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Clin Rehabil 2009; 23; 64
DOI: 10.1177/0269215508097861

The online version of this article can be found at:
http://cre.sagepub.com/cgi/content/abstract/23/1/64

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Clinical Rehabilitation 2009; **23:** 64–71

Botulinum toxin (Dysport) treatment of the spastic gastrocnemius muscle in children with cerebral palsy: a randomized trial comparing two injection volumes

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Received 15th July 2007; returned for revisions 18th July 2007; revised manuscript accepted 17th October 2007.

Objective: To compare the effect of equivalent doses in two different volumes of botulinum toxin type A (Dysport) on gastrocnemius spasticity.
Design: Single-blind, randomized, controlled trial.
Setting: Hospital rehabilitation department
Subjects: Twenty-two children with spastic diplegic or quadriplegic cerebral palsy.
Intervention: High (500 U/5 mL) and low (500 U/1 mL)-volume preparations of Dysport were injected into the gastrocnemius muscles, each child randomly receiving one preparation in the right and the other in the left leg.
Main measures: Dynamic ankle joint range of motion (ROM), passive ROM of the ankle joint, modified Ashworth Scale scores, and the areas of the compound muscle action potential assessed before treatment and at four and eight weeks post treatment.
Results: Both legs improved significantly. The mean (SD) improvements between baseline and the end of follow-up were 19.7 (10.83) degrees for dynamic ROM, 8.4 (9.19) degrees for passive ROM, −1.3 (0.6) for modified Ashworth Scale scores, and -9.4 (11.41) mV-ms for compound muscle action potential in the high-volume group; and 13.5 (10.45) degrees for dynamic ROM, 7.4 (7.88) for passive ROM, −0.9 (0.5) for modified Ashworth Scale scores, and −5.9 (7.50) mV-ms for areas of compound muscle action potential in the low-volume group.
The high-volume preparation yielded significantly greater improvement in dynamic ROM ($P < 0.001$), muscle tone ($P < 0.001$), and lower compound muscle action potential area ($P = 0.006$).
Conclusions: A high-volume preparation of Dysport is more effective than a low volume in reducing spasticity in the gastrocnemius muscle.

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10.1177/0269215508097861

Introduction

Botulinum toxin type A effectively reduces muscle spasticity in cerebral palsy by blocking the presynaptic release of acetylcholine into the neuromuscular junction.[1–5] Two formulations (Botox) and (Dysport), which differ in their composition and pharmacological properties, are currently approved for human use.[6]

Although higher doses of botulinum toxin A yield greater improvement in muscle tone, there is a risk of systemic side-effects and antibody formation.[7] Those risks, plus the high cost of the drug, have stimulated interested in finding ways to enhance the effect of botulinum toxin A per unit dose. One approach is to add a larger diluent volume to deliver the same dose, which may facilitate spread of the toxin to more motor endplates, resulting in greater reduction of muscle tone.

Two animal studies have demonstrated that a higher volume preparation of botulinum toxin A increased muscle paralysis.[8,9] However, human trials have had conflicting results. Some studies have found no difference,[10,11] while another reported superior results with high-volume preparations of botulinum toxin A.[12] Lack of randomization[11] or small sample size[10,11] may have limited these studies, however. In addition, they have all used Botox. Varying volumes of Dysport have not been tested.

The purpose of our study was to compare the effect of equivalent doses of Dysport in high- or low-volume preparations to treat gastrocnemius spasticity in children with cerebral palsy.

Methods

Study participants and study design

Twenty-three children with cerebral palsy were recruited over an 11-month period. Inclusion criteria were a diagnosis of either diplegic or quadriplegic spastic motor cerebral palsy involving the gastrocnemius muscle (modified Ashworth Scale grades 2 to 3), age between 2 and 8 years, no previous botulinum toxin A treatment, and no injections of any kind for at least four months before study entry. Exclusion criteria included fixed ankle joint contracture (Ashworth Scale grade 4); obvious atrophy of the calf muscles; previous foot, leg, or ankle surgery; or use of oral antispastic medication.

The hospital ethics committee approved the study protocol, and written informed consent was obtained from the parents or legal guardians of all of the children.

Intervention

After completing the baseline evaluation, one leg of each eligible child was randomly allocated to receive a high-volume botulinum toxin A preparation, while the other leg was allocated to receive a low-volume injection. An investigator not involved in subject recruitment developed a computer-generated random allocation schedule and placed the assignments in sealed, opaque, sequentially numbered envelopes. As each child was entered in the trial, the next envelope in the sequence was opened, indicating which leg was to receive which preparation.

The concentration of the high-volume preparation was 500 units of Dysport in 5 mL of normal saline, and the low-volume 500 units of Dysport in 1 mL of normal saline. The dose (10–15 units/kg) was determined according to the recommendations of previous studies,[2,13] the child's weight, and the severity of spasticity, with the dose for each child being the same in both legs. Injections were performed under sterile conditions using a 27-gauge 1.5-inch needle without local or general anaesthesia. The gastrocnemius muscle was palpated while being passively stretched and the needle was inserted between the two upper quadrants as described previously,[14] a site originally used for phenol and alcohol blocks and corresponding to the motor points of the muscle.[15] To minimize variability, all dilutions and injections were performed by the same investigator who had no part in assessment and had no contact with either therapists or assessors. Children and caregivers were not blinded, but they were requested not to discuss their allocation with the blinded assessor and therapists.

Each child was asked whether the injection site was painful immediately, 1 hour, and one day after the injections. After botulinum toxin A treatment, the children continued participating in routine physical therapy throughout the study period.

Measurements

Clinical assessments were performed before the injections and at four and eight weeks after treatment by a different investigator who was blinded to the volume of injection in each leg. The primary outcome measure was dynamic range of motion (ROM) at the ankle joint with the knee extended, as assessed by the modified Tardieu Scale. For this measurement, children lay supine with the knee in full extension and the subtalar joint stabilized. The ankle joint was flexed as fast as possible and the first catch angle measured by manual goniometry. The degrees of dorsiflexion from the joint's neutral position were recorded as a positive number and the degrees of plantar-flexion as a negative number.

Assessment of passive ROM of the ankle joint was made with the patient supine and relaxed. With the knee joint full extended, the ankle joint was dorsiflexed slowly until the maximum ROM was obtained.

Muscle tone was graded on an ordinal scale from 0 to 4 according to the modified Ashworth Scale.[16] This is a six-point ordinal scale of muscle tone measuring resistance during passive muscle stretching. Scores range from 0 (no increase in muscle tone) to 4 (rigid) and include a rating of 1+.

An electrophysiology study was performed with Keypoint electromyography apparatus (Dantec Electronics, Skovlunde, Denmark). The compound muscle action potential of the gastrocenimus muscle was recorded by conventional surface techniques. An active surface electrode was taped over the greatest circumference of the muscle and an indifferent electrode over the Achilles tendon. The posterior tibial nerve was stimulated by applying supramaximal electric shocks in the popliteal fossa to generate a maximal motor response in the gastrocnemius muscle. The action potential areas were calculated automatically.

Adverse events associated with botulinum toxin A injection were also assessed at each follow-up session.

Statistical analysis

Data of baseline characteristics were recorded as means (standard deviation) for continuous variables and as numbers with percentages for categorical data. The baseline and post-treatment variables of the two groups were compared using a paired *t*-test and Wilcoxon signed-rank test.

To have an 80% chance of detecting 10 degrees difference between the two preparation treatment volumes in the dynamic ankle joint ROM, with an assumed post-treatment ROM standard deviation of 15 degrees and a drop-out rate of 20%, 23 legs were needed in each group. These calculations were based on findings from a previous study comparing the efficacy of varying botulinum toxin A doses.[17]

Analysis of efficacy included all subjects who completed the follow-up visits. The change in outcome variables after injection was calculated. Each variable (dynamic and passive ankle joint ROM, the action potential area, and the Ashworth Scale scores) was compared between the two dilution volumes across all the post-injection assessments using mixed models (repeated-measures analysis of variance) analysis. The mixed model included group and time as fixed effects and subjects as random effect. The interaction between group and time was also evaluated. Planned contrast tests were used to compare the difference in outcome variables in the legs with both preparations at each follow-up assessment.

The paired *t*-test and Wilcoxon signed-rank test were used to examine within-group differences between baseline values and at four weeks and eight weeks post injection, with Bonferroni adjustments made for multiple comparisons. McNemar's test was used to compare pain perception between the high-volume and low-volume preparations.

All statistical analysis was conducted with the SAS software package. A P-value of <0.05 was considered statistically significant.

Results

Of the 23 children enrolled, 22 completed follow-up at four and eight weeks (Figure 1). Of these 22, there were 18 boys and 4 girls, 20 with spastic diplegia and 2 with spastic quadriplegia, and 3 who walked with aid and 19 who walked independently. Their mean age was 55 months $\pm$ 19.4 and mean weight 18 kg $\pm$ 4.1. The mean dose of

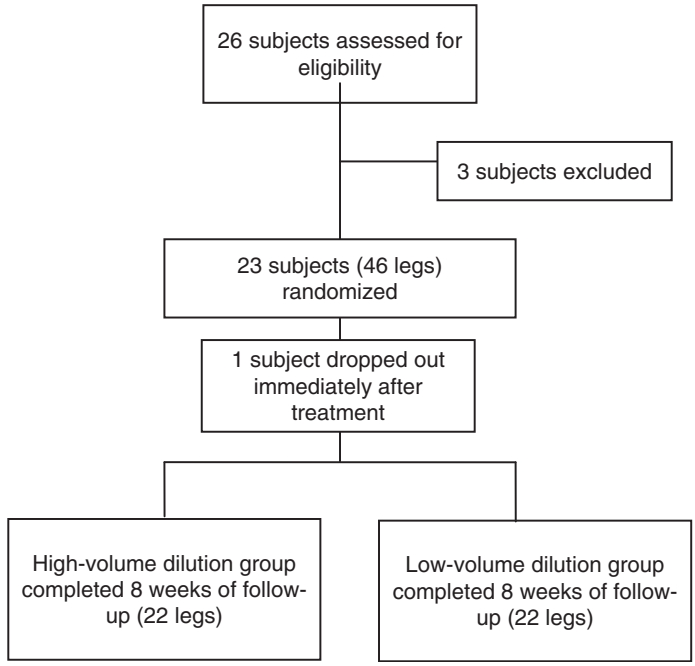

Figure 1 Flowchart of study participants.

botulinum toxin A per gastrocnemius muscle was 183 units ± 59.7.

At baseline, there were no significant differences in dynamic and passive ankle joint ROM, the action potential area, and the Ashworth Scale scores between the legs that received high- or low-volume preparations (Table 1). The dynamic ROM increased and the Ashworth Scale scores decreased from baseline to eight weeks for both groups; the passive ROM increased at four weeks and then declined slightly at eight weeks. The mean action potential areas in both groups were lowest at four weeks and had partially recovered by eight weeks after injection (Table 2).

The high-volume preparation resulted in significantly greater improvement in dynamic ROM, reduction in muscle tone and denervation of the injected muscle as demonstrated by the mixed-model analysis (significant group effect for the dynamic ROM, $P < 0.001$; the action potential areas, $P = 0.006$; and the Ashworth Scale scores, $P < 0.001$). Planned contrast analysis showed a significantly greater dynamic ROM (95% confidence interval (CI) for difference: 5.6 to 13.5), lower action potential area (95% CI for difference: −9.5 to −2.4), and lower Ashworth Scale scores (95% CI for difference: −0.7 to −0.3) in the high-volume group at four weeks. The differences at eight weeks

Table 1 Baseline measurements

Variables	High-volume[a]	Low-volume[b]	95% CI for differences between groups
Dynamic ROM (degrees)	−16.9 (8.65)	−15.8 (10.48)	−5.1–2.8
CMAP area (mV-ms)	36.7 (14.98)	36.4 (13.27)	−3.3–3.8
Passive ROM (degrees)	4.3 (10.53)	3.3 (7.38)	−4.1–2.0
MAS	2.8 (0.39)	2.8 (0.39)	−0.2–0.2

The data are given as mean (SD).
[a]Legs in high-volume dilution preparations.
[b]Low-volume: Legs in low-volume dilution preparations.
Dynamic ROM, dynamic range of motion at ankle joint; Passive ROM, passive range of motion at ankle joint; CMAP, compound muscle action potential; MAS, modified Ashworth Scale.

Table 2 Outcome measurements at baseline and after treatment

Outcome variables	Baseline		Week 4		Week 8		Within-group differences (week 8–baseline)	
	High-volume[a]	Low-volume[b]	High-volume[a]	Low-volume[b]	High-volume[a]	Low-volume[b]	High-volume[a]	Low-volume[b]
Dynamic ROM (degrees)	−16.9 (8.65)	−15.8 (10.48)	1.1 (9.28)	−8.4 (9.82)	2.8 (6.84)	−2.3 (8.05)	19.7 (10.83)	13.5 (10.45)
CMAP area (mV-ms)	36.7 (14.48)	36.4 (13.27)	23.4 (9.62)	29.3 (10.98)	27.3 (11.07)	30.5 (12.17)	−9.4 (11.41)	−5.9 (7.50)
Passive ROM (degrees)	4.3 (10.53)	3.3 (7.48)	13.5 (8.20)	12.0 (7.83)	12.7 (6.16)	10.7 (7.05)	8.4 (9.19)	7.4 (7.88)
MAS	2.8 (0.39)	2.8 (0.39)	1.5 (0.36)	1.9 (0.42)	1.5 (0.36)	1.9 (0.34)	−1.3(0.6)	−0.9 (0.5)

The data are given as mean (SD).
[a]Legs receiving a high-volume preparation.
[b]Legs receiving a low-volume preparation.
Dynamic ROM, dynamic range of motion at ankle joint; Passive ROM, passive range of motion at ankle joint; CMAP, compound muscle action potential; MAS, modified Ashworth Scale.

remained significant for dynamic ROM (95% CI for difference: 1.1 to 8.2), and Ashworth Scale scores (95% CI for difference: −0.6 to −0.2), but the action potential areas were no longer statistically different (95% CI for difference: −6.8 to 0.3).

Statistical analysis showed significant group by time interactions for dynamic ROM ($P = 0.001$) and the Ashworth Scale score of ($P = 0.005$). Contrast testing indicated the high-volume preparation yielded greater improvement in dynamic ROM and reduction in the Ashworth Scale scores at four weeks post injection.

Mixed-model analysis showed significant differences over time for all outcome variables (all P-values <0.001). Compared with baseline, both preparations resulted in significantly greater dynamic and passive ROM and reduction in the Ashworth Scale scores and action potential areas at four and eight weeks post injection ($P < 0.001$ for all comparisons).

These results indicate that both preparations of botulinum toxin A yielded significant reduction of spasticity that persisted through to the eight-week endpoint of the study. However, the high-volume preparation yielded greater improvement.

The children did not report a significantly different incidence of pain in the high- and low-volume legs immediately, 1 hour, or 1 day after injection ($P = 0.5$, 0.25, 0.5 respectively) (Table 3). No evidence of systemic botulism or adverse effects was reported.

Discussion

This prospective randomized study demonstrated that a high-volume injection of Dysport yielded a significantly greater reduction in spasticity than did a low-volume injection of an equivalent dose into the gastrocnemius muscles of children with cerebral palsy. Both preparations were beneficial in reducing spasticity, but the high-volume injection produced a greater benefit. There was no significantly greater frequency of injection site pain with the high-volume injection.

Theoretically, a greater injection volume would allow diffusion of the drug over a greater area,[18] resulting in blockade of a greater number of motor endplates and thus a greater reduction in

Table 3 Number of patients reporting pain at the injection site in legs

Time after injection	High-volume[a]	Low-volume[b]	*P*-value
Immediately	18 (81%)	16 (72%)	0.5
1 hour later	6 (26%)	3 (14%)	0.25
1 day later	3 (14%)	1 (5%)	0.5

[a]Legs receiving a high-volume preparation.
[b]Legs receiving a low-volume preparation.

spasticity.[19] Shaari and Sanders studied various doses of botulinum toxin A in rats, finding that a higher dose affected a larger area, a result they attributed to the larger volume injected.[9] Kim and co-workers gave rabbits 10 units of botulinum toxin A diluted in either 0.1 or 0.5 mL of normal saline and found that the larger volume led to greater muscle paralysis.[8] Francisco and co-workers compared high-volume (100 units/2 mL) and low-volume (100 units/1 mL) dilutions of botulinum toxin A in adults with spasticity of the wrist and finger flexors after brain injury and found no difference in therapeutic effect.[10] Similarly, Lee and colleagues found no significant difference in the action potential areas of the gastrocnemius in patients with cerebral palsy injected with either 100 units/4 mL or 100 units/1 mL of botulinum toxin A.[11] On the other hand, Gracies and co-workers found 100 units/5 mL of botulinum toxin A to be more effective than 100 units/1 mL in patients with spastic biceps muscles after strokes.[12] These studies are somewhat difficult to compare because of the variation in injected volumes and the different size muscles tested. We suspect that smaller muscles and smaller differences in the volumes may minimize any disparity in therapeutic effect. The higher volume base diffusion pharmacologic properties of Dysport may also partially account for the conflicting results between our study and those of others.[20]

There are two potential problems with a large-volume injection of botulinum toxin A. The agent could diffuse beyond the limits of the injected muscle, with unintended effects on the adjacent musculature or other adverse effects.[21] However, one would expect the low concentration in a higher volume to lessen the concentration gradient between the injected muscle and the neighbouring tissue. Also, the rapid, high-affinity binding of botulinum toxin A to the neuromuscular junction decreases the risk that the toxin will spread outside the injected muscle. We did not observe any adverse events in our study, either in adjacent muscles or systemically. Another potential disadvantage is greater discomfort with a larger volume needing to be injected.[22] Theoretically, the larger volume might increase the tissue pressure locally and activate more nociceptors at the injection site, but we did not find significant differences in the children's pain perception between the two legs, which is in agreement with the results of previous studies.[11, 23]

Because spasticity is difficult to quantify, several measurements are used to assess the treatment effect of botulinum toxin A in cerebral palsy. These include modified Ashworth Scale,[24] ROM,[25] dynamic muscle length,[26] muscle strength[27] and the Physician Rating Scale.[28] Of these, the dynamic gastrocnemius muscle length seems to be the most sensitive measure of the response to botulinum toxin A injection.[29] This is generally assessed clinically by using the dynamic ankle joint ROM. The Tardieu Scale for dynamic ankle ROM may be more useful for quantifying the neural and peripheral components of spasticity and for measuring the dynamic component of muscle spasticity.[29] It also has good intra- and inter-observer reliability and validity to detect changes in spasticity in cerebral palsy.[30] The method is quick and easy to perform in the outpatient setting. By using this scale, we demonstrated that the high-volume botulinum toxin A preparation yielded greater improvement in dynamic ankle dorsiflexion. Botulinum toxin A has a major effect on the dynamic spastic component but only limited effect on passive ROM, since the latter depends of resistance produced by muscle or joint connective tissue.[31] This is consistent with our results demonstrating that, compared with the low-volume injection, the high-volume botulinum toxin A preparation yielded greater improvement in dynamic but not in passive ankle dorsiflexion. Passive ROM improved about equally in both legs and more likely resulted from the physical therapy the children continued to receive throughout the study.

The area of an action potential is proportional to the number of motor units not blocked by botulinum toxin A. This is a simple and quite objective method for quantifying the degree of muscle paralysis following botulinum toxin A injection.[32] Our findings correlate well with the observations of Hamjian and co-workers who found the peak decline in the action potential areas around day 21 post injection.[33] In our study at eight weeks, although the action potential areas were beginning to increase, dynamic ROM had still improved slightly over the four-week measurement. Thus, even though the paralytic effect of botulinum toxin A may be declining, the residual muscle paralysis may continue to allow more effective physical therapy.[34]

Our study was limited in that only the assessor was blinded; both parent and child knew which volume was injected into which leg. We also did not formally test the reliability of the dynamic and passive ROM and spasticity assessments. However, all measurements were performed using a standardized procedure by the same experienced investigator. In order to control for other confounding factors, we had strict inclusion criteria, so our results are not necessarily broadly applicable to all children with spastic cerebral palsy. We were also unable to compare changes in overall functioning, as the subjects served as their own control, each one receiving both preparations of the drug. Assessment of overall function would require two groups of subjects, each group receiving one or the other of the two preparations.

To the best of our knowledge, this is the first randomized controlled study using Dysport to compare the effect of equivalent doses of botulinum toxin A in two different volumes on gastrocnemius spasticity in children with cerebral palsy. Although, cost-effectiveness and functional improvement are subjects requiring further investigation, we believe our study lays the groundwork for such investigations by demonstrating that a high-injection volume measurably reduces spasticity in the gastrocnemius to a greater extent than a low-volume injection. This approach is worth studying in larger, diverse groups of patients, as it has the potential to reduce costs and perhaps lessen the incidence of antibodies against botulinum toxin A.

Clinical messages

- High- and low-volume botulinum toxin injections both improve gastrocnemius muscle spasticity in children with cerebral palsy.
- High-volume botulinum toxin injections are more effective than low-volume in reducing gastrocnemius spasticity in cerebral palsy.
- High-volume botulinum toxin injections are well-tolerated and safe.

Acknowledgements

We thank Dr Mary Jeanne Buttrey for comments on the manuscript.

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