

Title Page

Exploratory randomized double-blind placebo controlled trial of botulinum therapy on grasp release after Stroke (PrOMBIS)

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Includes:

Table e-1: Injection report detail

Table e-2: adverse events

Appendix e-1: supplementary methods

Search Terms

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Botulinum Toxins, Type A

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Abstract

Background: OnabotulinumtoxinA injections improve upper limb spasticity after stroke but their effect on arm function remains uncertain.

Objective: To determine whether a single treatment with OnabotulinumtoxinA injections combined with upper-limb physiotherapy improves grasp release compared to physiotherapy alone after stroke. Methods: 28 patients, at least one month post-stroke, were randomised to receive either OnabotulinumtoxinA or placebo injections to the affected upper limb followed by standardized upper-limb physiotherapy (10 sessions over four weeks). The primary outcome was time to release grasp during a functionally relevant standardized task. Secondary outcomes included measures of wrist and finger spasticity and strength using a customised servomotor, clinical assessments of stiffness (modified Ashworth Scale - MAS), arm function (Action Research Arm Test-ARAT, Nine Hole Peg Test), arm use (Arm Measure of Activity - ArMA), goal attainment scale (GAS) and quality of life (EQ5D).

Results: There was no significant difference between treatment groups in grasp release time 5 weeks post injection (placebo median = 3.0s, treatment median = 2.0s; $t(24) = 1.20$, $p=0.24$; treatment effect -0.44, 95% CI -1.19 to 0.31). None of the secondary measures passed significance after correcting for multiple comparisons. Both groups achieved their treatment goals (placebo=65%, treatment=71%), and made improvements on the ARAT (placebo +3, treatment +5) and in active wrist extension (placebo +9 degrees, treatment +11 degrees).

Conclusions: In this group of stroke patients with mild to moderate spastic hemiparesis, a single treatment with OnabotulinumtoxinA did not augment the

improvements seen in grasp release time after a standardized upper-limb physiotherapy programme.

Clinical trial registration: EudraCT 2009-009357-22

Introduction

Upper-limb disability is problematic for many stroke survivors. Clinical and experimental observation suggest that disordered grasp release is a significant contributory factor. Although difficulties with grasp release have been shown to be primarily due to extensor weakness and impaired motor control,^{1,2} there is some evidence that persistent flexor activation may also contribute.² In the clinical setting, rehabilitation specialists increasingly postulate that temporarily reducing finger flexor over activation could enhance interventions to improve hand function after stroke.

Sustained or intermittent involuntary muscle activation after stroke is a manifestation of spasticity, as recently defined by Pandyan et al.³ Spasticity is common after stroke^{4,5} and its treatment is increasingly integrated in stroke rehabilitation and care. Systematic review provides strong evidence that Botulinum neurotoxin type A (OnabotulinumtoxinA) reduces spasticity and improves passive function; for example, easing opening of the hand for cleaning and skin care.⁶ Observational studies without controls or blinding have shown some promising results in improving active function of the upper limb.⁷⁻⁹ However, effects have been variable and significant changes are yet to be shown in placebo randomised controlled trials.¹⁰⁻¹² According to previous reviews, the lack of effect could reflect study limitations and recommendations were made that future research should measure change in specific active functional tasks or outcomes, and that an objective quantification of spasticity should be used.¹²⁻¹⁵

PROMBIS (Predicting Outcomes and Measuring benefit of Botulinum therapy in Stroke) is an exploratory randomized, double-blind, placebo-controlled parallel-group trial to investigate the effect of OnabotulinumtoxinA on one such specific outcome: grasp-release time. We hypothesized that the active treatment group would

demonstrate a statistically significant improvement in grasp release time compared to the control group.

Primary objective. To determine whether targeted OnabotulinumtoxinA injections (BOTOX[®], Allergan Limited, Marlow, UK) combined with standardized physiotherapy treatment of the upper limb after stroke will reduce grasp release time, a quantitative measure of active upper limb function.

Methods

Study design. A parallel group double-blind randomised controlled trial (RCT) with subjects randomly allocated in a 1:1 ratio between treatment and placebo groups. Participants performed baseline assessment and were then given injections of either placebo or OnabotulinumtoxinA in week 0, followed by ten sessions of intensive standardized physiotherapy over four weeks.¹⁶ Outcomes were then re-assessed at weeks 5, 9 and 13.

Standard protocol approvals, registrations, and patient consent. The trial was undertaken at The National Hospital for Neurology and Neurosurgery from 2009 to 2014 and the protocol and all amendments approved by the local review board. Written informed consent was obtained from all participants at the screening assessment. The study is registered on the EU Clinical Trial Register (EudraCT: 2009-009357-22) and is reported here in accordance with the CONSORT guidelines for reporting of RCTs.¹⁷

Participants. Patients presenting to focal spasticity clinics at the National Hospital for Neurology and Neurosurgery were screened for eligibility by the multidisciplinary team (MDT) including members of the independent research team. Inclusion criteria were: 1) confirmed diagnosis of stroke more than one month previously; 2)

established focal finger or wrist spasticity that the MDT felt could be interfering with active grasp and release function and had the potential to benefit from treatment with OnabotulinumtoxinA (this included an assessment on whether the potential participant presented with sufficient residual strength and motor control for rehabilitation to be effective); 3) score of 2 or more in the modified Ashworth scale in the joints of interest and 4) ability to transport the assessment cup to at least one of the target positions and release it at baseline. Exclusion criteria were:

OnabotulinumtoxinA injections to any site within the previous three months; contraindications to OnabotulinumtoxinA; fixed contracture in the upper limb; additional neurological impairment not related to stroke; uncontrolled upper limb pain; cognitive impairment preventing informed consent or the ability to follow task instructions.

Participants were not required to be naïve to OnabotulinumtoxinA treatment.

Randomization and blinding. Randomization was performed by an additional statistician prior to trial commencement using a block randomisation process to ensure equal numbers in each treatment arm (the trial statistician was blind to group allocation). Details of the blinding procedures including allocation concealment are provided in appendix e-1 (supplementary materials).

Intervention. *Drug intervention:* Injection sites were identified with standard neurophysiological technique (electromyography and electrical stimulation) using a portable handheld device (Clavis; Medtronic, Minneapolis, USA)¹⁸. The doses and distribution of the injections were guided by the clinical and neurophysiological evaluation (including the magnitude of the audible stretch response and degree of resting muscle over-activity) as per standard clinical practice.

Allergan Botox™, diluted as 100 units in 2 mls of saline, or a saline placebo was injected through a fine bore EMG needle electrode into the muscles identified by the

multidisciplinary assessment as likely to be hindering function. Treatment and placebo solutions looked identical and were reconstituted out of sight of the injecting doctor, treating physiotherapist, and the participant.

Physiotherapy intervention: The standardized physiotherapy intervention has previously been described in detail¹⁶. The original protocol consisted of daily sessions over 10 consecutive working days. For this study, it was modified to occur over 4 weeks to focus training during the peak action of the drug and reflect current clinical practice of outpatient therapy provision. The total session time ranged from 45 minutes up to 1.5 hours to accommodate each patient's need to complete the tasks, rest and stretch without affecting the overall intensity (repetitions) of the therapy.

In summary, the protocol included both strength training (three different muscle groups) and functional task practice (three different tasks). Strength training consisted of three sets of 10 repetitions of wrist extension, finger extension and grip strength at 60–80% of maximal isometric voluntary contraction measured in mid-range and was recalibrated every 3 training days.

Functional training tasks were chosen by the subject relevant to their personal treatment goals. The intervention was tailored to the individual's impairment level so that the intensity of intervention was standardized despite differing impairment levels at enrolment.

Participants were encouraged to stretch whenever needed throughout the strength and functional training.

Primary outcome variable We used a functionally relevant grasp-release task that measured the time taken to release grasp after moving an instrumented cup to a target. Grasp release timing data was calculated from first contact of the cup on the target until all digits were released from the cup. All electronic data were collected in a custom software programme (LabVIEW, National Instruments Corporation, Austin, Texas) then exported and further analysed with MATLAB scripts (The MathWorks Inc, Massachusetts, USA).

The targets were spread over 4 positions (2 in near space at 50% of arm length and 2 in far space at 90% of arm length) and two heights (low, at two cm above the table surface, and high, which was individually adjusted so that the subject required 90 degrees of shoulder flexion to place the ulnar border of their hand on the target surface). Test-retest variability varied from excellent (in the near and low target, ICC 0.8) to moderate (in the low far target, ICC 0.54).¹⁹ The participants could have assistance to place the cup into the hand but were required to be able to transport the cup to at least one target and release it unassisted. The cup weighed 300g and measured 124mm in height, with a base diameter of 58mm and top diameter of 88mm. For more details see Appendix e-1 (supplementary materials).

Secondary outcome variables Reliable measurement of spasticity remains controversial.^{3,20} Spasticity is defined as “disordered sensori-motor control, resulting from an upper motor neurone lesion, presenting as intermittent or sustained involuntary activation of muscles”.³ We employed the modified Ashworth scale (MAS)²¹ as it has been widely used in previous studies and would allow for comparisons. The MAS was converted to a 6-point score from 0 to 5. However, as the Modified Ashworth scale assesses overall passive stiffness²² and has been

shown to be a subjective measure^{21,22} a servomotor (Kollmorgen D062M Direct Drive) was used to measure spasticity objectively. The motor was programmed to measure torque in response to slow and fast passive movements. In healthy control subjects there is no significant difference between these measurements.¹⁹ In people with upper motor neuron lesions such as stroke, involuntary activation of the flexor muscles in response to the fast stretch of the motor (spasticity) can be objectively quantified as an increase in torque. The technique provides quantification of stretch reflex mediated activity which is proportional to and correlates with the size of the EMG response in people with Upper Motor Neuron Syndrome.²³

The servomotor held the subjects' arm firmly in position with the axis of either the metacarpophalangeal (MCP) joints or the wrist joint aligned to the axis of the motor. Wrist stretches started in 10 degrees of flexion moving through 20 degrees to 10 degrees extension. Finger stretches started in 45 degrees flexion, moving back 20 degrees. The fingers were measured together as the apparatus was not capable of measuring individual fingers. Subjects were instructed to relax and let the motor move their arm without resistance. Passive stiffness of the flexor muscles was measured by recording mean torque over 5 slow stretches (20 deg at 6 deg.s⁻¹) while EMG recordings confirmed that the muscle was not activated. There was no reflex activity at this slow speed and therefore the measure represents soft tissue stiffness without the reflex (spasticity) component. Total stiffness, which includes passive and reflex components, was measured by recording mean torque over 5 fast stretches (20 deg at 300 deg.s⁻¹) while EMG recordings identified and quantified the presence and duration of reflex response (spasticity). For more details of the methodology see Appendix e-1 (supplementary materials).

Isometric wrist and finger flexion and extension strengths were measured against the stationary servomotor with joints at zero degrees. Grip strength was measured by dynamometer. Wrist and finger range of movement was measured using a standard goniometer and positioning.

Functional tests comprised the Nine Hole Peg Test²⁴ and the Action Research Arm Test (ARAT)²⁵. The Arm Measure of Activity (ArMA)²⁶ and EQ5D²⁸ quality of life measure were completed. The ArMA is a self-reported functional scale with passive and active subscales that use a Likert scoring system between 0 (no difficulty) and 4 (unable to do task).

Finally, two personalised functional goals, including a grasp release goal, were also agreed with each participant.

Statistical analysis. Primary outcome analysis was performed using Analysis of Covariance (ANCOVA). Week 5 measurements were used as the outcome measure, with baseline score as the covariate, and a complete case analysis performed. All subjects completed baseline and immediate outcome measurement sessions. Data required log transformation to achieve normal distribution prior to analysis. Missing values, where subjects were unable to complete the grasp release task at all target positions, were imputed as per trial protocol using the maximum grasp release time recorded in the dataset at that given time point. The baseline adjusted mean difference between groups was reported along with corresponding 95% confidence interval.

Secondary analyses analysed combined data from all time periods. Generalised estimating equation (GEE) models were fitted to account for clustering of patients over time. Baseline values were included as one of the covariates in the model.

Assumptions were checked, and where they were not met, data transformation was performed prior to analysis. Where data transformation was not possible, a non-parametric test was used.

Results

Participants. Twenty-eight participants were recruited in total and randomized, 14 to each arm of the trial. The flow of participants is shown in figure 1 (CONSORT diagram). Baseline characteristics are presented in table 1. There were no significant differences between the groups at baseline. Most participants had experience of OnabotulinumtoxinA treatment (not within the preceding three months) but none had previously received an intensive upper-limb specific intervention. The treatment group received a total of 115 units (range 40 - 190) and a median of 5 muscles were injected per participant (range 1 to 8). The placebo group received 151 units (range 55 - 290) with a median of 7 muscles injected per participant (range 4 - 12). Table 4 gives a summary of the number of participants injected broken down by muscle. Please see e-Table 1 in the supplementary information for a more detailed breakdown of dose and muscles injected per participant.

Safety. There were two unrelated serious adverse events during the trial. One occurred after consent but prior to administration of the trial drug (ocular migraine, treated initially as a possible ocular TIA) and the other occurred 4 months after injection (kidney vasculitis). Neither subject was withdrawn. The emergency unblinding procedure was not used. Minor adverse reactions were reported as expected (detail in supplementary material Table e-2).

Primary outcome. There was no statistically significant difference in grasp release time in all four positions between treatment groups at follow-up at week 5,

(Placebo median = 3.0s, treatment median = 2.0s; $t(24) = 1.20$, $P=0.24$). Treatment effect was -0.44, and the 95% confidence interval -1.19 to 0.31. Within group median change for placebo was -606ms and treatment -1200ms. This finding remained true even when the low-near position (easiest target to reach) was considered in isolation (Placebo median = 4.0s, treatment median = 2.0s; $t(24) = 0.65$, $p=0.52$; treatment effect -0.30 CI -1.27 to 0.67). Within group median change for placebo was -423ms and treatment -2364ms. See table 2 and figure 2.

Secondary outcomes. There was no significant difference between groups on any of the secondary outcomes after correction for multiple comparisons (see table 3).

The Modified Ashworth group sum of scores decreased a total of 3 and 11 levels respectively for wrist and finger flexors in the placebo group, versus 11 and 19 in the active treatment group. Physiological measures of stiffness (slow motor) were not significantly different between groups in the wrist or fingers (for detailed values see Table 3); there was similarly no group difference on change in spasticity measures (fast motor) for either of the joints tested.

Wrist and finger extension strength improved with no significant difference between groups (placebo +0.04Nm and +0.05Nm; treatment +0.31Nm and +0.37Nm respectively). Extrinsic grip strength was maintained in the treatment group at 5 weeks despite the weakening effect of OnabotulinumtoxinA injections (see table 3).

Active wrist extension range of movement improved in both groups (placebo +9 degrees, treatment +11 degrees), as did active finger extension range of movement (placebo +1.96 degrees, treatment +11.65 degrees). Both groups also achieved treatment goals (placebo 65%, active 71%), and improved on the ARAT (placebo +3, treatment +5), and the ArMA active subsection (-1.0 in the placebo group versus -

7.5 in the active treatment group). There was no change in either group for the 9HPT or the EQ5D.

Discussion

In this randomized placebo-controlled proof-of-concept trial, clinically and physiologically measured hypertonia decreased and function improved regardless of whether participants received active treatment with OnabotulinumtoxinA or placebo. There was no statistically significant difference between the OnabotulinumtoxinA and placebo groups on any measure of impairment, functional outcome or goal attainment. Although all measures were in favor of the OnabotulinumtoxinA group, this was not statistically significant.

Although previous botulinum toxin studies have shown a significant reduction in spasticity measures,^{12,13} thus far translation to improvement in active function has not been clearly shown.^{12,13,27} Despite that, clinicians often propose OnabotulinumtoxinA injections to temporarily weaken overactive flexors and give a 'window of opportunity' for active physiotherapy to strengthen extensor muscles and improve hand function. This treatment approach requires residual extensor strength and some level of voluntary control over the extensors in order to strengthen and train grasp release function. In this trial, we wanted to test this physiologically plausible principle using a scientifically robust but also pragmatic design. We opted for a relevant quantitative variable (grasp release time) as our primary outcome and we included physiological measures of spasticity, as recently recommended for new trials in this area.^{14,15} Moreover, this is the first study to use a standardized physiotherapy program. Our results did not support our hypothesis; in other words, a single treatment with OnabotulinumtoxinA group targeting the wrist and finger flexors

does not seem to offer additional improvement in grasp release after 4 weeks of a targeted physiotherapy intervention when compared to a placebo group.

We acknowledge that the current study with its relatively small sample size does not provide a definitive answer on the usefulness of OnabotulinumtoxinA as an adjunct to improve active function in stroke patients with upper limb spasticity. A recent review has also shown no effect on active function at immediate outcome.²⁷ However, the authors acknowledge several limitations including the fact that adjuvant treatment may help optimize voluntary control, which was not controlled for in the included studies (16 out of 35 studies had no adjuvant treatment, quality and standardization of adjuvant therapy was unclear in the others). They also acknowledge that there was insufficient evidence for effects at a functional level and that the diversity of clinical measures used may have contributed to this. Our study has attempted to address some of these issues with standardized adjuvant therapy and the attempt to target and accurately measure one important functional activity (grasp release) that might be expected to benefit.

A few other points merit discussion. In contrast with previous studies,^{12,13} clinical spasticity measures (MAS) did not differ between the groups. The same was true for the servomotor measures that are more likely to measure spasticity rather than overall stiffness. This lack of difference may, in part, be related to the adjuvant standardized physiotherapy regime employed in this study, as more than half of participants in Gracies et al¹² were not receiving any therapy; in others, patients received unspecified and non-standardized interventions, introducing a potential confounder into that data.^{13,28} It is also possible that the population investigated here, who were required to be able to complete a grasp release task for inclusion, represent a less severely impaired sample than the Gracies et al study.¹² Patients

with more severe spasticity, i.e. unable to extend their fingers enough at baseline, may have shown better spasticity outcomes after the injections, but whether that would have translated to functional gains is debatable. As shown for other interventions,²⁹ response is more likely when patients have baseline voluntary activity, particularly active finger extension. Indeed both groups did improve in strength and arm function measures, arguably due to the physiotherapy intervention. Whether this represents a ceiling effect or simply highlights that strength and motor control are stronger modifiers of outcome than spasticity in this group of patients cannot be answered from our current study. A different study design including patients with variable impairments as well as a considerably longer intervention could address that in the future.

Another potential problem is the fact that patients were invited to enter if they were at least 1 month post stroke with no upper limit; this was again a pragmatic representation of the population referred to spasticity services. The vast majority of the patients were at least 6 months post stroke at enrollment (only one patient in the placebo group was enrolled at 3 months), which is the accepted cut-off point for chronic stroke and thus heterogeneity associated with plasticity related recovery should be minimal.

In this study we have implemented a single treatment with OnabotulinumtoxinA and 4 weeks of standardized therapy and all our patients improved. It is possible that 2 or more cycles of OnabotulinumtoxinA treatment could be more effective as some recent observational studies have suggested^{30,31} by consolidating the effects on spasticity and/or allowing more opportunity for retraining. The history of previous OnabotulinumtoxinA injections was not addressed in this study but may also have had potential to influence the outcome.

The intensity of physiotherapy is another important factor. We have opted for a pragmatic physiotherapy intervention but longer and more intensive therapy may have yielded better results. Looking from the opposite angle, one wonders if OnabotulinumtoxinA treatment without the same level of physiotherapy intervention (as often happens in clinical practice) may have shown stronger effects of the toxin per se and be more relevant to the current reality in stroke rehabilitation. Finally, we have investigated a specific functional outcome aiming to address a common clinical question for chronic stroke patients with moderate to mild spasticity and our results were in line with those using more traditional functional outcome measures (e.g. ARAT); it is conceivable that another parameter may prove more responsive or sensitive to change. The slightly higher test-retest variability associated with far targets during the grasp release task suggests more cautious interpretation of this specific subset of results.

We selected predominantly distal injection sites. In some cases proximal spasticity might have contributed to difficulty in the grasp release task. If participants were struggling with the reach part of the task, as many did, the additional effort could have made grasp release even more challenging. Targeting additional, including more proximal, targets thought clinically to be interfering with function may prove a better model for future trials.^{12,31,32} Likewise the measures of impairment gathered here (stiffness, spasticity, and muscle strength) have all been recorded in non-functional laboratory conditions, different to those encountered during activities of daily living. Functional use of the arm and hand often involves reaching out and requires proximal muscle activity to take the weight of the arm or stabilise it in space. This effort could alter the impact of the impairment.^{33,34}

Muscle stiffness and spasticity in the fingers were measured in mid-range to prevent contractures or tightness at end of range from interfering with measurement. Other studies though have tested throughout the available range and this could represent a more accurate measurement of stiffness within the functional range of movement^{35,36}; this becomes even more important as the interval from the stroke increases.³⁷ Accurate measurement of spasticity remains a challenge^{3,20} and requires careful consideration when used for patient selection or interpretation of functional change in research studies.³

Conclusion

In this group of stroke patients, addition of a single treatment with OnabotulinumtoxinA injection(s) aiming to reduce spasticity in the forearm flexors, was not significantly better than physiotherapy alone in improving grasp release time or other active functional outcomes.

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Conflict of interest statement

R. Farrell has received honoraria or consultancy fees from GW Pharma, Canbex Pharmaceuticals Ltd, Biogen Idec, Merck, Allergan PLC.

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Table 1: baseline patient demographics

Variable		Group		
		All subjects	Treatment	Placebo
Sex	Male	19	9	10
	Female	9	5	4
Age (years)	Mean	49	50	48
	St Dev	16	18	14
	Range	18-82	21-82	18-68
Time since stroke (months)	Mean	66	83	50
	St Dev	92	118	46
	Range	3-456	7-456	3-145
Stroke type	Haemorrhage	10 (35.7%)	4 (28.6%)	6 (42.9%)
	Infarction	16 (57.1%)	10 (71.4%)	6 (42.9%)
	Both	1 (3.6%)	0 (0%)	1 (7.1%)
	Unknown	1 (3.6%)	0 (0%)	1 (7.1%)
Stroke location	Cortical	10 (35.7%)	5 (35.7%)	5 (35.7%)
	Subcortical	8 (28.6%)	6 (42.9%)	2 (14.3%)
	Both	2 (7.1%)	1 (7.1%)	1 (7.1%)
	Unknown	8 (28.6%)	2 (14.3%)	6 (42.9%)
Barthel score	Mean	92	90	94.6
	St Dev	9	11.6	4.6
	Range	65-100	65-100	85-100

Table 2: treatment effects on primary outcomes

Primary outcomes	Baseline		5 weeks		Mean change from baseline		Significance
	Treatment	Placebo	Treatment	Placebo	Treatment	Placebo	p value
Mean grasp release all 4 positions (ms)	7089 (10326.81)	9100 (12394.31)	7285 (15361.01)	11478 (16108.19)	+196	+2378	0.24
Mean Grasp release (near and low) (ms)	8735 (11295.79)	9061 (11302.19)	7613 (15284.44)	10200 (15367.63)	-1122	+1139	0.52

Values are mean (SD) unless otherwise indicated, significance levels were $p < 0.05$ for primary outcomes

Table 3: treatment effects on secondary outcomes

Secondary outcomes	Baseline		5 weeks		Mean change from baseline		Significance
	Treatment	Placebo	Treatment	Placebo	Treatment	Placebo	p value
Wrist stiffness (slow : fast) (Nm.deg ⁻¹)	0.03 : 0.052 (0.016 : 0.02)	0.036 : 0.072 (0.024 : 0.041)	0.037 : 0.052 (0.017 : 0.019)	0.034 : 0.056 (0.029 : 0.033)	+0.007 : 0.0	-0.02 : -0.016	0.783 : 0.583
Finger stiffness (slow : fast) (Nm.deg ⁻¹)	0.042 : 0.043 (0.027: 0.014)	0.02 : 0.023 (0.014 :0.005)	0.026 : 0.029 (0.021 : 0.02)	0.023 : 0.026 (0.028 : 0.021)	+0.006 : -0.014	+0.003 : +0.003	0.067 : 0.046
MAS wrist flexors (group sum of scores)	32	35	21	32	-11	-3	N/A
MAS finger flexors (group sum of scores)	33	30	14	19	-19	-11	N/A
Wrist extensor strength (Nm)	2.29 (2.21)	1.34 (1.24)	2.60 (2.40)	1.38 (1.25)	0.31	0.04	0.221
Finger extensor strength (Nm)	0.43 (0.55)	0.51 (0.42)	0.80 (0.97)	0.56 (0.43)	0.37	0.05	0.884
Grip strength (N)	109.33	83.72	110.53	104.53	1.00	20.81	0.486

PROMBIS RCT

Wrist extension ROM (passive : active) (degrees of motion)	-62.0 : -32.15 (19.30 : 26.68)	-57.93 : -28.64 (20.5 :20.20)	-68.69 : -42.92 (22.75 :16.61)	-62.0 : -37.85 (21.80 : 18.51)	-6.69 : -10.77	-4.07 : -9.21	0.454 : 0.198
Wrist flexion ROM (passive : active) (degrees of motion)	66.43 : 45.46 (19.2 : 13.94)	77.93 : 52.0 (18.47 : 16.38)	71.54 : 44.92 (22.26 : 15.88)	74.0 : 52.0 (15.79 : 19.02)	+5.11 : -0.54	-3.93 : +7.08	0.101 : 0.121
Finger extension ROM (passive : active) (degrees of motion)	-29.14 : 25.73 (26.52 : 26.76)	-53 : 6.67 (24.74 : 37.61)	-39.58 : 14.08 (26.83 : 25.13)	-53.5 : 4.71 (18.78 : 27.94)	-10.44 : -11.65	-0.5 : -1.96	0.025 : 0.178
Finger flexion ROM (passive : active) (degrees of motion)	92.79 : 77.57 (8.23 : 10.17)	96.07 : 75.93 (10.02 : 16.34)	94.0 : 79.08 (6.71: 15.27)	94.5 : 79.71 (6.52 : 16.91)	+1.21 : +1.51	-1.57 : +3.78	0.504 : 0.644
NHPT (pegs per second)	0.04 (0.09)	0.03 (0.07)	0.04 (0.1)	0.03 (0.08)	0	0	0.345
Action Research Arm Test (ARAT) (range 0-57, 57= full function)	24.14 (0.80)	23.43 (9.97)	29.23 (9.76)	25.57 (10.38)	5.09	2.14	0.480
ArMA passive (range 0-28, 0=full function)	6.50 (2.93)	7.50 (4.31)	6.00 (3.77)	8.50 (3.95)	-0.5	+1.0	0.250
ArmA active (range 0-52, 0=full function)	41.00 (7.55)	38.50 (6.09)	33.50 (10.47)	37.50 (11.3)	-7.50	-1.0	0.979

PROMBIS RCT

GAS: Goal 1 (range -2 to +2)*	-1 (0)	-1 (0)	1.77 (0.6)	1.64 (0.63)	+2.77	+2.64	N/A
GAS: Goal 2 (range -2 to +2)*	-1 (0)	-1 (0)	2.23 (0.83)	2.64 (0.84)	+3.23	+3.64	N/A
EQ5D (range	0.62 (0.14)	0.637 (0.17)	0.61 (0.16)	0.68 (0.18)	-0.01	+ 0.043	0.225

Values are mean (SD) unless otherwise indicated, significance levels were $p < 0.001$ for secondary outcomes (adjusted for multiple comparisons). Key: Stiffness slow = passive stiffness of soft tissue with no reflex component measured with the motor at 6 degrees/second, Stiffness fast = measures the reflex component (spasticity) with the motor at 300 degrees/second, Nm.deg^{-1} = Newton metres per degree, MAS= Modified Ashworth Scale converted to a six point score (range 0 to 5), Nm = Newton Metre, N = Newtons, ROM = range of movement; extension movements are represented by negative numbers and a negative mean change represents an **improvement** in range, flexion movements are represented by positive numbers and a positive mean change represents an improvement in range; *GAS = Goal Attainment Score – a score of 0 or above indicates that the goal was achieved as planned

	Treatment n=14	Placebo n=14	Total n=28
FDS	11	14	25
FDP	1	2	3
FPL	3	6	9
FPB	0	1	1
FCR	9	10	19
FCU	7	9	16
ECR	0	1	1
ECU	0	1	1
Lumbricals	4	2	6
Pronator Teres	2	4	6
Biceps	0	2	2
Adductor Policis	0	1	1
Thenar Eminance	0	1	1

Table 4: count of number of subjects injected for each muscle

Key: FDS = flexor digitorum superficialis; FDP = flexor digitorum profundus; FCR = flexor carpi radialis; FCU flexor carpi ulnaris; ECR = extensor carpi radialis; ECU = extensor carpi ulnaris; FPL = flexor pollicis longus; FPB = flexor pollicis brevis.

Figures

Uploaded as separate TIF files as required by journal. Labels and legends:

Figure 1: Consort diagram

Figure 2: Time taken to release grasp (mean +/- SEM)

Online supplements

Table e-1: Injection report detail – which muscles were injected and doses

Table e-2: adverse reactions

Appendix e-1: supplementary methods