

Consensus Statement Regarding Storage and Reuse of Previously Reconstituted Neuromodulators

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BACKGROUND Legacy recommendations suggest that vials of botulinum toxin be used within 24 hours of reconstitution and in a single patient. Current standard of care is consistent with storage after reconstitution and use of a single vial for several patients.

OBJECTIVE To develop expert consensus regarding the effectiveness and safety of storage and reuse of botulinum toxin.

MATERIALS AND METHODS The American Society for Dermatologic Surgery authorized a task force of content experts to review the literature and provide guidance. Data extraction was followed by clinical question review, a consensus Delphi process, and validation of the results by peer review.

RESULTS After 2 rounds of Delphi process, the task force concluded by unanimous consensus and with the highest level of confidence that a vial of toxin reconstituted appropriately can, for facial muscle indications, be (1) refrigerated or refrozen for at least 4 weeks before injection without significant risk for contamination or decreased effectiveness and (2) used to treat multiple patients, assuming appropriate handling.

CONCLUSION The standard of care, which allows for use of botulinum toxin more than 24 hours after reconstitution and in more than 1 patient per vial, is appropriate and consistent with the safe and effective practice of medicine.

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The purpose of this guideline document is to review the available evidence regarding storage and reuse of previously reconstituted vials of botulinum toxin Type A and to offer clinical recommendations. This is a new guideline topic, and no prior, revised, or updated American Society for Dermatologic Surgery guidelines specifically address this topic.

Scope of the Guidelines

These guidelines are intended to address the time since reconstitution with saline during which botulinum toxin Type A can be safely and effectively used for clinical indications in adult humans and the extent to which a single prepackaged sterile vial of such toxin can be safely and effectively reconstituted for use on more than 1 such patient.

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Safety and effectiveness is reviewed in the context of cutaneous and subcutaneous injection of reconstituted botulinum toxin to induce temporary chemo-denervation of affected musculature, particularly the procerus, corrugator, frontalis, and orbicularis oculi muscles of the upper face.

Major outcomes considered are (1) whether potency of the toxin remains unimpaired for some duration after reconstitution, and if so, what is the best estimate of this duration and (2) whether reaccessing of the same vial of reconstituted toxin to treat another patient be performed without increasing risk for contamination.

Ancillary issues that will not be addressed include the use of various nonsaline diluents for reconstitution; the particulars of the manual technique of reconstitution; comparative effectiveness, potency, and area of diffusion associated with different botulinum toxins; concentration of the reconstituted solution (i.e., dilution level); and appropriate dosage.

Intended users of this guideline may be physicians from various medical specialties, including dermatology, otolaryngology, ophthalmology, plastic surgery, and neurology, as well as nurses and ancillary medical personnel.

Methods

Search Strategy

Electronic databases (MEDLINE, EMBASE, and CINAHL) were searched for a 15-year period, 1999 to 2014, for the English language articles. The search term “botulinum toxin,” was used, serially, in conjunction with the following key words: “safe,” “safety,” “sterile,” “sterility,” “effective,” “effectiveness,” “efficacy,” “storage,” “store,” “refrigerate,” and “refrigerated.” A total of 3,167 articles were detected, and consensus review of the available abstracts by 2 data extractors (M.A. and K.M.) reduced this number to 114. Manual review (i.e., hand searching) of the full text of these articles by the same data extractors resulted in 42 selected source

documents, which were reviewed by the members of the guidelines task force.

Data Extraction

A systematic review of the source documents was performed to analyze the evidence. This was performed by a specially constituted task force of 5 content experts proposed by the executive director of the ASDS and ratified by the executive committee of the ASDS. Assessment of the quality and strength of the evidence was performed by the full task force, the ASDS Task Force on Guidelines for the Use and Storage of Neurotoxins, by a committee-based expert consensus process. All source materials were made available to each task force member, and the evidence was discussed during a series of face-to-face meetings and conference calls.

Clinical Questions and Review Process

To simplify the development of this guideline, 4 questions were presented to the task force that comprised the expert panel. Experts were asked to restrict themselves to responses that would be appropriate for all botulinum toxin Type A products available in the United States:

1. How long can reconstituted botulinum toxin Type A be stored without risk for contamination?
2. How long can reconstituted botulinum toxin Type A be stored without loss of clinical effectiveness?
3. Can 1 vial of reconstituted botulinum toxin Type A be safely used for the treatment of more than 1 patient?
4. Can 1 vial of reconstituted botulinum toxin Type A be effectively used for the treatment of more than 1 patient?

Recommendations were formulated by expert consensus based on the Delphi method, and 2 rounds of Delphi process were performed. To address the primary clinical questions in a systematic manner, the following functional questions were posed to the members of the task force: (1) is reconstituted botulinum toxin effective (i.e., experiences no clinically significant loss of potency) for more than 24 hours since reconstitution?, (2) if yes to Question 1, how long

in days is the duration since reconstitution for which there is 95% confidence that there is no clinically significant loss of potency, (3) can a single vial of reconstituted botulinum toxin be used without elevated risk for contamination for the treatment of more than 1 patient, (4) if yes to Question 3, how many patients can be treated from a single vial without elevated risk for contamination?, (5) If yes to Questions 1 and 3, is there any interaction between the duration that a reconstituted vial is stored and the number of patients who can be treated from the same vial (e.g., hypothetically, would increased storage time decrease the number of patients who could be treated without risk for contamination).

Consensus Process

Final recommendations required unanimous agreement of all guidelines task force members. Additionally, individual task force members were asked to rate each of the final recommendations on a scale of 1 to 5 (1 = least confident and 5 = most confident) regarding their personal assessment of the quality and relevance of the supporting evidence.

Validation Process

Validation of this guideline before submission for publication was performed by internal and external peer review. Internal peer review included review by members of the ASDS guideline committee not involved with production of these guidelines. External peer review was by content experts, both dermatologists and non-dermatologists, identified by the ASDS Board of Directors.

Literature Summary

An abridged summary of the relevant medical literature as assessed by the task force is as follows:

Human Studies of Delayed Use of Refrigerated Toxin in Aesthetic Facial Applications

In a randomized controlled study, 105 subjects were treated at the glabella from botulinum toxin vials reconstituted 8 hours, 8 days, or 15 days before injection; no difference was found by 5 blinded raters

evaluating motility at 5 time points culminating at 112 days after injection, and no microbial pathogens were cultured from any of the vials at the end of the study.^{1,2} A study with comparable design compared the rater-measured clinical effectiveness of treatment every 2 weeks after injection until 4 months for 88 subjects receiving glabella toxin from vials reconstituted the day before treatment or 1, 2, 3, 4, 5, or 6 weeks before injection; again, no differences in clinical effect across groups were observed.³

Human Studies of Delayed Use of Refrigerated and Refrozen Toxin in Aesthetic Facial Applications

As early as 1993, Greene⁴ noted that partially used reconstituted vials of botulinum toxin can be frozen at -20°C , rethawed, and reused for “days to several weeks” without loss of clinical effectiveness for the treatment of blepharospasm. Similarly, Greene did not detect any instances of contamination of toxin used in this manner or any infections attributable to such. A prospective cohort study of 118 facial injection sites in 80 subjects compared toxin reconstituted within 4 hours and toxin refrozen for up to 180 days after reconstitution and found no difference in rated clinical effectiveness or frequency of adverse events (AEs).⁵ A split-face double-blind randomized controlled trial (RCT) of 30 subjects injected with fresh toxin at 1 lateral canthus and reconstituted toxin stored for 1 week on the other side did not detect any rater-assessed differences in clinical appearance or in neuroconduction parameters at any of the 6 time points through 18 weeks after injection.⁶ In another split-face randomized controlled study of lateral canthal injections, 45 patients received fresh toxin to 1 side and toxin reconstituted 2 hours earlier to the contralateral side; neither blinded raters nor patient self-report revealed any difference in clinical effectiveness during the 3-month follow-up.⁷ A double-blind, randomized control, split-forehead trial of 40 subjects entailed injection of toxin for the reduction of frontalis-mediated horizontal lines, with substances compared including freshly reconstituted toxin, toxin stored at 4°C (i.e., refrigerated) for 2 weeks, and toxin stored at -20°C (i.e., frozen) for 2 weeks, and then thawed before use; no difference in the degree or

duration of effect was detected at time points up to 120 days after injection.⁸

Based on at least 5 RCTs,^{9–13} botulinum toxin reconstituted with preservative-containing saline has been shown to be effective in reducing injection pain, with approximately a 50% reduction detected on numerical pain scales. Preserved saline includes benzyl alcohol, which has anesthetic properties. There is a single report of contact dermatitis in fragrance-sensitive patients associated with reconstitution of botulinum toxin with preserved saline, but no indication of increased risk associated with storage or reuse of toxin.¹⁴

Human Studies of Delayed Use of Refrigerated and Refrozen Toxin in Non-facial Applications

A similar study of the same 3 formulations injected at the extensor digitorum brevis (EDP) that measured compound muscle action potential amplitude and area, before and after injection, detected no quantitative difference in activity across formulations at any of the 6 time points after injection.¹⁵ A 94-subject rater-blinded randomized control study of EDP action potentials compared toxin used within 2 hours, 72 hours, 1, 2, 3, or 4 weeks after reconstitution; at 1 week after injection, the point of maximum effect, there was no difference in quantitative paralysis among patients injected with each of the 5 toxin types, and the duration of effective paralysis did not differ during 16 weeks of follow-up.¹⁶ In a crossover study of 43 patients with adductor spasmodic dysphonia injected with either freshly reconstituted or refrozen and thawed toxin, there was no significant difference in postinjection duration of action, self-rated satisfaction, or AEs.¹⁷

Animal Studies of Delayed Use of Refrigerated and Refrozen Toxin in Aesthetic Facial Applications

Another study placed refrigerated reconstituted toxin in a mechanized refrigerator that agitated the vial by inverting and straightening it continuously 30 times a minute; among groups of mice injected intraperitoneally with the toxin at 8 time points ranging from 1 to 42 days after agitation began, exactly half of each

group (4/8 mice) died within 48 hours of injection.¹⁸ Injection of toxin into the right anterior auricular muscle of white rabbits performed with freshly reconstituted toxin, and toxin refrozen for 2, 6, and 12 weeks after reconstitution showed similar potency based on nerve conduction studies and visual inspection but more rapid return to baseline by visual inspection in the 12-week group and by nerve conduction in the 6- and 12-week groups.¹⁹ In another rabbit auricular muscle study, nerve ultrastructure assessed by electron microscopy was not different at 12 weeks after injection in ears that had received freshly reconstituted toxin versus reconstituted toxin refrigerated for 2 weeks.²⁰

Human Studies of Contamination Risk During Delayed and Multiple Use of Refrigerated Toxin

Several other studies have also assessed potential risk for contamination of reconstituted botulinum toxin. In 1 study, 11 consecutive vials of reconstituted toxin were exposed to room air for 4 hours, then refrigerated for 3 to 5 days, and then cultured for possible microbial contamination, which was not detected.²¹ In another study, 127 consecutive reconstituted vials, used to treat 1 to 3 patients each over the course of a week, were then refrigerated and reaccessed repeatedly by different personnel who withdrew and discarded small aliquots with different syringes, with the average vial undergoing 4.5 access procedures over 7 weeks; at the end of the study, each vial was cultured, and no microbial contaminants were detected.²²

FDA Adverse Event Reports, Industry Reports, and Physician Consensus Reports and Surveys

Among serious AEs associated with botulinum toxin Type A reported to FDA during the first 13.5 years when the drug was FDA approved for any indication (i.e., December 1989 to May 2003),²³ there were 28 deaths, all associated with therapeutic rather than cosmetic uses. At present, there is no evidence that any serious or nonserious AEs were associated with the use of toxin vials more than 4 hours after reconstitution or for more than 1 patient each. Roger Aoki, then a researcher at Allergan, the manufacturer of 1 type of botulinum toxin Type A, has reported that reconstituted toxin is stable with refrigeration for up to

5 weeks.¹³ A 25-member multispecialty multi-profession expert consensus panel²⁴ convened in 2004 found that 73% of members routinely stored toxin for greater than 4 hours after reconstitution, and the remainder stored toxin in this manner occasionally. Of 322 board-certified dermatologists responding to a survey, 68.6% reported routinely storing toxin for more than 1 week after reconstitution and routinely using each vial for more than 1 patient and 67.0% believed toxin could be safely stored and reused for 1 to 4 weeks; none of the respondents, who had been in clinical practice for a median duration of 15 years, reported any local infections associated with cosmetic toxin injection.²⁵

Major Recommendations

Specific recommendations emerged from the 2 rounds of Delphi process described above and were then adopted unanimously (i.e., full consensus) with the highest level of confidence (i.e., mean confidence score of 5) by the members of the task force. Given that the preponderance of the relevant evidence reviewed described facial aesthetic use of botulinum toxin, the recommendations below are for facial muscle indications, but the task force believes that they are likely valid for therapeutic applications as well:

1. A vial of botulinum toxin A, reconstituted in a clinically appropriate manner, can be refrigerated or refrozen for at least 4 weeks before injection without clinically significant risk for contamination or decreased effectiveness.
2. A vial of botulinum toxin A, reconstituted in a clinically appropriate manner, can be used to treat multiple patients, assuming appropriate handling.

The phrase “in a clinically appropriate manner” is used to denote usual and customary practice, as the precise method of reconstitution is not the subject of this guideline. Similarly, “assuming appropriate handling” is intended to mean that reconstituted vials of toxin used for multiple patients should be, in general, managed and handled in the same manner as vials used for a single patient.

Use of preservative-containing saline to reconstitute botulinum toxin is routine, has been definitively shown to be effective in reducing patient discomfort, and is not associated with AEs.

Rating Scheme for the Strength of the Recommendations

Grading of the strength of recommendations and quality of evidence in this clinical guideline was performed in accordance with modification of the GRADE system developed by the American College of Chest Physicians.

The ASDS task force determined that supporting evidence for the recommendations made included multiple RCTs without important limitations and overwhelming evidence from observational studies. Additionally, the task force determined that the benefits of the recommendations clearly outweighed the risks and burdens, which were deemed to be exceedingly low.

As a result, the recommendations were assigned the grade of 1A/strong recommendation, high-quality evidence. The implication of this grade is that the task force believes that these recommendations can apply to most patients in most circumstances without reservation.

Potential Benefits

Potential benefits of this guideline include improved access to medication, reduced cost and wastage, and potentially improved patient quality of life. No formal cost analysis was performed.

Potential Harms, Contraindications, and Qualifying Statements

The potential harms including loss of medication effectiveness and risk for contamination or associated infection were rated as exceedingly low by the task force. In particular, contamination and/or infection associated with storage of reconstituted toxin for more than 24 hours before use has never been reported in RCTs, observational studies, FDA AE reports, survey data, or clinical simulations.

The task force issued the following qualifying statements: (1) although it is likely that reconstituted toxin can be refrigerated or refrozen for longer than 4 weeks before use, there is insufficient evidence for a strong recommendation in this regard, (2) apart from practical considerations regarding the limited volume of material available for use, there is no specific limit on the number of patients who can be treated with a single vial of reconstituted botulinum toxin.

Consensus Statement Approval

This consensus statement was approved by the ASDS Task Force on Guidelines for the Use and Storage of Neuromodulators on March 15, 2014, and was adopted as amended by the ASDS Board of Directors on May 12, 2014, whereupon it was immediately in effect.

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