

Botulinum toxin type A in the treatment of patients with cervical dystonia

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Abstract: Dystonia is an involuntary movement involving twisting and turning of agonist and antagonist muscles. Cervical dystonia is isolated to neck musculature. Botulinum toxin type A is a safe and effective treatment of this disabling and often painful syndrome. Three forms of botulinum toxin type A are available worldwide to treat patients with cervical dystonia. This is a review of the studies of botulinum toxin type A to treat cervical dystonia.

Keywords: dystonia, botulinum toxin type A, cervical dystonia

Introduction

Dystonia is an involuntary movement involving twisting and turning of agonist and antagonist muscles.¹ Cervical dystonia (CD) is a dystonic disorder of the neck musculature, characterized by tilting, turning of the neck.² CD is often painful and leads to disability interfering with working, driving, reading and other activities of daily living.³ In a 1988 retrospective chart review, the prevalence of CD is estimated to be 9/100,000.⁴ More recently, the Dystonia Medical Research Foundation estimates that 250,000 people suffer from CD but many more are suspected to be undiagnosed and/or untreated (<http://www.dystonia-foundation.org/>).

Idiopathic CD generally presents slowly over several years in mid to late adulthood but the age of onset may vary widely. Secondary causes of CD include tardive dystonia after exposure to dopamine blocking medications and post-traumatic dystonia.⁵ While CD presents with an involuntary tilt or turn of the neck muscles, the patients also frequently experience pain. Regardless of the etiology, CD often results in permanent disability due to decreased range of motion, involuntary movements and intractable pain.⁶ All forms of CD can benefit from selective injection of botulinum toxin into overactive muscles of the neck.

Before the introduction of botulinum toxin (BoNT) to treat CD, therapy was limited to oral medication and surgery. Oral medications included anticholinergics, benzodiazepines, and antispasticity medications.⁷ Oral medications are often associated with side effects of sedation, dry mouth, and withdrawal symptoms. In the past, surgery for CD included selective denervation and myotomies,^{8–10} but more recently have been replaced with deep brain stimulation (DBS) for refractory cases.¹¹ In 2007 the experienced Canadian group published a series of 10 patients with medical refractory CD who improved on with stimulation of bilateral globus pallidus internus.¹¹ In a larger study, 40 patients with primary segmental or generalized dystonia treated with DBS were randomly assigned to receive either neurostimulation or sham stimulation for 3 months. After 3 months significant improvement in the Burke-Fahn-Marsden Dystonia Rating Scale was noted in the stimulations group compared to the sham stimulations group ($p < 0.001$). Adverse events included lead infection, lead dislodgment and dysarthria.¹²

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The introduction of botulinum toxin type A (BoNT-A) in 1989 in the US dramatically improved the care of patients with CD. The addition of botulinum toxin type B (BoNT-B) in 2000 as an alternative treatment to type BoNT-A further expanded the resources to treat this disabling disease.^{13,14} In 2008 the Therapeutics and Technology Assessment Subcommittee of the American Academy of Neurology concluded that BoNT is established as safe and effective for the treatment of CD based on seven Class I studies.¹⁵ Based upon a review of the literature, the expert group recommended that BoNT injection should be offered as a treatment option to patients with CD and that BoNT is probably more efficacious and better tolerated in patients with CD than treatment with trihexyphenidyl (see Table 1). The reports also noted that there were no data to compare BoNT with any form of surgical treatment of CD.¹⁵

Serotypes of botulinum toxin

The bacterium, *Clostridium botulinum* produces seven immunologically distinct serotypes but only serotype A^{16–18} and B^{14,19} have been used routinely in clinical practice to treat patients with CD. Serotype F has limited availability in Japan.^{20,21} Worldwide there are three forms of botulinum toxin type A (BTX-A): BOTOX[®] (Allergan, Irvine, CA), Dysport[®] (Ipsen), and Xeomin[®] (Merz Pharmaceuticals GmbH, Frankfurt, Germany) commercially available. The different brands vary by commercial processing, strength and amounts of protein. Each of the type A serotypes is a unique form of botulinum toxin. Of the non A serotypes, serotype B is available as botulinum toxin type B (BoNT-B) (marketed as Myobloc[™] in the US and NeuroBloc[®] elsewhere). The development of new formulations or different serotypes of botulinum toxin is an active research area and more options for patients may be forthcoming.

Each of the seven serotypes of botulinum toxin is synthesized as a single-chain polypeptide with a molecular weight of 150 kDa.²² To activate the toxin, the disulfide bond holding the heavy chain and light chain together must be cleaved,²³ followed by internalization of the light chain into the cytosol. The entire process is called “chemodenervation”.^{24,25}

The pharmacologic activity of the specific light chain of the serotype is specific to which proteins it interacts with at the neuromuscular junction.²⁶ Regardless of the type of serotype used, once the toxin is recognized at the presynaptic terminal and internalized by endocytosis and translocated to the cytosol. Subsequently at the presynaptic nerve terminal the light chain catalyses a zinc-dependent protein cleavage.

The result deactivates components of the “SNARE” (soluble N-ethyl-maleimide sensitive factor attachment protein Receptor complex).^{26–28}

Different components of the SNARE are required for successful release of the acetylcholine vesicle.²⁶ Of the seven serotypes, there are differences at the level of acceptor binding and substrate interference in the SNARE complex. The light chain of each serotype acts at a distinct site on one or more of the proteins required for vesicle release. The light chain of BTX type A and E targets the cytoplasmic protein SNAP-25 while the light chain of serotype B toxin specifically targets VAMP/synaptobrevin.^{29,30,31} Even when the same protein is affected, the different serotypes (for example A and E) affect it at a different sites of the same protein. Clinically, treatment with BoNT-A relaxes muscles, but the dose required and side effects may vary by formulation.

The bacterial proteins in BoNTs have the potential to elicit immunologic responses when injected in humans. Neither the incidence rates for neutralizing antibodies nor the clinical meaningfulness of the presence of antibodies have been clearly established. The exact cause of antibody formation to BoNT in a particular patient is unknown but studies have linked the development of neutralizing antibodies to the formulations and its protein load, the dose used per treatment cycle, the total cumulative dose given to a patient and the frequency of repeated injections.^{17,32,33}

Antibodies to BoNT have been measured by determining the clinic response in the patient. For example in the Frontalis Antibody Test (FTAT) or Unilateral Brow Injection (UBI) either the Frontalis or corrugator muscle are injected with a small amount of BoNT-A capable of eliciting a clinical response.^{32,34–36} Others have used the change in a Compound Motor Action Potential (CMPA) after treatment of the extensor digitorum brevis (EDB) in the foot with toxin.³⁷ Still others have used the Mouse Protection Assay (MPA) to determine the ability of antibodies in the patient to protect a toxin naïve mouse when exposed to a fatal dose of toxin.^{34,38,39} Regardless of the method, the secondary development of antibodies in patients with a previous response to BoNT remains a concern for patients and physicians using this effective form of treatment. Once neutralizing antibodies develop, a sustained benefit to that serotype is rarely experienced by the patient.⁴⁰

Early experience with BoNT-A (Dysport and BOTOX[®]) in CD suggests that the incidence of neutralizing antibodies ranges from 4% to 17%, depending on the formulation used.^{26,41} A new formulation of BOTOX[®] with a lower protein load has been in use since 1998.³⁸ A prospective longitudinal

Table 1 BoNT-A for cervical dystonia. Adapted with permission from Simpson DM, Blitzer A, Brashear A, et al. Assessment: Botulinum neurotoxin for the treatment of movement disorders (an evidence-based review): report of the Therapeutics and Technology Assessment Subcommittee of the American Academy of Neurology 3. *Neurology*. 2008;70(19):1699–1706. ¹⁵ Copyright © 2008 Lippincott Williams & Wilkins

Reference	Design	Cohort size	Treatment (serotype/brand/dose)	Follow-up	Outcome measures/ (1-primary, 2-secondary)	Adverse events	Results/effect size
57	Randomized, double-blind, placebo-controlled	55 BoNT-naive	A/Botox®: 118 U	3 mo	Head turn rest/walk Patient assessment Pain scale Functional capacity	11 with pain ≥ 24 h at injection site 3 with dysphagia, fatigue, weakness, increased spasms	61% improved during double blind phase
52	Randomized, double-blind, placebo-controlled	75 BoNT-naive	A/Dysport Total 250, 500, or 1000 U	2 mo	Tsui scale; Global impression	Neck weakness, voice change, and dysphagia	Improvement ($p \leq 0.05$) for 500 U, 1000 U at week 4, not at week 8. Subjective improvement in 45%–50%
58	Randomized, double-blind	66 BoNT-naive	A/Dysport 292 U vs Trihexyphen idyl//mean = 16.25 mg	3 mo	Tsui scale, general health perception, TWSTRS disability and pain	More AEs in trihexyphenidyl: dry mouth, forgetfulness, fatigue; 3 with neck weakness	BoNT-A improved more on Disability of TWSTRS or Tsui
34	Randomized, double-blind, placebo-controlled	80 Prior Botox®-experi-benefit from 80–250 U	A/Dysport 500 U	4 mo	Total TWSTRS	blurred vision and neck weakness than placebo	Mean improvement in TWSTRS = 9.9 for BTX and 3.8 for placebo ($p = 0.01$)

Abbreviations: AE, adverse events; TWSTRS, Toronto western spasmodic torticollis rating scale; U, unit; BoNT, botulinum toxin; BoNT-A, botulinum toxin type A.

study in 326 subjects with CD found the incidence of neutralizing antibodies to be 1.2%.³³ Only 4 of 326 subjects tested positive for antibodies in the MPA; 3 of these subjects stopped responding clinically to BoNT-A (of whom one also showed clinical resistance in the FTAT) and one continued to respond. The formulation for Dysport[®] has not been reported to have changed. Limited studies with Dysport[®] have reported its incidence of neutralizing antibody formation to be between 0% and 3%.²⁶ No clinical immunogenicity data have been published to date for Xeomin[®].⁴³

Botulinum toxin for treatment of CD

Because of the existence of three formulations of BoNT-A, all different formulations, trade names will be used in this section to discern which formulations are being discussed.

Studies of botulinum toxin type A (formulated as BOTOX[®])

BOTOX[®] (sold in the US since the late 1980s) was pioneered by Dr Allan Scott, a pediatric ophthalmologist, for the treatment of strabismus.⁴⁴ Originally labeled Oculinum, once marketed by Allergan, Inc., the formulation quickly became known by its trade name, BOTOX[®]. Off-label treatment of patients with CD with BOTOX[®] started in the late 1980s and continued until FDA approval in 2000. Despite the acceptance of the safety and efficacy of BOTOX[®], only a limited number of large multi-center trials in the treatment of CD with BOTOX[®] have been published (see Table 1).

The largest trial of BoNT-A (formulated as BOTOX[®]) studied 170 subjects with CD at 21 centers is published in abstract form and the package insert.⁴¹ Prior BOTOX[®] responders who continued to receive a good response to injections were included in the study. A unique outcome measure, the Cervical Dystonia Severity Scale (CDSS), similar to a large protractor, measured the change in turn, tilt or shift after treatment.⁴⁵ Subjects were treated with BOTOX[®] and followed for 12 weeks with the CDSS. At 12 weeks after the first injection, the patients who responded to the BOTOX[®] and demonstrated 20° or greater change in head position from normal were randomized to receive either BOTOX[®] or placebo. The difference between the BTX-A treated and placebo-treated groups on the CDSS was 1.03 to 3.13 (corresponding to an improvement of approximately 5.15° to 10.65° in head position) ($p \leq 0.046$) at weeks 2, 4, 6, 8, and 10. In addition, a Physician's Global Assessment score was statistically significant at weeks 2, 4, 6, and 8. According to the package insert, the common adverse events for the BTX-A

treated patients were upper respiratory infections, neck pain, back pain, dysphagia, and rhinitis. This is the largest reported multi-center, double-blind, placebo-controlled trial with BOTOX[®] in patients with CD.^{2,41,46,47} However, this study has only been reported in abstract form and the information listed above is available in the package insert approved by the US Food and Drug Administration.⁴¹

Older studies noted improvement in patients with CD treated with BOTOX[®]. In 1990, Jankovic et al reported follow-up of 202 out of 232 subjects with medically intractable CD for at least 3 months and up to 4 years, during which time they received 1074 injections of BTX-A.¹⁸ Seventy-one percent reported improvement of symptoms and, in those with pain, 76% had almost complete relief of their pain. Side effects included mild dysphagia and neck weakness. In series of smaller studies, Gelb et al administered injections to 20 patients with CD and demonstrated subjective improvement in 80% of subjects.⁴⁸ Blackie et al reported the results of 19 patients who participated in a double-blind placebo-controlled trial of BTX-A in patients with CD, demonstrating a mean improvement in neck posture in 83% if the treatment periods had a mean duration of 12 weeks.²⁵ A recent long-term study⁴⁷ also found that efficacy (measured as global response) and duration of effect tend to increase over time. In a longitudinal study of dose consistency, Brashear et al⁴⁸ found that dose and dose interval were consistent over a 2-year period.⁴⁹

Studies of CD with botulinum toxin type A (formulated as Dysport[®])

Botulinum toxin type A (Dysport[®]) is currently available outside the US and is labeled in European countries for treatment of CD.^{50,51} Efficacy and safety of Dysport[®] for treatment of CD has been studied in fixed dose⁵² and variable dose³⁴ protocols.

In a large European multi-center study, 75 patients were randomly assigned to receive treatment with placebo or total doses of 250, 500, and 1000 Dysport[®] units (U) divided between two muscles, the splenius capitus and the contralateral sternocleidomastoid. Those treated with 500 or 10000 U demonstrated significant changes in the modified Tsui score at week 4 versus placebo ($p \leq 0.05$). Additionally positive dose-response relations were found for the degree of subjective patient report, duration of improvement, and improvement on a clinical global rating scale. A dose-response relationship for treatment-related adverse events occurred with increasing doses. The authors suggested that a starting dose of 500 U of Dysport[®] should be used to lower

the risk of neck muscle weakness and voice changes and to assure some efficacy. The starting recommendations were 500 U with titration upwards to 1000 U as needed.⁵²

In the first multi-center, double-blind, randomized, controlled trial in the US, 80 patients with CD were randomly assigned to receive one treatment with Dysport® (500 U) or placebo.³⁴ Over the 20-week trial, Dysport® demonstrated more improvement in the total Toronto Western Spasmodic Torticollis Rating Scale score than placebo at weeks 4, 8, and 12. The median duration of response to Dysport® was 18.5 weeks. Side effects were generally similar in the two treatment groups; only blurred vision and weakness occurred significantly more often with Dysport®.

In the dose-ranging study increasing doses of Dysport® resulted in improved duration of response but with increasing overall incidence of adverse events (AEs) (37%, 65%, and 83% with 250, 500, and 1000 U, respectively).⁴⁹ The most common AEs were dry mouth (21%, 18%, and 33% with 250, 500, and 1000 U, respectively), neck muscle weakness (11%, 12%, and 56% with 250, 500 and 1000 U, respectively), and dysphagia (21%, 29%, and 39% with 250, 500, and 1000 U, respectively).⁵²

A study comparing trihexyphenidyl tablets versus injections of Dysport® in 64 patients with CD demonstrated significant changes in favor Dysport® on the disability section of the Toronto Western Spasmodic Torticollis Rating Scale (TWSTRS-Disability) (primary outcome), Tsui Scale, and the General Health Perception Subscale.⁵³ More patients treated with Dysport had an improvement of at least 3 points on the TWSTRS-Disability subscale and on the Tsui Scale than those treated with oral medication. In addition, adverse effects were significantly less frequent in the Dysport group than the trihexyphenidyl treated group.⁵³

Those physicians who have both BOTOX® and Dysport® available in their countries need to have a clear understanding of the differences in dosing. While these are both from the type A serotype, they are formulated differently and human studies demonstrate different doses needed for treatment. In a comparative, double-blind study of Dysport® and BOTOX®, patients with prior exposure to BOTOX® demonstrated that a Dysport® dose of 3 times the BOTOX® dose demonstrated similar efficacy with no difference in safety profiles (58% of the Dysport® group reporting adverse events versus 69% of the BOTOX® group [$p = 0.85$]).⁵⁴ The duration of effect, assessed by time to pretreatment, was also similar (mean [SD]; Dysport®, 83.9 [13.6] days; BOTOX®, 80.7 [14.4] days; $p = 0.85$).⁵⁵ Of note is that the large clinical trial (which led to labeling of BOTOX® for CD in the US)

had a average dose of 232 U. BOTOX® and Dysport® are different medications and are dosed differently. The difference between the recommended dose of 500 U of Dysport® by Wissel et al⁵¹ and the average dose of BOTOX® of 232 U listed in the package insert should be clearly understood by treating physicians who have access to both drugs.

Studies of BoNT-A (formulated as Xeomin®)

Xeomin® is a newly formulated purified, freeze-dried BTX-A reportedly free of complexing proteins. No placebo-controlled trial of Xeomin® in patients with CD has been performed. However, in a head to head comparative trial, of BOTOX® and Xeomin®, both drugs were comparable in the treatment of patients with CD.⁴³ A total of 466 patients with CD were recruited from 51 European centers to participate in the non-inferiority study. Using the TWSTRS total rating scale as the primary outcome, the study demonstrated no difference in efficacy between the two drugs on day 28 after injection. At baseline, patients in both groups had a median TWSTRS Severity score of 18 points and scores improved by a median of 11 points in both groups at day 28 after treatment. Both treatment groups improved compared to baseline (p value for change in TWSTRS Severity score was $p \leq 0.0001$).⁴³ The authors propose that the clinical effects of the two drugs were comparable in the patients studied.

Technique of botulinum toxin injections for CD

The technique of botulinum toxin injection remains both a science and an art. An understanding of the anatomy of the neck remains essential. The interaction of the neck musculature and head movement determines which muscles are involved in the primary problem movement in the individual patients (see Table 2 for a list of muscles typically involved in these neck movements). Moreover, the patient may have a compensatory movement used to ally some of the discomfort of CD. Many practitioners use a series of clinical observations and palpation to determine which muscles to treat and how much BoNT-A to use in a particular patient.²

The use of electromyography remains controversial. While those who inject BTX-A for CD may not consistently use electromyography, Comella et al reported a significantly greater magnitude of improvement in those in whom the neck muscles treated were selected by clinical and electromyographic guidance than those in whom only clinical examination was used.⁵⁶ In the Therapeutics and Technology Assessment of the American Academy of

Table 2 Typical muscles involved in cervical dystonia (adapted from Brashear⁵⁹)

Predominant movement	Muscles involved
Turn (torticollis)	Ipsilateral Splenius/semispinalis capitis Contralateral sternocleidomastoid
Tilt (laterocollis)	Ipsilateral sternocleidomastoid Ipsilateral Splenius/semispinalis capitis Ipsilateral Scalene complex Ipsilateral Levator scapulae Ipsilateral Posterior paravertebrals
Shoulder elevation	Ipsilateral Levator scapulae Ipsilateral Trapezius
Retrocollis	Bilateral Splenius/semispinalis capitis Bilateral Upper Trapezius Bilateral Deep Posterior Paravertebrals
Anterocollis	Bilateral Sternocleidomastoid Bilateral Scalene complex Bilateral Submental complex

Neurology, the authors concluded that the use of EMG remained controversial.¹⁵

Regardless of which formulation is used to treat patients, the practitioner should understand the muscles involved in the primary head movements of tilt, turn, anterocollis, retrocollis, and shoulder elevation (see Table 2 for a list of common muscles treated with BoNT-A). Focusing on the primary movement provides the patient with relief while limiting the side effects from treating non-involved muscles.

Summary

BoNT-A is a safe and effective therapy for CD. Three formulations are available worldwide, but as of 2008 only BOTOX[®] is available in the US. The dosages between the formulations of Dysport and BOTOX[®] are different and the dosage of Xeomin[®] in preliminary studies mirrors BOTOX[®] dosing. The use of BoNT-A has dramatically improved treatments for patients with CD. Physicians treating patients with CD will need to be attentive to the burgeoning literature on all three forms of BoNT-A, including variations in dosing, efficacy, and side effects.

Disclosures

Dr Brashear is a consultant for Allergan and Merz. Dr Brashear has current research relationships with Allergan, Merz and Ipsen Pharmaceuticals. In the past Dr Brashear also has had consulting and research relationships with Solstices and Elan. Over the last 5 years she has served on advisory

boards for Allergan, Solstice and Merz. Dr Brashear is not an employee nor owns stock in any of these companies. She does not receive patent royalties for any work with botulinum neurotoxins.

References

- Fahn S, Bressman SB, Marsden CD. Classification of dystonia. *Adv Neurol*. 1998;78:1–10.
- Brashear A. The botulinum toxins in the treatment of cervical dystonia. *Semin Neurol*. 2001;21(1).
- Jankovic J, Leder S, Warner D, Schwartz K. Cervical dystonia: clinical findings and associated movement disorders. *Neurology*. 1991;41(7):1088–1091.
- Nutt JG, Muenter MD, Melton LJ III, Aronson A, Kurland LT. Epidemiology of dystonia in Rochester, Minnesota 5. *Adv Neurol*. 1988;50:361–365.
- Geyer HL, Bressman SB. The diagnosis of dystonia 2. *Lancet Neurol*. 2006;5(9):780–790.
- Jankovic J, Brin MF. Botulinum toxin: historical perspective and potential new indications. *Muscle Nerve Suppl*. 1997;6(45):5129–5145.
- Bressman SB. Dystonia update. *Clin Neuropharmacol*. 2000;23(5):239–251.
- Ward AB, Molenaers G, Colosimo C, Berardelli A. Clinical value of botulinum toxin in neurological indications. *Eur J Neurol*. 2006;13 (Suppl 4):20–26.
- Duvoisin RC. Spasmodic torticollis: the role of surgical denervation. *Mayo Clin Proc*. 1991;66(4):433–435.
- Braun V, Richter HP. Selective peripheral denervation for spasmodic torticollis: 13-year experience with 155 patients. *J Neurosurg*. 2002;97(Suppl 2):207–212.
- Hung SW, Hamani C, Lozano AM, et al. Long-term outcome of bilateral pallidal deep brain stimulation for primary cervical dystonia 10. *Neurology*. 2007;68(6):457–459.
- Kupsch A, Benecke R, Muller J, et al. Pallidal deep-brain stimulation in primary generalized or segmental dystonia 2. *N Engl J Med*. 2006;355(19):1978–1990.
- Brin MF, Lew MF, Adler CH, et al. Safety and efficacy of NeuroBloc (botulinum toxin type B) in type A-resistant cervical dystonia. *Neurology*. 1999;53(7):1431–1438.
- Brashear A, Lew MF, Dykstra DD, et al. Safety and efficacy of NeuroBloc (botulinum toxin type B) in type A-responsive cervical dystonia 8. *Neurology*. 1999;53(7):1439–1446.
- Simpson DM, Blitzer A, Brashear A, et al. Assessment: Botulinum neurotoxin for the treatment of movement disorders (an evidence-based review): report of the Therapeutics and Technology Assessment Subcommittee of the American Academy of Neurology 3. *Neurology*. 2008;70(19):1699–1706.
- Scott AB, Magoon EH, McNeer KW, Stager DR. Botulinum treatment of strabismus in children. *Trans Am Ophthalmol Soc*. 1989;87:174–80.
- Greene P, Fahn S, Diamond B. Development of resistance to botulinum toxin type A in patients with torticollis. *Mov Disord*. 1994;9(2):213–217.
- Jankovic J, Schwartz K. Botulinum toxin injections for cervical dystonia. *Neurology*. 1990;40(2):277–280.
- Lew MF, Adornato BT, Duane DD, et al. Botulinum toxin type B: a double-blind, placebo-controlled, safety and efficacy study in cervical dystonia 3. *Neurology*. 1997;49(3):701–707.
- Greene PE, Fahn S. Use of botulinum toxin type F injections to treat torticollis in patients with immunity to botulinum toxin type A. *Mov Disord*. 1993;8(4):479–483.
- Houser MK, Sheean GL, Lees AJ. Further studies using higher doses of botulinum toxin type F for torticollis resistant to botulinum toxin type A. *J Neurol Neurosurg Psychiatry*. 1998;64(5):577–580.
- Dasgupta BR, Berry LJ, Boroff DA. Purification of Clostridium botulinum type A toxin. *Biochim Biophys Acta*. 1970;214(2).

23. Brin MF, Comella CL, Jankovic J, Lai F, Naumann M. Long-term treatment with botulinum toxin type A in cervical dystonia has low immunogenicity by mouse protection assay 1. *Mov Disord.* 2008;23(10):1353–1360.
24. Brin MF. Botulinum toxin: chemistry, pharmacology, toxicity, and immunology. *Muscle Nerve Suppl.* 1997;6(68):S146–S148.
25. Blackie JD, Lees AJ. Botulinum toxin treatment in spasmodic torticollis. *J Neurol Neurosurg Psychiatry.* 1990;53(8):640–643.
26. Brashear A. Clinical comparisons of botulinum neurotoxin formulations 1. *Neurologist.* 2008;14(5):289–298.
27. Xiao AY, Wei L, Xia S, Rothman S, Yu SP. Ionic mechanism of ouabain-induced concurrent apoptosis and necrosis in individual cultured cortical neurons. *J Neurosci.* 2002;22(4):1350–1362.
28. Brin MF, K Roger Aoki. Botulinum toxin Type A: Pharmacology. In: Mayer NSD, editor. New York: WeMove; 2002. p 110–124.
29. Burgen AS, Dickens F, Zatman LJ. The action of botulinum toxin on the neuro-muscular junction 1. *J Physiol.* 1949;109(1–2):10–24.
30. Setler PE. Therapeutic use of botulinum toxins: background and history 1. *Clin J Pain.* 2002;18(Suppl 6):S119–S124.
31. Setler P. The biochemistry of botulinum toxin type B 2. *Neurology.* 2000;55 12(Suppl 5):S22–S28.
32. Comella CL, Jankovic J, Shannon KM, et al. Comparison of botulinum toxin serotypes A and B for the treatment of cervical dystonia. *Neurology.* 2005;65(9):1423–1429.
33. Brin MF, Comella CL, Jankovic J, Lai F, Naumann M. Long-term treatment with botulinum toxin type A in cervical dystonia has low immunogenicity by mouse protection assay 1. *Mov Disord.* 2008;23(10):1353–1360.
34. Truong D, Duane DD, Jankovic J, et al. Efficacy and safety of botulinum type A toxin (Dysport) in cervical dystonia: results of the first US randomized, double-blind, placebo-controlled study. *Mov Disord.* 2005;20(7):783–791.
35. Hanna PA, Jankovic J. Mouse bioassay versus Western blot assay for botulinum toxin antibodies: correlation with clinical response. *Neurology.* 1998;50(6):1624–1629.
36. Hanna PA, Jankovic J, Vincent A. Comparison of mouse bioassay and immunoprecipitation assay for botulinum toxin antibodies. *J Neurol Neurosurg Psychiatry.* 1999;66(5):612–616.
37. Hamjian JA, Walker FO. Serial neurophysiological studies of intramuscular botulinum-A toxin in humans. *Muscle Nerve.* 1994;17(12):1385–1392.
38. Jankovic J, Vuong KD, Ahsan J. Comparison of efficacy and immunogenicity of original versus current botulinum toxin in cervical dystonia. *Neurology.* 2003;60(7).
39. Jankovic J, Hunter C, Dolimbek BZ, et al. Clinico-immunologic aspects of botulinum toxin type B treatment of cervical dystonia. *Neurology.* 2006;67(12):2233–2235.
40. Dressler D, Munchau A, Bhatia KP, Quinn NP, Bigalke H. Antibody-induced botulinum toxin therapy failure: can it be overcome by increased botulinum toxin doses? *Eur Neurol.* 2002;47(2): 108–121.
41. Allergan I. Package Insert, BOTOX (Botulinum toxin Type A Purified Neurotoxin Complex). 2000. Irvine Allergan Inc. 2000.
42. Elan Pharmaceuticals I. Package Insert Myobloc Botulinum toxin type B injectable solution. 2000. South San Francisco Elan, 2000.
43. Benecke R, Jost WH, Kanovsky P, Ruzicka E, Comes G, Grafe S. A new botulinum toxin type A free of complexing proteins for treatment of cervical dystonia. *Neurology.* 2005;64(11):1949–1951.
44. Scott AB, Kraft SP. Botulinum toxin injection in the management of lateral rectus paresis. *Ophthalmology.* 1985;92(5):676–683.
45. O'Brien C, Brashear A, Cullis P, et al. Cervical dystonia severity scale reliability study 9. *Mov Disord.* 2001;16(6):1086–1090.
46. Adler CH, Kumar R. Pharmacological and surgical options for the treatment of cervical dystonia. *Neurology.* 2000;55(12):S9–S14.
47. Brashear A. The safety and tolerability of botulinum toxins for the treatment of cervical dystonia. *Expert Opin Drug Saf.* 2005;4(2).
48. Gelb DJ, Lowenstein DH, Aminoff MJ. Controlled trial of botulinum toxin injections in the treatment of spasmodic torticollis. *Neurology.* 1989;39(1):80–84.
49. Brashear A, Bergan K, Wojcieszek J, Siemers ER, Ambrosius W. Patients' perception of stopping or continuing treatment of cervical dystonia with botulinum toxin type A. *Mov Disord.* 2000;15(1):150–154.
50. Haussermann P, Marzoch S, Klinger C, Landgrebe M, Conrad B, Ceballos-Baumann A. Long-term follow-up of cervical dystonia patients treated with botulinum toxin A. *Mov Disord.* 2004;19(3):303–308.
51. Wissel J, Kanovsky P, Ruzicka E, et al. Efficacy and safety of a standardised 500 unit dose of Dysport (clostridium botulinum toxin type A haemagglutinin complex) in a heterogeneous cervical dystonia population: results of a prospective, multicentre, randomised, double-blind, placebo-controlled, parallel group study. *J Neurol.* 2001;248(12):1073–1078.
52. Poewe W, Deuschl G, Nebe A, et al. What is the optimal dose of botulinum toxin A in the treatment of cervical dystonia? Results of a double blind, placebo controlled, dose ranging study using Dysport. German Dystonia Study Group. *J Neurol Neurosurg Psychiatry.* 1998;64(1):13–7.
53. Brans JW, Aramideh M, Koelman JH, Lindeboom R, Speelman JD, Ongerboer de Visser BW. Electromyography in cervical dystonia: changes after botulinum and trihexyphenidyl. *Neurology.* 1998;51(3):815–818.
54. Ranoux D, Gury C, Fondarai J, Mas JL, Zuber M. Respective potencies of Botox and Dysport: a double blind, randomised, crossover study in cervical dystonia. *J Neurol Neurosurg Psychiatry.* 2002;72(4):459–462.
55. Odergren T, Hjaltason H, Kaakkola S, et al. A double blind, randomised, parallel group study to investigate the dose equivalence of Dysport and Botox in the treatment of cervical dystonia 3. *J Neurol Neurosurg Psychiatry.* 1998;64(1):6–12.
56. Comella CL, Buchman AS, Tanner CM, Brown-Toms NC, Goetz CG. Botulinum toxin injection for spasmodic torticollis: increased magnitude of benefit with electromyographic assistance. *Neurology.* 1992;42(4):878–882.
57. Greene P, Kang U, Fahn S, Brin M, Moskowitz C, Flaster E. Double-blind, placebo-controlled trial of botulinum toxin injections for the treatment of spasmodic torticollis. *Neurology.* 1990;40(8):1213–1218.
58. Brans JW, Lindeboom R, Snoek JW, et al. Botulinum toxin versus trihexyphenidyl in cervical dystonia: a prospective, randomized, double-blind controlled trial. *Neurology.* 1996;46(4):1066–72.
59. Brashear A. Treatment of Cervical Dystonia with Botulinum Toxin. Blitzer A (ed). [15],122–7. 6-1-0004. Elsevier. Operative Techniques in Otolaryngology Head and Neck Surgery. Friedman Michael.

