**BOTOX 100 Units (N=169) %**	**Placebo (N=151) %**	**BOTOX 100 Units (N=94) %**	**Placebo (N=86) %**
Urinary tract infection	21	7	30	13	38	19
Urinary retention	6	0.6	8	0	9	1

Observed effectiveness was comparable between these age groups in placebo-controlled clinical studies.

10 OVERDOSAGE

Excessive doses of BOTOX (onabotulinumtoxinA) for injection may be expected to produce neuromuscular weakness with a variety of symptoms.

Symptoms of overdose are likely not to be present immediately following injection. Should accidental injection or oral ingestion occur or overdose be suspected, the person should be medically supervised for several weeks for signs and symptoms of systemic muscular weakness which could be local, or distant from the site of injection *[see Boxed Warning and Warnings and Precautions (5.1, 5.6)]*. These patients should be considered for further medical evaluation and appropriate medical therapy immediately instituted, which may include hospitalization.

If the musculature of the oropharynx and esophagus are affected, aspiration may occur which may lead to development of aspiration pneumonia. If the respiratory muscles become paralyzed or sufficiently weakened, intubation and assisted respiration may be necessary until recovery takes place. Supportive care could involve the need for a tracheostomy and/or prolonged mechanical ventilation, in addition to other general supportive care.

In the event of overdose, antitoxin raised against botulinum toxin is available from the Centers for Disease Control and Prevention (CDC) in Atlanta, GA. However, the antitoxin will not reverse any botulinum toxin-induced effects already apparent by the time of antitoxin administration. In the event of suspected or actual cases of botulinum toxin poisoning, please contact your local or state Health Department to process a request for antitoxin through the CDC. If you do not receive a response within 30 minutes, please contact the CDC directly at 1-770-488-7100. More information can be obtained at http://www.cdc.gov/mmwr/preview/mmwrhtml/mm5232a8.htm.

11 DESCRIPTION

OnabotulinumtoxinA is a sterile, vacuum-dried purified botulinum toxin type A, produced from fermentation of Hall strain *Clostridium botulinum* type A, and intended for intramuscular, intradetrusor and intradermal use. It is purified from the culture solution by dialysis and a series of acid precipitations to a complex consisting of the neurotoxin, and several accessory proteins. The complex is dissolved in sterile sodium chloride solution containing Albumin Human and is sterile filtered (0.2 microns) prior to filling and vacuum-drying.

The primary release procedure for BOTOX uses a cell-based potency assay to determine the potency relative to a reference standard. The assay is specific to AbbVie's products BOTOX and BOTOX Cosmetic. One Unit of BOTOX corresponds to the calculated median intraperitoneal lethal dose (LD_{50}) in mice. Due to specific details of this assay such as the vehicle, dilution scheme, and laboratory protocols, Units of biological activity of BOTOX cannot be compared to nor converted into Units of any other botulinum toxin or any toxin assessed with any other specific assay method. The specific activity of BOTOX is approximately 20 Units/nanogram of neurotoxin protein complex.

Each vial of BOTOX (onabotulinumtoxinA) for injection contains either 100 Units of Clostridium botulinum type A neurotoxin complex, 0.5 mg of Albumin Human, and 0.9 mg of sodium chloride; or 200 Units of Clostridium botulinum type A neurotoxin complex, 1 mg of Albumin Human, and 1.8 mg of sodium chloride in a sterile, vacuum-dried form without a preservative.

12 CLINICAL PHARMACOLOGY

12.1 Mechanism of Action

BOTOX blocks neuromuscular transmission by binding to acceptor sites on motor or autonomic nerve terminals, entering the nerve terminals, and inhibiting the release of acetylcholine. This inhibition occurs as the neurotoxin cleaves SNAP-25, a protein integral to the successful docking and release of acetylcholine from vesicles situated within nerve endings. When injected intramuscularly at therapeutic doses, BOTOX produces partial chemical denervation of the muscle resulting in a localized reduction in muscle activity. In addition, the muscle may atrophy, axonal sprouting may occur, and extrajunctional acetylcholine receptors may develop. There is evidence that reinnervation of the muscle may occur, thus slowly reversing muscle denervation produced by BOTOX.

When injected intradermally, BOTOX produces temporary chemical denervation of the sweat gland resulting in local reduction in sweating.

Following intradetrusor injection, BOTOX affects the efferent pathways of detrusor activity via inhibition of acetylcholine release.

12.3 Pharmacokinetics

Using currently available analytical technology, it is not possible to detect BOTOX in the peripheral blood following intramuscular injection at the recommended doses.

13 NONCLINICAL TOXICOLOGY

13.1 Carcinogenesis, Mutagenesis, Impairment of Fertility

Carcinogenesis

Long term studies in animals have not been performed to evaluate the carcinogenic potential of BOTOX.

Mutagenesis

BOTOX was negative in a battery of in vitro (microbial reverse mutation assay, mammalian cell mutation assay, and chromosomal aberration assay) and in vivo (micronucleus assay) genetic toxicology assays.

Impairment of Fertility

In fertility studies of BOTOX (4, 8, or 16 Units/kg) in which either male or female rats were injected intramuscularly prior to mating and on the day of mating (3 doses, 2 weeks apart for males: 2 doses, 2 weeks apart for females) to untreated animals, reduced fertility was observed in males at the intermediate and high doses and in females at the high dose. The no-effect doses for reproductive toxicity (4 Units/kg in males, 8 Units/kg in females) are approximately equal to the human dose of 400 Units, on a body weight basis (Units/kg).

13.2 Animal Toxicology and/or Pharmacology

In a study to evaluate inadvertent peribladder administration, bladder stones were observed in 1 of 4 male monkeys that were injected with a total of 6.8 Units/kg divided into the prostatic urethra and proximal rectum (single administration). No bladder stones were observed in male or female monkeys following injection of up to 36 Units/kg (~12X the highest human bladder dose) directly to the bladder as either single or 4 repeat dose injections or in female rats for single injections up to 100 Units/kg (~33X the highest human bladder dose [200 Units], based on Units/kg).

14 CLINICAL STUDIES

14.1 Overactive Bladder (OAB)

Two double-blind, placebo-controlled, randomized, multi-center, 24-week clinical studies were conducted in patients with OAB with symptoms of urge urinary incontinence, urgency, and frequency (Studies OAB-1 and OAB-2). Patients needed to have at least 3 urinary urgency incontinence episodes and at least 24 micturitions in 3 days to enter the studies. A total of 1105 patients, whose symptoms had not been adequately managed with anticholinergic therapy (inadequate response or intolerable side effects), were randomized to receive either 100 Units of BOTOX (n=557), or placebo (n=548). Patients received 20 injections of study drug (5 Units of BOTOX or placebo) spaced approximately 1 cm apart into the detrusor muscle.

In both studies, significant improvements compared to placebo in the primary efficacy variable of change from baseline in daily frequency of urinary incontinence episodes were observed for BOTOX 100 Units at the primary time point of week 12. Significant improvements compared to placebo were also observed for the secondary efficacy variables of daily frequency of micturition episodes and volume voided per micturition. These primary and secondary variables are shown in Table 25 and Table 26, and Figure 7 and Figure 8.

Table 25: Baseline and Change from Baseline in Urinary Incontinence Episode Frequency, Micturition Episode Frequency and Volume Voided Per Micturition, Study OAB-1

	BOTOX 100 Units (N=278)	Placebo (N=272)	Treatment Difference	p-value
Daily Frequency of Urinary Incontinence Episodes[a]				
Mean Baseline	5.5	5.1		
Mean Change[*] at Week 2	-2.6	-1.0	-1.6	
Mean Change[*] at Week 6	-2.8	-1.0	-1.8	
Mean Change[*] at Week 12[**]	-2.5	-0.9	-1.6 (-2.1, -1.2)	<0.001
Daily Frequency of Micturition Episodes[b]				
Mean Baseline	12.0	11.2		
Mean Change[†] at Week 12[**]	-1.9	-0.9	-1.0 (-1.5, -0.6)	<0.001
Volume Voided per Micturition[b] (mL)				
Mean Baseline	156	161		
Mean Change[†] at Week 12[**]	38	8	30 (17, 43)	<0.001

[*] Least squares (LS) mean change, treatment difference and p-value are based on an ANCOVA model with baseline value as covariate and treatment group and investigator as factors. Last observation carried forward (LOCF) values were used to analyze the primary efficacy variable.

[†] LS mean change, treatment difference and p-value are based on an ANCOVA model with baseline value as covariate and stratification factor, treatment group and investigator as factors.

[**] Primary timepoint

[a] Primary variable

[b] Secondary variable

Table 26: Baseline and Change from Baseline in Urinary Incontinence Episode Frequency, Micturition Episode Frequency and Volume Voided Per Micturition, Study OAB-2

	BOTOX 100 Units (N=275)	Placebo (N=269)	Treatment Difference	p-value
Daily Frequency of Urinary Incontinence Episodes[a]				
Mean Baseline	5.5	5.7		
Mean Change[*] at Week 2	-2.7	-1.1	-1.6	
Mean Change[*] at Week 6	-3.1	-1.3	-1.8	
Mean Change[*] at Week 12[**]	-3.0	-1.1	-1.9 (-2.5, -1.4)	<0.001
Daily Frequency of Micturition Episodes[b]				
Mean Baseline	12.0	11.8		
Mean Change[†] at Week 12[**]	-2.3	-0.6	-1.7 (-2.2, -1.3)	<0.001
Volume Voided per Micturition[b] (mL)				
Mean Baseline	144	153		
Mean Change[†] at Week 12[**]	40	10	31 (20, 41)	<0.001

[*] LS mean change, treatment difference and p-value are based on an ANCOVA model with baseline value as covariate and treatment group and investigator as factors. LOCF values were used to analyze the primary efficacy variable.

[†] LS mean change, treatment difference and p-value are based on an ANCOVA model with baseline value as covariate and stratification factor, treatment group and investigator as factors.

[**] Primary timepoint

[a] Primary variable

[b] Secondary variable

Figure 7: Mean Change from Baseline in Daily Frequency of Urinary Incontinence Episodes Following Intradetrusor Injection in Study OAB-1

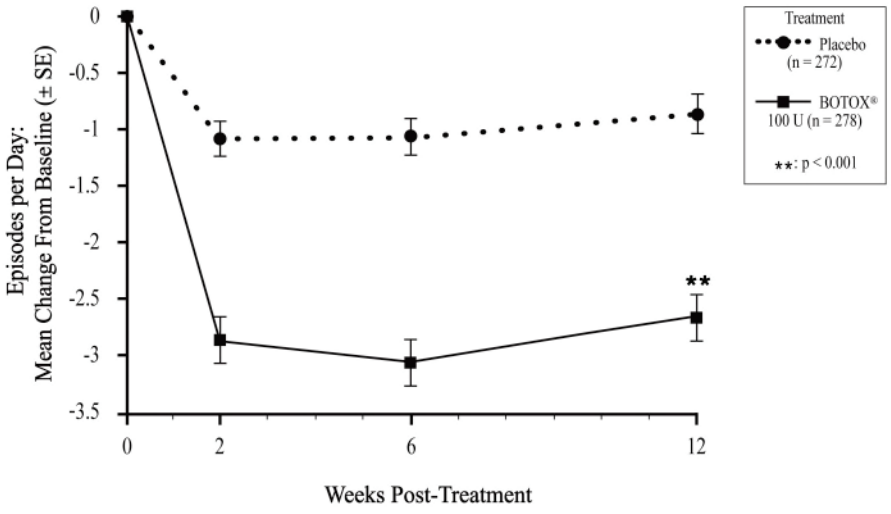
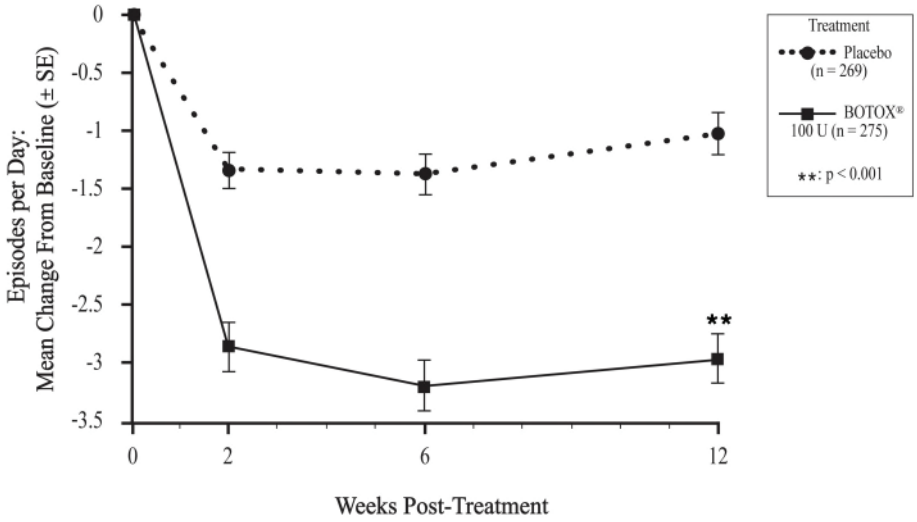


Figure 8: Mean Change from Baseline in Daily Frequency of Urinary Incontinence Episodes Following Intradetrusor Injection in Study OAB-2



The median duration of response in Study OAB-1 and OAB-2, based on patient qualification for re-treatment, was 19-24 weeks for the BOTOX 100 Unit dose group compared to 13 weeks for placebo. To qualify for re-treatment, at least 12 weeks must have passed since the prior treatment, post-void residual urine volume must have been less than 200 mL and patients must have reported at least 2 urinary incontinence episodes over 3 days.

14.2 Adult Detrusor Overactivity Associated with a Neurologic Condition

Two double-blind, placebo-controlled, randomized, multi-center clinical studies were conducted in patients with urinary incontinence due to detrusor overactivity associated with a neurologic condition who were either spontaneously voiding or using catheterization (Studies NDO-1 and NDO-2). A total of 691 spinal cord injury (T1 or below) or multiple sclerosis patients, who had an inadequate response to or were intolerant of at least one anticholinergic medication, were enrolled. These patients were randomized to receive either 200 Units of BOTOX (n=227), 300 Units of BOTOX (n=223), or placebo (n=241).

In both studies, significant improvements compared to placebo in the primary efficacy variable of change from baseline in weekly frequency of incontinence episodes were observed for BOTOX (200 Units) at the primary efficacy time point at week 6. Increases in maximum cystometric capacity and reductions in maximum detrusor pressure during the first involuntary detrusor contraction were also observed. These primary and secondary endpoints are shown in Table 27 and Table 28, and Figure 9 and Figure 10.

No additional benefit of BOTOX 300 Units over 200 Units was demonstrated.

Table 27: Baseline and Change from Baseline in Weekly Urinary Incontinence Episode Frequency, Maximum Cystometric Capacity and Maximum Detrusor Pressure during First Involuntary Detrusor Contraction (cmH_2O) Study NDO-1

	BOTOX 200 Units	Placebo	Treatment Difference*	p-value*
Weekly Frequency of Urinary Incontinence Episodes[a]				
N	134	146		
Mean Baseline	32.3	28.3		
Mean Change* at Week 2	-15.3	-10.0	-5.3	—
Mean Change* at Week 6**	-19.9	-10.6	-9.2 (-13.1, -5.3)	p<0.001
Mean Change* at Week 12	-19.8	-8.8	-11.0	—
Maximum Cystometric Capacity[b] (mL)				
N	123	129		
Mean Baseline	253.8	259.1		
Mean Change* at Week 6**	135.9	12.1	123.9 (89.1, 158.7)	p<0.001
Maximum Detrusor Pressure during First Involuntary Detrusor Contraction[b] (cmH_2O)				
N	41	103		
Mean Baseline	63.1	57.4		
Mean Change* at Week 6**	-28.1	-3.7	-24.4	—

* LS mean change, treatment difference and p-value are based on an analysis using an ANCOVA model with baseline weekly endpoint as covariate and treatment group, etiology at study entry (spinal cord injury or multiple sclerosis), concurrent anticholinergic therapy at screening, and investigator as factors. LOCF values were used to analyze the primary efficacy variable.
** Primary timepoint
[a] Primary endpoint
[b] Secondary endpoint

Table 28: Baseline and Change from Baseline in Weekly Urinary Incontinence Episode Frequency, Maximum Cystometric Capacity and Maximum Detrusor Pressure during First Involuntary Detrusor Contraction (cmH_2O) in Study NDO-2

	BOTOX 200 Units	Placebo	Treatment Difference*	p-value*
Weekly Frequency of Urinary Incontinence Episodes[a]				
N	91	91		
Mean Baseline	32.7	36.8		
Mean Change* at Week 2	-18.0	-7.9	-10.1	—
Mean Change* at Week 6**	-19.6	-10.8	-8.8 (-14.5, -3.0)	p=0.003
Mean Change* at Week 12	-19.6	-10.7	-8.9	—
Maximum Cystometric Capacity[b] (mL)				
N	88	85		
Mean Baseline	239.6	253.8		
Mean Change* at Week 6**	150.8	2.8	148.0 (101.8, 194.2)	p<0.001
Maximum Detrusor Pressure during First Involuntary Detrusor Contraction[b] (cmH_2O)				
N	29	68		
Mean Baseline	65.6	43.7		
Mean Change* at Week 6**	-28.7	2.1	-30.7	—

* LS mean change, treatment difference and p-value are based on an analysis using an ANCOVA model with baseline weekly endpoint as covariate and treatment group, etiology at study entry (spinal cord injury or multiple sclerosis), concurrent anticholinergic therapy at screening, and investigator as factors. LOCF values were used to analyze the primary efficacy variable.
** Primary timepoint
[a] Primary endpoint
[b] Secondary endpoint

Figure 9: Mean Change from Baseline in Weekly Frequency of Urinary Incontinence Episodes During Treatment Cycle 1 in Study NDO-1

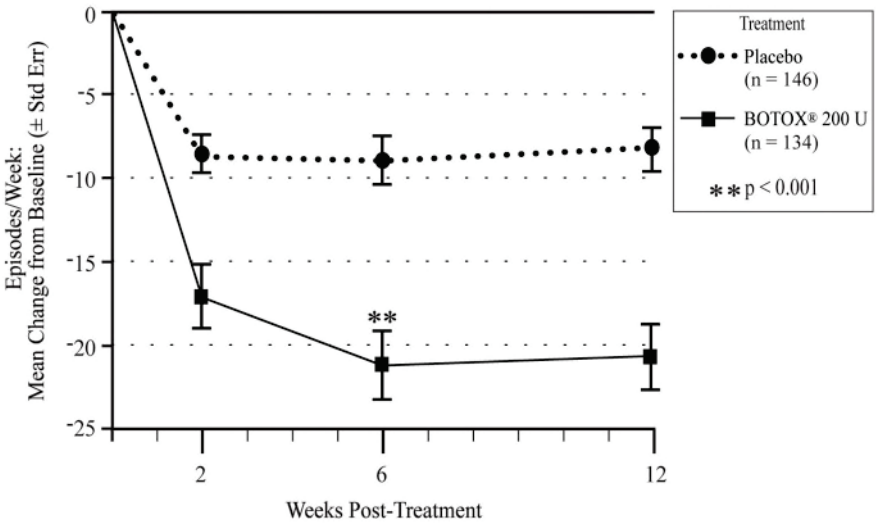
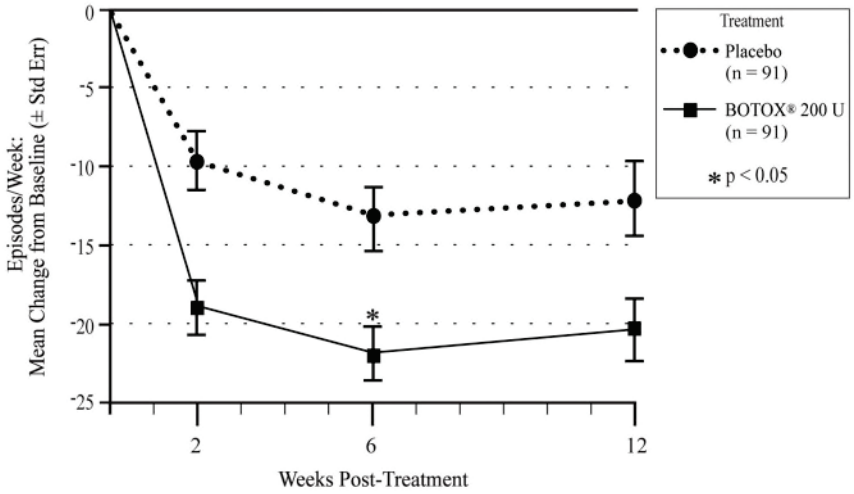


Figure 10: Mean Change from Baseline in Weekly Frequency of Urinary Incontinence Episodes During Treatment Cycle 1 in Study NDO-2



The median duration of response in study NDO-1 and NDO-2, based on patient qualification for re-treatment was 295-337 days (42-48 weeks) for the 200 Units dose group compared to 96-127 days (13-18 weeks) for placebo. Re-treatment was based on loss of effect on incontinence episode frequency (50% of effect in Study NDO-1; 70% of effect in Study NDO-2).

A placebo-controlled, double-blind randomized post-approval 52 week study (Study NDO-3) was conducted in MS patients with urinary incontinence due to neurogenic detrusor overactivity who were not adequately managed with at least one anticholinergic agent and not catheterizing at baseline. These patients were randomized to receive either 100 Units of BOTOX (n=66) or placebo (n=78).

Significant improvements compared to placebo in the primary efficacy variable of change from baseline in daily frequency of incontinence episodes were observed for BOTOX (100 Units) at the primary efficacy time point at week 6. Increases in maximum cystometric capacity and reductions in maximum detrusor pressure during the first involuntary detrusor contraction were also observed. These primary and secondary endpoints are shown in Table 29.

Table 29: Baseline and Change from Baseline in Daily Urinary Incontinence Episode Frequency, Maximum Cystometric Capacity and Maximum Detrusor Pressure during First Involuntary Detrusor Contraction (cmH_2O) in Study NDO-3

	BOTOX 100 Units	Placebo	Treatment Difference*	p-value*
Daily Frequency of Urinary Incontinence Episodes[a]				
N	66	78		
Mean Baseline	4.2	4.3		
Mean Change* at Week 2	-2.9	-1.2	-1.7	—
Mean Change* at Week 6**	-3.4	-1.1	-2.3 (-3.0, -1.7)	p<0.001
Mean Change* at Week 12	-2.7	-1.0	-1.8	—
Maximum Cystometric Capacity[b] (mL)				
N	62	72		
Mean Baseline	248.9	245.5		
Mean Change* at Week 6**	134.4	3.5	130.9 (94.8, 167.0)	p<0.001
Maximum Detrusor Pressure during First Involuntary Detrusor Contraction[b] (cmH_2O)				
N	25	51		
Mean Baseline	42.4	39.0		
Mean Change* at Week 6**	-19.2	2.7	-21.9 (-37.5, -6.3)	

* LS mean change, treatment difference and p-value are based on an analysis using an ANCOVA model with baseline daily endpoint as covariate and treatment group and propensity score stratification as factors. LOCF values were used to analyze the primary efficacy variable.
** Primary timepoint
[a] Primary endpoint
[b] Secondary endpoint

The median duration of response in study NDO-3, based on patient qualification for re-treatment was 362 days (52 weeks) for the BOTOX 100 Units dose group compared to 88 days (13 weeks) for placebo. To qualify for re-treatment, at least 12 weeks must have passed since the prior treatment, post-void residual urine volume must have been less than 200 mL and patients must have reported at least 2 urinary incontinence episodes over 3 days with no more than 1 incontinence-free day.

14.3 Pediatric Detrusor Overactivity Associated with a Neurologic Condition

Study 191622-120 (NCT01852045) was a multicenter, randomized, double-blind, parallel-group clinical study conducted in patients 5 to 17 years of age with urinary incontinence due to detrusor overactivity associated with a neurologic condition and using clean intermittent catheterization. A total of 113 patients (including 99 with spinal dysraphism such as spina bifida, 13 with spinal cord injury and 1 with transverse myelitis) who had an inadequate response to or were intolerant of at least one anticholinergic medication were enrolled. The median age was 11 years (range: 5 to 17 years), 49% were female; 75% were White, 10% were Black. These patients were randomized to 50 Units, 100 Units or 200 Units, not to exceed 6 Units/kg body weight. Patients receiving less than the randomized dose due to the 6 Units/kg maximum, were assigned to the nearest dose group for analysis. The sample size for BOTOX 50 Units, 100 Units, and 200 Units were 38, 45 and 30, respectively. Prior to treatment administration, patients received anesthesia based on age and local site practice. One hundred and nine patients (97.3%) received general anesthesia or conscious sedation and 3 patients (2.7%) received local anesthesia.

The study results demonstrated within group improvements in the primary efficacy variable of change from baseline in daytime urinary incontinence episodes (normalized to 12 hours) at the primary efficacy time point (Week 6) for all 3 BOTOX treatment groups. Additional benefits were seen with BOTOX 200 Units for measures related to reducing maximum bladder pressure when compared to 50 Units. The decrease in maximum detrusor pressure (MDP) during the storage phase (MDP defined as the highest value in the Pdet channel during the storage phase [e.g., the greater of the following: the maximum Pdet during the highest amplitude IDC, the maximum Pdet during a terminal detrusor contraction, the Pdet at the end of filling, or the highest Pdet at any other time during the storage phase]) for BOTOX 200 Units at Week 6 was greater than the decrease observed for 50 Units. Within group improvements for the primary and secondary endpoints for the 200 Units dose group are shown in Table 30.

The efficacy of BOTOX 6 U/kg for pediatric patients with NDO weighing less than 34 kg was comparable to that of BOTOX 200 U.

Table 30: Baseline and Change from Baseline in Daily Daytime Frequency of Urinary Incontinence Episodes, Urine Volume at First Morning Catheterization, Maximum Detrusor Pressure during the Storage Phase (cmH_2O), and Maximum Cystometric Capacity (mL) in Study191622-120

	BOTOX 200 U N=30
Daily average frequency of daytime urinary incontinence episodes[a]	
Mean Baseline	3.7
Mean Change* at Week 2 (95% CI)	-1.1 (-1.7, -0.6)
Mean Change* at Week 6** (95% CI)	-1.3 (-1.8, -0.9)
Mean Change* at Week 12 (95% CI)	-0.9 (-1.5, -0.4)
Urine Volume at First Morning Catheterization (mL)[b]	
Mean Baseline	187.7
Mean Change* at Week 2 (95% CI)	63.2 (27.9, 98.6)
Mean Change* at Week 6** (95% CI)	87.5 (52.1, 122.8)
Mean Change* at Week 12 (95% CI)	45.2 (10.0, 80.5)
Maximum Detrusor Pressure (PdetMax) During the Storage Phase (cm H2O)[b]	
Mean Baseline	56.7
Mean Change* at Week 6** (95% CI)	-27.3 (-36.4, -18.2)
Maximum Cystometric Capacity (mL) (MCC)[b]	
Mean Baseline	202.3
Mean Change* at Week 6** (95% CI)	63.6 (29.0, 98.1)

CI = Confidence Interval
* LSmean change and 95% CI are based on an ANCOVA model with baseline value as covariate and treatment group, age (< 12 years or >= 12 years), baseline daytime urinary incontinence episodes (<= 6 or > 6) and anticholinergic therapy (yes/no) at baseline as factors.
** Primary timepoint
[a] Primary endpoint
[b] Secondary endpoint

The median duration of response in this study, based on patient qualification for re-treatment was 207 days (30 weeks) for BOTOX 200 Units dose group. To qualify for re-treatment, patients must have reported at least 2 urinary incontinence episodes over 2 days and at least 12 weeks have passed from the prior bladder injection.

14.4 Chronic Migraine

BOTOX was evaluated in two randomized, multi-center, 24-week, 2 injection cycle, placebo-controlled double-blind studies. Study 1 and Study 2 included chronic migraine adults who were not using any concurrent headache prophylaxis, and during a 28-day baseline period had ≥15 headache days lasting 4 hours or more, with ≥50% being migraine/probable migraine. In both studies, patients were randomized to receive placebo or 155 Units to 195 Units BOTOX injections every 12 weeks for the 2-cycle, double-blind phase. Patients were allowed to use acute headache treatments during the study. BOTOX treatment demonstrated statistically significant and clinically meaningful improvements from baseline compared to placebo for key efficacy variables (see Table 31).

Table 31: Week 24 Key Efficacy Variables for Study 1 and Study 2

Efficacy per 28 days	Study 1		Study 2	
	BOTOX (N=341)	Placebo (N=338)	BOTOX (N=347)	Placebo (N=358)
Change from baseline in frequency of headache days	-7.8*	-6.4	-9.2*	-6.9
Change from baseline in total cumulative hours of headache on headache days	-107*	-70	-134*	-95

* Significantly different from placebo (p≤0.05)

Patients treated with BOTOX had a significantly greater mean decrease from baseline in the frequency of headache days at most timepoints from Week 4 to Week 24 in Study 1 (Figure 11), and all timepoints from Week 4 to Week 24 in Study 2 (Figure 12), compared to placebo-treated patients.

Figure 11: Mean Change from Baseline in Number of Headache Days for Study 1

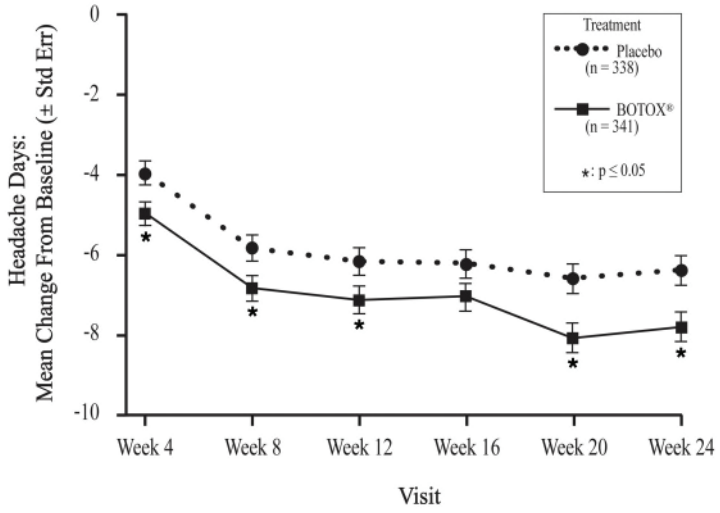
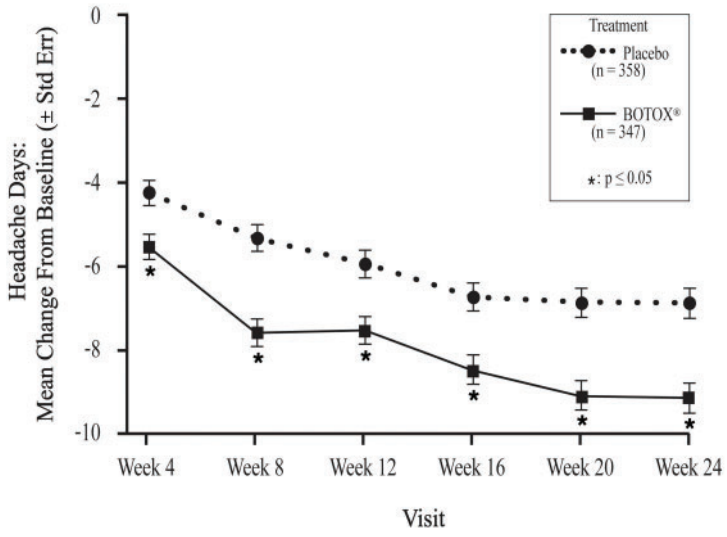


Figure 12: Mean Change from Baseline in Number of Headache Days for Study 2



14.5 Adult Spasticity

Adult Upper Limb Spasticity

The efficacy of BOTOX for the treatment of adult upper limb spasticity was evaluated in several randomized, multi-center, double-blind, placebo-controlled studies (Studies 1 through 6).

Study 1 included 126 adult patients (64 BOTOX and 62 placebo) with upper limb spasticity (Ashworth score of at least 3 for wrist flexor tone and at least 2 for finger flexor tone) who were at least 6 months post-stroke. BOTOX (a total dose of 200 Units to 240 Units) and placebo were injected intramuscularly (IM) into the flexor digitorum profundus, flexor digitorum sublimis, flexor carpi radialis, flexor carpi ulnaris, and if necessary into the adductor pollicis and flexor pollicis longus (see Table 32). Use of an EMG/nerve stimulator was recommended to assist in proper muscle localization for injection. Patients were followed for 12 weeks.

Table 32: BOTOX Dose and Injection Sites in Study 1

Muscles Injected	Volume (mL)	BOTOX (Units)	Number of Injection Sites
Wrist			
Flexor Carpi Radialis	1	50	1
Flexor Carpi Ulnaris	1	50	1
Finger			
Flexor Digitorum Profundus	1	50	1
Flexor Digitorum Sublimis	1	50	1
Thumb			
Adductor Pollicis[a]	0.4	20	1
Flexor Pollicis Longus[a]	0.4	20	1

[a] Injected only if spasticity is present in this muscle

The primary efficacy variable was wrist flexors muscle tone at week 6, as measured by the Ashworth score. The Ashworth Scale is a 5-point scale with grades of 0 [no increase in muscle tone] to 4 [limb rigid in flexion or extension]. It is a clinical measure of the force required to move an extremity around a joint, with a reduction in score clinically representing a reduction in the force needed to move a joint (i.e., improvement in spasticity).

Key secondary endpoints included Physician Global Assessment, finger flexors muscle tone, and thumb flexors tone at Week 6. The Physician Global Assessment evaluated the response to treatment in terms of how the patient was doing in his/her life using a scale from -4 = very marked worsening to +4 = very marked improvement. Study 1 results on the primary endpoint and the key secondary endpoints are shown in Table 33.

Table 33: Primary and Key Secondary Endpoints by Muscle Group at Week 6 in Study 1

	BOTOX (N=64)	Placebo (N=62)
Median Change from Baseline in Wrist Flexor Muscle Tone on the Ashworth Scale†[a]	-2.0*	0.0
Median Change from Baseline in Finger Flexor Muscle Tone on the Ashworth Scale††[b]	-1.0*	0.0
Median Change from Baseline in Thumb Flexor Muscle Tone on the Ashworth Scale††[c]	-1.0	-1.0
Median Physician Global Assessment of Response to Treatment††	2.0*	0.0

† Primary endpoint at Week 6
†† Secondary endpoints at Week 6
* Significantly different from placebo ($p \leq 0.05$)
[a] BOTOX injected into both the flexor carpi radialis and ulnaris muscles
[b] BOTOX injected into the flexor digitorum profundus and flexor digitorum sublimis muscles
[c] BOTOX injected into the adductor pollicis and flexor pollicis longus muscles

Study 2 compared 3 doses of BOTOX with placebo and included 91 adult patients [BOTOX 360 Units (N=21), BOTOX 180 Units (N=23), BOTOX 90 Units (N=21), and placebo (N=26)] with upper limb spasticity (expanded Ashworth score of at least 2 for elbow flexor tone and at least 3 for wrist flexor tone) who were at least 6 weeks post-stroke. BOTOX and placebo were injected with EMG guidance into the flexor digitorum profundus, flexor digitorum sublimis, flexor carpi radialis, flexor carpi ulnaris, and biceps brachii (see Table 34).

Table 34: BOTOX Dose and Injection Sites in Study 2 and Study 3

Muscles Injected	Total Dose			Volume (mL) per site	Injection Sites (n)
	BOTOX low dose (90 Units)	BOTOX mid dose (180 Units)	BOTOX high dose (360 Units)		
Wrist					
Flexor Carpi Ulnaris	10 Units	20 Units	40 Units	0.4	1
Flexor Carpi Radialis	15 Units	30 Units	60 Units	0.6	1
Finger					
Flexor Digitorum Profundus	7.5 Units	15 Units	30 Units	0.3	1
Flexor Digitorum Sublimis	7.5 Units	15 Units	30 Units	0.3	1
Elbow					
Biceps Brachii	50 Units	100 Units	200 Units	0.5	4

The primary efficacy variable in Study 2 was the wrist flexor tone at Week 6 as measured by the expanded Ashworth Scale. The expanded Ashworth Scale uses the same scoring system as the Ashworth Scale, but allows for half-point increments.

Key secondary endpoints in Study 2 included Physician Global Assessment, finger flexors muscle tone, and elbow flexors muscle tone at Week 6. Study 2 results on the primary endpoint and the key secondary endpoints at Week 6 are shown in Table 35.

Table 35: Primary and Key Secondary Endpoints by Muscle Group and BOTOX Dose at Week 6 in Study 2

	BOTOX low dose (90 Units) (N=21)	BOTOX mid dose (180 Units) (N=23)	BOTOX high dose (360 Units) (N=21)	Placebo (N=26)
Median Change from Baseline in Wrist Flexor Muscle Tone on the Ashworth Scale†[b]	-1.5*	-1.0*	-1.5*	-1.0
Median Change from Baseline in Finger Flexor Muscle Tone on the Ashworth Scale††[c]	-0.5	-0.5	-1.0	-0.5
Median Change from Baseline in Elbow Flexor Muscle Tone on the Ashworth Scale††[d]	-0.5	-1.0*	-0.5[a]	-0.5
Median Physician Global Assessment of Response to Treatment	1.0*	1.0*	1.0*	0.0

† Primary endpoint at Week 6
†† Secondary endpoints at Week 6
* Significantly different from placebo ($p \leq 0.05$)
[a] $p=0.053$
[b] Total dose of BOTOX injected into both the flexor carpi radialis and ulnaris muscles
[c] Total dose of BOTOX injected into the flexor digitorum profundus and flexor digitorum sublimis muscles
[d] Dose of BOTOX injected into biceps brachii muscle

Study 3 compared 3 doses of BOTOX with placebo and enrolled 88 adult patients [BOTOX 360 Units (N=23), BOTOX 180 Units (N=23), BOTOX 90 Units (N=23), and placebo (N=19)] with upper limb spasticity (expanded Ashworth score of at least 2 for elbow flexor tone and at least 3 for wrist flexor tone and/or finger flexor tone) who were at least 6 weeks post-stroke. BOTOX and placebo were injected with EMG guidance into the flexor digitorum profundus, flexor digitorum sublimis, flexor carpi radialis, flexor carpi ulnaris, and biceps brachii (see Table 34).

The primary efficacy variable in Study 3 was wrist and elbow flexor tone as measured by the expanded Ashworth score. A key secondary endpoint was assessment of finger flexors muscle tone. Study 3 results on the primary endpoint at Week 4 are shown in Table 36.

Table 36: Primary and Key Secondary Endpoints by Muscle Group and BOTOX Dose at Week 4 in Study 3

	BOTOX low dose (90 Units) (N=23)	BOTOX mid dose (180 Units) (N=21)	BOTOX high dose (360 Units) (N=22)	Placebo (N=19)
Median Change from Baseline in Wrist Flexor Muscle Tone on the Ashworth Scale†b	-1.0	-1.0	-1.5*	-0.5
Median Change from Baseline in Finger Flexor Muscle Tone on the Ashworth Scale††c	-1.0	-1.0	-1.0*	-0.5
Median Change from Baseline in Elbow Flexor Muscle Tone on the Ashworth Scale†d	-0.5	-0.5	-1.0*	-0.5

† Primary endpoint at Week 4
†† Secondary endpoints at Week 4
* Significantly different from placebo ($p \leq 0.05$)
[b] Total dose of BOTOX injected into both the flexor carpi radialis and ulnaris muscles
[c] Total dose of BOTOX injected into the flexor digitorum profundus and flexor digitorum sublimis muscles
[d] Dose of BOTOX injected into biceps brachii muscle

Study 4 (NCT01153815) included 170 adult patients (87 BOTOX and 83 placebo) with upper limb spasticity who were at least 6 months post-stroke. In Study 4, patients received 20 Units of BOTOX into the adductor pollicis and flexor pollicis longus (total BOTOX dose = 40 Units in thumb muscles) or placebo (see Table 37). Study 5 (NCT00460564) included 109 patients with upper limb spasticity who were at least 6 months post-stroke. In Study 5, adult patients received 15 Units (low dose) or 20 Units (high dose) of BOTOX into the adductor pollicis and flexor pollicis longus under EMG guidance (total BOTOX low dose = 30 Units, total BOTOX high dose = 40 Units), or placebo (see Table 37). The duration of follow-up in Study 4 and Study 5 was 12 weeks.

Table 37: BOTOX Dose and Injection Sites in Studies 4 and 5

Muscles Injected	Study 4		Study 5				Number of Injection Sites for Studies 4 and 5
	BOTOX (Units)	Volume (mL)	BOTOX low dose (Units)	BOTOX high dose (Units)	Volume low dose (mL)	Volume high dose (mL)	
Thumb							
Adductor Pollicis	20	0.4	15	20	0.3	0.4	1
Flexor Pollicis Longus	20	0.4	15	20	0.3	0.4	1

The results of Study 4 for the change from Baseline to Week 6 in thumb flexor tone measured by modified Ashworth Scale (MAS) and overall treatment response by Physician Global Assessment at week 6 are presented in Table 38. The MAS uses a similar scoring system as the Ashworth Scale.

Table 38: Efficacy Endpoints for Thumb Flexors at Week 6 in Study 4

	BOTOX (N=66)	Placebo (N=57)
Median Change from Baseline in Thumb Flexor Muscle Tone on the modified Ashworth Scale††a	-1.0*	0.0
Median Physician Global Assessment of Response to Treatment††	2.0*	0.0

†† Secondary endpoints at Week 6
* Significantly different from placebo ($p \leq 0.001$)
[a] BOTOX injected into the adductor pollicis and flexor pollicis longus muscles

In Study 5, the results of the change from Baseline to Week 6 in thumb flexor tone measured by modified Ashworth Scale and Clinical Global Impression (CGI) of functional assessment scale assessed by the physician using an 11-point Numeric Rating Scale [-5 worst possible function to +5 best possible function] are presented in Table 39.

Table 39: Efficacy Endpoints for Thumb Flexors at Week 6 in Study 5

	BOTOX low dose (30 Units) (N=14)	Placebo low dose (N=9)	BOTOX high dose (40 Units) (N=43)	Placebo high dose (N=23)
Median Change from Baseline in Thumb Flexor Muscle Tone on the modified Ashworth Scale†††[a]	-1.0	-1.0	-0.5*	0.0
Median Change from Baseline in Clinical Global Impression Score by Physician ††	1.0	0.0	2.0*	0.0

†† Secondary endpoint at Week 6
††† Other endpoint at Week 6
* Significantly different from placebo ($p \leq 0.010$)
[a] BOTOX injected into the adductor pollicis and flexor pollicis longus muscles

Study 6 (NCT03261167) enrolled 124 post-stroke adult patients with upper limb spasticity. In Study 6, 61 patients received 160 Units BOTOX divided among 3 elbow flexors (biceps brachii, brachioradialis, and brachialis) and 63 patients received placebo (see Table 40). EMG, nerve stimulation, or ultrasound techniques were recommended to assist in proper muscle localization for injections. The duration of follow-up was 12 weeks.

Table 40: BOTOX Dose and Injection Sites in Study 6

Muscles Injected	BOTOX 160 U (Units)	Volume (mL)	Number of Injection Sites
Elbow Biceps Brachii	70	1.4	2
Brachioradialis	45	0.9	1
Brachialis	45	0.9	1

The change from baseline in elbow flexor tone measured by modified Ashworth Scale at Week 6 is presented in Table 41.

Table 41: Primary Efficacy Endpoint Results for Elbow Flexors at Week 6 in Study 6

	BOTOX 160 U (N=61)	Placebo (N=63)
Mean Change from Baseline in Elbow Flexor Muscle Tone on the modified Ashworth Scale at Week 6	-1.09*	-0.71

*nominal p value <0.05

Adult Lower Limb Spasticity
The efficacy and safety of BOTOX for the treatment of adult lower limb spasticity was evaluated in Study 7, a randomized, multi-center, double-blind, placebo-controlled study. Study 7 included 468 post-stroke adult patients (233 BOTOX and 235 placebo) with ankle spasticity (modified Ashworth Scale ankle score of at least 3) who were at least 3 months post-stroke. A total dose of 300 Units of BOTOX or placebo were injected intramuscularly and divided between the gastrocnemius, soleus, and tibialis posterior, with optional injection into the flexor hallucis longus, flexor digitorum longus, flexor digitorum brevis, extensor hallucis, and rectus femoris (see Table 42) with up to an additional 100 Units (400 Units total dose). The use of electromyographic guidance or nerve stimulation was required to assist in proper muscle localization for injections. Patients were followed for 12 weeks.

Table 42: BOTOX Dose and Injection Sites in Study 7

Muscles Injected	BOTOX (Units)	Number of Injection Sites
Mandatory Ankle Muscles Gastrocnemius (medial head)	75	3
Gastrocnemius (lateral head)	75	3
Soleus	75	3
Tibialis Posterior	75	3
Optional Muscles Flexor Hallucis Longus	50	2
Flexor Digitorum Longus	50	2
Flexor Digitorum Brevis	25	1
Extensor Hallucis	25	1
Rectus Femoris	100	4

The co-primary endpoints were the average of the change from baseline in modified Ashworth Scale (MAS) ankle score at Week 4 and Week 6, and the average of the Physician Global Assessment of Response (CGI) at Week 4 and Week 6. The CGI evaluated the response to treatment in terms of how the patient was doing in his/her life using a 9-point scale from -4=very marked worsening to +4=very marked improvement.

Statistically significant between-group differences for BOTOX over placebo were demonstrated for the co-primary efficacy measures of MAS and CGI (see Table 43).

Table 43: Co-Primary Efficacy Endpoints Results in Study 7 (Intent-To-Treat Population)

	BOTOX 300 to 400 Units (N=233)	Placebo (N=235)
Mean Change from Baseline in Ankle Plantar Flexors on the modified Ashworth Scale		
Week 4 and 6 Average	-0.8*	-0.6
Mean Clinical Global Impression Score by Investigator		
Week 4 and 6 Average	0.9*	0.7

* Significantly different from placebo ($p<0.05$)

Compared to placebo, significant improvements in MAS change from baseline for ankle plantar flexors (see Figure 13) and CGI (see Figure 14) were observed at Week 2, Week 4, and Week 6 for patients treated with BOTOX.

Figure 13: Modified Ashworth Scale Ankle Score for Study 7 – Mean Change from Baseline by Visit

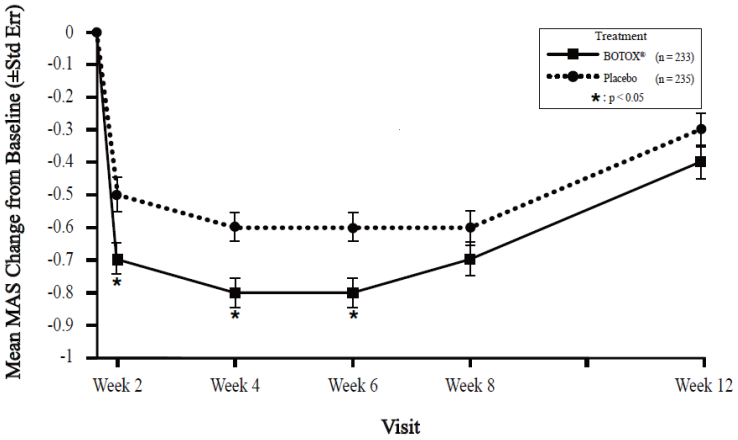
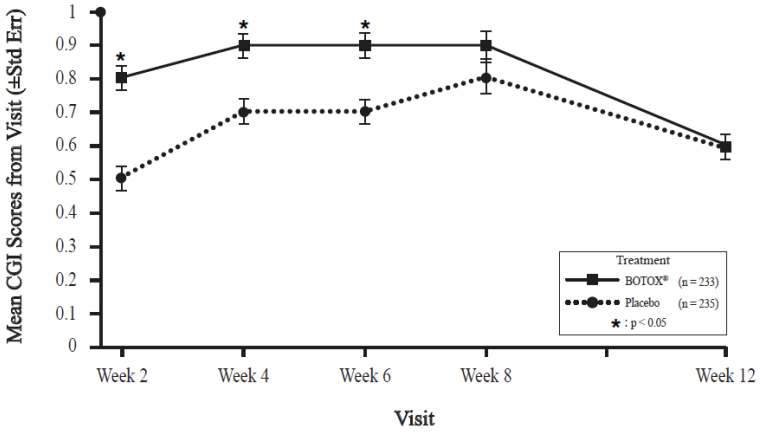


Figure 14: Clinical Global Impression by Physician for Study 7 – Mean Scores by Visit



14.6 Pediatric Spasticity

Pediatric Upper Limb Spasticity

The efficacy and safety of BOTOX for the treatment of upper limb spasticity in pediatric patients 2 to 17 years of age was evaluated in Study 1 (NCT01603602), a randomized, multi-center, double-blind, placebo-controlled study. Study 1 included 234 pediatric patients (78 BOTOX 3 Units/kg, 77 BOTOX 6 Units/kg, and 79 placebo) with upper limb spasticity (modified Ashworth Scale elbow or wrist score of at least 2) because of cerebral palsy or stroke. A total dose of 3 Units/kg BOTOX (maximum 100 Units), 6 Units/kg BOTOX (maximum 200 Units), or placebo was injected intramuscularly and divided between the elbow or wrist and finger muscles (see Table 44). Electromyographic guidance, nerve stimulation, or ultrasound techniques were used to assist in muscle localization for injections. Patients were followed for 12 weeks after injection.

Table 44: BOTOX Dose and Injection Sites in Study 1

Muscles Injected	BOTOX 3 Units/kg* (maximum Units per muscle)	BOTOX 6 Units/kg** (maximum Units per muscle)	Number of Injection Sites
Elbow Flexor Muscles			
Biceps	1.5 Units/kg (50 Units)	3 Units/kg (100 Units)	4
Brachialis	1 Unit/kg (30 Units)	2 Units/kg (60 Units)	2
Brachioradialis	0.5 Units/kg (20 Units)	1 Unit/kg (40 Units)	2
Wrist and Finger Muscles			
Flexor carpi radialis	1 Unit/kg (25 Units)	2 Units/kg (50 Units)	2
Flexor carpi ulnaris	1 Unit/kg (25 Units)	2 Units/kg (50 Units)	2
Flexor digitorum profundus	0.5 Units/kg (25 Units)	1 Unit/kg (50 Units)	2
Flexor digitorum sublimis	0.5 Units/kg (25 Units)	1 Unit/kg (50 Units)	2

* Did not exceed a total dose of 100 Units
** Did not exceed a total dose of 200 Units

The co-primary endpoints were the average of the change from baseline in modified Ashworth Scale (MAS) principal muscle group score (elbow or wrist) at Week 4 and Week 6, and the average of the Clinical Global Impression of Overall Change by Physician (CGI) at Week 4 and Week 6. The CGI evaluated the response to treatment in terms of how the patient was doing in his/her life using a 9-point scale (-4=very marked worsening to +4=very marked improvement).

Compared to placebo, significant improvements in MAS change from baseline were observed at all timepoints for BOTOX-treated patients (see Table 45, Figure 15 and Figure 16). Although CGI scores numerically favored BOTOX over placebo, the difference was not statistically significant.

Table 45: Co-Primary Efficacy Endpoints Results in Study 1 (Pediatric Upper Limb Spasticity, Modified Intent-To-Treat Population)

	BOTOX 3 Units/kg (N=78)	BOTOX 6 Units/kg (N=77)	Placebo (N=79)
Mean Change from Baseline in Principal Muscle Group (Elbow or Wrist) on the modified Ashworth Scale			
Week 4 and 6 Average	-1.92*	-1.87*	-1.21
Mean Clinical Global Impression Score			
Week 4 and 6 Average	1.88	1.87	1.66

*Nominal p value <0.05

Figure 15: Modified Ashworth Scale Score for Study 1 (Pediatric Upper Limb Spasticity, Modified Intent-To-Treat Population) – Mean Change from Baseline by Visit

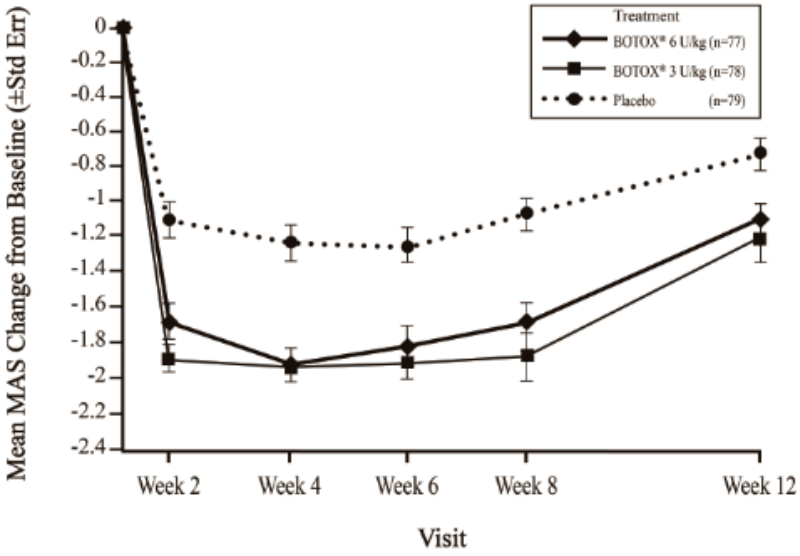
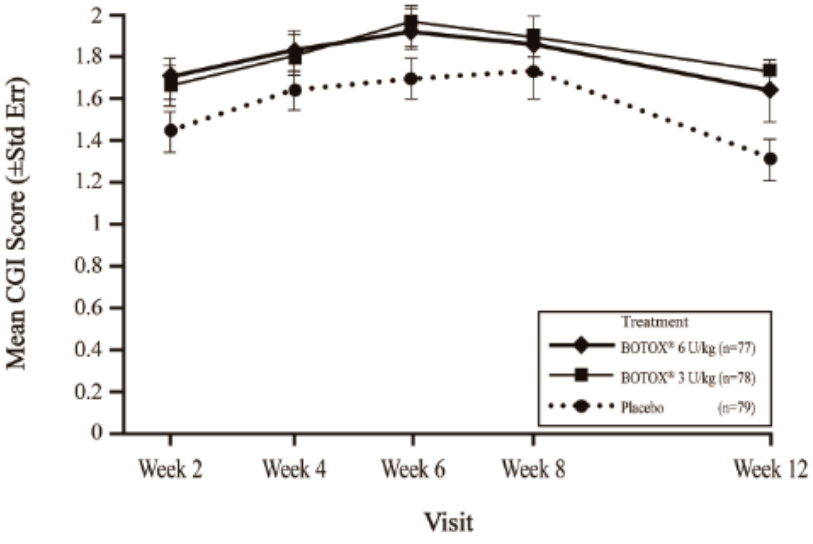


Figure 16: Clinical Global Impression of Overall Change for Study 1 (Pediatric Upper Limb Spasticity, Modified Intent-To-Treat Population) – Mean Scores by Visit



Pediatric Lower Limb Spasticity

The efficacy and safety of BOTOX for the treatment of lower limb spasticity in pediatric patients 2 to 17 years of age was evaluated in Study 2 (NCT01603628), a randomized, multi-center, double-blind, placebo-controlled study. Study 2 included 381 pediatric patients (125 BOTOX 4 Units/kg, 127 BOTOX 8 Units/kg, and 129 placebo) with lower limb spasticity (modified Ashworth Scale ankle score of at least 2) because of cerebral palsy. A total dose of 4 Units/kg BOTOX (maximum 150 Units), 8 Units/kg BOTOX (maximum 300 Units), or placebo was injected intramuscularly and divided between the gastrocnemius, soleus, and tibialis posterior (see Table 46). Electromyographic guidance, nerve stimulation, or ultrasound techniques were used to assist in muscle localization for injections. Patients were followed for 12 weeks after injection.

Table 46: BOTOX Dose and Injection Sites in Study 2

Muscles Injected	BOTOX 4 Units/kg* (maximum Units per muscle)	BOTOX 8 Units/kg** (maximum Units per muscle)	Number of Injection Sites
Mandatory Ankle Muscles Gastrocnemius medial head	1 Unit/kg (37.5 Units)	2 Units/kg (75 Units)	2
Gastrocnemius lateral head	1 Unit/kg (37.5 Units)	2 Units/kg (75 Units)	2
Soleus	1 Unit/kg (37.5 Units)	2 Units/kg (75 Units)	2
Tibialis Posterior	1 Unit/kg (37.5 Units)	2 Units/kg (75 Units)	2

* did not exceed a total dose of 150 Units
** did not exceed a total dose of 300 Units

The co-primary endpoints were the average of the change from baseline in modified Ashworth Scale (MAS) ankle score at Week 4 and Week 6, and the average of the Clinical Global Impression of Overall Change by Physician (CGI) at Week 4 and Week 6. The CGI evaluated the response to treatment in terms of how the patient was doing in his/her life using a 9-point scale (-4=very marked worsening to +4=very marked improvement).

Statistically significant differences between BOTOX and placebo were demonstrated for the MAS and CGI for the 8 Units/kg dose only (see Table 47).

Table 47: Co-Primary Efficacy Endpoints Results in Study 2 (Pediatric Lower Limb Spasticity, Modified Intent-To-Treat Population)

	BOTOX 4 Units/kg (N = 125)	BOTOX 8 Units/kg (N=127)	Placebo (N=129)
Mean Change from Baseline in Plantar Flexors on the modified Ashworth Scale			
Week 4 and 6 Average	-1.01**	-1.06*	-0.80
Mean Clinical Global Impression Score			
Week 4 and 6 Average	1.49	1.65*	1.36

* Significantly different from placebo ($p<0.05$)
** Nominal p value <0.05

Compared to placebo, improvements in mean change from baseline for the MAS, and mean CGI score for lower limb spasticity were observed at timepoints up to Week 12 for BOTOX-treated patients (see Figure 17 and Figure 18).

Figure 17: Modified Ashworth Scale Ankle Score for Study 2 (Pediatric Lower Limb Spasticity, Modified Intent-To-Treat Population) – Mean Change from Baseline by Visit

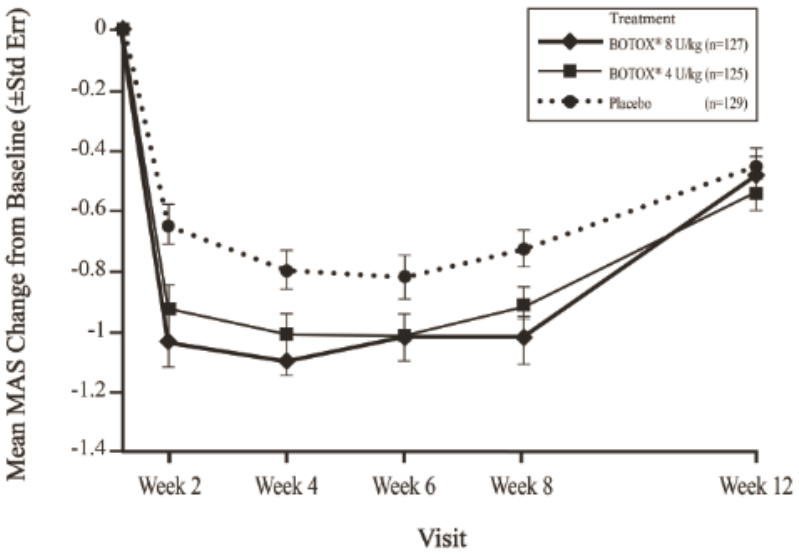
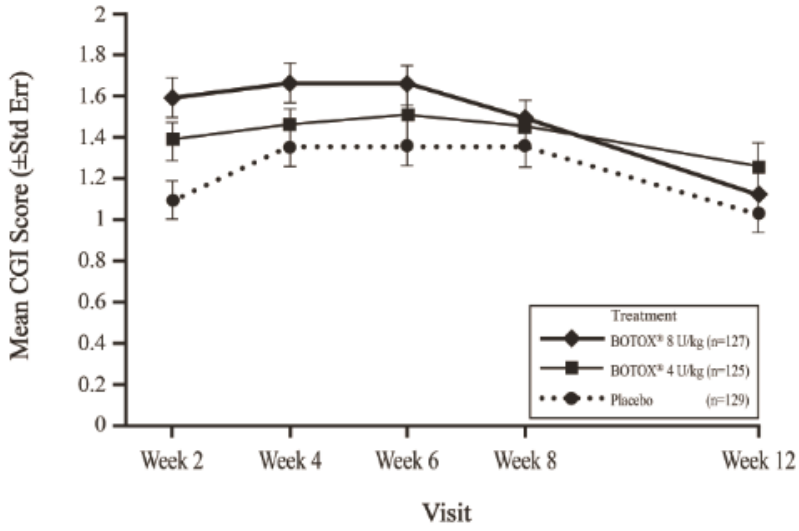


Figure 18: Clinical Global Impression of Overall Change for Study 2 (Pediatric Lower Limb Spasticity, Modified Intent-To-Treat Population) – Mean Scores by Visit



14.7 Cervical Dystonia

A randomized, multi-center, double-blind, placebo-controlled study of the treatment of cervical dystonia was conducted. This study enrolled adult patients with cervical dystonia and a history of having received BOTOX in an open label manner with perceived good response and tolerable side effects. Patients were excluded if they had previously received surgical or other denervation treatment for their symptoms or had a known history of neuromuscular disorder. Subjects participated in an open label enrichment period where they received their previously employed dose of BOTOX. Only patients who were again perceived as showing a response were advanced to the randomized evaluation period. The muscles in which the blinded study agent injections were to be administered were determined on an individual patient basis.

There were 214 subjects evaluated for the open label period, of which 170 progressed into the randomized, blinded treatment period (88 in the BOTOX group, 82 in the placebo group). Patient evaluations continued for at least 10 weeks post-injection. The primary outcome for the study was a dual endpoint, requiring evidence of both a change in the Cervical Dystonia Severity Scale (CDSS) and an increase in the percentage of patients showing any improvement on the Physician Global Assessment Scale at 6 weeks after the injection session. The CDSS quantifies the severity of abnormal head positioning and was newly devised for this study. CDSS allots 1 point for each 5 degrees (or part thereof) of head deviation in each of the three planes of head movement (range of scores up to theoretical maximum of 54). The Physician Global Assessment Scale is a 9 category scale scoring the physician's evaluation of the patients' status compared to baseline, ranging from –4 to +4 (very marked worsening to complete improvement), with 0 indicating no

change from baseline and +1 slight improvement. Pain is also an important symptom of cervical dystonia and was evaluated by separate assessments of pain frequency and severity on scales of 0 (no pain) to 4 (constant in frequency or extremely severe in intensity). Study results on the primary endpoints and the pain-related secondary endpoints are shown in Table 48.

Table 48: Efficacy Outcomes of the Phase 3 Cervical Dystonia Study (Group Means)

	Placebo (N=82)	BOTOX (N=88)	95% CI on Difference
Baseline CDSS	9.3	9.2	
Change in CDSS at Week 6	-0.3	-1.3	(-2.3, 0.3)[a,b]
% Patients with Any Improvement on Physician Global Assessment	31%	51%	(5%, 34%)[a]
Pain Intensity Baseline	1.8	1.8	
Change in Pain Intensity at Week 6	-0.1	-0.4	(-0.7, -0.2)[c]
Pain Frequency Baseline	1.9	1.8	
Change in Pain Frequency at Week 6	-0.0	-0.3	(-0.5, -0.0)[c]

[a] Confidence intervals are constructed from the analysis of covariance table with treatment and investigational site as main effects, and baseline CDSS as a covariate.
[b] These values represent the prospectively planned method for missing data imputation and statistical test. Sensitivity analyses indicated that the 95% confidence interval excluded the value of no difference between groups and the p-value was less than 0.05. These analyses included several alternative missing data imputation methods and non-parametric statistical tests.
[c] Confidence intervals are based on the t-distribution.

Exploratory analyses of this study suggested that the majority of patients who had shown a beneficial response by week 6 had returned to their baseline status by 3 months after treatment. Exploratory analyses of subsets by patient sex and age suggest that both sexes receive benefit, although female patients may receive somewhat greater amounts than male patients. There is a consistent treatment-associated effect between subsets greater than and less than age 65. There were too few non-Caucasian patients enrolled to draw any conclusions regarding relative efficacy in racial subsets.

In this study the median total BOTOX dose in patients randomized to receive BOTOX (N=88) was 236 Units, with 25th to 75th percentile ranges of 198 Units to 300 Units. Of these 88 patients, most received injections to 3 or 4 muscles; 38 received injections to 3 muscles, 28 to 4 muscles, 5 to 5 muscles, and 5 to 2 muscles. The dose was divided amongst the affected muscles in quantities shown in Table 49. The total dose and muscles selected were tailored to meet individual patient needs.

Table 49: Number of Patients Treated per Muscle and Fraction of Total Dose Injected into Involved Muscles

Muscle	Number of Patients Treated in this Muscle (N=88)	Mean % Dose per Muscle	Mid-Range of % Dose per Muscle*
Splenius capitis/cervicis	83	38	25-50
Sternocleidomastoid	77	25	17-31
Levator scapulae	52	20	16-25
Trapezius	49	29	18-33
Semispinalis	16	21	13-25
Scalene	15	15	6-21
Longissimus	8	29	17-41

* The mid-range of dose is calculated as the 25th to 75th percentiles.

There were several randomized studies conducted prior to the double-blind, placebo-controlled study, which were supportive but not adequately designed to assess or quantitatively estimate the efficacy of BOTOX.

14.8 Primary Axillary Hyperhidrosis

The efficacy and safety of BOTOX for the treatment of primary axillary hyperhidrosis were evaluated in two randomized, multi-center, double-blind, placebo-controlled studies. Study 1 included adult patients with persistent primary axillary hyperhidrosis who scored 3 or 4 on a Hyperhidrosis Disease Severity Scale (HDSS) and who produced at least 50 mg of sweat in each axilla at rest over 5 minutes. HDSS is a 4-point scale with 1 = “underarm sweating is never noticeable and never interferes with my daily activities”; to 4 = “underarm sweating is intolerable and always interferes with my daily activities”. A total of 322 patients were randomized in a 1:1:1

ratio to treatment in both axillae with either 50 Units of BOTOX, 75 Units of BOTOX, or placebo. Patients were evaluated at 4-week intervals. Patients who responded to the first injection were re-injected when they reported a re-increase in HDSS score to 3 or 4 and produced at least 50 mg sweat in each axilla by gravimetric measurement, but no sooner than 8 weeks after the initial injection.

Study responders were defined as patients who showed at least a 2-grade improvement from baseline value on the HDSS 4 weeks after both of the first two treatment sessions or had a sustained response after their first treatment session and did not receive re-treatment during the study. Spontaneous resting axillary sweat production was assessed by weighing a filter paper held in the axilla over a period of 5 minutes (gravimetric measurement). Sweat production responders were those patients who demonstrated a reduction in axillary sweating from baseline of at least 50% at week 4.

In the three study groups the percentage of patients with baseline HDSS score of 3 ranged from 50% to 54% and from 46% to 50% for a score of 4. The median amount of sweat production (averaged for each axilla) was 102 mg, 123 mg, and 114 mg for the placebo, 50 Units and 75 Units groups respectively.

The percentage of responders based on at least a 2-grade decrease from baseline in HDSS or based on a >50% decrease from baseline in axillary sweat production was greater in both BOTOX groups than in the placebo group ($p<0.001$), but was not significantly different between the two BOTOX doses (see Table 50).

Duration of response was calculated as the number of days between injection and the date of the first visit at which patients returned to 3 or 4 on the HDSS scale. The median duration of response following the first treatment in BOTOX treated patients with either dose was 201 days. Among those who received a second BOTOX injection, the median duration of response was similar to that observed after the first treatment.

In study 2, 320 adults with bilateral axillary primary hyperhidrosis were randomized to receive either 50 Units of BOTOX (n=242) or placebo (n=78). Treatment responders were defined as subjects showing at least a 50% reduction from baseline in axillary sweating measured by gravimetric measurement at 4 weeks. At week 4 post-injection, the percentages of responders were 91% (219/242) in the BOTOX group and 36% (28/78) in the placebo group, $p<0.001$. The difference in percentage of responders between BOTOX and placebo was 55% (95% CI=43.3, 65.9).

Table 50: Study 1 - Study Outcomes

Treatment Response	BOTOX 50 Units (N=104)	BOTOX 75 Units (N=110)	Placebo (N=108)	BOTOX 50-placebo (95% CI)	BOTOX 75-placebo (95% CI)
HDSS Score change ≥2 (n)[a]	55% (57)	49% (54)	6% (6)	49.3% (38.8, 59.7)	43% (33.2, 53.8)
>50% decrease in axillary sweat production % (n)	81% (84)	86% (94)	41% (44)	40% (28.1, 52.0)	45% (33.3, 56.1)

[a] Patients who showed at least a 2-grade improvement from baseline value on the HDSS 4 weeks after both of the first two treatment sessions or had a sustained response after their first treatment session and did not receive re-treatment during the study.

14.9 Blepharospasm

Botulinum toxin has been investigated for use in patients with blepharospasm in several studies. In an open label, historically controlled study, 27 patients with essential blepharospasm were injected with 2 Units of BOTOX at each of six sites on each side. Twenty-five of the 27 patients treated with botulinum toxin reported improvement within 48 hours. One patient was controlled with a higher dosage at 13 weeks post initial injection and one patient reported mild improvement but remained functionally impaired.

In another study, 12 patients with blepharospasm were evaluated in a double-blind, placebo-controlled study. Patients receiving botulinum toxin (n=8) improved compared with the placebo group (n=4). The effects of the treatment lasted a mean of 12 weeks.

One thousand six hundred eighty-four patients with blepharospasm who were evaluated in an open label trial showed clinical improvement as evaluated by measured eyelid force and clinically observed intensity of lid spasm, lasting an average of 12 weeks prior to the need for re-treatment.

14.10 Strabismus

Six hundred seventy-seven patients with strabismus treated with one or more injections of BOTOX were evaluated in an open label trial. Fifty-five percent of these patients improved to an alignment of 10 prism diopters or less when evaluated six months or more following injection.

16 HOW SUPPLIED/STORAGE AND HANDLING

16.1 How Supplied

BOTOX (onabotulinumtoxinA) for injection is a sterile, vacuum-dried powder supplied in a single-dose vial in the following sizes:
100 Units NDC 0023-1145-01
200 Units NDC 0023-3921-02

BOTOX cartons have features to alert users if contents may have been compromised. Each BOTOX vial label and carton also contains the U.S. License number: 1889 *[see Dosage and Administration (2.1)]*.

Do not use the product and contact AbbVie for additional information at 1-800-678-1605 if the labeling is not described as above.

16.2 Storage and Handling

Unopened vials of BOTOX should be stored in a refrigerator between 2° to 8°C (36º to 46ºF) for up to 36 months. Do not use after the expiration date on the vial. Reconstituted BOTOX may be stored in a refrigerator (2° to 8°C) for up to 24 hours until time of use *[see Dosage and Administration (2.2)]*.

17 PATIENT COUNSELING INFORMATION

Advise the patient or caretaker to read the FDA-approved patient labeling (Medication Guide).

Swallowing, Speaking or Breathing Difficulties, or Other Unusual Symptoms
Advise patients or their caretaker(s) to inform their doctor or pharmacist if they develop any unusual symptoms (including difficulty with swallowing, speaking, or breathing), or if any existing symptom worsens *[see Boxed Warning and Warnings and Precautions (5.1, 5.6)]*.

Ability to Operate Machinery or Vehicles
Advise patients or their caretaker(s) that if loss of strength, muscle weakness, blurred vision, dizziness, or drooping eyelids occur, they should avoid driving a car or engaging in other potentially hazardous activities.

Voiding Symptoms after Bladder Injections
After bladder injections for urinary incontinence, advise patients to contact their physician if they experience difficulties in voiding or burning sensation upon voiding.

Manufactured by: AbbVie Inc.
1 N Waukegan Rd. North Chicago, IL. 60064
U.S. License Number 1889

abbvie

v11.0USPI1145

MEDICATION GUIDE
BOTOX®
BOTOX® Cosmetic
(Boe-tox)
(onabotulinumtoxinA)
for injection, for intramuscular, intradetrusor, or intradermal use

What is the most important information I should know about BOTOX and BOTOX Cosmetic?

BOTOX and BOTOX Cosmetic may cause serious side effects that can be life threatening, including:

- **Problems breathing or swallowing**
- **Spread of toxin effects**

These problems can happen hours, days, to weeks after an injection of BOTOX or BOTOX Cosmetic. Call your doctor or get medical help right away if you have any of these problems after treatment with BOTOX or BOTOX Cosmetic:

- **Problems swallowing, speaking, or breathing. These problems can happen hours, days, to weeks after an injection of BOTOX or BOTOX Cosmetic** usually because the muscles that you use to breathe and swallow can become weak after the injection. Death can happen as a complication if you have severe problems with swallowing or breathing after treatment with **BOTOX** or **BOTOX Cosmetic**.
  - People with certain breathing problems may need to use muscles in their neck to help them breathe. These people may be at greater risk for serious breathing problems with **BOTOX** or **BOTOX Cosmetic**.
  - Swallowing problems may last for several months. People who cannot swallow well may need a feeding tube to receive food and water. If swallowing problems are severe, food or liquids may go into your lungs. People who already have swallowing or breathing problems before receiving **BOTOX** or **BOTOX Cosmetic** have the highest risk of getting these problems.
- **Spread of toxin effects.** In some cases, the effect of botulinum toxin may affect areas of the body away from the injection site and cause symptoms of a serious condition called botulism. The symptoms of botulism include:
  - loss of strength and muscle weakness all over the body
  - double vision, blurred vision and drooping eyelids
  - hoarseness or change or loss of voice (dysphonia)
  - trouble saying words clearly (dysarthria)
  - loss of bladder control
  - trouble breathing
  - trouble swallowing

These symptoms can happen hours, days, to weeks after you receive an injection of **BOTOX** or **BOTOX Cosmetic**.

These problems could make it unsafe for you to drive a car or do other dangerous activities. See "What should I avoid while receiving **BOTOX** or **BOTOX Cosmetic**?"

There has not been a confirmed serious case of spread of toxin effect away from the injection site when **BOTOX** has been used at the recommended dose to treat chronic migraine, severe underarm sweating, blepharospasm, or strabismus, or when **BOTOX Cosmetic** has been used at the recommended dose to treat frown lines, crow’s feet lines, forehead lines, or vertical bands connecting the jaw and neck.

What are BOTOX and BOTOX Cosmetic?

BOTOX is a prescription medicine that is injected into muscles and used:

- to treat overactive bladder symptoms such as a strong need to urinate with leaking or wetting accidents (urge urinary incontinence), a strong need to urinate right away (urgency) and urinating often (frequency) in adults when another type of medicine (anticholinergic) does not work well enough or cannot be taken.
- to treat leakage of urine (incontinence) in adults with overactive bladder due to neurologic disease when another type of medicine (anticholinergic) does not work well enough or cannot be taken.
- to treat overactive bladder due to a neurologic disease in children 5 years of age and older when another type of medicine (anticholinergic) does not work well enough or cannot be taken.
- to prevent headaches in adults with chronic migraine who have 15 or more days each month with headache lasting 4 or more hours each day.
- to treat increased muscle stiffness in people 2 years of age and older with spasticity.
- to treat the abnormal head position and neck pain that happens with cervical dystonia (CD) in adults.
- to treat certain types of eye muscle problems (strabismus) or abnormal spasm of the eyelids (blepharospasm) in people 12 years of age and older.

BOTOX is also injected into the skin to treat the symptoms of severe underarm sweating (severe primary axillary hyperhidrosis) when medicines used on the skin (topical) do not work well enough.

BOTOX Cosmetic is a prescription medicine for adults that is injected into muscles and used for a short period of time (temporary) to improve the look of:

- moderate to severe frown lines between the eyebrows (glabellar lines)
- moderate to severe crow's feet lines
- moderate to severe forehead lines
- moderate to severe vertical bands connecting the jaw and neck (platysma bands)

You may receive treatment for frown lines, crow's feet lines, forehead lines, and vertical bands connecting the jaw and neck at the same time.

It is not known whether **BOTOX** is safe and effective in people younger than:

- 18 years of age for treatment of overactive bladder with urinary incontinence
- 5 years of age for the treatment of overactive bladder due to a neurologic disease
- 18 years of age for treatment of chronic migraine
- 16 years of age for treatment of cervical dystonia
- 18 years of age for treatment of hyperhidrosis
- 12 years of age for treatment of strabismus or blepharospasm
- 2 years of age for treatment of spasticity

BOTOX Cosmetic is not recommended for use in children younger than 18 years of age.

It is not known whether **BOTOX** and **BOTOX Cosmetic** are safe and effective to prevent headaches in people with migraine who have 14 or fewer headache days each month (episodic migraine).

It is not known whether **BOTOX** and **BOTOX Cosmetic** are safe and effective for severe sweating anywhere other than your armpits.

It is not known if **BOTOX Cosmetic** is safe and effective for use more than 1 time every 3 months.

Who should not receive BOTOX or BOTOX Cosmetic?

Do not receive **BOTOX** or **BOTOX Cosmetic** if you:

- are allergic to any of the ingredients in **BOTOX** or **BOTOX Cosmetic**. See the end of this Medication Guide for a complete list of ingredients in **BOTOX** and **BOTOX Cosmetic**.
- had an allergic reaction to any other botulinum toxin product such as Myobloc® (rimabotulinumtoxinB), Dysport® (abobotulinumtoxinA), Xeomin® (incobotulinumtoxinA), Jeuveau® (prabotulinumtoxinA-xvfs), Daxxify® (daxibotulinumtoxinA-lanm), or Letybo® (letibotulinumtoxinA-wlbg). This may not be a complete list of all botulinum toxin products.
- have a skin infection at the planned injection site.
- are being treated for urinary incontinence and have a urinary tract infection (UTI).
- are being treated for urinary incontinence and find that you cannot empty your bladder on your own (only applies to people who are not routinely catheterizing).

What should I tell my doctor before receiving BOTOX or BOTOX Cosmetic?

Tell your doctor about all your medical conditions, including if you:

- have a disease that affects your muscles and nerves (such as amyotrophic lateral sclerosis [ALS or Lou Gehrig's disease], myasthenia gravis or Lambert-Eaton syndrome). See "What is the most important information I should know about **BOTOX** and **BOTOX Cosmetic**?"
- have allergies to any botulinum toxin product.
- had any side effect from any botulinum toxin product in the past.
- have or have had a breathing problem, such as asthma or emphysema.
- have or have had swallowing problems.
- have or have had bleeding problems.
- have plans to have surgery.
- had surgery on your face.
- have weakness of your forehead muscles, such as trouble raising your eyebrows.
- have drooping eyelids.
- have any other change in the way your face normally looks.
- have symptoms of a urinary tract infection (UTI) and are being treated for urinary incontinence. Symptoms of a urinary tract infection may include pain or burning with urination, frequent urination, or fever.
- have problems emptying your bladder on your own and are being treated for urinary incontinence.
- are pregnant or plan to become pregnant. It is not known if **BOTOX** or **BOTOX Cosmetic** can harm your unborn baby.
- are breastfeeding or plan to breastfeed. It is not known if **BOTOX** or **BOTOX Cosmetic** passes into breast milk.

Tell your doctor about all the medicines you take, including prescription and over-the-counter medicines, vitamins and herbal supplements. Using **BOTOX** or **BOTOX Cosmetic** with certain other medicines may cause serious side effects. **Do not start any new medicines until you have told your doctor that you have received BOTOX or BOTOX Cosmetic in the past.**

Especially tell your doctor if you:

- have received any other botulinum toxin product in the last four months.
- have received injections of botulinum toxin, such as Myobloc® (rimabotulinumtoxinB), Dysport® (abobotulinumtoxinA), Xeomin® (incobotulinumtoxinA), Jeuveau® (prabotulinumtoxinA-xvfs), Daxxify® (daxibotulinumtoxinA-lanm), or Letybo® (letibotulinumtoxinA-wlbg) in the past. This may not be a complete list of all botulinum toxin products. Be sure your doctor knows exactly which product you received.
- have recently received an antibiotic by injection.
- take muscle relaxants.
- take an allergy or cold medicine.
- take a sleep medicine.
- take anti-platelets (aspirin-like products) or anti-coagulants (blood thinners).

Ask your doctor if you are not sure if your medicine is one that is listed above.

Know the medicines you take. Keep a list of your medicines with you to show your doctor and pharmacist each time you get a new medicine.

How will I receive BOTOX or BOTOX Cosmetic?

- **BOTOX** or **BOTOX Cosmetic** is an injection that your doctor will give you.
- **BOTOX** is injected into your affected muscles, skin, or bladder.
- **BOTOX Cosmetic** is injected into your affected muscles.
- Your doctor may change your dose of **BOTOX** or **BOTOX Cosmetic**, until you and your doctor find the best dose for you.
- **Your doctor will tell you how often you will receive your dose of BOTOX or BOTOX Cosmetic injections.**

What should I avoid while receiving BOTOX or BOTOX Cosmetic?

BOTOX and **BOTOX Cosmetic** may cause loss of strength or general muscle weakness, vision problems, or dizziness within hours to weeks of taking **BOTOX** or **BOTOX Cosmetic**. **If this happens,**

do not drive a car, operate machinery, or do other dangerous activities. See "What is the most important information I should know about **BOTOX** and **BOTOX Cosmetic**?"

What are the possible side effects of BOTOX and BOTOX Cosmetic?

BOTOX and BOTOX Cosmetic can cause serious side effects. See "What is the most important information I should know about **BOTOX** and **BOTOX Cosmetic**?"

Other side effects of BOTOX and BOTOX Cosmetic include:

- dry mouth.
- discomfort or pain at the injection site.
- tiredness.
- headache.
- neck pain.
- eye problems: double vision, blurred vision, decreased eyesight, drooping eyelids, swelling of your eyelids, and dry eyes.
- drooping eyebrows.
- urinary tract infection in both children and adults being treated for urinary incontinence.
- painful urination in adults being treated for urinary incontinence.
- bacteria, white blood cells, and blood in the urine of children being treated for urinary incontinence.
- inability to empty your bladder on your own and are being treated for urinary incontinence. If you have difficulty fully emptying your bladder after getting **BOTOX**, you may need to use disposable self-catheters to empty your bladder up to a few times each day until your bladder is able to start emptying again.
- allergic reactions. Symptoms of an allergic reaction to **BOTOX** or **BOTOX Cosmetic** may include: itching, rash, red itchy welts, wheezing, asthma symptoms, or dizziness or feeling faint. Tell your doctor or get medical help right away if you are wheezing or have asthma symptoms, or if you become dizzy or faint.
- upper respiratory tract infection.

Tell your doctor if you have any side effect that bothers you or that does not go away.

These are not all the possible side effects of **BOTOX** and **BOTOX Cosmetic**. For more information, ask your doctor or pharmacist.

Call your doctor for medical advice about side effects. You may report side effects to FDA at 1-800-FDA-1088.

General information about the safe and effective use of BOTOX and BOTOX Cosmetic:

Medicines are sometimes prescribed for purposes other than those listed in a Medication Guide.

This Medication Guide summarizes the most important information about **BOTOX** and **BOTOX Cosmetic**. If you would like more information, talk with your doctor. You can ask your doctor or pharmacist for information about **BOTOX** and **BOTOX Cosmetic** that is written for health professionals.

What are the ingredients in BOTOX and BOTOX Cosmetic?

Active ingredient: onabotulinumtoxinA

Inactive ingredients: human albumin and sodium chloride

Manufactured by: AbbVie Inc.
1 N Waukegan Rd. North Chicago, IL 60064
U.S. License Number 1889

abbvie Allergan Aesthetics an AbbVie company

V8.0MG1145

EXHIBIT 5 to Brin Declaration

toxins

MDPI

Review

Update on Non-Interchangeability of Botulinum Neurotoxin Products

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Abstract: The growing use of botulinum neurotoxins (BoNTs) for medical and aesthetic purposes has led to the development and marketing of an increasing number of BoNT products. Given that BoNTs are biological medications, their characteristics are heavily influenced by their manufacturing methods, leading to unique products with distinct clinical characteristics. The manufacturing and formulation processes for each BoNT are proprietary, including the potency determination of reference standards and other features of the assays used to measure unit potency. As a result of these differences, units of BoNT products are not interchangeable or convertible using dose ratios. The intrinsic, product-level differences among BoNTs are compounded by differences in the injected tissues, which are innervated by different nerve fiber types (e.g., motor, sensory, and/or autonomic nerves) and require unique dosing and injection sites that are particularly evident when treating complex therapeutic and aesthetic conditions. It is also difficult to compare across studies due to inherent differences in patient populations and trial methods, necessitating attention to study details underlying each outcome reported. Ultimately, each BoNT possesses a unique clinical profile for which unit doses and injection paradigms must be determined individually for each indication. This practice will help minimize unexpected adverse events and maximize efficacy, duration, and patient satisfaction. With this approach, BoNT is poised to continue as a unique tool for achieving individual goals for an increasing number of medical and aesthetic indications.

Keywords: botulinum; progenitor toxin complex; SNAP-25; manufacturing; potency; reference standard; duration; immunogenicity

Key Contribution: Given the many new BoNT products on the market and in development, this paper summarizes the unique features that make BoNTs non-interchangeable and highlights the importance of using each product according to its own specifications as supported by clinical studies.

check for updates

Citation: Brin, M.F.; Nelson, M.; Ashourian, N.; Brideau-Andersen, A.; Maltman, J. Update on Non-Interchangeability of Botulinum Neurotoxin Products. *Toxins* **2024**, *16*, 266. https://doi.org/10.3390/toxins16060266

Received: 2 May 2024
Revised: 1 June 2024
Accepted: 5 June 2024
Published: 10 June 2024

1. Introduction

Botulinum neurotoxins (BoNTs) are locally injectable biological medications that are used to treat medical and aesthetic indications. Over the last four decades, clinical use has expanded from the treatment of strabismus and blepharospasm with onabotulinumtoxinA (onabotA) [1,2] to a variety of other neurologic, urologic, dermatologic, and aesthetic indications [3,4]. The growing popularity of BoNT injections has led to a marked increase in the number of available products since the original approval of onabotA in 1989 (Table 1).

As additional BoNT products enter the market, it is increasingly important for clinicians to be aware of differences in formulations, doses, serotypes, and immunogenicity that can impact safety and efficacy [5]. The intrinsic differences among BoNTs impart unique physiochemical characteristics that result in distinct interactions with the tissue microenvironments into which they are injected. Additionally, units of BoNT products are

not interchangeable due to differences in the assays used to measure unit potency, including methods for determination of potency reference standards. The non-interchangeability of units among BoNT products led the United States Food and Drug Administration (FDA) to adopt unique, established nonproprietary names for each BoNT product (Table 1). In 2000, with the introduction of a serotype B product, the US FDA began requiring a statement in the prescribing information of each product indicating that units are not interchangeable or convertible among BoNTs. Our initial review published in 2014 examined some of the reasons for non-interchangeability of BoNTs [5]. In view of additional data, coupled with new products on market and in development, the current update seeks to clarify and expand on the basis of non-interchangeability of BoNTs within this new milieu.

Table 1. BoNT products commercially available or in development in selected regions worldwide.

Trade Name(s)	Nonproprietary USAN Name	Manufacturer	Serotype	Complex Size or NT Only	Formulation	Selected Regions Approved *
Commercially available						
BOTOX®, BOTOX® Cosmetic, Vistabel®, Vistabex® [4,6]	OnabotulinumtoxinA	Allergan/AbbVie	A	~900 kDa	In 100 U vial • 900 µg sodium chloride • 500 µg human serum albumin Finishing: vacuum dried	USA, Canada, EU, China, Japan, South Korea, Brazil
Dysport®, Azzalure® [7,8]	AbobotulinumtoxinA	Ipsen	A	~400 kDa **	In 500 U vial • 2.5 mg lactose • 125 µg human serum albumin Finishing: lyophilized	USA, Canada, EU, China, South Korea, Brazil
Xeomin®, Boucouture® [9]	IncobotulinumtoxinA	Merz	A	~150 kDa	In 100 U vial • 4.7 mg sucrose • 1 mg human serum albumin Finishing: Lyophilized	USA, Canada, EU, Japan, South Korea, Brazil
Nabota®, Jeuveau®, Nuceiva® [10,11]	PrabotulinumtoxinA	Evolus/Daewoong	A	~900 kDa	In 100 U vial • 900 µg sodium chloride • 500 µg human serum albumin Finishing: vacuum dried	USA, Canada, EU, South Korea, Brazil
Daxxify™ [12]	DaxibotulinumtoxinA-lanm	Revance	A	~150 kDa	In 100 U vial • 0.14 mg L-histidine • 0.65 mg L-histidine-HCl monohydrate • 0.1 mg polysorbate 20 • 11.7 µg RTP004 peptide • 36 mg trehalose dihydrate Finishing: lyophilized	USA

Table 1. *Cont.*

Trade Name(s)	Nonproprietary USAN Name	Manufacturer	Serotype	Complex Size or NT Only	Formulation	Selected Regions Approved *
Myobloc® [13,14]	RimabotulinumtoxinB	Solstice	B	~700 kDa	In 5000 U vial • 5.8 mg sodium chloride • 470 µg human serum albumin • 2.7 mg sodium succinate Finishing: liquid	USA, Canada
Alluzience™ (EU) [8,15]	AbobotulinumtoxinA solution for injection	Ipsen	A	~400 kDa	• 1.55 mg L-histidine • 4.0 mg sucrose • 8.76 mg sodium chloride • 0.10 mg polysorbate-80 0.10 mg, hydrochloric acid to Finishing: liquid (in water for injection	EU
Neuronox®/Meditoxin®	Unassigned	Medytox	A	NR	Information from the manufacturer could not be identified.	South Korea, Brazil
Innotox®	Unassigned	Medytox	A	NR	Information from the manufacturer could not be identified.	South Korea (approved in 2018; product not available at the time of manuscript submission)
Botulax® (Korea) [16], Letybo® [17] EU: 50 U vial only	LetibotulinumtoxinA	Hugel	A	NR	In 100 U vial • 0.9 mg sodium chloride • 0.5 mg HSA Finishing: lyophilized	Canada, EU, China, South Korea, USA
Relatox® [18]	None established	Microgen	A	NR	In 100 U vial: • 6 mg gelatin • 12 mg maltose Finishing: lyophilized	Russia
Hutox® (Liztox®) [19]	None established	Huons	A	900 kDa	NR	South Korea

Table 1. *Cont.*

Trade Name(s)	Nonproprietary USAN Name	Manufacturer	Serotype	Complex Size or NT Only	Formulation	Selected Regions Approved *
Lantox® (Hengli®, Prosigne®, Lantox®, Lazox® Redux®, Liftox®) [20–22]	None established	Lanzhou	A	900 kDa	In 100 U vial: • 5.0 mg gelatin • 25 mg dextran • 25 mg sucrose Finishing: lyophilized	EU, China, South Korea, Brazil
In Development						
NR [23,24]	RelabotulinumtoxinA	Galderma	A	~150 kDa	• Saline phosphate buffer (salt amounts not reported) Finishing: liquid	
NR [25,26]	TrenibotulinumtoxinE	Allergan Aesthetics, an AbbVie company	E	NR	NR	

HCl = hydrochloric acid; HSA = human serum albumin; kDa = kilodalton; mg = milligram; NR = not reported; NT = neurotoxin; U = unit; USAN = United States Adopted Name; µg = microgram. * Approved for one or more indications in the listed countries/regions as of August 2023 based on a search of publicly available information and, for Korean approvals, Wee and Park, 2022 [27]. Specificity of indications and trade names vary from country to country based on local regulatory approvals. See local prescribing information for current indication specifics, including any limitations of use, warnings and precautions, dosage and administration, and adverse reactions. ** The molecular size of the abobotulinumtoxinA neurotoxin complex has been reported to be heterogeneous [28].

2. Properties of Botulinum Neurotoxins

2.1. Structure

BoNTs are large, multi-domain proteins synthesized by various strains of *Clostridium botulinum* bacteria and are among the most potent substances known, active in the nanogram range [29]. BoNTs are produced as progenitor toxin complexes (PTCs) consisting of a ~150 kDa neurotoxin protein component in association with different-sized naturally occurring neurotoxin associated proteins (NAPs). Each strain produces a non-toxin, non-hemagglutinin (NTNH) protein, that binds in a handshake-like configuration to BoNT, which stabilizes both proteins against low pH and proteases [30–32]. Some strains also produce non-toxin hemagglutinin (HA) or other proteins that associate with the neurotoxin/NTNH to form larger complexes [33,34]. As described in the Manufacturing section, the NAPs are retained in some of the BoNT/A products and removed in others.

Clostridium botulinum strains produce seven classical immunologically distinct serotypes of BoNTs, referred to as types A through G [35–37]. Various strains produce different BoNT serotypes, as well as PTCs of different sizes [32]. In 1946, the highest-molecular-weight complex of type A was reported to be ~900 kDa, calculated based on analytical centrifugation [38]. Later studies identified different-sized BoNT/A PTCs as the medium (M) ~300 kDa complex, large (L) ~500 kDa complex, and the extra large (LL) ~900 kDa complex based on gel filtration chromatography and sucrose density gradient centrifugation [39,40]; the BoNT/A 900 kDa complex has also been reported using size exclusion high-performance liquid chromatography [41]. The M-PTC is made up of BoNT and NTNHA, and the L- and LL-PTCs are formed by the association of various HAs (Figure 1). Some serotypes do not have the HA genes (BoNT/E and/F) and may only form the M-PTC [32]. The NAPs play various roles in BoNT's activity, including protecting the neurotoxin from degradation [42] and potentially reducing exposure to the immune system [43]. Additional actions of the NAPs are described in a subsequent section (Role of NAPs in the Pharmacodynamic Action of BoNTs).

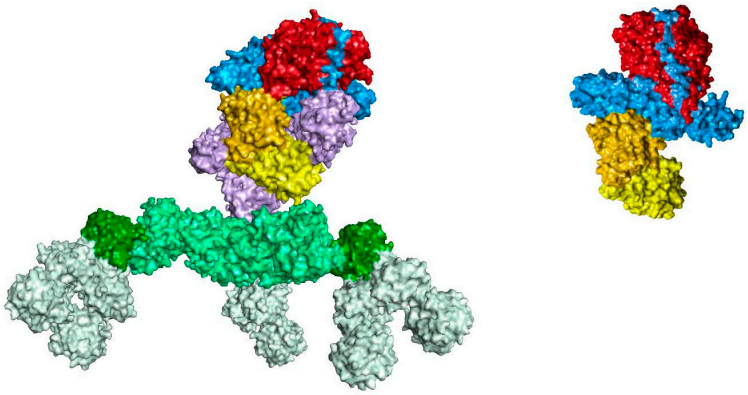

Figure 1. Structure of the BoNT/A LL-progenitor toxin complex (PTC; (**left**)) and the BoNT/A neurotoxin protein component (**right**). The complex shown here comprises the neurotoxin protein component (red/blue/yellow/gold), the non-toxin non-hemagglutinin protein (NTNH; purple), and several hemagglutinin (HA) proteins (shades of green). Images created by Lance Steward (AbbVie/Allergan Aesthetics) with Discovery Studio 2017 R2 (BIOVIA, Dassault Systèmes). Neurotoxin component image based on PDB ID 3BTA; Lacy et al. [44]. LL-PTC based on PDB IDs 4LO4, 4LO7, 4LO8, 4LO0 (RCSB.org; accessed on 29 February 2024); Lee et al. [45] and PDB ID 3V0A (RCSB.org; accessed on 29 February 2024); Gu et al. [31]).

2.2. Mechanism of Action

For all BoNT serotypes, the ~150 kDa neurotoxin component is made up of two protein chains: a ~50 kDa light chain and a ~100 kDa heavy chain. These chains are linked by a disulfide bridge [46]. Specific locations or domains within the ~50 kDa light chain and a ~100 kDa heavy chain of the neurotoxin component mediate different aspects

of the BoNT multi-step mechanism of action, which have inspired the moniker "modular nanomachine" [47].

The overall mechanism of action involves binding to nerve terminals, internalization into the neuron, translocation of the light chain, and cleavage of one or more proteins in the SNARE (soluble N-ethylmaleimide sensitive factor attachment protein receptor) complex that mediates vesicular fusion with the plasma membrane, resulting in inhibition of neurotransmitter release from the neuron.

The mechanism of action of BoNT/A has been well studied and characterized. The first step involves dual binding of the C-terminal portion of the BoNT/A ~100 kDa heavy chain to low affinity gangliosides (lipid-carbohydrate molecules) on the surface of nerve terminals and to a higher-affinity synaptic vesicle protein, SV2, that becomes accessible during vesicular neurotransmitter release (Figure 2) [33,48].

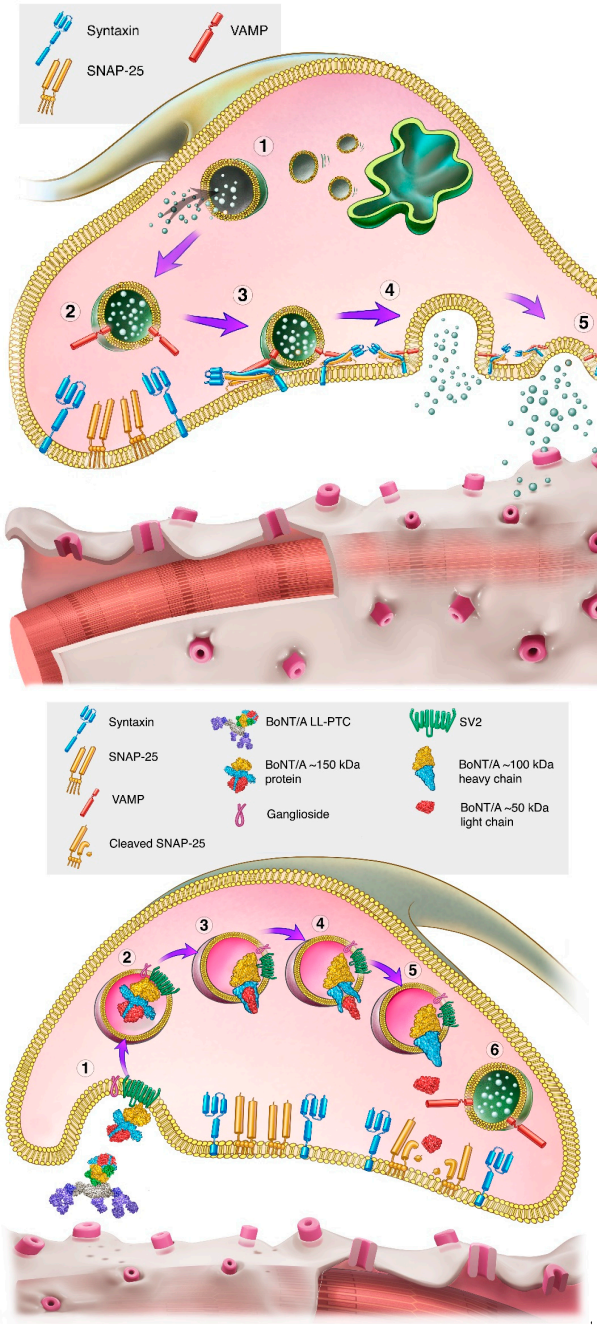

Figure 2. Mechanism of action of BoNT/A. The **top** panel shows fusion of cholinergic synaptic vesicles with the motor nerve terminal membrane in the absence of BoNT/A. Vesicles bud off the early endosome

and are loaded with acetylcholine (1). The vesicle approaches the nerve terminal membrane (2), where the SNARE proteins in the vesicle membrane (VAMP; red) and neuronal membrane (syntaxin—blue; SNAP-25—gold) assist with vesicle docking and fusion (3). Acetylcholine is released into the synaptic cleft (4) where it binds to cholinergic receptors on motor neurons (5). The **bottom** panel shows the mechanism by which BoNT/A inhibits cholinergic neurotransmission. BoNT/A binds to gangliosides and the protein SV2 in the nerve terminal membrane (1). BoNT/A is then internalized into the neuron via receptor-mediated endocytosis (2). The BoNT/A heavy chain translocates the light chain of the protein across the synaptic vesicle membrane into the cytoplasm (3–5), where the light chain functions as a zinc-dependent protease, cleaving SNAP-25 and preventing vesicle fusion and hence acetylcholine release (6).

After binding, BoNT/A is internalized into nerve cells via receptor-mediated endocytosis, where it temporarily resides within vesicles. The N-terminal portion of the BoNT/A heavy chain translocates the ~50 kDa light chain of the protein across the vesicle membrane (Figure 2) [49]. The disulfide bridge is then reduced, enabling the release of the light chain into the cytosol [46] where the ~50 kDa BoNT/A light chain cleaves synaptosomal associated protein-25 kDa (SNAP-25)—part of the SNARE complex.

Cleavage of SNAP-25 inhibits synaptic vesicle fusion with the neuronal membrane, thereby inhibiting vesicular neurotransmitter release such as occurs at the neuromuscular junction. It also inhibits other cellular processes that require synaptic vesicle fusion with membranes, including the insertion of protein receptors and channels from the vesicle into the membrane [50].

Although the general mechanism of action of BoNTs is well characterized, several detailed questions remain, such as the specific mode of endocytosis responsible for neurotoxin internalization (e.g., clathrin-mediated endocytosis, ultrafast endocytosis, and/or activity-dependent bulk endocytosis) and the specific localization of the light chains of various BoNT serotypes following translocation across the endocytotic vesicle membrane (e.g., continued association with the vesicle versus diffusion within the cytosol) [51].

In addition to its action on motor neurons, BoNT/A inhibits the release of pain-related peptides such as substance P and calcitonin gene-related peptide (CGRP) from sensory neurons [52,53]. BoNT/A further prevents plasma membrane trafficking of transient receptor potential (TRP) receptors, which are important in pain [54,55].

Additionally, BoNT/A binds to fibroblast growth factor receptor 3 (FGFR3) in motor neurons [56] and increases FGFR3 dimerization, a marker of ligand–receptor binding [57]. The contribution of FGFR3 binding to the actions of BoNT/A requires further investigation.

2.3. Serotypes

Although all BoNT serotypes exhibit the same general mechanism of action, their specific features and actions differ, which influence their clinical properties as described later. The serotypes have related but non-identical primary structures (amino acid sequences) [58], which determine secondary (local protein folding), tertiary (overall 3-dimensional structure), and quaternary structures (arrangement of protein chains) that are essential for biological activity.

BoNT serotypes have different binding affinities for specific gangliosides on the nerve membrane, and the synaptic protein receptor varies by serotype [59]. For instance, BoNT/A binds primarily to synaptic vesicle protein 2C (SV2C), BoNT/E to SV2A and SV2B, and BoNT/B and/G to synaptotagmin [59]. BoNT/C1 appears to lack a protein receptor, instead binding to two gangliosides to mediate cell entry [60]. Inside neurons, each BoNT serotype cleaves a unique point on one or more SNARE proteins, resulting in the generation of different sized protein fragments (Figure 3) [33]. For instance, like serotype A, BoNT serotypes C1 and E also cleave SNAP-25, but at different sites than type A. Serotypes B, D, F, and G cleave vesicle associated membrane protein (VAMP)/synaptobrevin at specific sites; type C1 also cleaves syntaxin. Readers are referred to several expert reviews for details on mechanism of action [33,49].

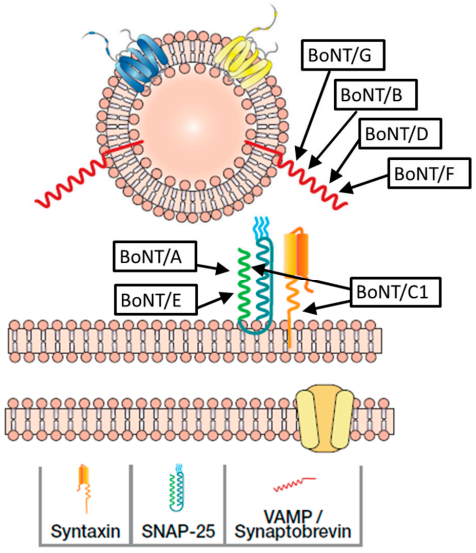

Figure 3. SNARE cleavage sites of different BoNT serotypes. BoNT serotypes B, D, F, and G all cleave VAMP/synaptobrevin at different sites. BoNT serotypes A, C1, and E all cleave SNAP-25 at different sites. Serotype C1 also cleaves syntaxin. Image modified from: Burstein et al., 2014 [61].

2.4. Role of NAPs in the Pharmacodynamic Action of BoNTs

BoNT/A is naturally expressed in *Clostridium botulinum* as PTCs, with the complex conferring a thermodynamically stable structure [62] that protects the 150 kDa neurotoxin in harsh environments [63,64]. The NAPs have been shown to protect BoNT from proteolysis [42] and alter the secondary structure of neurotoxin conformation [62].

Emerging evidence suggests that the NAPs may play a fundamental, and intracellular, role in BoNT pharmacology. In particular, HA34, the most abundant HA in the BoNT/A LL-PTC [41], binds to the neurotoxin with a KD of 0.1–0.4 μM [65,66]. HA34 itself has been shown to increase BoNT/A endopeptidase activity in vitro and in synaptosomes [65]. In a comprehensive study in human bronchial epithelial cells, HA34 increased the kinetics of BoNT/A binding to the cell surface and trafficked with BoNT/A into the same intracellular vesicles [66]. HA34 has also been demonstrated to bind to cell-expressed carbohydrates [67] and synaptotagmin II (a synaptic vesicle protein and calcium sensor that is also the protein receptor for BoNT serotypes B and G in presynaptic axon terminals) [68].

In 1970, prior to the advent of therapeutic BoNTs, Carl Lamanna, Leonardo Spero and Edward Schantz reported in vivo preclinical experiments evaluating the biology of the ~900 kDa BoNT complex and 150 kDa isolated neurotoxin [69]. They assessed the time-to-death for both compounds when administered to mice intravenously and intraperitoneally. Their results demonstrated that the time-to-death dose response curves were neither overlapping nor parallel for either injection route, leading them to conclude that "these findings preclude a rapid conversion of the large to the small molecule under physiological conditions, but they are consistent with the hypothesis that the time the toxin takes to escape from body fluids to reach specific receptor sites is influenced by molecular dimensions and the related property of diffusion rate". In a post hoc analysis, Allergan demonstrated that the biologic effects reported by Lamanna et al. were statistically different (Allergan Data on File).

Overall, the in vitro and in vivo preclinical studies support the concept of the neurotoxin complex as a critical biological component in the pharmacotherapy of BoNTs.

3. Transforming BoNTs into Medications

Given that BoNTs are large, complex biological products, transforming them into medications is more involved than for conventional small-molecule drugs. Small-molecule drugs are produced via a series of chemical reactions and their structures can be fully defined, which allows generic versions to be produced. In contrast, biological products

such as BoNTs are produced by living organisms and then manufactured into medicines via complex and highly controlled processes during which they are subject to post-translational structural modifications that lead to intrinsic heterogeneity [70]. The manufacture of BoNT complexes is even more challenging because of the multiple proteins involved and protein–protein interactions. For these reasons, generic biologics are not possible.

Given that biological medications cannot be generics, the term biosimilars is used to describe biological medications deemed highly similar, but not identical to, the original innovator (reference) product, and that show no clinically meaningful differences in terms of safety, purity, and potency [71]. Notably, the concept "clinically meaningful" is notoriously difficult to define, particularly as it depends on the perspectives of different stakeholders (e.g., patients, caregivers, insurers, etc.) [72]. Although some of the BoNT/As in Table 1 have similarities, there are currently no BoNT biosimilars. For this reason, each BoNT product is referred to in the United States by a unique United States Adopted Name (USAN) nonproprietary name (Table 1). The following text outlines the manufacturing process for BoNTs, noting the variations at each step that can affect the nature of the final products.

3.1. Bacterial Strain

The manufacturing process for each commercial BoNT product is distinct and proprietary, beginning with the master cell bank containing the *C. botulinum* bacterial strain. The single commercial BoNT product based on the B serotype is produced from the *C. botulinum* type B Bean strain [13]. Some of the available BoNT type A products are based on "a Hall strain" as noted in the next paragraph (e.g., onabotA, incobotulinumtoxinA (incobotA), abobotulinumtoxinA (abobotA), daxibotulinumtoxinA (daxibotA)) [3,7,9,73], whereas others are based on different strains (e.g., letibotulinumtoxinA (letibotA): CBFC26 strain) [74,75].

In the early 1900s, bacteriologist Ivan Hall isolated and preserved a number of different *C. botulinum* strains from several sources [76]. and eventually distributed them to various academic institutions where they were sub-cultured. These strains became known as "the Hall strain", even though they are not identical [77–79]. A comparison of four different bacterial strains producing BoNT type A, three of which were identified as Hall strains, found differences in neurotoxin gene sequence, gene content, and genome arrangement [78]. Even minor differences in the amino acid sequence can substantially alter in vitro and in vivo properties of BoNTs, including onset and duration of effect [80,81].

3.2. Fermentation

C. botulinum bacteria produce BoNT when they are fermented under appropriate conditions. Fermentation is an anaerobic metabolic process used by bacteria and yeast to generate energy for cell growth; fermentation is best known as the process by which yeast produce wine from grapes and beer from grains. In the case of *C. botulinum* bacteria, neurotoxin is produced when the bacteria are cultured and maintained under conducive conditions. Each manufacturer uses its own proprietary fermentation method, including the constituents of the fermentation media, which includes nutrients such as carbon, nitrogen, and hydrolysate (amino acid) sources. The growth conditions and duration of fermentation may vary. Fermentation conditions such as glucose concentration and temperature will affect production of BoNT/A [82] and may be expected to have different quality attributes and yield between manufacturers.

3.3. Purification

The next step in BoNT manufacturing is purification of the proteins from the fermentation broth. The purification methods used for each product are proprietary and contribute to the specific characteristics of the drug substance (e.g., complex size, protein configuration). Purification is accomplished by crystallization for onabotA [83] and chromatography for many other BoNTs, including abobotA [84], incobotA [85], rimabotulinumtoxinB (rimabotB) [13,82], daxibotA [73], and letibotA [74]. However, even within a given purification method such as chromatography, the specific methods used for each

product can differ, such as processing reagents, etc. Given that the purification methods are proprietary, it is not possible to compare specific procedures across manufacturers. Notably, manufacturers intentionally design their procedures to retain some, all, or none of the NAPs (Table 1; Figure 1).

Purification results in the drug substance—an active ingredient that is intended to furnish pharmacological activity [86]. For BoNT products, NAPs are part of the drug substance if they are retained during purification. In vitro, NAPs increase the stability of the ~150 kDa neurotoxin component at a range of physiologically relevant temperatures and pH values [87] and may reduce exposure to the immune system [43]. Additional roles of the NAPs are described in a previous section (Role of NAPs in the Pharmacodynamic Action of BoNTs). The drug substance is stored in the manufacturer's drug substance-specific formulation and aliquots are subsequently used in the manufacture of drug products.

The manufacturing processes described to this point are designed to obtain BoNT proteins, with or without NAPs, that retain their secondary, tertiary, and quaternary structures (when present). As with all proteins, even small changes in the manufacturing process can lead to changes in protein biochemistry or structure, impacting function and biological activity [88]. This is not merely theoretical: manufacturing changes have led to unexpected and consequential alterations with protein therapies including alglucosidase alfa (upscaling production resulted in glycosylation differences that necessitated a new biological license application) [89] and a human growth hormone product (increased antibody formation occurred due to host cell protein contamination) [90,91]. Preservation of protein biochemistry and structure is therefore one reason that the manufacturing processes for biological proteins must be strictly controlled and monitored. In-process testing throughout the manufacturing process is utilized to characterize protein integrity, purity and activity during processing.

3.4. Unit Testing Procedures

For chemically synthesized drugs such as acetaminophen, amounts are measured in mass or weight in milligrams, micrograms, or nanograms. However, weights are not adequate measures of potency of BoNTs because of the complexities inherent in large proteins and their manufacture. The clinically relevant measure for BoNTs is not the weight of the substance present (e.g., in a vial or syringe) but rather the ability of that substance to affect biological processes. As such, BoNTs are measured in units of biological activity that are not interchangeable among different products. The specific features of unit testing, including differences among manufacturers' assays that lead to potency differences among BoNTs, are described in a subsequent section (Botulinum Neurotoxin Potency).

3.5. Excipients and Formulation—Generating the Drug Product

The final step in the manufacturing process of BoNTs is fashioning the drug substance into a form that can be used by clinicians. The finished dosage form of a medication is the drug product [86].

BoNT drug products include the drug substance along with excipients. Excipients are substances that are appropriately evaluated for safety and intentionally added to the drug substance [92], to maintain integrity/stability and enable delivery of the drug substance (Table 1).

Several currently available solid BoNT preparations (onabotA, abobotA, incobotA, prabotA) include large proteins such as albumin as excipients. In the initial formulation of onabotA for clinical use, albumin was used to help ensure stability during the reconstitution process [83], increase the amount of physical substance in the vials, and help prevent BoNT from adhering to surfaces such as glass. The large protein, gelatin, is included in the BoNT/A products from Lanzhou and Microgen (Table 1). Sugars are also used as bulking agents to enhance product stability or provide structure in lyophilized preparations, such as the lactose in abobotA, sucrose in incobotA, maltose in Relatox®, and trehalose dihydrate in daxibotA.

Surfactants are excipients that help prevent large proteins binding to contact surfaces or reduce interfacial tension at the liquid/air interface (e.g., polysorbate-20, daxibotA;

polysorbate-80, and liquid abobotA). BoNT products also contain a tonicity agent (e.g., sodium chloride, or trehalose) to help control osmolarity of the injected substance (i.e., prevent hypo- or hypertonicity).

The recently introduced daxibotA contains the proprietary cell-penetrating peptide RTP004. A study by Malmirchegini et al. found that the proprietary peptide prevented the neurotoxin from thermal aggregation in solution and adsorption to the vial surface [93]. Conversely, two groups reported that cell penetrating peptides did not have significant effects on the adsorption or aggregation of BoNT/A [94,95]. As such, the role of the proprietary peptide in daxibotA remains unclear.

A finishing process results in the final commercial form as solids or liquids. BoNTs formulated as solids require some method of drying to deliver the final drug product. AbobotA and letibotA are freeze-dried, and several other products (e.g., incobotA, prabotulinumtoxinA (prabotA) [in Korea]; BoNT/As from Microgen and Lanzhou) are lyophilized. Freeze drying and lyophilization (often used interchangeably) are processes in which the liquid is frozen and the ice evaporated under low pressure. OnabotA is vacuum dried, in which the liquid is removed under reduced air pressure without the freezing step.

The first liquid BoNT formulation to be approved in the US was rimabotB, which is formulated as a buffered solution of pH = 5.6 [14]. A liquid abobotA is approved in EU for glabellar lines, and additional liquid formulations are in development by multiple manufacturers (Table 1). Liquid formulations may reduce the time and burden associated with reconstitution in clinics, as well as the potential for medication errors. However, liquid formulations limit the ability to modify injected product concentration when needed to individualize patient care. Overall, the variety of BoNT product formulations provides options for clinicians.

3.6. Pre-Release Unit Testing

After compounding and finishing, BoNT products are tested for unit activity prior to release for clinical use (Figure 4). Pre-release assay conditions differ from those used for drug substance testing, as drug substance concentration and drug product excipients can influence assay performance [96]. The potential for interaction of unique excipients with LD50 and cell-based potency assays (CBPAs) adds a layer of complexity to the differences among BoNT product Unit assignment.

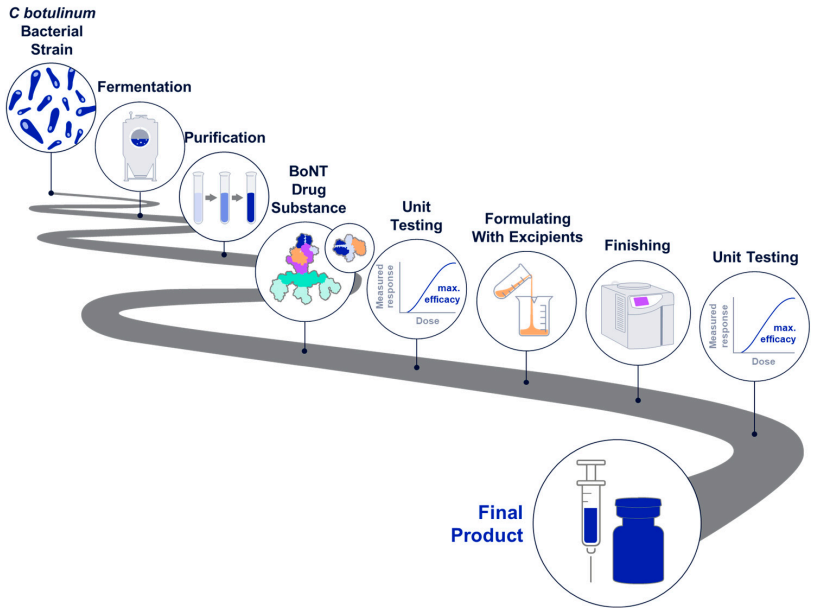

Figure 4. Overview of manufacturing process for BoNTs. This graphic shows the main steps of manufacture for BoNTs. Differences among products can occur at each step, as described in the text.

4. Botulinum Neurotoxin Potency

Potency in pharmacology is defined as the concentration or amount of drug needed to produce a defined effect [97,98]. As noted previously, potency for BoNTs is measured in units of biological activity rather than in weight. In biological activity assays, a number (unit) is assigned to a functional effect. For BoNTs, units of biological activity are specific to each product and manufacturer and are not interchangeable. The non-interchangeability of units is based on differences in unit testing procedures implemented by manufacturers, which includes differences in determining the potency of reference standards. Per the European Pharmacopoeia, potency of BoNT/A products for injection must be confirmed in a mouse model of toxicity or by in vivo/ex vivo methods validated with respect to the mouse LD50 assay (see next section on Definition of a Unit) [99]. Advances in technology have permitted the development of cell-based potency assays for BoNT potency testing while ensuring the pharmaceutical quality of the product (see Cell Based Potency Assays).

4.1. Definition of a Unit

The mouse LD50 (mLD50) has been the standard method for BoNT unit testing against which newer methods (e.g., CBPAs) are anchored. One unit is defined as the median lethal dose in mice following intraperitoneal (IP) injection. Mouse LD50 testing is performed under controlled conditions to promote consistency (e.g., animal strain, age, sex, diet, temperature, caging, season, liquid used to dilute the product, etc.) [14,100,101]. Although methods of mLD50 testing are standardized for a specific product, procedures vary between companies. For instance, all BoNT manufacturers use proprietary reference standards (see Potency Reference Standards section) and many use different assay protocols, including reconstitution agents, dilution schemes, etc., resulting in units that are not comparable or interchangeable. Indeed, different assays are needed to accommodate the formulation differences among BoNT products.

The influence of assay conditions on mLD50 outcomes has been studied by Sesardic and colleagues working at the National Institute for Biological Standards and Control in Potters Bar, Hertfordshire, United Kingdom [96,102]. Their studies found a differential effect of assay conditions on BoNT/A products from different manufacturers [96]. For instance, the strain of mice used and the addition of gelatin phosphate to the dilution buffer had a greater effect on the mLD50 values of some BoNT preparations than others [96]. For this reason, they concluded that mLD50 tests and the units obtained are specific to each BoNT manufacturer and only apply to each individual BoNT product. Sesardic and colleagues also found inter-and intra-laboratory variation in mLD50 values, that was improved by the use of a reference standard [102].

4.2. Potency Reference Standards

Potency reference standards are an important aspect of unit testing that renders BoNT units unique and non-interchangeable. Reference standards are certified materials or substances whose properties are sufficiently well established that they can be used for calibration of an apparatus, assessment of a measurement method, and assigning values to materials [103]. Potency reference standards are used to calibrate assays that measure the biological activity of BoNT. Each company creates and maintains its own proprietary potency reference standard that defines a unit and against which the potency of each BoNT lot intended for commercial use is measured and compared. Consequently, the units of each BoNT remain relative to each manufacturer's specific proprietary qualified reference standard according to international guidelines. Small proteins such as insulin have international standards against which the potencies of products from different manufacturers are compared [104]. International reference standards are also available for some large proteins such as infliximab, but these standards are to be used only for each manufacturer's quality control purposes and not to compare different products [105].

The non-interchangeability of units or unit doses among BoNT products is a critical clinical concept given the increasing number of BoNTs that are now commercially

available or in development worldwide (Table 1). Non-interchangeability of units means that 100 units of one BoNT product are not the same and do not have the same potency as 100 units of another product because each BoNT is unique due to the differences in manufacturing process and units of each are determined by different assays and internal reference standards.

4.3. Cell Based Potency Assays

Although the mLD50 has been the basis of BoNT potency testing for decades, some BoNT manufacturers have sought to reduce animal use and develop methods that can accommodate high-volume testing. Today, proprietary CBPAs are increasingly used to assess potency of BoNTs in place of the mLD50 [7,9,106]. The potency reference standards used for these assays are qualified based on mLD50 tests and thus trace their lineage back to that test. For CBPAs to be approved by regulatory agencies, they must be rigorously developed and cross validated against product-specific mLD50 tests. Additionally, CBPAs must recapitulate all steps in the mechanism of action of BoNTs, including binding, internalization, translocation of the light chain, and SNARE protein cleavage [107].

Development of a BoNT CBPA is a difficult undertaking due to the specificity and sensitivity required to detect the minute amounts of BoNTs in medicinal products. Like mLD50 tests, CBPAs are impacted by a large number of factors, including the (1) type of cells, (2) number of cells, (3) conditions/media used to grow and maintain cells, (4) treatment times, (5) incubation times, (6) diluent that maintains integrity of the BoNT sample and is compatible with cell viability, (7) antibody or other molecular reporters used to detect SNAP-25 cleavage, and (8) antibody amount (if antibodies are used) (Figure 5) [106]. These factors are optimized differently by individual manufacturers in their own proprietary assays. For example, differences in the sensitivity and specificity of antibodies against SNARE protein fragments used by different manufacturers can lead to more or less BoNT required to meet the definition of a unit in a specific assay.

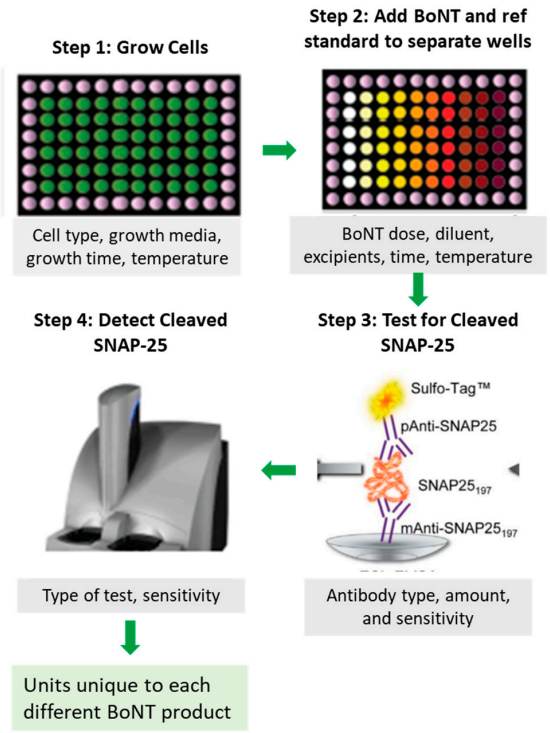

Figure 5. Factors that affect the development of cell-based potency assays (CBPAs). The development of CBPAs is influenced by many different factors, some examples of which are listed under each step. Variations in these factors influence assay performance. This graphic depicts the general steps in the Allergan/AbbVie CBPA. Modified from Rupp et al. 2021 [108] and Fernandez Salas et al. 2012 [106].

4.4. Examples of Differences in Potency among BoNTAs

Given the differences in mLD50 tests, CBPAs, and reference standards described, it is expected that BoNT products would yield different potencies when compared at the same number of labeled units. This has been demonstrated in two recent studies in which onabotA displayed greater potency than incobotA [109] and prabotA [108] in the Allergan/AbbVie CBPA. In the CBPA, incobotA demonstrated reduced relative potency compared with onabotA, showing in a 1.3-fold difference [109]. A separate study that compared prabotA with onabotA in the CBPA also found a reduced relative potency of prabotA compared with onabotA, showing a 1.3-fold difference [108]. Thus, although incobotA and prabotA were labeled as 100 U and tested as 100 U in their own manufacturers' potency assays, they measured less than 100 U when tested in the onabotA potency assay. This means that more than 1 U incobotA and prabotA are needed to achieve the same biologic effect as 1 U of onabotA.

4.5. No Fixed Dose Ratios

The lack of a standardized unit for BoNT products has led to attempts at defining dose conversion ratios. Preclinical studies conducted over the past several decades have demonstrated that dose ratios of BoNT products vary in different experimental models and for different outcome measures [110–113]. These preclinical studies, conducted under controlled conditions, demonstrate that there is no fixed dose ratio across the range of doses and no single dose ratio is accurate to compare BoNT products.

The preclinical findings are supported by clinical evidence, as shown in Tables 2 and 3 for several BoNT products. Ratios of onabotA:abobotA doses in clinical studies have ranged from 1:1.2 to 1:13, with blepharospasm showing the most variability (Table 2). In cervical dystonia, hemifacial spasm, and spasticity, onabotA:abobotA doses have tended to be used at ratios ranging from 1:2.5 to 1:6 (Table 2). Ratios of onabotA:incobotA doses in clinical studies have ranged from 1:1 to 1:2.5, with the majority of ratios >1 and none of the ratios <1 (Table 3).

Table 2. Ratios of doses studied or derived from onabotA:abobotA in clinical studies. Adapted from Ferrari et al., 2018 [114].

Indication	Author/Publication	RatioI (ona-botA:abobotA)
Blepharospasm	Bentivoglio et al., 2012 [115]	1:1.2–1:13.3
	Bihari, 2005 [116]	1:4–1:5
	Dodel et al., 1997 [117]	1:4–1:6
	Kollewe et al., 2015 [118]	1:2.3
	Marion et al., 1995 [119]	1:3
	Marchetti et al., 2005 [120]	1:3–1:11
	Nussgens and Roggenkämper, 1997 [121]	1:4
	Sampaio et al., 1997 [122]	1:4
Cervical dystonia	Bihari, 2005 [116]	1:4–1:5
	Dodel et al., 1997 [117]	1:4–1.6
	Marchetti et al., 2005 [120]	1:3–1:11
	Misra et al., 2012 [123]	3.1:1
	Odergren et al., 1998 [124]	1:3
	Ranoux D et al., 2002 [125]	1:3–1:4
	Rystedt A et al., 2015 [126]	1.7:1
	Van den Bergh and Lison, 1998 [127]	1:2.5
	Yun et al., 2015 [128]	1:2.5
Hemifacial spasm	Bihari, 2005 [116]	1:4–1:5
	Dodel et al., 1997 [117]	1:4–1:6
	Marion et al., 1995 [119]	1:3
	Van den Bergh and Lison, 1998 [127]	1:2.5
Spasticity	Bhakta et al., 1996 [129]	1:4–1:5
	Keren-Capelovitch et al., 2010 [130]	1:2.5
	Rasmussen et al., 2000 [131]	1:4

Table 3. Ratios of doses studied or derived from onabotA:incobotA in clinical studies.

Indication	Author/Publication	Ratio (onabotA:incobotA)
Blepharospasm	Bladen et al., 2020 [132]	1:1
	Juarez et al., 2011 [133]	1:1.2
	Kent et al., 2021 [134]	1:1.37
	Kollewe et al., 2015 [118]	1:1.2
	Roggenkämper et al., 2006 [135]	1:1
	Saad and Gourdeau, 2014 [136]	1:1
Cervical dystonia	Benecke et al., 2005 [137]	1:1
	Dressler et al., 2014 [138]	1:1
	Juarez et al., 2011 [133]	1:1.2
	Kent et al., 2021 [134]	1:1.21
Hemifacial spasm	Bladen et al., 2020 [132]	1:1
	Juarez et al., 2011 [133]	1:1.1
Spasticity	Italian Society of Pharmacology, 2017 [139]; Ferrari et al., 2018 [114]	1:1.5–1:2.5
Glabellar lines	Banegas et al., 2013 [140]	1:1
Blepharospasm	Kollewe et al., 2015 [118]	1:2

5. Botulinum Neurotoxin Dose Response

Dose response is a basic principle of pharmacology in which responses to medications are evaluated in relation to increasing doses. Due to the nature of biological systems, responses often increase to a finite point beyond which they do not increase further regardless of the increase in dose [141]. At extremely high doses, therapeutic responses can even decline, for example, due to toxicity, adverse events, or other factors related to the system under study [142].

Like most medications, BoNTs exhibit a dose response relationship. As doses are increased, more SNAP-25 is cleaved [105,143], leading to greater inhibition of neurotransmitter release [143,144]. BoNT dose response has been documented in non-human preclinical studies in which a wide range of doses can be evaluated [145], as well as in clinical studies evaluating a narrower range of doses for many of the BoNT products (e.g., onabotA [146], abobotA [147,148], incobotA [149], daxibotA [150]; see Figure 6 for an example). In the dose-ranging glabellar line studies, efficacy/duration responses appear to plateau at the highest doses, representing the peak pharmacological effect and/or duration [146,150], an observation consistent with typical dose–response kinetics characterized by a "ceiling effect" or maximal attainable response [151]. Conversely, in several studies that evaluated only two doses or two dose ranges of incobotA [9] and daxibotA [152] for cervical dystonia, responses appeared to be similar across doses. In these studies, it is unclear whether the data plateaus represent a ceiling effect or whether higher doses would produce greater efficacy or longer duration.

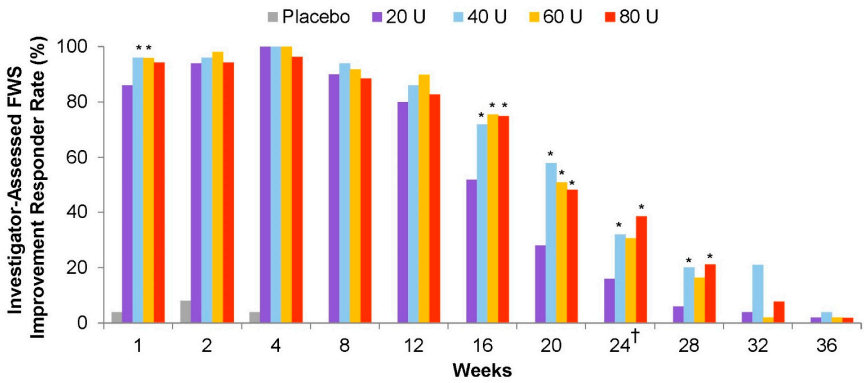

Figure 6. Dose response of onabotA duration. Proportion of responders at each visit (subjects with a ≥1-grade Facial Wrinkle Scale [FWS] improvement from baseline at maximum frown), assessed by

investigator. Data are from all randomized and treated subjects with a baseline and at least 1 postbaseline efficacy assessment, only in the double-blind phase. * $p < 0.05$ vs. onabotA 20 U by the Cochran Mantel-Haenszel test stratified by center. † Primary time point. Figure reproduced from Joseph et al., 2022 [146] (Creative Commons CC by NC; https://creativecommons.org/licenses/by-nc/4.0/).

Dose response can be assessed not only for efficacy and duration, but also for adverse events, reflecting safety. In preclinical studies, dose response curves generated for different variables have permitted comparison of safety margins among BoNTs. In such studies, the safety margin of onabotA has been reported to be significantly higher than those of abobotA and rimabotB [110,111]. The difference in preclinical safety margins is supported by clinical studies that have found differences in adverse events among these BoNT products [121,153].

6. BoNT Onset of Effect

In clinical use, BoNT/A has been reported to exert noticeable effects within 24 h of intramuscular injection [154,155], with >90% of individuals treated for facial lines reporting effects within 3 days [156]. The onset of BoNT/B is also evident within 1–3 days of intramuscular injection [157,158]. Type E has been reported to have an onset of 1 to 2 days in humans and a more robust effect with a recombinant BoNT/E than abobotA at earlier timepoints [159]. The onset of efficacy (at least 2-grade FWS improvement from baseline) of an investigational BoNTE product (trenibotulinumtoxinE) was demonstrated at 8 h after administration, which was the earliest time point assessed [26].

The faster onset of type E has been suggested to relate to a cellular mechanism, in which it is translocated more rapidly from the endosome into the cytoplasm of neurons where it exerts its action (see Figure 2) [160,161]. The more rapid translocation may be due to BoNT/E's structure, which leads the protein to adopt a position in the endosomal vesicle that permits faster entry into the neuronal cytoplasm [162]. BoNT/E's more rapid onset of action and shorter duration of action (see next section) may be desirable for certain clinical uses such as previewing aesthetic results and inhibiting muscle contractions immediately prior to or following surgical procedures.

7. BoNT Efficacy and Duration of Action

In clinical pharmacology, efficacy refers to a medication's therapeutic effect and duration broadly refers to the length of the therapeutic effect. Several general concepts may be considered in clinical scenarios relevant to efficacy and duration of action of BoNTs: (1) intrinsic factors attributable to the products themselves (e.g., serotype, formulation, and unit potency); (2) peripheral neuron type (e.g., motor, autonomic, and nociceptors); and (3) study-level differences (e.g., patient population, doses, injection paradigm, duration assessments, etc.) (Table 4).

Table 4. Factors that affect BoNT efficacy and duration.

Intrinsic Product Factors
Serotype/subtype Formulation Unit potency
Peripheral neuron type
Motor (alpha and gamma) Autonomic (sympathetic and parasympathetic) Nociceptor
Study-level factors
Patient population Dose Injection paradigm Efficacy and duration assessments Rating scales/raters Follow-up timepoint frequency

7.1. Influence of Intrinsic Product Differences

7.1.1. Serotype/Subtype

The various BoNT serotypes bind to different receptors, which include synaptic vesicle proteins and gangliosides (see Mechanism of Action section) [59]. The distribution and affinity of receptors influence the activity of BoNTs [59]. Within a given BoNT serotype, different subtypes can exhibit different potencies, as has been documented in laboratory models [80].

The fundamental differences in duration among BoNT serotypes are driven by intracellular biology. Duration differences among BoNT serotypes have been well documented in isolated nerve cell cultures [143,163], which are studied under controlled conditions and permit dose response assessments. In such studies, serotypes B and E exhibit a shorter duration than serotype A [143,163]. The shorter durations of serotypes B and E compared with type A have been confirmed in human studies [26,153,159,164].

The mechanism underlying the long duration of type A has been under investigation for several decades and likely involves multiple factors, including (1) a longer persistence of the type A light chain in nerve endings [143,163] and (2) the site at which it cleaves SNAP-25 (synaptosomal associated protein-25 kD) [165].

Persistence of Type A Light Chain

As described in the mechanism of action section of this paper, the light chain of the BoNT protein translocates across the endosomal membrane into the cytoplasm to cleave one or more SNARE proteins (Figure 2). In the cytoplasm, the type A light chain has a long-lasting effect that is not observed with types B or E because it is not degraded as quickly [143].

Although the reason for this slower degradation is not yet fully established, it has been suggested that different intracellular light chain localization may make them differentially accessible for degradation [166]. The light chains of the various serotypes congregate at different regions inside the cell: the type A light chain associates with cytoskeletal proteins (septins) and localizes to the neuronal membrane; the type B light chain is dispersed throughout the neuron; and the type E light chain is located mainly in the cytosol [166–168].

The type E light chain is rapidly marked for degradation by the ubiquitin–proteasome system—one of the main protein degradation systems in the cell—via the attachment of ubiquitin proteins that mark it for elimination [169]. The type A light chain is also ubiquitinated; however, the BoNT/A light chain recruits and directly binds a deubiquitinating enzyme (VCIP135), which removes ubiquitin [170]. This antagonistic relationship between ubiquitination and deubiquitination ultimately slows the protein's degradation [170]. Another deubiquitinating enzyme (USP9X) may also indirectly contribute to prolonging the

longevity of BoNT/A in cells [170,171]. Either or both mechanisms (light chain localization to the membrane and ubiquitination/other intracellular clearance pathways) may be responsible for the slower degradation of the type A light chain and its continued protease activity.

Site of SNAP-25 Cleavage

Studies suggest that at least one additional mechanism may contribute to the longer duration of BoNT/A than/E, namely, the site at which it cleaves SNAP-25. Although BoNT/A and/E both cleave SNAP-25, they target different sites: BoNT/E cleaves SNAP-25 between amino acids 180 and 181, whereas BoNT/A cleaves SNAP-25 between amino acids 197 and 198 [172]. The larger SNAP-25 protein fragment left by BoNT/A (i.e., amino acids 1-197) can continue to incorporate with VAMP and syntaxin into the SNARE complex [173,174]. The SNARE complex with the truncated SNAP-25 fragment generated by BoNT/A cleavage enables vesicle docking at the membrane but does not permit fusion and therefore is not functional—it does not mediate exocytosis [175,176]. Through this mechanism, the truncated SNAP-25 may exert a "dominant negative" effect, preventing any remaining full SNAP-25 protein from forming SNARE complexes and interfering with SNARE functions [33,174]. In contrast, the truncated SNAP-25 fragment generated by BoNT/E cleavage (i.e., amino acids 1–180) is smaller and not does incorporate into SNARE complexes [177].

7.1.2. Formulation and Unit Potency

Even among BoNT products of the same serotype, other product factors such as formulation and units influence efficacy and duration. For instance, in preclinical studies where the effects of other variables are minimized, some BoNT/A products exhibit different durations at the same labeled unit doses [113]. In clinical studies, unit doses of BoNT/A products needed to achieve a comparable duration can vary several-fold [121,124].

7.2. Influence of Peripheral Neuron Type

Duration of effect may depend on the indication and type of peripheral neurons present in the tissue. Following injection into skeletal muscles at labeled doses, BoNT/A products generally exhibit durations of approximately 3 to 4 months in placebo controlled studies [4,7,9,11]. When injected into smooth muscles innervated by autonomic cholinergic nerves for the treatment of incontinence due to neurogenic detrusor overactivity, onabotA (200 U) shows a mean duration of 42 to 48 weeks [4]. In overactive bladder, the median time to re-treatment for onabotA (100 U) is 19 to 24 weeks [4]. Following intradermal injection into the axilla, where glands receive autonomic cholinergic innervation, onabotA (50 U) shows a mean duration of 28.7 weeks [4], with more than 22% of patients reporting a response for at least 52 weeks [178,179].

The long duration of BoNT/A in the bladder has been attributed to a lack of axonal sprouting following injections [180], although this explanation remains theoretical, particularly in view of the preclinical evidence that neuronal sprouts from the alpha motor neurons innervating skeletal muscle are relatively ineffectual for functional contractions [181]. In tissue from individuals with palmar hyperhidrosis, initial axonal sprouting was observed 3 months following BoNT/A injection but was not linked to chemodenervation and was not followed by reinnervation of the sweat gland [182]. The authors interpreted this to suggest that an imbalance between chemodenervation and sprouting in palmar tissue may underlie the long duration observed. This is consistent with other work showing that early neuronal sprouts are poorly functional in terms of neurotransmission [181].

7.3. Influence of Study-Level Factors

In addition to differences in the intrinsic properties of BoNTs and their interactions with various tissue types, differences among BoNTs can be observed at the clinical study level that may or may not reflect differences in the underlying intrinsic properties of

the individual medications. Clinical study-level variables can affect conclusions about the comparability of BoNTs and include the patient population studied (e.g., indication, inclusion/exclusion criteria), differences in doses and treatment paradigms, assessment methods (e.g., rating scales and raters), and follow-up timepoints (Table 4).

7.3.1. Patient Population

Various BoNT/A products are indicated for multiple conditions that differ in their complexity. For instance, several BoNT/A products are indicated for glabellar lines, a common condition that is treated by injecting a defined set of facial muscles. Some BoNT/A products are also indicated for spasticity, a condition that varies in its presentation, with patients experiencing involvement of different combinations of, for instance, finger, elbow, shoulder, ankle, and other limb flexors and extensors. Treatment of spasticity must be individualized, with different doses of each product selected and administered to the muscles involved, in support of a pre-defined treatment goal. Assessing efficacy and duration in complex conditions like spasticity can be challenging and it is difficult to compare between studies.

Even among patients with the same medical or aesthetic condition, variations in severity and muscle mass may affect BoNT pharmacodynamics and consequently efficacy and duration. Weight can be a factor in some studies (e.g., pediatrics) due to weight-based dosing guidelines.

In designing clinical trials with the intent of identifying a responsive population, studies may focus on patients who are the most likely to benefit from treatment. The responsive population may be refined over time as lessons are learned from clinical studies and practice. Consequently, efficacy and duration in early studies may appear to be lower than in later studies in which less responsive groups of patients have been excluded. This is evident in the cervical dystonia literature in which early studies of onabotA and abobotA allowed enrollment of patients with predominant retrocollis [183] and anterocollis [184], which are more difficult to treat with BoNTs due to their complexity and the involvement of deeper or poorly accessible muscles [185,186]. More recent studies with incobotA [187] and daxibotA [152] excluded these patients. This natural progression in trial populations means that later studies can be enriched with patients who are more likely to respond to treatment and may result in more favorable outcomes such as a greater improvement in scores or a longer duration of effect.

Another example in which the patient population can influence efficacy and duration outcomes can be found in the glabellar lines literature, with some studies including a higher population of subjects with moderate as opposed to severe lines at baseline. An outcome of a none or mild on a glabellar lines rating scale will be easier to achieve over a longer period in subjects with less severe lines at baseline. Other endpoints such as a 2-point improvement on a glabellar lines rating scale may favor subjects with severe lines at baseline. Potential bias can be avoided by enrolling an equal proportion of subjects with moderate and severe lines at baseline, but different enrollment strategies may be used based on the experimental question.

7.3.2. Dose Differences

Units of BoNT products are not interchangeable and it can therefore be difficult to determine the comparability of doses across studies. Dose differences are relevant because efficacy and duration increase with dose up to a maximal point (see Dose–response section). As noted previously, in placebo controlled studies at labeled doses, BoNT/A products generally exhibit durations of approximately 3 to 4 months following injection into skeletal muscles [4,7,9,11]. In a dose-ranging study of daxibotA for the treatment of glabellar lines, the median durations of a $\geq$1-point improvement from baseline on the Investigator Global Assessment-Facial Wrinkle Scale were 20.0 weeks with 20 U, 23.6 weeks with 40 U, and 20.9 weeks with 60 U [150]. The 40 U dose of daxibotA was selected for phase 3 development in glabellar lines because it had the most favorable risk:benefit profile [150].

Increasing the doses of several other BoNT/A products increases their durations of action, as documented for glabellar lines (see Dose Response section) [146,147,149], and cervical dystonia [188], with the exception of the 150 kDa products incobotA [9] and daxibotA [152] at the doses tested in registration clinical trials for cervical dystonia.

7.3.3. Treatment Paradigms

Prospective clinical studies typically have highly structured protocols that may not reflect clinical practice. This includes treatment paradigms, which are often standardized for the particular BoNT and indication under study.

Given that units of BoNT products are not interchangeable and each product has different physicochemical properties due to its unique formulation, the doses and muscles injected can vary from one product to the next. For instance, the doses, numbers, and locations of recommended muscles and injection sites in upper limb spasticity are different for onabotA [4] and abobotA [7], making it difficult to design studies with protocols optimized for each. These differences are confirmed by a recent analysis of real-world studies in upper limb spasticity that found that the two products were not only injected at different doses, but also at different ratios of doses per muscle, indicating that the products are not used at a set dose ratio in clinical practice [189]. This study of adult post-stroke patients also found that a higher number of muscles were injected with onabotA than abobotA [189]. These differences are underscored by the different injection paradigms outlined in the product labels, which for onabotA includes 13 potential muscles to be injected in upper limb spasticity [4] and for abobotA includes 9 potential muscles [7]. Given these differences, comparing the products in spasticity using the same injection paradigm may favor one product over another. For instance, a recent study uses the same injection sites for both onabotA and abobotA in upper limb spasticity, limits injection sites to 5 pre-identified muscles for both, and avoids the finger flexors [190].

7.3.4. Assessments of Efficacy and Duration

Efficacy and duration of response can be assessed using many different methods that vary from statistical estimates to more clinically relevant improvement that is important to patients. Efficacy may be expressed in terms of responders at various timepoints throughout the study, which can then be used to describe duration. Different assessments and definitions of response often give different estimates of efficacy and duration, underscoring the importance of carefully considering the assessments that underlie the reported outcomes in each individual study.

The glabellar lines literature provides an example of the many different definitions that have been used to define a responder, which in turn affects the estimated duration that is based on responder rate. For instance, a study of abobotA specified response as days to return to grade 2 (moderate) or 3 (severe) on a 4-point categorical scale [147], whereas a study of incobotA specified time to return to baseline [191]. Studies of incobotA [149], daxibotA [150], and onabotA [192] specified at least a 1-point improvement from baseline as the definition of response.

Duration can be measured as point estimates of responder rates at study visits or through statistical analyses such as the Kaplan–Meier method. In the aforementioned glabellar lines studies, responder data were estimated using the Kaplan–Meier method, a time-to-event analysis. In the context of BoNT duration estimates, the defined event is loss of response based on the definition of a responder used in the study. The Kaplan–Meier plots for BoNT durations are graphical representations that use horizontal lines to show the probability of maintaining response and vertical lines to show loss of response. Duration in Kaplan–Meier analyses is often reported as the median—the timepoint at which the likelihood of continuing to respond (i.e., not experiencing the event—loss of response) is 50%. Like all measures, the Kaplan–Meier analysis has some limitations, and its use in the context of BoNT duration has been questioned by the FDA because different criteria may be used to define the endpoint event [8]. Another limitation of the Kaplan–Meier in the

context of BoNT duration studies is that it only includes subjects who were considered responders at a given timepoint early in the study (e.g., 4 weeks post-treatment) and excludes non-responders. The method is additionally limited by the frequency of study visits, such that subjects are assumed to be responders until the visit date at which they did not meet the response criteria. This can overestimate the duration of response for subjects who lose response between visits because they are considered responders until their next visit. Additionally, it is important to verify that the definition of clinical response applied to the duration analysis is the same as that used for determining efficacy. However, an important advantage of the Kaplan–Meier method is that it captures data from patients who left the study (known as censored), which are typically shown as tick marks or dots on the horizontal lines of the graph.

Different assessments of duration are also evident in the spasticity literature, making it problematic to compare across studies. For instance, time to re-treatment has often been used as an estimate of duration but can itself be defined in various ways. In clinical practice, patients typically request retreatment prior to returning to baseline and injection sessions are routinely scheduled a certain number of months apart. Thus, time to re-treatment does not necessarily measure continued clinical effect, even though it can be useful for some purposes. In the setting of a clinical trial, a more systematized framework is generally required for a structured analysis. In the phase 3 study of abobotA for upper limb spasticity, subjects qualified for re-treatment only when their scores on the modified Ashworth scale returned to baseline [193]. Other studies have required that subjects meet two assessment criteria instead of one, as in the case of a daxibotA phase 2 study for spasticity in which subject scores must have returned to baseline on the modified Ashworth scale and to zero or less on the Physician Global Impression of Change scale, or until the subject requested retreatment [194]. Dual criteria such as these are more difficult to meet and prolong the recorded duration.

Overall, the many different assessment methods for evaluating efficacy and duration of BoNT clinical effects necessitate careful inspection of the criteria used to define response. The different assessment methods further contribute to the difficulty comparing duration across studies. The statistically driven Kaplan–Meier analysis provides different insights into clinical trial outcomes than, for instance, time to retreatment, which itself provides different insights than return to baseline on a clinical rating scale. The assessments may be useful for different purposes but care must be taken not to directly compare data derived from such different methods.

7.3.5. Rating Scales and Raters

The rating scale utilized is another study-level variable that can affect duration results. An example is from the glabellar lines literature, in which most BoNT products have been assessed on a 4-point categorical rating scale refined from the original Facial Wrinkle Grading System published by Keen, Blitzer, Brin, and others in 1994 [195]. The current 4-point scales were developed for registration purposes and are accompanied by proprietary, validated photonumeric guides used to inform the facial wrinkle ratings. Given that each of these guides uses different photos to define their ratings, they are not identical, making it difficult to draw conclusions about BoNT comparability across studies. In the therapeutic literature, different scales have been used to evaluate cervical dystonia (e.g., Cervical Dystonia Severity Scale [183], Toronto Western Spasmodic Torticollis Scale [187]) and spasticity (e.g., Ashworth scale [196], modified Ashworth scale [190], modified Tardieu scale [197], individual goal attainment scales [198]).

Across aesthetic and therapeutic uses, ratings may be performed by investigators who administer the injections, independent raters, and/or by the subjects themselves. The latter are included based on a growing recognition of the importance of patient perception of treatment effects and the lack of complete concurrence between patients and clinicians. Some studies measure duration based on the ratings of either investigators or subjects, whereas others use a composite of both investigator and subject ratings [199]. If duration is

estimated based on loss of response on only one rating scale, it is likely to be shorter than if duration is estimated based on loss of response on two ratings scales or across 2 raters (i.e., composite endpoint), due to the lack of complete concurrence in ratings. Again, this complicates comparison of duration across studies.

7.3.6. Follow-Up Timepoints

Follow-up timepoints also differ in BoNT studies, with variations in both number of timepoints assessed, time between assessments, and the duration of follow up. Studies designed with numerous follow-up timepoints enhance the precision of the comparison, regardless of the assessment method used [5].

7.4. Summary of Study-Level Variables

Comparing efficacy and duration from different studies, particularly if they used different rating scales and assessments, can be misinterpreted as one product having higher efficacy or a longer duration than another. Study-level variables can affect conclusions not only about efficacy and duration, but also about onset, adverse events, and other outcomes. These challenges are likely to intensify as more BoNT products enter the market.

Prospective studies designed to directly compare BoNT products are preferable to comparing across different studies because, in the former, the interventions are subject to the same protocol, tested in the same subject populations, and evaluated using the same outcome measures. However, prospective comparison studies have challenges of their own. In such studies, it can be difficult to select comparable doses of BoNT products given that units are not interchangeable and BoNT onset and duration vary with dose. Treatment paradigms, including number and location of muscles injected, vary between BoNT products, making it difficult to design studies with protocols optimized for each BoNT product. As noted above, these studies often have highly structured protocols that do not reflect clinical practice.

Although randomized, double-blind trials have the advantage of minimizing the effects of pre-existing differences and expectation on outcomes, designs can differ based on the types of conclusions that the authors seek to draw, such as non-inferiority or superiority studies. Non-inferiority trials are designed to show that the effect of one treatment is not inferior to that of an active comparator treatment by more than a specified statistical margin [200]. Conclusions about equivalence, or equipotency, cannot be made based on non-inferiority clinical trials not designed for that purpose, although such claims have erroneously been made in the BoNT literature [137,201,202].

Overall, study-level variables can influence conclusions about the comparability of BoNTs that may or may not be due to intrinsic product-level differences. It can be difficult to determine the source of the differences observed and it is important for clinicians to be aware that such ambiguity exists. Table 5 lists study-level variables to consider in comparing efficacy and duration of different BoNTs.

Table 5. Study-level variables to consider in comparing efficacy and duration of different BoNTs.

Patient population	Are there differences in clinical presentation, severity, or duration of disease, or of pre-existing conditions/comorbidities? Is a more or less responsive group included or excluded? Disease severity/complexity may influence overall efficacy, which can influence efficacy and duration.
Doses	Does the study account for dose–response effects in comparing products, in addition to non-interchangeability of units when evaluating efficacy and duration? Higher doses may lead to increased efficacy and longer durations.
Injection paradigm	Are the muscles and injection sites optimized for each of the BoNT products?
Efficacy and duration assessments	Are the definitions of efficacy and duration the same/comparable for the different products? For example, a definition that requires two raters to agree on an outcome is more difficult to achieve and will lead to longer duration than that of a single rater.
Rating scales/raters	Are the same rating scales being used? Who is doing the rating (e.g., investigator, subject)? Different scales and raters may yield different apparent responder rates and durations. Patient perception is an important outcome.
Follow-up timepoints	Is the number of timepoints adequate to provide a full assessment of duration? More follow-up timepoints give a more precise estimate of duration.

8. BoNT Safety and Adverse Events

The approved BoNT products are generally well-tolerated at approved doses. Adverse events differ based on indication and may be due to relaxation of the treated target muscle, local diffusion away from the injection area, or spread distant from the site of injection. Diffusion of BoNT products is based on intrinsic, product-level characteristics and has been discussed in the literature; readers are referred to several reviews [5,203]. Product-level local and distant diffusion characteristics influence the safety margin in preclinical models and adverse event profile in clinical situations [5].

In addition, in clinical practice, safety is linked to the dose administered. Unit potency, also an intrinsic BoNT product factor, is therefore critical to product safety: Dose confusion among products can have serious consequences for patients such that doses that are too low will not produce optimal efficacy and doses that are greater than desired can increase the risk of adverse events.

As described for efficacy and duration, extrinsic study-level factors can influence adverse event rates for BoNT products. An example of study-level factors affecting adverse event rates in clinical trials can be found in the cervical dystonia literature, where patients with pre-existing dysphagia are sometimes included or excluded from studies. Given that pre-existing dysphagia has been found to persist after BoNT treatment [204], excluding such patients can lead to a lower rate of post-treatment dysphagia obtained in the study. Another example comes from the neurogenic detrusor overactivity literature. A proportion of patients with this condition require clean intermittent catheterization to completely drain urine from the bladder and help prevent infection. Studies that include only patients who require catheterization at baseline do not count the need for catheterization as an adverse event [205]. In contrast, studies that include patients who do not catheterize at baseline report new catheterization as an adverse event [206,207]. These observations illustrate that, when comparing across clinical studies, it is important to consider the inclusion criteria of the study population and whether this may have influenced the adverse event rates obtained.

9. Immunogenicity

All foreign proteins injected into the body have the potential to induce the development of antibodies. In the case of BoNTs, the development of antibodies that interfere with clinical response is an uncommon occurrence [208–213], likely due to the extremely high

potency and low amounts of protein injected. It will nevertheless be important to monitor the immunogenicity of newer BoNT products.

Antibodies that develop in response to protein therapies such as BoNTs are classified as neutralizing—meaning that they interfere with the drug's action—and non-neutralizing—meaning that they do not interfere with the drug's action. In some cases, antibodies that develop against the BoNT protein can be neutralizing, whereas antibodies that develop against the NAPs are non-neutralizing [214]. Some tests assess both neutralizing and non-neutralizing antibodies (e.g., the enzyme-linked immunosorbent assay [ELISA]), whereas in vivo tests such as the mouse protection assay assess only neutralizing antibodies [214,215].

9.1. Factors Affecting Neutralizing Antibody Formation

Over the years, numerous studies have evaluated factors that affect neutralizing antibody formation with the three established type A products (onabotA, abobotA, incobotA) and these have been reviewed [208]. Past studies found that the incidence of neutralizing antibody formation increased with the cumulative dose and number/frequency of injection visits [216,217]. However, in more recent studies, few subjects developed neutralizing antibodies regardless of number of treatment cycles or indication, suggesting that current treatment practices—which have been informed by the aforementioned past studies—contribute to the current low rates [208–213].

A limited number of studies have reported that switching to different BoNT/A products is a risk factor for neutralizing antibody formation [218,219]. Another study reported that switching secondary non-responders from one BoNT/A preparation to another reinstituted at least a partial response [220]. Given the extremely low rates of neutralizing antibody formation with the established BoNT/A products and the variability in responses even in patients who have developed neutralizing antibodies, potential reasons for these partial responses are that the doses and muscles involved were re-evaluated and updated or that patients' titers varied over time, resulting in clinical non-response after one series of injections and clinical response after another.

9.2. Secondary Non-Response: Usually Not Due to Neutralizing Antibodies

The term secondary nonresponse has been used to describe inadequate or non-response to a pharmacological intervention following previously successful treatment. Secondary non-response can refer to a lack of pharmacological response to the medication or a suboptimal clinical response for other reasons. However, lack of perceived response to BoNTs is not usually due to a lack of pharmacological effect resulting from antibodies [212,213,221]. Instead, it is typically caused by inadequate dose, inappropriate muscle selection, complex movement patterns, and/or dynamic disease changes (e.g., contractures, abnormal postures, change in pattern of muscle contractions) [212,221,222]. A small study of cervical dystonia patients found that nonoptimal doses and muscles injected were the top two causes of secondary non-response [221]. Thus, an important first step in managing secondary non-response to BoNTs is to re-assess the doses and muscles injected.

Patient perceptions may also be a factor in secondary nonresponse. Patients sometimes interpret partial response as nonresponse [221]. Such patients may experience a therapeutic response but do not believe they are responding due to expectations. Patients may not recall how severe their condition was at baseline. After the first treatment, improvements from baseline are typically very evident to patients but improvements between the subsequent treatments may not be as noticeable because the change is not the same magnitude, as they did not return to baseline between treatments. Although secondary non-response to BoNTs can also be caused by the development of neutralizing antibodies, the vast majority of patients do not have them [213,221,223].

Additionally, the presence of neutralizing antibodies does not always lead to non-response. Studies have repeatedly shown that patients with neutralizing antibodies frequently show continued clinical response to BoNTs [213,218,224,225]. A study that followed

2240 patients who received up to 16 treatments with onabotA found that 11/2240 (0.49%) converted from antibody negative at baseline to positive at one or more post-treatment time points, but only three were clinically non-responsive at some point following a positive neutralizing antibody test [224]. This study also showed that neutralizing antibody status can vary over time, as only 4/2240 had a positive test at the final post-treatment study visit. A change in antibody status over time has also been reported by others [213,215,226] and, notably, in a recent meta-analysis of 5876 subjects treated with onabotA across 10 therapeutic and aesthetic indications in which 0.5% developed neutralizing antibodies at some point but only 0.3% remained positive at the end of the study [213].

9.3. Clinical Implications of Neutralizing Antibody Formation?

There is an imperfect relationship between neutralizing antibodies and clinical non-response [210,213,224,225]. Based on manufacturer's sponsored clinical trials, doses, and injection intervals, the rates of neutralizing antibody formation with the established BoNT/A products are low: reported as 0% with onabotA in lateral canthal lines [3] and 1.2% in cervical dystonia [4]; 0% with abobotA in glabellar lines and less than 3% in cervical dystonia [7]; 0% with incobotA in glabellar lines and 1.8% in cervical dystonia [9]; 0% with daxibotA in glabellar lines and 0.5% in cervical dystonia [12]; and 0.14% with prabotA in glabellar lines [11]. The rate of neutralizing antibody formation with rimabotB is reported as 18% or less of cervical dystonia patients in the registration studies [13].

Several recent studies that have compared rates of neutralizing antibody formation across different BoNT products are retrospective chart reviews that were not designed for direct comparison. For example, different BoNT products were administered for different durations in at least one study [227], whereas others had large differences in patient numbers per group [218,219]. Moreover, doses were not controlled in these retrospective studies. These limitations preclude conclusions about relative immunogenicity among the different BoNTs.

Overall, the rates of neutralizing antibody formation with the established BoNT/A products are low with current labeled treatment recommendations.

10. Summary

An increasing number of BoNTs are currently available and in clinical development, some of which include innovations in serotype and formulation. BoNT products are not interchangeable due to differences introduced at each step of the manufacturing process: bacterial strain, fermentation, purification, excipients, finishing, and unit potency testing, all of which affect clinical profile. Of paramount importance for clinicians, units of BoNT products are not interchangeable due to differences in the assays used to measure unit potency, including different potency reference standards. Each BoNT has its own dosing information based on clinical studies in each indication; there are no established fixed inter-product dose ratios. Understanding the unique features of each BoNT is essential to optimizing patient experience, including efficacy, safety, and patient satisfaction.

In addition to the aforementioned intrinsic product-level differences between BoNTs, study-level differences contribute to the variability among products. Study outcomes such as efficacy and duration depend critically on the assessments and definitions of response. Moreover, all BoNT products exhibit dose responses—an observation that must be considered when comparing clinical properties such as duration. These study-level differences compound the intrinsic product-level differences, leading to unique clinical characteristics for each BoNT (Figure 7).

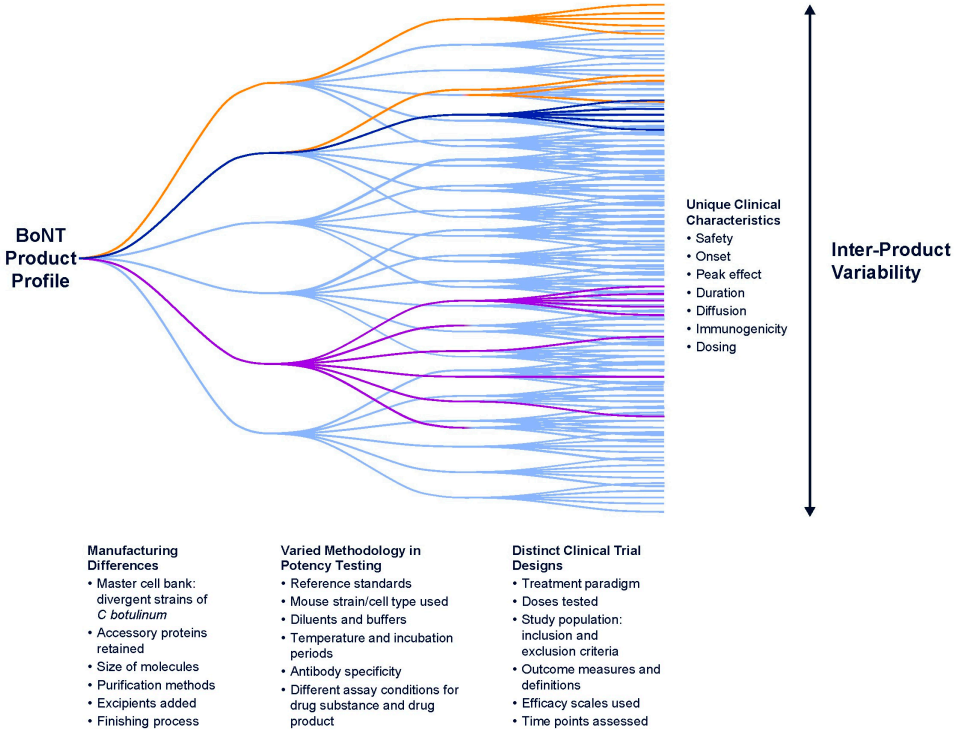

Figure 7. Lack of interchangeability among BoNT products. This graphic illustrates the divergence of BoNT products from manufacturing, through unit potency testing, to clinical trial designs. Individual lines illustrate the multiple points of divergence, with the highlighted orange, purple, and blue lines exemplifying different paths that products may take throughout the process. The intrinsic, product-level differences observed in the manufacturing and unit potency testing stages are compounded by study-level differences in clinical trial designs that ultimately lead to a unique set of clinical characteristics for each BoNT product.

Non-interchangeability of BoNT products is recognized by regulatory agencies in major markets worldwide, which require a statement in the labels of all approved BoNT products indicating that units are not interchangeable or convertible among different BoNT products. To reinforce the individual potencies of BoNT products and prevent medication errors, the US Food and Drug Administration (FDA) requires each BoNT product to have its own unique nonproprietary name [228,229].

Given the non-interchangeability of BoNTs, issues related to non-medical switching among products take on added importance. As new BoNT manufacturers enter the market and existing manufacturers continue to negotiate pricing with institutions, governments and insurers, patients may be increasingly compelled to switch to products that are less expensive. This non-medical switching can disrupt the benefits of ongoing treatment, requiring clinicians to alter doses and injection sites to re-establish stable regimens for each patient, particularly because there are no fixed inter-product dose ratios. Such switching can also increase the potential for medical errors, adverse events, and cessation of treatment [230].

Overall, the growing number of BoNT products available or in development make this an intriguing time for BoNT therapy. This also includes emerging BoNT products with formulations designed for different onset of action and/or duration. The unique properties across this category of therapeutics highlight the importance for clinicians to recognize that each BoNT must be used according to its own specifications as supported by clinical studies, which will help decrease the potential for unexpected adverse events and maximize efficacy, duration, and patient satisfaction. With these considerations in mind, BoNT therapy has an exciting future of helping an increasing number of individuals achieve their treatment goals.

Author Contributions: Conceptualization, M.F.B., M.N. and N.A.; methodology, M.F.B., J.M., M.N., N.A. and A.B.-A.; data curation, M.F.B., J.M., M.N., N.A. and A.B.-A.; writing—original draft preparation, M.F.B., J.M., M.N., N.A. and A.B.-A.; writing—review and editing, M.F.B., J.M., M.N., N.A. and A.B.-A.; visualization, M.F.B., J.M., M.N., N.A. and A.B.-A.; project administration, M.F.B. and M.N. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: This review was based on publicly available sources as cited in the reference list. No new data were created as part of this review.

Acknowledgments: Writing assistance was provided to the authors by Mary Ann Chapman, Visage Communications, and was funded by AbbVie. The authors gratefully acknowledge the following individuals who reviewed and provided comments on this manuscript: Martin Gastens, Carsten Jahn, Hemant Patel, David Rupp, Claire Spronk, and Lance Steward. The authors thank Levent Efe, CMI, for the development of Figure 2, funded by AbbVie. Data visualization direction and figure development assistance for Figures 4 and 7 were provided by Justin Sodano and Ashley Wirsing of Peloton Advantage, LLC (an OPEN Health company) and funded by Allergan Aesthetics, an AbbVie Company.

Conflicts of Interest: Mitchell F. Brin, Mariana Nelson, Amy Brideau-Andersen and John Maltman are employees of AbbVie and receive stock as part of their compensation. Nazanin Ashourian is an employee of HireGenics and consultant of AbbVie. Nazanin Ashourian is a former employee of AbbVie and may hold AbbVie stock.

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EXHIBIT 6 to Brin Declaration

J Neural Transm (2009) 116:437–441
DOI 10.1007/s00702-009-0200-6

BASIC NEUROSCIENCES, GENETICS AND IMMUNOLOGY - ORIGINAL ARTICLE

Experience with long-term treatment with albumin-supplemented botulinum toxin type A

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Received: 29 September 2008 / Accepted: 12 February 2009 / Published online: 25 March 2009

Abstract In earlier studies, we have demonstrated the efficacy of albumin-supplemented botulinum toxin type A (ASBTA) in principle. Here, we present long-term data from 106 patients who received ASTBA over 5–10 years for the treatment of cervical dystonia, blepharospasm and hemifacial spasm. Vials of Dysport® were diluted in 0.1% albumin solution to a concentration of 25 units/ml. Overall patients and indications, the mean latency to response was 7.1 ± 2.2 days, the mean duration of response was 12.3 ± 3.1 weeks and the mean global clinical improvement (scale 0–3) was 2.6 ± 0.2. Only one patient had neutralizing antibodies against BoNT-A. Side effects were less frequent than known for conventional BoNT-A and generally mild. These findings were confirmed by analysis of data of 71 patients who have been reconverted from ASBTA to conventional dilutions of Dysport® or Botox®. We conclude that long-term treatment with ASBTA is effective, safe and help to reduce costs.

Keywords Botulinum toxin type A · Cervical dystonia · Blepharospasm · Hemifacial spasm · Low-dose therapy

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Introduction

BoNT-A binds specifically to cholinergic nerve terminals and inhibits acetylcholine release by interfering with vesicle docking proteins. These effects make BoNT-A useful for the treatment of disorders with muscular and glandular hyperactivity such as dystonia, spasticity, hyperhidrosis, and sialorrhea (Giess et al. 2000; Naumann 2001; Jost 2006).

Unwanted side effects and the likelihood of immunoresponses with antibody production may be reduced by low-dose application of BoNT-A (Goschel et al. 1997; Wohlfarth et al. 1997). Several studies suggest that lower toxin doses than usually advocated may be sufficient to provide satisfying improvement (D'Costa and Abbott 1991; Viriyavejakul et al. 1998; Laubis-Herrmann et al. 2002; Suputtitada et al. 2004; Heckmann and Plewig 2005; Suputtitada and Suwanwela 2005). Therefore, approaches allowing a dose reduction without decrease of therapeutic efficacy would be desirable. We could recently show that an appropriate way to reduce BoNT-A doses without losing efficacy is supplementation by human albumin giving satisfactory results in cervical dystonia (CD), blepharospasm (BSP) and hemifacial spasm (HFS) over 2 years (Rollnik et al. 2000). This has been confirmed in a single-blinded, crossover study using albumin-supplemented BoNT-A (ASBTA) in different dilutions and in patients with various forms of dystonia (Bigalke et al. 2001). Since the close of this study in 1996, ASBTA treatment has been continued in patients who wished to retain it and in a small number of newly recruited patients. We can now present a long-term follow-up of 106 patients receiving ASBTA over periods of 5–10 years for CD, BSP, and HFS. We also present data of 71 patients who, according to their wish, had been reconverted from ASBTA to conventionally diluted Dysport® or

Botox®. To the best of our knowledge, comparable information has not been reported to date.

Patients and methods

Patient data were retrieved from the database of our movement disorders clinic containing BoNT-A treatment details of all patient contacts such as commercial product, type of dilution, doses, efficacy and side effects as indicated by the patients. Data of 177 patients were evaluated (see Table 1). All patients were treated with ASBTA for at least 5 years. 110 patients had participated in a controlled study closing in 1996 (Rollnik et al. 2000) and 67 patients were included afterwards. Five patients were lost to follow-up because they have moved to other cities. During the study period more than 100 new patients were treated with full-dose Dysport® and thus were not included in the present analysis.

Group I consisted of 106 patients with CD ($n = 62$), BSP ($n = 25$) or HFS ($n = 19$) treated with ASBTA for the whole time. For all the patients, vials of Dysport® containing 500 MU BoNT-A were reconstituted with 20 ml saline supplemented with 0.1% human serum albumin resulting in a concentration of 25 units/ml. Forty-two of these patients had been on conventional BoNT-A treatment when they entered the study with ASBTA and 64 received it as primary treatment. ASBTA treatment was approved by our ethical committee. Patients were informed of the special preparation of BoNT-A different from the manufacturer's recommendation and gave written informed consent prior to each treatment session.

Group II consisted of 71 patients with CD ($n = 50$), BSP ($n = 11$) or HFS ($n = 10$) who, according to their wish, were stepwise reconverted from ASBTA to conventionally diluted Dysport® or Botox®. All these patients wished the reconversion since they had preferred to be treated according to the manufacturer's recommendations.

Electromyographic (EMG) guidance was generally used in CD with injections performed at sites of maximum EMG activity. In BSP and HFS, injections were carried out without EMG. BoNT-A dose per treatment and per treated muscle, global clinical improvement (GCI), side effects, latency to response and duration of response were recalled from our database. The latency to response was measured as time (days) between the treatment and first significant improvement of symptoms rated by patients and was asked at each visit. The duration of response was calculated as time between the first improvement after injection and reappearance of symptoms. This period was also asked at each visit and was expressed in weeks. The GCI which resembled measures described and used by other groups (Jankovic and Schwartz 1995; Rollnik et al. 2000; Mejia et al. 2005) and consisted of a 0–3 scale (0 = no effect, 1 = slight, 2 = moderate and 3 = marked improvement in severity and function) was rated by the patients at each contact. BoNT-A doses were redefined at each treatment according to the efficacy of the last injections, duration of response and eventual side effects. All patients who reported a less than satisfactory response to their BoNT-A injections (peak effect rating of 0 or 1) on two consecutive visits were tested for neutralizing antibodies (NAb) in the Institute of Toxicology, Medical School Hannover, using the mouse diaphragm assay (Goschel et al. 1997). Statistical analysis of 1,231 treatments was carried out using SPSS v 14.0 (SPSS, Chicago, IL).

Table 1 Demographic data of patients

Diagnosis	Number	Gender (female/male)	Age in years (mean)
Group I			
Cervical dystonia	62	38/24	24–85 (59 ± 14)
Blepharospasm	25	20/5	47–92 (70 ± 11)
Hemifacial spasm	19	14/5	39–90 (69 ± 12)
Group II			
Cervical dystonia	50	30/20	28–74 (60 ± 11)
Blepharospasm	11	8/3	51–92 (72 ± 12)
Hemifacial spasm	10	7/3	39–87 (67 ± 10)

Data are given as mean value ± SD

Results

Group I

For all patients and indications, the mean duration of ASBTA treatment was 8.1 years (range 5–10 years). The mean latency to response was 7.1 ± 2.2 days, the mean duration of response was 12.3 ± 3.1 weeks and the mean GCI was 2.6 ± 0.2. Four patients (3.7%) became secondary non-responders, only one of these patients (0.94%) showed neutralizing antibodies against BoNT-A. Details for the different indications are provided in the following sections (Table 2).

Sixty-two patients with CD were treated with ASBTA. Injections were applied to splenius capitis, sternocleidomastoid, trapezius, and levator scapulae muscles. The mean total dose for all muscles was 184 ± 59 units, the doses for the single muscles are listed in Table 3. The mean latency to response was 6.2 ± 4.1 days, the mean duration of response was 11.3 ± 4.2 weeks with a mean GCI of 2.5 ± 0.3. Side effects included dysphagia (3.5% of

Table 2 Treatment data of ASTBA patients (group I) for all indications (mean value ± SD)

Diagnosis	Mean dose (units)	Latency to response (days)	Duration (weeks)	GCI
Cervical dystonia ($n = 62$)	184 ± 59	6.2 ± 4.1	11.3 ± 4.2	2.5 ± 0.3
Blepharospasm ($n = 25$)	50 ± 25	6.4 ± 4.2	13.7 ± 7.3	2.6 ± 0.4
Hemifacial spasm ($n = 19$)	25 ± 12	8.2 ± 3.1	11.2 ± 4.3	2.5 ± 0.3

Table 3 Treated muscles in cervical dystonia with corresponding ASBTA-dosage

Muscle	Mean dose	Range
Splenius capitis muscle	72 ± 35	25–150
Levator scapulae muscle	43 ± 15	2–75
Trapezius muscle	49 ± 14	25–75
Sternocleidomastoid muscle	28 ± 10	12.5–50

treatments), neck muscle weakness (2.5%) and pain at the injection site (1.5%), which were not severe and did not require treatment, especially not in the case of dysphagia.

Twenty-five patients with BSP were treated with ASBTA. The orbicularis oculi muscles were bilaterally injected with a mean dose of 13 units (range 7.5–20) BoNT-A per injection site giving a mean total dose of 50 ± 25 units. The mean latency to response was 6.4 ± 4.2 days, the mean duration of response was 13.7 ± 7.3 weeks. The mean GCI was 2.6 ± 0.4 and was fairly stable over time. Relevant dose changes were not necessary. The rate of documented side effects was low and included ptosis (1%) and dry eye (0.8%).

Nineteen patients with HFS were treated with ASBTA. The orbicularis oculi and the orbicularis oris muscle were injected with mean doses of 18 units (range 5–25) and 7.5 units (range 5–10), respectively giving a mean total dose of 25 ± 12 units. The mean latency to response was 8.2 ± 3.1 days and the mean duration of response 11.2 ± 4.3 weeks. The mean GCI was 2.5 ± 0.3 and was fairly stable over time. Side effects included ptosis (0.7%), dry eye (0.9%) and facial weakness (0.7%).

Group II (reconversion)

Seventy-one patients were reconverted to conventionally diluted Dysport® ($n = 47$) or Botox® ($n = 24$) after having been treated with ASBTA for 3–5 years. Data of these patients were compared between ASBTA and the subsequent conventional BoNT-A treatment after reconversion. Taking all indications together, the GCI was very similar for ASBTA (GCI: 2.5 ± 0.3), conventional

Table 4 Treatment data for group II, reconversion from: (a), (b)

	Cervical dystonia ($n = 40$)	Blepharospasm ($n = 3$)	Hemifacial spasm ($n = 4$)
(a) ASBTA to Dysport®			
Mean dose (ASBTA/Dysport®) (units)	203 ± 66/445 ± 135	46 ± 19/102 ± 28	23 ± 8/61 ± 9
Ratio (ASBTA/Dysport®)	1:2.3 ± 0.7	1:2.4 ± 0.3	1:2.6 ± 0.2
Side effects (ASBTA/Dysport®) (%)	1.6/5.2	1.5/5.1	1.3/4.8
Latency to response (ASBTA/Dysport®) (days)	7.5 ± 0.8/7.3 ± 1.2	7.1 ± 0.6/7.2 ± 0.8	7.0 ± 0.9/7.5 ± 1.1
Duration (ASBTA/Dysport®) (weeks)	12.5 ± 0.7/11.2 ± 0.3	11.9 ± 0.6/12.4 ± 0.7	11.8 ± 0.5/12.2 ± 0.9
GCI (ASBTA/Dysport®)	2.4 ± 0.4/2.5 ± 0.2	2.7 ± 0.5/2.7 ± 0.3	2.5 ± 0.3/2.6 ± 0.4
	Cervical dystonia ($n = 10$)	Blepharospasm ($n = 8$)	Hemifacial spasm ($n = 6$)
(b) ASBTA to Botox®			
Mean dose (ASBTA/Dysport®) (units)	141 ± 59/181 ± 58	37 ± 11/48 ± 13	21 ± 6/31 ± 5
Ratio (ASBTA/Dysport®)	1:1.4 ± 0.3	1:1.3 ± 0.2	1:1.5 ± 0.3
Side effects (ASBTA/Dysport®) (%)	1.5/4.6	1.6/4.9	1.1/4.5
Latency to response (ASBTA/Dysport®) (days)	8.3 ± 0.5/7.9 ± 1.0	7.5 ± 2.1/7.1 ± 1.3	6.8 ± 1.5/7.2 ± 0.9
Duration (ASBTA/Dysport®) (weeks)	12.6 ± 0.8/12.1 ± 0.7	12.5 ± 2.1/12.2 ± 1.7	12.9 ± 1.8/11.8 ± 0.5
GCI (ASBTA/Dysport®)	2.6 ± 0.2/2.5 ± 0.3	2.4 ± 0.3/2.7 ± 0.4	2.6 ± 0.2/2.4 ± 0.2

Data are given as mean value ± SD

Dysport® (GCI: 2.4 ± 0.2) and Botox® (GCI: 2.3 ± 0.3). The latency to response showed no significant difference between ASBTA (6.9 ± 1.8 days), Dysport® (7.2 ± 2.5 days) and Botox® (7.1 ± 3.1 days). The duration of response was also very similar, 12.1 ± 2.8 weeks for ASBTA, 11.5 ± 1.9 weeks for Dysport® and 11.9 ± 2.4 weeks for Botox®. Side effects were reported in 1.5, 4.7 and 4.4% of treatment sessions with ASBTA, Dysport® and Botox®, respectively and were significantly less frequent with ASBTA than with the other two applications ($P < 0.001$). The above general statements hold also true if the single diagnoses are considered separately. Details are listed in Table 4a, b.

Discussion

The efficacy of ASBTA has already been demonstrated in our previous open label, non-controlled studies (Rollnik et al. 2000; Bigalke et al. 2001). The core finding of the present retrospective analysis is that ASBTA is reliably effective also in the long-term use over periods of up to 10 years. Although this has been shown only for patients with CD, BSP and HFS it appears reasonable to assume that ASBTA would be similarly effective also in other conditions treatable by BoNT-A. Moreover, we used Dysport® to produce ASBTA and comparable studies using other commercial BoNT-A products are, to our knowledge, not available. Dose reductions might be possible; however, also for other brands.

The essential aspect of ASBTA treatment is that it is a low-dose treatment allowing reduction of BoNT-A doses to less than a half of those applied with conventional dilutions of Dysport®. The mechanisms allowing dose reduction are still not well understood. In a previous study (Bigalke et al. 2001) we have concluded that albumin supplementation may prevent toxin loss caused by adhesion to the syringe walls. The mean dose of ASBTA in CD was 184 units and compares with doses of conventional Dysport® in other long-term observations between 500 units (recommended starting dose) (Poewe et al. 1998; Truong et al. 2005) and 1,072 units (Kessler et al. 1999; Haussermann et al. 2004; Marchetti et al. 2005). The dose relationships are similar for BSP (Marchetti et al. 2005; Bhidayasiri et al. 2006) and HFS (Elston 1992; Yu et al. 1992; Van den Bergh et al. 1995; Jitpimolmard et al. 1998) considering the literature. For our clinic, the relation between ASBTA and conventional Dysport® expressed in units was around 1–2.5 for all diagnoses. The relevant reduction of the amount of BoNT-A necessary to achieve therapeutic efficacy could help to lower costs and makes ASBTA especially interesting if one considers the increasing number of indications of BoNT-A and its use in economically disadvantaged countries. Our results suggest pharmacological advantages concerning side effects and immunogenicity that will need to be confirmed in randomized, blinded trials. There are some limitations of this study. Specifically, the study was non-randomized and open label. This precludes definitive comparisons about efficacy, safety, and immunogenicity of low-dose versus full-dose preparations.

Side effects reported by our patients were transitory and there were no systemic ones. In CD, dysphagia was indicated in 3.5% of treatment sessions, neck muscle weakness in 2.5% and pain at the injection site in 1.5%. These rates are lower than those usually described in the literature with dysphagia ranging from 12 up to 80% (Moore and Blumhardt 1991; Kessler et al. 1999; Haussermann et al. 2004; Marchetti et al. 2005; Truong et al. 2005) neck muscle weakness from 16 to 62% (Kessler et al. 1999; Haussermann et al. 2004; Truong et al. 2005) and pain at injection side from 4.7 to 38% (Kessler et al. 1999; Truong et al. 2005). In BSP, ptosis with ASBTA was documented in 1% of the patients and dry eye in 0.8% which compares with ptosis in 2.8–12% and dry eye in 7.5% described in a Cochrane-Review (Costa et al. 2005a). None of our patients complained of diplopia. The situation in HFS is very similar according to another Cochrane-Review (Costa et al. 2005b). The head-to-head comparison between the ASTBA patients and those reconverted to conventional BoNT-A (group II) revealed a generally lower rate of side effects for ASBTA (1.5%) than for Dysport® (4.7%) or Botox® (4.4%). Taken together, ASBTA was associated in this long-term observation with fewer side effects than conventional BoNT-A treatment and there were no specific or additional side effects. Although the injected volumes were higher by a factor of 2–3 when using ASBTA versus conventional Dysport®, only few patients (1.5%) reported slight to moderate pain at the injection site and there was no clinical indication of diffusion to remote sites or muscles as was already shown in our previous studies (Rollnik et al. 2000; Bigalke et al. 2001).

ASBTA carried a relatively small risk of immunoresistance due to development of NAb. Using the mouse diaphragm assay, NAb were detected in only one of four secondary non-responders having CD (0.94% of all patients) which is lower than in other long-term studies (Greene et al. 1994; Kessler et al. 1999; Haussermann et al. 2004; Mejia et al. 2005) and in our own experience. Direct comparison with the existing literature, however, is difficult as commercial BoNT-A products have been changed over time, especially the Botox®-preparation. The introduction of the present formulation of Botox® (Aoki 1999) in 1998 resulted in a relevant decrease in Nab-formation (Jankovic et al. 2003). The low BoNT-A doses necessary when using ASBTA may, however, very well be advantageous in the prevention of

antibody development (Greene et al. 1994; Jankovic 2004).

In conclusion, this study shows that long-term treatment with ASBTA on the basis of Dysport® is effective and safe. This has been proven for CD, BSP and HFS. It would be worthwhile to test ASBTA also in other BoNT-A treatable conditions and also for other commercial products. This could help to reduce costs, side effects and immunoresistance.

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EXHIBIT 7 to Brin Declaration (REDACTED)

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NOT SUBJECT TO FOIA; PRE-DISCLOSURE NOTIFICATION REQUIRED

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October 16, 2025

Re. Inflation Reduction Act: Exclusion of BOTOX® (onabotulinumtoxinA), A Human Plasma-Derived Biological Product, from Medicare Drug Price Negotiation Program.

Dear Mr. Stuart and Ms. Kelley:

AbbVie Inc. (AbbVie) requests a meeting with the Centers for Medicare & Medicaid Services (CMS) before October 31, 2025, to discuss why BOTOX® (onabotulinumtoxinA) is ineligible for selection under the Inflation Reduction Act of 2022 (IRA). BOTOX falls squarely within the IRA's plasma-derived product exclusion and, therefore, is expressly excluded from the IRA's definition of a negotiation-eligible "qualifying single source drug." CMS, therefore, cannot lawfully include BOTOX in the Selected Drug List or any list of the 50 top negotiation-eligible drugs. AbbVie seeks a meeting with CMS to confirm BOTOX's exclusion status and provide any additional information CMS may need regarding this matter.

I. BACKGROUND.

A. The IRA's Plasma-Derived Biological Products Exclusion.

The IRA, as codified in sections 1191 through 1198 of the Social Security Act (SSA or Act), established the Medicare Drug Price Negotiation Program (the Program) to set pricing controls for "selected drugs," which may include biological products. CMS's authority to select drugs and biological products for inclusion in the Program is limited by the specific eligibility requirements under section 1192(e), including the exclusions set forth in section 1192(e)(3) of the Act.

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Only a "qualifying single source drug" (QSSD) may qualify as a "negotiation-eligible drug"—and in turn, a "selected drug."[1] The Act, however, expressly excludes certain products from the definition of QSSD, and thus from the IRA's pricing-control provisions. One of these exclusions applies to "plasma-derived products."[2] Under the Act, "the term 'qualifying single source drug' does not include. . . a biological product that is derived from human whole blood or plasma."[3]

B. BOTOX IS AN INNOVATIVE BIOLOGICAL PRODUCT CONTAINING HSA, WHICH IS DERIVED FROM HUMAN PLASMA AND IS CRITICAL TO ENSURING THE SAFETY AND EFFICACY OF THE PRODUCT.

BOTOX® is a biological product containing two proteins, onabotulinumtoxinA (onabotA) and Human Serum Albumin (HSA), the latter of which is a protein present in human blood plasma and derived from human blood donors.[4] BOTOX is licensed under a biologics license application (BLA 103000) and marketed under the Public Health Service Act (PHSA) in three strengths, with the 100U product and 200U product labeled for therapeutic use. Each BOTOX product contains onabotA and HSA, and the FDA-approved prescribing information (PI) for BOTOX expressly states "[t]his product contains albumin, a derivative of human blood."[5]

BOTOX is an exceptional biological product due to the structural complexity and extraordinary potency of its active ingredient (onabotA) and the critical interrelationship between onabotA and HSA.[6] At the time of its approval, BOTOX was the first botulinum toxin-containing product approved anywhere in the world, pioneering the regulatory precedent and standards for approval of extraordinarily potent botulinum neurotoxins. BOTOX is approved for nine therapeutic indications spanning both adult and pediatric populations, including chronic Migraine, Spasticity, Overactive Bladder, Cervical Dystonia, and Neurogenic Detrusor Overactivity. [7]

BOTOX is an American innovation that embodies AbbVie's commitment to domestic manufacturing—including the safe and responsible handling of highly potent botulinum

[1] *See generally* SSA § 1192.

[2] SSA § 1192(e)(3)(C).

[3] *Id.*

[4] BOTOX contains three total ingredients – the third is sodium chloride, which is added as a tonicity agent to help control osmolarity. *See e.g., See e.g.*, Brin MF, Nelson M, Ashourian N, Brideau-Andersen A, Maltman J., *Update on Non-Interchangeability of Botulinum Neurotoxin Products*, 16 TOXINS 266, at 12 (Jun 2024), https://pubmed.ncbi.nlm.nih.gov/38922160/.

[5] PI at Section 5.14, https://www.rxabbvie.com/pdf/botox_pi.pdf.

[6] OnabotA is a botulinum neurotoxin protein complex consisting of ~150 kDa neurotoxin protein component in association with various other neurotoxin accessory proteins (NAPs). Brin, *infra* note 4, at 6.

[7] Refer to the BOTOX® (onabotulinumtoxinA) Prescribing Information for the full indications statements; *see e.g.*, BOTOX® (onabotulinumtoxinA) Product Monograph,

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neurotoxins and the plasma-derived HSA. The research and development leading to the initial approval of BOTOX was conducted in the United States. In fact, the onabotA drug substance has been manufactured in the United States from its inception to the present, and the HSA in BOTOX is sourced from American donors. AbbVie has made and continues to make significant investments to safeguard the manufacture and distribution of neurotoxins, as BOTOX is highly regulated by several different federal agencies through the Federal Select Agent Program, and also because the biological product contains blood components (through inclusion of HSA), which requires compliance with FDA regulations applicable to blood and blood products located at 21 C.F.R. Part 640.[8]

1. HSA Plays a Critical Function in Managing the Activity of onabotA and Directly Contributes to the Safety and Efficacy of BOTOX.

HSA is an integral component of BOTOX, which contains approximately 100,000 times more HSA by mass than onabotA. This ratio of HSA to onabotA manages the extreme potency of onabotA, which directly contributes to the overall safety and efficacy of BOTOX. The table below illustrates the relative mass of BOTOX's three ingredients.

BOTOX (Unit/vial)	OnabotulinumtoxinA*	Human Albumin	Sodium Chloride
50U	50U (2.5 ng)	0.25 mg	0.45 mg
100U	100U (5 ng)	0.5 mg	0.9 mg
200U	200U (10 ng)	1.0 mg	1.8 mg

*The amount of onabotA included in each vial is measured by units of activity, however, the BOTOX PI states "approximately 20Units/nanogram of neurotoxin protein complex."

Because onabotA is dosed in miniscule amounts (*i.e.*, nanograms), it is essential to minimize the loss, denaturation, or inactivation of the toxin protein throughout the manufacture, storage, and use of BOTOX. HSA provides these effects by protecting the onabotA from environmental factors that can affect the quality, stability, and performance of the biological product.[9] Specifically, the HSA in BOTOX minimizes protein aggregation (*i.e.*, protein "clumping") that would reduce the activity of onabotA and mitigates the increased risk of an unwanted immune response. HSA also limits protein loss via adsorption (*i.e.*, "sticking" to surfaces or syringes), which can contribute to

https://www.botoxone.com/content/dam/botoxone/pdf/BOTOX%20Product%20Monograph%20-%20All%20Indications.pdf.

[8] AbbVie also continues to invest heavily in therapeutic neurotoxin innovation through advancement of manufacturing capabilities and platform modernization. Since 2023, AbbVie has been building additional toxin research facilities in the United States and hiring teams to support its long-term vision to address unmet patient needs and provide toxin innovation that will serve future generations of patients.

[9] *See e.g.,* S. Sattler, et al., *A Narrative Literature Review of the Established Safety of Human Serum Albumin Use as a Stabilizer in Aesthetic Botulinum Toxin Formulations Compared to Alternatives*, TOXINS (Oct 2023) (HSA is able to provide protection from aggregation, non-specific adsorption, and oxidation, as well as improve solubility and consistency), https://pmc.ncbi.nlm.nih.gov/articles/PMC10610632/.

inconsistency in delivered dose. And HSA protects against toxin oxidation, which impacts the biological activity of onabotA. Given the minute quantities of onabotA in each BOTOX vial, any impact to the biological activity or amount of the toxin protein can be disproportionately consequential. These challenges, attributed to the structural complexity and properties of onabotA, demonstrate the critical role HSA plays in the safety and efficacy of BOTOX.

2. HSA Impacts the Activity of BOTOX.

HSA also uniquely affects the activity of BOTOX in other ways. Multiple studies in scientific literature discuss the critical role HSA plays in impacting the potency of toxins like onabotA.[10] Preclinical and clinical studies in humans from the early 2000s consistently observed that increasing concentrations of HSA increased the paralytic effects of toxins such as onabotA. Given the limitations in scientific techniques, however, these studies attributed the potential benefit effect to more efficient toxin extraction out of vial and syringe (*e.g.*, less toxin loss via adsorption).

Expanding upon these observations, AbbVie conducted additional research using state of the art techniques to further investigate the role of HSA and onabotA in BOTOX. [redacted] As detailed more fully in the attached "Scientific Addendum", [redacted] demonstrate HSA's impact on the activity of BOTOX by:

(1) Increasing the binding activity of onabotA – which is critical to BOTOX's mechanism of action;

(2) Impacting onabotA peak effect and duration (e.g., muscle contraction strength); and,

(3) Increasing onabotA toxin availability within the injected muscle.

Accordingly, due to the distinctive attributes of onabotA and the interactions between HSA and onabotA, HSA is an integral component of BOTOX both quantitatively (*i.e.*, by comprising most

[10] The scientific literature discusses the unique relationship between botulinum neurotoxins and HSA. *See e.g.*, A. Kutschenko, et al., *The role of human serum albumin and neurotoxin associated proteins in the formulation of BoNT/A products*, 168 TOXICON 158, 161-62 (Oct 2019) (observing increasing doses of HSA resulted in dose-dependency of the activity of certain botulinum toxin type A toxins); B. Mohammadi, et al., *Experience with long-term treatment with albumin-supplemented botulinum toxin type A*, 116 J. NEURAL TRANSMISSION 437, 440 (2009) (observing HSA supplementation enabled halving the dose of a certain botulinum toxin type A product, while achieving similar therapeutic effect and reducing side effects compared to conventional dosing in cervical dystonia patients over 5-10 years); J. D. Rollnik, et al., *Low-Dose Treatment of Cervical Dystonia, Blepharospasm and Facial Hemispasm with Albumin-Diluted Botulinum Toxin Type A under EMG Guidance*, 43 Eur. Neurol 9, at 11 (2000) (concluding that "low-dose, albumin-diluted botulinum toxin" is "effective (and even preferred)" in study involving 115 patients with cervical dystonia, blepharospasm, and hemifacial spasm).

of the total protein in the product) and qualitatively, due to the unique role it plays in the biological activity of BOTOX.

II. BOTOX IS PRECLUDED FROM IRA SELECTION BECAUSE IT IS A BIOLOGICAL PRODUCT THAT IS DERIVED FROM HUMAN PLASMA.

A. The Plain Text of the IRA Makes Clear that BOTOX Is Ineligible for Selection under the Program.

To determine whether BOTOX is a "qualifying single source drug" under the IRA, CMS must "start, as always, with the language of the statute."[11] The IRA contains two separate statutory provisions relevant to identifying a QSSD: (1) general statutory eligibility criteria; and (2) specific statutory exclusions. [12]

Section 1192(e)(1) sets forth the baseline requirements for QSSDs "[i]n [g]eneral," explaining that "the term ''qualifying single source drug'" encompasses certain "drug products" and certain "biological products." A "biological product" to qualify as a QSSD must be "licensed under section 351(a) of the Public Health Service Act and … marketed under section 351 of such Act;" "at least 11 years [must] have elapsed since the date of such licensure;" and it can "not [be] the reference product for any biological product that is licensed and marketed under section 351(k) of such Act."[13] BOTOX clearly meets these statutory QSSD threshold requirements because BOTOX is licensed and marketed pursuant to section 351(a) of the PHSA under BLA No. 103000, as reflected in FDA's "Purple Book Database of Licensed Biological Products";[14] more than 11 years have elapsed since licensure;[15] and BOTOX is not the reference product for any biosimilar product licensed under Section 351(k) of the PHSA.[16]

The baseline QSSD requirements, however, are "subject to" the statutory exclusions set forth in section 1192(e)(3).[17] Section 1192(e)(3) provides that the "the term 'qualifying single source drug' does not include" certain drugs and biological products. In other words, even if a "drug product" or "biological product" meets the general requirements outlined in 1192(e)(1), it

[11] *See Universal Health Servs. v. United States ex rel. Escobar,* 579 U.S. 176, 187 (2016); *accord Twitter, Inc. v. Taamneh,* 598 U.S. 471, 484 (2023).

[12] SSA §§ 1192(e)(1), 1192(e)(3).

[13] "A drug product," by contrast, qualifies as a QSSD if it is "approved under section 505(c) of the Federal Food, Drug, and Cosmetic Act and is marketed pursuant to such approval," "at least 7 years … have elapsed since the date of such approval," and it "is not the listed drug for any drug that is approved and marketed under section 505(j) of such Act." SSA § 1192(e)(1)(A).

[14] BOTOX is listed in FDA's "Purple Book Database of Licensed Biological Products." See https://purplebooksearch.fda.gov/productdetails?query=103000

[15] *Id.*

[16] SSA § 1192(e)(1)(B).

[17] SSA § 1192(e)(1).

nevertheless does *not* qualify as a QSSD if it falls within any of section 1192(e)(3)'s exclusions. One of those exclusions is for "plasma-derived products," defined as any "biological product that is derived from human whole blood or plasma."[18] With respect to BOTOX, then, the relevant question, is whether the biological product, is "derived from"—*i.e.*, "come[s] or [is] obtained from"[19]—"human whole blood or plasma." It clearly is, because the HSA in BOTOX comes from whole blood collected from human donors.[20] A "biological product" is therefore "derived from human whole blood or plasma" if it contains an ingredient sourced from human whole blood or plasma like the HSA in BOTOX. Accordingly, because under any plain reading of the statutory language, BOTOX is "plasma-derived," it does not qualify as a QSSD and is thus ineligible to be selected under the Program.

HSA is a critical component of BOTOX, integral to the safe and effective use of the biological product. It cannot be said that the HSA is present in BOTOX in token amounts, or as a pretext. As discussed in the attached Scientific Addendum, HSA modulates the peak effect, duration, and diffusion properties of onabotA. Data from several mutually-reinforcing studies show that the combination of HSA and onabotA increases the binding of onabotA to neuronal receptors that are involved in the BOTOX mechanism of action. This evidence reinforces that the presence of HSA in BOTOX results in the biological product attributes that are essential to how BOTOX works.[21] In addition to this unique pharmacological role, the HSA in BOTOX also ensures dosage consistency by minimizing the loss, denaturation, or inactivation of the onabotA protein complex. Given the minute quantities of onabotA in each BOTOX vial, even slight changes in the biological activity of onabotA due to toxin loss or inactivation can have a substantial impact.

B. CMS Guidance Reinforces that BOTOX is a Plasma-Derived Product Ineligible for Selection.

In addition to the clear text of the IRA, agency practice and guidance likewise support the conclusion that BOTOX is ineligible for selection as a negotiation eligible drug because it is a plasma-derived biological product.

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[18] SSA § 1192(e)(3)(C).

[19] "Derive from something," Cambridge Dictionary, https://dictionary.cambridge.org/us/dictionary/english/derive-from (last visited Aug. 12, 2025); *see also* The New Oxford American Dictionary (2001) (defining "derive something from" as "obtain something from (a specified source)").

[20] Botox PI at Section 5.12 ("this product contains albumin, a derivative of human blood"); *see also* 67 Fed. Reg. 66718, 66774 (Nov. 1, 2003) (recognizing that human albumin is "plasma-derived"). An alternative reading of the exemption that requires every component of a biological product to be plasma-derived would be nonsensical given that biological products that are clearly intended to be covered by this exclusion often contain non-plasma components. *See, e.g.,* Kcentra (Prothrombin Complex Concentrate (Human)) Package Insert at 17 (containing sodium chloride, sodium citrate, hydrochloric acid, and sodium hydroxide in the vial along with components that are plasma derived).

[21] *See supra* at footnote 10.

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In its September 2025 "Final Guidance on the Medicare Drug Price Negotiation Program" ("IPAY 2028 Final Guidance"), CMS describes a QSSD as a bundle of certain finished biological products sharing the same active ingredient from the same applicant.[22] Specially, CMS says "a potential" QSSD means "[f]or biological products, all dosage forms and strengths of the biological product with the same active ingredient [from] the same holder of a BLA, inclusive of products that are marketed pursuant to different BLAs."[23] Critically, CMS asserts that section 1192(d)(3)(B) enables the Agency to aggregate data for the purpose of identifying negotiation-eligible drugs.[24] Per its Guidance, CMS accomplishes this by identifying, as a threshold matter, potential products comprising a single QSSD by aggregating individual National Drug Codes (NDCs) corresponding to all product(s) from the same BLA holder containing the same active ingredient(s).[25] Under this approach, a QSSD must include dosage forms, formulations, strengths, and routes of administration of products from that manufacturer containing the same active ingredient.[26] In other words, a QSSD "***means*** the specific constituent dosage forms and strengths (at the NDC-9 or NDC-11 level) that are identified as aggregated under the [BLA(s)] for the . . . active ingredient."[27]

In using NDC codes to identify eligible "biological products" under Section 1192(e)(1), CMS confirms that it looks at finished dosage forms, strengths and formulations when identifying which biological products to aggregate into individual QSSDs. This conclusion is supported by statements made on behalf of the Secretary and the Agency in pending litigation. For example, in defending its decision to aggregate insulin aspart products from Novo Nordisk, CMS asserted: "CMS's selection of Novolog derives from CMS's determination that ***all the selected formulations*** of Novo's insulin aspart constitute a single 'qualifying single source drug.'"[28] The government further explained that "if one company produces several forms of a drug with the same active ingredient (and no additional active ingredients . . .), these various forms will be considered collectively under the provisions of the IRA that require aggregation across dosage forms, package

[22] CMS, Final Guidance on the Medicare Drug Price Negotiation Program, 159-173 (2025) https://edit.cms.gov/files/document/ipay-2028-final-guidance.pdf.

[23] *Id.* at 165 (footnote omitted).

[24] *Id.* at Section 30.1 (pages 164-65).

[25] *See id*. at 166. Every human prescription product approved in the U.S. is assigned an NDC which consists of a unique, three-segment, number that corresponds to the labeler, product, and trade package size. The labeler code (first segment) is assigned by FDA and includes any entity that manufacturers or distributes a product. The product code (second segment), identifies a specific strength, dosage form, and formulation for a particular entity. The package code (third segment), identifies package sizes and types. *See, e.g.*, FDA, *National Drug Code Database Background Information* (Mar. 20, 2017), https://www.fda.gov/drugs/development-approval-process-drugs/national-drug-code-database-background-information.

[26] *Id.* at 165 n.75; *see also id.* at 166 (Table 1 example identifies "12 [t]otal NDCs included in [a] single potential [QSSD].").

[27] *Id.* at 165 n.75 (emphasis added).

[28] Mem. in Opp'n to Pls.' Mot. for Summ. J. and in Supp. of Defs.' Cross-Mot., at 15, *Novo Nordisk Inc. v. Becerra*, 3:23-cv-20814-ZNQ-JBD (D.N.J. Jan. 26, 2024), Dkt. No. 37-1 (emphasis added).

types, and formulations."[29] CMS thus recognizes that a "biological product" is a finished dosage form. Accordingly, under this interpretation, a "biological product" must encompass the various ingredients in each of the different dosage forms, strengths, and formulations aggregated by CMS.

Consistent with this approach, CMS must likewise assess, when applying the Section 1192(e)(3)(C) plasma-derived exclusion, whether a particular QSSD "does not include" a dosage form, strength, or formulation of a biological product because it is "derived from human whole blood or plasma." With respect to BOTOX, all forms of the biological product contain the critical component HSA so none can be selected for negotiation.

This conclusion also aligns with CMS' specific discussion of the plasma-derived product exclusion in the recent Guidance:

> For purposes of this exclusion, a plasma-derived product is a licensed biological product that is derived from human whole blood or plasma, as indicated on the approved product labeling. CMS will refer to product information available on the FDA Approved Blood Products website, including the list of fractionated plasma products, and will refer to databases such as FDALabel and the FDA Online Label Repository to verify if the product is derived from human whole blood or plasma. CMS also will consult with FDA, as appropriate.[30]

CMS further clarifies, in the question and answer section, that a product need not appear on the FDA's website list of Fractionated Plasma Products in order to qualify for the exclusion. For example, CMS notes that cellular and gene therapy products—which are not listed on that FDA website—may qualify for the exclusion.[31] This conclusion makes sense, because FDA's list of Fractionated Plasma Products includes recombinant forms of plasma-derived products and therefore, cannot be a definitive resource for application of the statutory exclusion.[32]

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[29] *See, e.g.*, Brief for Appellees at 16, *Novo Nordisk Inc. v. U.S. Dep't of Health & Hum. Servs.*, No. 24-2510 (3rd Cir. Dec. 16, 2024) (emphasis added); *see also id.* at 20 ("CMS grouped these different *forms* of the drug together in the selection and negotiation process")(emphasis added); *id.* at 42 ("The IRA thus contemplates that there may be multiple *formulations* and package types of a selected drug and directs CMS to apply the negotiated price to each formulation.") (emphasis added); *id.* at 47 ("CMS reasonably determined that the relevant date is the earliest approval date of a *product* in the set, Revised Guidance 101, ensuring that the introduction of variations of the drug do not alter its eligibility.") (emphasis added).

[30] Final Guidance on the Medicare Drug Price Negotiation Program, *supra* note 22, at 173 (footnotes omitted).

[31] *Id.* at 27.

[32] *See, e.g.*, Atryn, Antithrombin (Recombinant) PI at 10 (noting that the product is "produced by recombinant DNA technology using genetically engineered goats"); Vonvendi (von Willebrand Factor (Recombinant)) PI at 14 (product is "a purified rVWF expressed in Chinese Hamster Ovary (CHO) cells" and "is produced and formulated without the addition of any exogenous raw materials of human or animal origin").

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Per the forgoing discussion in the Guidance, BOTOX is plasma-derived. First and foremost, CMS says it will look to a product's FDA-approved label. Here, BOTOX's FDA-approved product labeling—as posted on the cited FDALabel and the FDA Online Label Repository websites—expressly states that BOTOX "contains albumin, a derivative of human blood."[33] BOTOX's "approved product labeling" therefore confirms its plasma-derived status. Since all BOTOX products are plasma-derived due to the HSA contained therein, no currently licensed and marketed AbbVie onabotA biological product can be a QSSD.

For the foregoing reasons, BOTOX is a "biological product that is derived from human whole blood or plasma" and thus excluded from the QSSD definition. Because only a QSSD may qualify as a "negotiation-eligible drug"—and in turn, a "selected drug," no BOTOX product is eligible for selection.

III. Congress and CMS Have Recognized that Plasma-Derived Products Have Special Status Due to their Unique Sourcing and Manufacturing Considerations.

There is good reason for Congress's decision to exclude plasma-derived products such as BOTOX from the Program. Unlike other pharmaceutical products, plasma-derived therapies are reliant on donated human plasma, making such products subject to strict manufacturing regulations and uniquely sensitive to variable supply. Plasma is a blood component originating from the voluntary donation of biological material by donors. After donation, the production of medicines depends upon a complex, time-consuming, and highly regulated process for collecting, storing, testing, and processing donated blood plasma.

Congress and CMS have consistently recognized special status for products derived from human plasma. As Congress recognized through its enactment of the exclusion, imposing maximum fair prices on plasma-derived products would exacerbate these issues, because manufacturers would be even more limited in their ability to maintain the supply of plasma. CMS acknowledged the same when implementing the Medicare Inflation Rebate Program. In the Agency's Revised Guidance, CMS reduced the rebate amount for certain plasma products, given their "manufacturing complexity" and "unique reliance on donations of blood plasma."[34]

The HSA in BOTOX is derived from human plasma donated by Americans, making BOTOX subject to precisely the strict manufacturing regulations and unique sensitivities to variable supply that Congress and CMS intended to address by the plasma-derived QSSD exclusion. After donation, the production of BOTOX depends on a complex, time-consuming, and

[33] PI at Section 5.14.

[34] CMS, Medicare Part B Drug Inflation Rebates Paid by Manufacturers: Revised Guidance, Implementation of Section 1847A(i) of the Social Security Act, at 34 (Dec. 14, 2023); CMS, Medicare Part D Drug Inflation Rebates Paid by Manufacturers: Revised Guidance, Implementation of Section 1860D-14B of the Social Security Act, at 22 (Dec. 14, 2023).

highly regulated process for collecting, storing, testing, and processing donated blood plasma.[35] In fact, BOTOX manufacturing must comply with FDA regulations for blood and blood product at 21 C.F.R. Part 640, further evidencing that BOTOX is a plasma-derived biological product.

These complexities, coupled with the critical role HSA plays in BOTOX, makes it uniquely deserving of Congress's and CMS's special recognition.

* * *

[35] *See* 21 C.F.R. Part 640 (setting forth standards that apply to blood and blood products, including for the collection and processing of "source plasma" and human albumin). The HSA used in BOTOX is collected and produced in accordance with global regulatory standards. Donors are carefully screened and qualified, and donations are tested for multiple viruses and antigens. Detailed documentation ensures traceability of each donation from each donor for several years. To ensure a high level of safety, the donated plasma undergoes processes to remove and inactivate viruses, including fractionation and extended heat treatment. *See, e.g.*, Raman Malhotra et al., *Botulinum toxin and human serum albumin*. 121 ARCH OPHTHALMOL. 1661, 1662 (Reply by M. Brin, MD) (Nov. 2003), https://pubmed.ncbi.nlm.nih.gov/14609937/.

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For the reasons explained, BOTOX is a biological product that is derived from human plasma within the meaning of section 1192(e)(3)(C) and thus ineligible for selection under the Program. We request a meeting between AbbVie and CMS before October 31, 2025, to discuss BOTOX's status and any questions you may have.

Sincerely,

Johanna Corbin
Senior Vice President, Chief Patent and Innovation Counsel
Intellectual Property, Transactions, and Innovation

On behalf of AbbVie Inc.

cc:
Betsy Pelovitz
Acting Deputy Associate General Counsel,
Program Review Branch
Office of the General Counsel
U.S. Dep't of Health and Human Services
Betsy.Pelovitz@hhs.gov
330 Independence Ave., SW, Room 5326
Washington, DC 20201

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NOT SUBJECT TO FOIA; PRE-DISCLOSURE NOTIFICATION REQUIRED

October 16, 2025

Re. Inflation Reduction Act: Exclusion of BOTOX® (onabotulinumtoxinA), A Human Plasma-Derived Biological Product, from Medicare Drug Price Negotiation Program.

SCIENTIFIC ADDENDUM

Several studies were conducted to investigate the integral function of Human Serum Albumin ("HSA"), a protein present in human blood plasma and derived from human blood donors, with onabotulinumtoxinA ("onabotA"), a type A botulinum neurotoxin, as present in the BOTOX product formulation. [redacted] studies below demonstrated that HSA interacts with onabotA to meaningfully impact the toxin's pharmacological activity and accordingly contributes to the overall safety and efficacy profile of the biological product, BOTOX.

More specifically, these mutually-reinforcing results demonstrate the unique role that HSA plays in BOTOX. [redacted]

[redacted]

The summary below is intended to provide an overview of various experimental study results. Additional details regarding materials and methods, as well as comprehensive results, are available upon request.

[redacted]

Botulinum toxins, such as onabotA, exert their effects by binding to specific neuronal cell receptors at the neuromuscular junction. Once bound, the toxin is internalized into the neuron, where it inhibits the release of neurotransmitters, thus temporarily paralyzing the muscle and leading to the

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therapeutic effect of BOTOX. [redacted]

[redacted]

As detailed in the cover submission, besides its role in reducing protein oxidation and aggregation, HSA has a well understood role as a surfactant in reducing the loss of botulinum toxin protein through adsorption, which can occur when onabotA adheres to surfaces like vials and syringes, during and after reconstitution from a vacuum-dried powder into a liquid form for injection into a patient. Control of toxin protein adsorption is important to ensure consistent and accurate dosing, which directly affects the safety and efficacy of biological products. It is particularly important for products like BOTOX, where the toxin protein is dosed in minute quantities given the extreme potency of onabotA.

[redacted]

[redacted]

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[redacted]

- [redacted]
- [redacted]
- [redacted]

[redacted]

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- [redacted]

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EXHIBIT 8 to Brin Declaration (REDACTED)

Office of the General Counsel
U.S. Dep't of Health and Human Services
200 Independence Ave., S.W., Room 713-F
Washington, DC 20201

December 5, 2025

Dear Sir and/or Madam,

The Role of Human Serum Albumin on the Biological Activity of OnabotulinumtoxinA

I have been requested by AbbVie Inc. to provide an expert opinion for the Centers for Medicare & Medicaid Services ("CMS") on the potential direct role of Human Serum Albumin ("HSA") in the activity of BOTOX®. Below, I provide this opinion in the context of both historical clinical and non-clinical data in published, peer-review literature and recent findings by AbbVie when studying this relationship in detail, in the laboratory.

By way of context, I trained in Microbiology in the University of London and obtained my degree in 1975 and my doctorate in 1978. I have worked for over 40 years in the pharmaceutical industry on a wide range of biological products including botulinum toxin, microbial enzymes, vaccines, natural products and many recombinant proteins, all in the laboratory, development and also in production and quality. In particular, I have worked on botulinum toxin continuously, during this period, in roles and responsibilities including scientific and product support and technical expertise on botulinum toxin structure, function, physiology, assay technologies, quality, regulatory and production aspects. I have liaised with many regulatory authorities around the world, including the US Food and Drug Administration, European authorities and the Japanese PMDA. I have over 85 publications on botulinum toxin in peer-reviewed journals, book chapters, magazine articles and related publications. I am also a joint inventor on number of patent applications relating to botulinum toxins. I now provide consultancy services to companies in the botulinum toxin space covering numerous aspects related to the industry.

1. Physical Characteristics of BOTOX®

BOTOX consists of three ingredients: onabotulinumtoxinA ("onabotA"), HSA derived from human plasma, and sodium chloride. In order for AbbVie to manufacture the injectable BOTOX, with a potency suitable for therapeutic use, onabotA is extensively diluted and mixed together with HSA and sodium chloride, sterilised by filtration and then vacuum dried to yield BOTOX. There is at least 100,000 times more HSA than onabotA in BOTOX, a vast excess. The HSA is also an FDA regulated biologic that is purified from human plasma donations. There have been no changes to the BOTOX product formulation since the earliest product was approved, with a decades-long proof of product quality and efficacy having been established.

1.1 OnabotulinumtoxinA

OnabotA is a natural, biological protein toxin produced by the growth of the bacterium *Clostridium botulinum*. After growth of the bacteria under controlled conditions, the toxin is

purified to yield the onabotA Drug Substance, a very large complex of the neurotoxin molecule coupled to other natural proteins. These proteins form, together, a so-called toxin-protein complex.

1.2 Human Serum Albumin

When used in biological products, HSA has many functions, including as an antioxidant, inhibitor of protein aggregation, and to prevent proteins from adhering to the surfaces of the vial/syringe they are contained in.[1]

HSA was included in the formulation of the first available botulinum type A toxin ("toxin") products from the very beginning. Since the successful use of BOTOX for the treatment of a wide and expanding range of clinical conditions, follow-on products have adopted the same concentration of HSA in the vial as that leader product.[2] Five out of the six FDA approved toxin products contain HSA, to varying concentrations.[3]

2. Human Serum Albumin has a Recognized Influence on the Pharmacokinetics and Pharmacodynamics of Products

Since the 1940s, HSA has been widely recognized to interact with other components in a product formulation: these interactions have been extensively studied over the years.[4] HSA interactions with other components can affect drug distribution, metabolism and elimination.[5] During the manufacture of toxin products, as finished products in vials, the toxin molecules spend a considerable time, in the initial preparation phases, in solution with HSA and other excipients. The likelihood that some form of toxin–HSA interaction occurs is unsurprising, especially since the concentration of HSA is a hundred thousand times greater than that of the toxin (milligrams of HSA per mL compared to nanograms of toxin).

While HSA can interact with other components, interaction with receptors in the body may occur to exert a physiological or biological effect. For example, HSA could affect a natural receptor in the body which, in turn, may facilitate or enhance another drug binding to that receptor.

[1] Sattler, S., Gollomp, S., & Curry, A. (2023). A Narrative Literature Review of the Established Safety of Human Serum Albumin Use as a Stabilizer in Aesthetic Botulinum Toxin Formulations Compared to Alternatives. *Toxins (Basel)*, *15*(10). https://doi.org/10.3390/toxins15100619

[2] Sattler, S., Gollomp, S., & Curry, A. (2023). A Narrative Literature Review of the Established Safety of Human Serum Albumin Use as a Stabilizer in Aesthetic Botulinum Toxin Formulations Compared to Alternatives. *Toxins (Basel)*, *15*(10). https://doi.org/10.3390/toxins15100619 Table 1

[3] See, for example, *Ibid* Table 1

[4] Reported in Lindup, W. E. (1975). Drug-albumin binding. *Biochem Soc Trans*, *3*(5), 635–640. https://doi.org/10.1042/bst0030635

[5] Fanali, G., di Masi, A., Trezza, V., Marino, M., Fasano, M., & Ascenzi, P. (2012). Human serum albumin: from bench to bedside. *Mol Aspects Med*, *33*(3), 209–290. https://doi.org/10.1016/j.mam.2011.12.002

2.1 Initial Evidence that Human Serum Albumin Plays a Role in the Activity of Toxin

The first report that HSA may play a direct role in the activity of a toxin product clinically was described in 2000.[6] A German group described how 115 patients with varying clinical conditions were treated with a lower dose of a different toxin product Dysport® (abobotulinumtoxinA) than recommended by the manufacturers. For the study, this product was diluted with 0.1% HSA solution instead of the normal recommended diluent (sterile, preservative-free 0.9% saline). Their findings indicated that a lower dose therapy, when diluted with HSA solution, was both effective and even preferred by the patients.

The group continued their work in more detail later, using both a non-clinical model and a human foot muscle model example to identify more optimal, but lower, doses of Dysport.[7] Their final report, on 106 patients who had received an albumin-diluted Dysport over 5-10 years of treatment, came to the same conclusion, namely that an HSA-diluted product was safe and effective.[8] The authors speculated that the use of additional HSA prevented non-specific binding of Dysport to glass and plastic surfaces.

Studies on the potential role of HSA in toxin activity, using an *ex vivo* model of a mouse diaphragm phrenic nerve stimulation (where paralysis is due to toxin effect), have also been reported by the Hannover group.[9,10] This model has been used by the group to study the activity of toxin preparations since the 1980s.[11]

The *ex vivo* results obtained clearly demonstrated a relationship between HSA concentration and toxin activity, with a fixed concentration of toxin in each formulation, up to a maximum activity achieved at 0.8 mg/mL HSA. No activity was found in the absence of HSA which, according to the authors, was presumably due to the toxin binding to surfaces in the test system. Overall, the key role of HSA in the activity of toxin was demonstrated.

Importantly, while various theories were considered, these publications did not definitively identify the underlying mechanism for the relationship between HSA and toxin.

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[6] Rollnik, J. D., Matzke, M., Wohlfarth, K., Dengler, R., & Bigalke, H. (2000). Low-dose treatment of cervical dystonia, blepharospasm and facial hemispasm with albumin-diluted botulinum toxin type A under EMG guidance. An open label study [Clinical Trial]. *Eur Neurol*, *43*(1), 9–12. https://doi.org/10.1159/000008121

[7] Bigalke, H., Wohlfarth, K., Irmer, A., & Dengler, R. (2001). Botulinum A toxin: Dysport improvement of biological availability. *Exp Neurol*, *168*(1), 162–170. https://doi.org/10.1006/exnr.2000.7583

[8] Mohammadi, B., Kollewe, K., Wegener, M., Bigalke, H., & Dengler, R. (2009). Experience with long-term treatment with albumin-supplemented botulinum toxin type A . *J Neural Transm (Vienna)*, *116*(4), 437–441. https://doi.org/10.1007/s00702-009-0200-6

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[10] Kutschenko, A., Bigalke, H., Wegner, F., & Wohlfarth, K. (2019). The role of human serum albumin and neurotoxin associated proteins in the formulation of BoNT/A products. *Toxicon*, *168*, 158–163. https://doi.org/10.1016/j.toxicon.2019.07.005

[11] Bigalke, H., & Rummel, A. (2015). Botulinum Neurotoxins: Qualitative and Quantitative Analysis Using the Mouse Phrenic Nerve Hemidiaphragm Assay (MPN). *Toxins (Basel)*, *7*(12), 4895–4905. https://doi.org/10.3390/toxins7124855

2.2 New Studies by AbbVie on the Role of HSA in the Activity of onabotA

The detailed elucidation of the mechanism of action of toxin[12] has enabled AbbVie to carry out relevant studies on precisely *how* HSA might affect the biological activity of onabotA. I am not aware of any other published work studying this mechanism of action.

I have reviewed the study data from the three sets of work that AbbVie has provided to both myself and to CMS, including the Scientific Addendum submitted to CMS. I summarise here the findings, with my own interpretation.

[redacted]

This indicates to me that HSA enhances the binding of onabotA to the natural nerve receptors attached to the muscle being treated to an endogenous receptor, resulting in increased internalization of the toxin into the neuron.

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[redacted]

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[redacted]

Yours sincerely,

Dr. Andrew Pickett
Toxin Science Limited
Director and Founder

CURRICULUM VITAE

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Sweden

Tel.: +44 7921 164537

Email andy@toxinscience.com

DATE OF BIRTH: 3 August 1954

EDUCATION:

1965 - 1972	Surbiton County Grammar School
1972 - 1978	University of London (Queen Elizabeth College - now KQC College, Kensington Campus)

QUALIFICATIONS:

B.Sc.	(Second Class Honours Upper Division) Microbiology University of London, 1975
Ph.D.	Microbiology University of London, 1978

Also holding an academic position:

Adjunct Professor
Botulinum Research Center
Institute for Advanced Sciences
Bedford
MA, USA

TOXIN SCIENCE LIMITED January 2011 ONWARDS

TITLE Director and Founder

I founded the company in January 2011 focused on translating the science and technology of botulinum toxin in order to enable clinicians, dermatologists, plastic surgeons, rehabilitation specialists and medical students to have a clear understanding of the toxin products available, how they work and how they can be used. My speciality is translating the current, state-of-the-art toxin science into practical knowledge and subsequent benefits for clinicians. I have been recognised internationally for this work.

I have delivered over 500 lectures to audiences worldwide, including invited presentations at major international medical and aesthetic conferences. I have lectured at many specialist meetings, hospitals, universities and colleges, to both senior clinicians and trainees, and have been invited to present to many different clinical disciplines globally. I have also specialised in presenting to and training aesthetic clinicians mainly on botulinum toxin but also, from time to time, dermal fillers.

GALDERMA May 2014 to December 2017 (retired)

TITLE Senior Program Leader & Scientific Expert, Neurotoxins

COMPANY Galderma Aesthetic and Corrective (formerly Q-Med)

A fully-dedicated responsibility for the Galderma in-house Neurotoxin program and for providing market and technical support related to neurotoxins, worldwide. Close liaison with all markets, with Key Opinion Leaders and with the peer group of the toxin community was an important part of the responsibilities.

Member of Neurotoxin project End-to-End group (the group with senior responsibility for the in-house construction project for the new product program) and member of Toxin Product Steering Committee.

Q-MED/GALDERMA August 2011 to May 2014

TITLE Head of Development

COMPANY Galderma Aesthetic and Corrective (formerly Q-Med)

Initially responsible for the Development group, which was newly-established during 2011 following the acquisition of Q-MED by Galderma. The Development teams were responsible for Clinical Development, Project Management, Microbiology and Stability and Special Projects of all the Q-MED products in the Aesthetic and Corrective portfolio of the company.

The Development department carried out all the work required from the end of Research until first registration and launch of a new product. Market technical support on existing products was also an important part of the activities. All areas and products dealt with by Q-Med were included in the responsibility.

Total staff approximately 30 persons.

IPSEN BIOPHARM LIMITED

January 2007 to December 2010

TITLE	Senior Director, Biologicals Science and Technology
COMPANY	Ipsen Biopharm Limited
REPORTING TO	Vice President Manufacturing and Supply Organisation Biologicals Supply

MAIN RESPONSIBILITIES

- Provision of scientific and technical advice to MSO, Development and Commercial colleagues in support of business needs.
- Liaison with external industry scientific and regulatory experts, ensuring that Ipsen is appropriately represented at key meetings on an international basis.
- Acting as Ipsen's toxin scientific expert in all matters related to external agencies.
- Assisting as an expert in interactions with regulatory agencies on a worldwide basis at the highest level, to maintain a current awareness of all matters that will impact on the successful execution of the Company's regulatory strategy.
- Liaison with academic partners and centres of excellence in the toxin field.
- Technical review of Ipsen intellectual property portfolio within the toxins area, including recommendations and support for both new Ipsen applications and the opposition of competitor IP.
- Provision of scientific and technical support to both Ipsen's and other partner companies' commercial activities as business expands into new geographies and applications.
- Member of Neurology Portfolio Management Team
- Member of Dysport Publications Support Group
- Group size: 2 direct reports
- Budget responsibilities: £1.7m cost centre, £217k external projects

IPSEN BIOPHARM LIMITED

April 2005 to January 2007

TITLE	Senior Director, Biologicals Manufacturing Support and Technology
COMPANY	Ipsen Biopharm Limited
REPORTING TO	Vice President Manufacturing and Supply Organisation Biologicals Supply

MAIN RESPONSIBILITIES

This role was essentially the same as that within Technical Affairs.

The new department of Biologicals Manufacturing Support and Technology (BMST) was established to merge together the Technical Affairs team, based at Slough and the Technical Department, based at Wrexham. The role was overall management of the merged groups, based at Wrexham.

The total size of BMST was approximately 30 staff, including a number of contractors and 8 direct reports. The external projects budget was approximately €4.5m.

Further additions were made to the department during 2006 when Validation functions were reorganised on site. The elements of equipment validation and methods (analytical and microbiological) validation, including cold-chain validation, were incorporated into the department.

Additional Ipsen group responsibilities remained together with additional member of the Neurology Disease Area Team.

IPSEN LIMITED July 2000 to April 2005

TITLE Director of Technical Affairs

COMPANY Ipsen Limited

REPORTING TO Executive Vice President
Manufacturing and Supply Organisation
Ipsen Group

MAIN RESPONSIBILITIES

- Responsible for the Technical Affairs Department which provides technical support for biological products produced and marketed by Ipsen Group companies

- Overall management of a group of 14 staff, including Managers and Senior Managers

  Responsible for a range of industrial development and product support programs carried out worldwide, encompassing existing products

- Also responsible for certain biologicals production when performed by contract manufacturers outside of the Ipsen Group

- Technical Affairs areas of activity also include Packaging Development, for a range of Ipsen group products both marketed and newly licensed, together with Clinical Trials Supply management for one product

- Total direct budget responsibility in the order of Euros 3.5m for external development and product support activities (exclusing Department costs)

- Technical Affairs is responsible for the CMC (Chemistry, Manufacturing and Controls) sections of Marketing Authorisation dossiers and licence submissions together with the supporting data for the biological products. This requires very close liaison with Regulatory Affairs groups worldwide within the Group together with consultants, experts and advisors in specialist fields

- Member of the Dysport® Integration Team

- Chairman of the CMC Sub-Team for Dysport®

- Maintaining close liaison with Quality Assurance, Quality Control, Production, Business Development, Commercial, Clinical and Pharmacovigilance functions to ensure the achievement of Group objectives

- Responsible for presenting the CMC dossier and dealing with technical issues for all regulatory authorities world-wide for compliance with licensing requirements, especially MHRA (United Kingdom), FDA (United States) and HPB (Canada). Presentations have also been made to the IMB, AFSSAPS and other authorities

- Close liaison also with Group Research and Development, especially on the areas of biological toxin research and development

- Chairman of the Botulinum Toxin Patents Committee

- Member of the Dysport Publications Support Group

- Member of numerous Ipsen Group committees relating to various aspects of the biological products, including two Group Recall Committees for products

- Regular presenter of Dysport technical aspects to Group manager and employee meetings, groups of clinicians, outside companies and business partnerships

CENTRE FOR APPLIED MICROBIOLOGY AND RESEARCH

December 1997 to July 2000

TITLE	Production Manager
COMPANY	Centre for Applied Microbiology and Research [The operational site for the Microbiological Research Authority, then a Special Health Authority of the UK Department of Health: now part of Public Health England]
REPORTING TO	Director of Production [Member of the CAMR Management Executive Committee]

MAIN RESPONSIBILITIES

- Overall management of a group of over 100 staff, including up to 13 Unit Managers as direct reports
- Responsible for managing the development and production of licenced and unlicenced biopharmaceutical and biological products in CAMR manufacturing facilities
- Total direct budget responsibility in the order of £1.8m for both development and production activities, together with a further £1m staff costs
- Responsible for identification, budgeting and, in conjunction with Engineering and Facilities Management, implementation of capital requirements (1999/2000, £4m; 2000/2001, £4.5m)
- Management of all facility and human resources available within the Division to achieve product delivery targets
- Scheduling of development and production activities, in accordance with product delivery requirements
- Areas of responsibility include fermentation development and cGMP production, downstream processing development and cGMP production, protein purification development and cGMP production, freeze-drying development and cGMP production, product assembly, raw materials stores, finished product stores, dispatch
- Use of microorganisms of all types (bacteria, yeast and fungi) for production activities including both pathogenic (to ACDP Category III) and non-pathogenic strains, recombinant and non-engineered strains
- Maintaining close liaison with General Project Managers for planning and execution of development and manufacture of products
- Maintaining close liaison with Quality Assurance, Quality Control and Safety to ensure the satisfactory application of relevant regulatory standards to achieve production objectives
- Liaison with Business Managers to identify and develop new business opportunities
- Liaison with all relevant regulatory authorities world-wide for compliance with licensing requirements for biopharmaceutical manufacture, especially MCA (United Kingdom), FDA (United States) and HPB (Canada)

- Close liaison with customers at the initial stages of project discussion, costing, planning and scheduling through to final execution of agreed programs and delivery of products

- Technology transfer from customers and development of processes for production purposes, as required

- Liaison with Site Engineering functions, including all aspects from regular maintenance activities and PPM systems through to planning and execution of large facility re-development programs

SPEYWOOD PHARMACEUTICALS LIMITED

December 1991 to December 1997

TITLE: Director of Technical Affairs

COMPANY: Speywood Pharmaceuticals Limited, (now named Ipsen Limited) part of the Beaufour-Ipsen Group

REPORTING TO: Chief Executive Officer
[Until early 1997 Director of Research and Development, Speywood Group Ltd.]

MAIN RESPONSIBILITIES:

- Responsible for management of a group of 32 staff including senior managers, technicians and support staff in the Technical Affairs Group, including Quality.
- Total direct budget responsibility (1997) £0.75m together with an additional £1m project funding
- Provision, function, operation and management of Quality Assurance and Quality Control for all products within Speywood in accordance with all applicable company, national, international and regulatory standards.
- Product technical support for all products marketed or in late-stage development, through management of project teams, use of contract organisations or in-group resources.
- Transfer of production, quality and analytical technology from development groups and/or partner companies to manufacturing sites within the Speywood group or to external contractors, including for contract manufacture performed by Speywood.
- Management of product development and support projects wherever necessary to achieve program objectives, including programs in UK, US, Canada, Japan, Germany, Italy, and Holland.
- Manufacture (where contracted out) of clinical trial or other products in accordance with current GMP and all applicable company, national, international and regulatory standards.
- Manufacture of the company's products at the MRA Centre for Applied Microbiology and Research, Porton Down, Salisbury.
- Liaison with the Centre for Applied Microbiology and Research on all products sold by the company and, where appropriate, under development.
- Stock and despatch of company products distributed from the Centre for Applied Microbiology and Research.
- Continual liaison with Regulatory Affairs in group companies, including preparation of Chemistry, Manufacturing and Control (CMC) sections of regulatory dossiers.
- Preparation of responses to questions and issues raised by regulatory authorities worldwide; direct liaison and consultation with those authorities (including MCA, FDA, HPB (Canada), MHW (Japan), AMM (France) and CBG (Holland)).

- Co-ordinator of the Technical Committee of Speywood.

- Member of the Product Launch and Supply Group and Product Support Group, regular liaison meetings with the Centre for Applied Microbiology and Research).

- Project Manager for Acellular Pertussis Vaccine project (in collaboration with the Centre for Applied Microbiology and Research).

- Project Team Leader for Purified Porcine FVIII project

- Compliance Officer for licences related to Export of Goods Control Act

- Shipment (import and export) of specialised products, where applicable.

- Liaison with national and international organisations, groups, sites and personnel as required to fulfil project objectives.

- Development and management of product packaging presentations, from concept development to final approved artwork stage, through a designated Packaging Development Group.

- Continual liaison with Sales, Marketing, Development, Commercial and other company groups as required to carry out the responsibilities of the position.

ADDITIONAL RESPONSIBILITIES:

- Assessment of technology, projects and products, as requested by management.

- Provision of technical assistance to Group companies.

- Provision of technical assistance to the Project Engineering functions.

- Preparation and dissemination of technical reports, design studies, production/product assessments and other specialist evaluations, as requested by management.

- Maintenance of a detailed technical data base for the company, relating to all aspects of manufacture and associated areas.

- Industrial supervisor to staff undertaking various sponsored courses, to MSc and PhD level.

PORTON PRODUCTS LIMITED 1987 to 1991

TITLE: Director of Production/Director of Production Operations (from 1989), Porton Products Limited

RESPONSIBLE TO: Managing Director, Porton Products Limited

Overall responsibility for the company's products at the Centre for Applied Microbiology and Research, Porton Down, for the relevant manufacturing operations, including;

- Production scheduling.
- Production costing.
- Attainment of all QA/QC standards.
- Ensuring all operations met appropriate regulatory standards.
- Provision of all necessary process documentation.
- Planning equipment maintenance and replacement.
- Advising and assisting the Managing Director on delivery, product information, planning etc.
- Stock levels of products.
- Order processing, packaging and despatch.

In addition to these responsibilities, a number of other activities were undertaken, namely;

- Responsible (on a temporary basis from January 1988 for one year) for all regulatory affairs in the UK, including liaison with consultants, liaison with regulatory authorities, preparation of licence application dossiers (both UK and US FDA product and establishment licence applications) and co-ordination of activities.
- Member of the company Product Launch Group (in collaboration with the Centre for Applied Microbiology and Research) to co-ordinate all aspects of production and marketing of products following development and near to launch.
- Provision of technical assistance and advice to other Porton International Group companies.
- Provision of technical assistance to the Project Engineering Group, as appropriate.
- Supervision of specific projects and programs relating to production.
- Supervision of doctoral and graduate students.

PORTON PRODUCTS LIMITED 1985 to 1987

TITLE: Fermentation Operations Director, Porton International/ Porton Products Limited

RESPONSIBLE TO: Chief Executive/Board of Directors, Porton International

The position was based at the Public Health Laboratory Service Centre for Applied Microbiology and Research, Porton Down, attached to the Microbial Technology Laboratory as Head of Fermentation Production and Development, Large Scale (Process Plant Group).

The main responsibility was for the day-to-day management and organisation of a group of 23 staff, including senior staff, shift workers, laboratory staff, process workers, workshop employees and graduate students.

The Group carried out all development and production activities in large-scale fermentation and downstream processing for the products made in the laboratory and marketed by Porton Products. These processes included the use of bacteria, yeast and streptomycetes, either as normal strains or genetically-engineered, for the production of a range of licensed therapeutics, diagnostic enzymes or biochemicals.

In addition, other responsibilities were;

- Negotiation, installation, validation and implementation of a production process for the manufacture of recombinant growth hormone (Somatonorm) in conjunction with Kabi Vitrum AB of Sweden.

- Overall responsible (until end 1986) for the design of a Fermentation Production Plant (value £30m) for Porton Products Limited. Responsibilities included liaison with the Centre for Applied Microbiology and Research and consultants, ensuring the design met current and future requirements of the company, compliance with regulatory authorities, preparation of a submission to the US FDA and carrying out the submission and budget preparation.

- Liaison between Centre for Applied Microbiology and Research and Porton Products Limited.

- Technical and scientific adviser to Porton Capital Projects, part of the Porton International Group. Involvement with projects at all stages, from early discussions with clients or prospective partner companies, through the design stages to final validation, testing and handover to client.

BIOGEN SA, GENEVA 1981 to 1985

POSITION:	1981 - 1983	Fermentation Scientist
	1983 - 1985	Head of Fermentation Development (later Process Development)

RESPONSIBLE TO: Director of Research, Geneva.

Biogen SA was one of the early biotechnology companies involved in the research, development and (later) production of new generation therapeutics and diagnostics by recombinant DNA technology.

The Geneva laboratory was started in late 1980 and was located in part of the Battelle Institute, Geneva.

Initially, my responsibilities were as follows:

- Design and construction of laboratory and pilot plant facilities for the growth of recombinant bacteria, yeast and fungi.

- Establishment of a Group to support the functions of the laboratory. The group size grew from zero to 13 staff, including one graduate student, by 1983.

- Liaison with molecular biologists, protein chemists, chemists, cell biologists, business development and senior management in project team structures.

Following the establishment of the group and promotion to Head of Department in 1983, my responsibilities also included in addition;

- Budget formulation and monitoring.

- Design, construction and operation of a larger fermentation laboratory and pilot plant facility, including dedicated process development and downstream processing facilities and analytical capability.

- Assistance in the design of a large production facility to be located in Geneva (value $13m).

The fermentation systems and processes investigated included those for the production of alpha-interferons (seven types and hybrids), beta-interferon, gamma-interferon, bovine and porcine growth hormones, hepatitis B core and surface antigens, insulin, IGF-1, colony stimulating factors, foot and mouth disease antigens, gut hormones and interleukin-2. Fermentation systems were developed for the direct comparison of genetic constructs in the laboratory and at the large scale. Certain processes were also contracted to outside establishments, under my overall supervision. The processes led to manufacture of intermediate material for final purification of the proteins, eventually to be utilised in pre-clinical testing.

DISTA PRODUCTS LIMITED 1979 to 1980

POSITION: Fermentation Technologist

RESPONSIBLE TO: Technical Services Manager

The position was in a team involved in technical services associated with the production scale (170 cubic metre) fermentation of streptomycetes for the manufacture of human and veterinary antibiotics, specifically the macrolide antibiotic Tylosin used in animal feed additives.

The main objectives were to advise on methods to maintain and improve the productivity of a number of different processes. The approaches taken involved the application of control strategies, the investigation of process changes and critical parameters and identification and correction of potential problem areas.

In addition, specific projects were undertaken, namely;

- Commissioning of a nutrient feed system following installation.
- Training of operators to utilise new procedures.
- Commissioning of new facilities for medium preparation.
- Initiation of a plant-wide program to examine the source of contamination in production processes, using basic microbiological identification techniques.

Experience was gained in the daily running operations and procedures used in a large production plant. Also, a contribution was made to laboratory and pilot plant development programs.

POSTDOCTORAL FIELD OF STUDY

1978-1979 Postdoctoral Research Fellow in the Microbiology Department, Queen Elizabeth College (now part of King's College, London).

FIELD OF RESEARCH: Antibiotic production by a newly-isolated Streptomyces species.

PROJECT HEAD: Professor S J Pirt, Head of Department

This research was in collaboration with my wife then, Dr M A S Pickett and two members of the Chemistry Department at Queen Elizabeth College.

The research project was a continuation of work that had been in progress for several years. A detailed study of antibiotic production by a newly-isolated streptomyces species was undertaken. The organism was grown in batch culture and optimal nutrient conditions for antibiotic production ascertained. The organism was also grown in repeated fed-batch culture in a 3-litre, fully-instrumented apparatus and studies of the antibiotic production at different cycle times performed.

In addition the streptomycete was grown in repeated fed-batch culture in a 400-litre pilot plant over a period of some 100 hours. Experience was gained in the day-to-day running of the pilot plant, together with seed culture stages and subsequent bulk product recovery. Antibiotics were recovered from both the mycelium and the culture supernatant using various large-scale techniques.

Antimicrobial testing of purified antibiotic was carried out to determine the exact antimicrobial spectrum of the compound. Also, preliminary toxicological testing was carried out on mice and physiological studies on rats, in conjunction with the Physiology Department at Queen Elizabeth College.

The research was supported by the National Research Development Corporation.

POSTGRADUATE FIELD OF STUDY

TOPIC OF STUDY:	Growth and composition of *Escherichia coli* under well-defined transient conditions.
SUPERVISORS:	Dr M J Bazin (Queen Elizabeth College) and Dr H H Topiwala (Shell International Chemical Corporation).

The work was performed under an SRC-CASE award (jointly sponsored by the Science Research Council and Shell International Chemical Corporation).

The basis of the research was to examine the effects of varying but defined environmental conditions on the behaviour of a microorganism grown in a model system to simulate the effects found in large-scale reactor environments.

A detailed study of the behaviour of *Escherichia coli* when grown in chemostat culture, with one environmental parameter varied repetitively in a well-defined manner, was undertaken. The input limiting substrate concentration was altered in a square-wave pattern for long periods of time. Studies centred on the response of the organism to varying amplitudes at a constant concentration amplitude. The macromolecular and elemental composition and the extensive properties of the organism were examined during the cycles and as cycle-average samples. The organism was also examined by electron microscopy to ascertain the morphology of the cells and the degree of cell lysis.

Results were also compared to predictions obtained from mathematical simulations of the system. Two mathematical models were used in this context, being representative of 'structured' and 'unstructured' types. By examining the differences between theoretical predictions and actual behaviour, certain differences between the two models could be accounted for.

The experiments indicated that cells possess a limited, research transport and biosynthetic capacity to accommodate increases in the nutritional status of their environment. Above certain cycle frequencies, cells altered their composition (particularly RNA and protein content) to accommodate increased rates of change. Also, at varying cycle amplitudes the organism again responded by increasing protein and RNA content. The transient method of continuous culture therefore indicated a method to increase the intracellular concentration of components important in industrial fermentations.

ADDITIONAL INFORMATION

MEMBER OF THE MANAGEMENT BOARD, Botulinum Research Center, Institute for Advanced Studies, Bedford, MA USA

MEMBER OF THE BRITISH PHARMACOPOEIA PANEL OF EXPERTS BIO (Biological and Biotechnological products)
(1998 to November 2021)

MEMBER OF THE EDITORIAL BOARD AND TECHNICAL EDITOR:
Journal of Chemical Technology and Biotechnology
(January 1991 to December 1996).

MEMBERSHIP OF LEARNED SOCIETIES:

Member of the Microbiology Society

Member of the Society of Chemical Industry (SCI)

Member of International Society of Toxinology (IST)

Member of International Neurotoxin Association

REVIEWER — for journals Toxins, Aesthetic Surgery Journal, the Protein Journal, Plastic and Aesthetic Research and Clinical Neurology and Neurosurgery

LANGUAGES: — French spoken fluently

AWARDS

Appreciation award as Guest Lecturer from DASIL 3rd Annual Congress, 7th-10th September 2014, Sun City, South Africa

President's award 18th SIES-VALET International Congress of Aesthetic Medicine and Surgery, 27th February -1st March 2015, Bologna, Spain

Symposium Banquet Speaker for 11th Annual Symposium Institute of Advances Sciences, Botulinum Research Center, 16th-18th August 2017, Bedford, MA, USA

Symposium Speaker for 12th Annual Symposium Institute of Advances Sciences, Botulinum Research Center, 15th-17th August 2018, Bedford, MA, USA

PUBLICATIONS

R K POOLE and A M PICKETT
Persistence of Respiratory Oscillations in Deoxyadenosine-induced Synchronous cultures of *Schizosaccaromyces pombe*
Proceedings of the Society for General Microbiology III, February 1976, pp.86

R K POOLE and A M PICKETT
Which Cells are Selected from Exponential Cultures by Continuous-Flow Centrifugation? The Selection of Small Cells from Cultures of *Escherichia coli* and *Schizosaccaromyces pombe* that Exhibit Minimal Density Fluctuations During their Cell Cycles
Journal of General Microbiology **107**, 399-402 (1978)

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A new method of industrial bioreactor operation: The transient operation technique
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Laboratory Practice **28**, 253-255 (1979)

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Biotechnology and Bioengineering **XXII**, 1213-1224 (1980)

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1. Production of a Polyene Antifungal Agent (Elizabethin) and Actinomycin in Shake Flask Culture
Journal of Chemistry and Biotechnology **31**, 167-177 (1981)

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In Microbial Population Dynamics, ed. M J Bazin, CRC Press, Florida, 1982; pp.91-124

ANDREW PICKETT and NARENDRA LADWA
Updating Fermentation Process Control
Processing, October 1985, pp.23-24

J.C. MERCHUK, N.LADWA, A.CAMERON, M.BULMER and A.PICKETT
Concentric-Tube Airlift Reactors: Effects of Geometrical Design on Performance
AIChE Journal **40**, 1105-1117 (1994)

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Liquid Flow and Mixing in Concentric Tube Air-Lift Reactors
Journal of Chemical Technology and Biotechnology **66**, 174-182 (1996)

SOUCIE JM, ERDMAN DD, EVATT BL, ANDERSON LJ, TOROK TJ, EL-JAMIL M, BARNHART, E, TEPPER, M, BURRILL, HN, PICKETT, AM, MENGELING, WL
Investigation of porcine parvovirus among persons with hemophilia receiving Hyate:C porcine factor VIII concentrate
Transfusion **40** (6), 708-711 (2000)

HUBER A, PICKETT A, SHAKESHEFF KM
Reconstruction of spatially orientated myotubes in vitro using electrospun, parallel microfibre arrays
European Cells & Materials **14,** 56-63 (2007)

PUBLICATIONS RELATED TO BOTULINUM TOXIN

A.M.PICKETT and P.HAMBLETON
Dose standardisation of botulinum toxin
The Lancet **344**, 474 (1994).

P.HAMBLETON and A.M.PICKETT
Potency equivalence of botulinum toxin preparations
Journal of The Royal Society of Medicine **87**, 719 (1994).

A. PICKETT
A botulinum toxin purified and non-immunizing?
In *Les Amis du Clostridium Tome II: Dystonie, neurone et plasticité*. Solal, 1999, pp 113-121.

ANDY PICKETT, NAVEED PANJWANI & RODERIC O'KEEFFE
Potency of Type A Botulinum Toxin Preparations in Clinical Use
40th Meeting of the Interagency Botulism Research Co-ordinating Committee, Atlanta, November 2003.
[Presented as both a poster and a platform presentation]

ANDY PICKETT, NAVEED PANJWANI, RODERIC O'KEEFFE, SARAH SHIPLEY & B R SINGH
Characterisation and Consistency of Botulinum Type A Toxin-Haemagglutinin Complex used for Clinical Therapy
41st Meeting of the Interagency Botulism Research Co-ordinating Committee, Baltimore, October 2004.
[Presented as both a poster and a platform presentation]

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EXHIBIT 9 to Brin Declaration (REDACTED)

abbvie

CONTAINS COMMERCIALLY CONFIDENTIAL INFORMATION
NOT SUBJECT TO FOIA; PRE-DISCLOSURE NOTIFICATION REQUIRED

DECLARATION OF MARTIN GASTENS, PH.D.

1. I am currently the Vice President of Parenteral Product Development, Science & Technology at AbbVie. I oversee a team that is responsible for ensuring efficient development and technical commercial support of biological products that are manufactured to high quality standards and reliably delivered to patients. My team is also responsible for supporting process improvements, regulatory compliance, and technology transfer from research to commercial production of biological products.

2. In 1999, I received a biology degree from the Heinrich Heine University in Duesseldorf, Germany, followed by a Ph.D. in biology from the Institute of Medical Microbiology & Virology at the same University in 2003.

3. Since 2007, I have been employed by AbbVie (formerly Abbott) where I have held positions of increasing responsibility related to biologics quality control, product development, and operations. I have extensive experience in the pharmaceutical industry, with significant expertise in the unique scientific and operational challenges of manufacturing complex therapeutic proteins for development and commercial uses.

4. I have overseen the development of neurotoxins at AbbVie since July 2022 and provided technical commercial support for toxin biological products including BOTOX since June 2024.

5. I have been asked to provide a high-level summary of the BOTOX manufacturing process for the biological product, including the important role HSA plays in the BOTOX product.

I. BOTOX MANUFACTURING BACKGROUND.

6. Since its original approval, BOTOX has always contained three ingredients: onabotulinumtoxinA (onabotA), human serum albumin (HSA), and sodium chloride. Both onabotA and HSA are large, complex proteins.

7. Due to the structural and biochemical complexities of onabotA, the biological activity of BOTOX is influenced by its manufacturing environment, including the ingredients, processes, and conditions applied at each step. Since variations in the manufacturing process can impact the clinical characteristics of BOTOX, AbbVie takes great care to ensure the consistency of BOTOX throughout the manufacturing process. Every part is tightly controlled, with meticulous attention given to the materials and equipment used during each step.

8. HSA is a recognized source of variability due to its human origin and the chemical and physical conditions of the manufacturing process used to separate HSA from other plasma components.

9. Before any HSA raw material is used to manufacture BOTOX, AbbVie must confirm that the physical and chemical properties of the HSA meet pre-set specifications. Even

then, there are controls throughout the BOTOX manufacturing process to ensure that [redacted]

II. BOTOX SUPPLY IS HEAVILY RELIANT ON HSA.

10. Recognizing the integral role HSA has in BOTOX, AbbVie has proactively safeguarded its HSA supply by qualifying two suppliers under its biologics license application (BLA). This redundancy is designed to minimize the risk of BOTOX shortages due to issues with HSA availability or changes by HSA suppliers. [redacted]

11. [redacted] Plasma, the source from which HSA is derived, is a complex natural mixture containing various proteins and phospholipids from human donors. As such the composition of the final HSA material can be impacted by changes in the vendor's commercial process to extract and isolate the HSA protein. [redacted]

12. [redacted]

13. [redacted]

14. With no doubt, variability in HSA supply has directly affected BOTOX supply. The critical relationship between onabotA potency and HSA means that BOTOX supply is at risk without an adequate supply of an appropriately qualified HSA. Even when alternative HSA supply is available, switching suppliers requires a significant investment of time and resources due to the uniquely intricate relationship between onabotA and HSA.

III. RECENT FDA APPROVAL OF PROCESS CHANGES ALSO REENFORCES THE UNIQUE RELATIONSHIP BETWEEN ONABOTA AND HSA.

15. AbbVie continues to invest in efforts to optimize the BOTOX manufacturing process and explore ways to reduce batch-to-batch variability in the final biological product. Although these process improvements do not alter any critical quality attributes of BOTOX, they are important for ensuring reproducibility and consistent product characteristics to patients.

16. Earlier this year, FDA approved process modifications [redacted]

17. [redacted]

18. [redacted]

19. This emphasizes the highly sensitive and extremely dynamic relationship between onabotA and HSA, even minor changes to the toxin environment—such as the conditions under which HSA is added to onabotA—has a meaningful impact on BOTOX, the biological product.

M. Gastens

Martin Gastens, Ph.D.
Vice President
Parenteral Product Development, Science & Technology
AbbVie Deutschland GmbH & Co. KG

December 5, 2025