BMJ Open

Evaluating the effectiveness of two injection-site localization techniques for botulinum toxin injections, echography or electrostimulation: a single-blind, crossover randomized trial.

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2016-011751
Article Type:	Protocol
Date Submitted by the Author:	02-Mar-2016
Complete List of Authors:	MOREL, Claire; CHU Gabriel Montpied, Centre Hospitalier Universitaire de Clermont Ferrand, service Médecine Physique et de Réadaptation HAURET, Isabelle; Centre Médical Etienne Clementel ANDANT, Nicolas; Biostatistics Unit, Délégation Recherche Clinique & Innovation, CHU Clermont Ferrand Pereira, Bruno; University Hospital CHU Clermont-Ferrand, Coudeyre, Emmanuel; Centre Hospitalier Universitaire de Clermont-Ferrand, Médecine Physique et de Réadaptation
Primary Subject Heading:	Rehabilitation medicine
Secondary Subject Heading:	Neurology
Keywords:	botulinum toxin, tracking, spasticity, hemiplegia, tardieu scale, ankle dorsiflexion

Evaluating the effectiveness of two injection-site localization techniques for botulinum toxin injections, echography or electrostimulation: a single-blind, crossover randomized trial.

Claire Morel[1,2], Isabelle Hauret[3], Nicolas Andant[4], Bruno Pereira[4], and Emmanuel Coudeyre[1,2,5]

1 Service de Médecine Physique et de Réadaptation; CHU Clermont-Ferrand, 58 rue Montalembert, 63 000 Clermont-Ferrand, France

2 Université Clermont Auvergne, Clermont-Ferrand, France

3 Centre médical Etienne Clémentel, 63530 Enval, France

4 Biostatistics Unit, Délégation Recherche Clinique & Innovation (DRCI), CHU Clermont-Ferrand, 63 000 Clermont-Ferrand F-63003;

5 INRA, Unité de Nutrition Humaine (UNH, UMR 1019), CRNH Auvergne, Clermont-Ferrand, France

Corresponding author:

Emmanuel COUDEYRE, Service de Médecine Physique et Réadaptation, CHU Clermont-Ferrand, Hôpital Nord,

Route de Chateaugay, F-63118 Cébazat, France

ecoudeyre@chu-clermontferrand.fr

Key words: botulinum toxin, tracking, spasticity, hemiplegia, Tardieu scale, ankle dorsiflexion

Word count: 3694 words

ABSTRACT

Introduction

Botulinum toxin injections are an effective treatment for limb spasticity following stroke. Different tracking techniques are used for this purpose: palpation, electrostimulation, electromyography, and ultrasound. Yet very few studies have compared these different techniques, and none has successfully proven the superior efficacy of ultrasound-guided injections compared to another tracking method. The primary objective of our study was therefore to compare the efficacy of botulinum toxin injections depending on the tracking technique used: ultrasound *versus* electrostimulation.

Methods and analysis

This is a clinical, single-center, prospective, interventional, single-blind, crossover randomized trial. In total, 30 patients aged between 18 and 80 years old presenting with triceps surae spasticity (evaluated at 1+/4 on the modified Ashworth scale, Appendix 1) associated with hemiplegia sequelae will be included. The patients will be selected among those who attend for consultation the Physical Medicine and Rehabilitation Department of the Clermont-Ferrand University Hospital. One group will receive the botulinum toxin injection (Dysport®) guided by electrostimulation then ultrasound, the second group's botulinum toxin injections will be guided by ultrasound then electrostimulation. For each patient, the duration of study participation is 5 months. The primary endpoint is variation in passive ankle dorsiflexion range of motion at slow and high speeds (Tardieu scale, Appendix 2) with the knee straight.

Ethics and dissemination

This study has ethical approval form the CPP of Rhônes-Alpes region. Results will be published in a peer-reviewed journal.

Trial registration

ClinicalTrials.gov Identifier, NCT01935544

Strengths and limitations of this study:

The management of muscle spasticity proves a major challenge in hemiplegia cases, with botulinum toxin injections constituting the first-line treatment for local or loco-regional spasticity. To our knowledge, no study has yet successfully proven the benefits of ultrasound-guided botulinum toxin injections in terms of efficacy and patient comfort compared to other guiding techniques.

Concerning the limitations, it is a prospective study with inherent risks due to this type of studies such as lost to follow up bias.

1) INTRODUCTION

The management of muscle spasticity is a major challenge in hemiplegia cases, with botulinum toxin injections constituting the first-line treatment for local or loco-regional spasticity [1].

Yet there is a range of techniques involving different methods for both injection and tracking. The most commonly-used tracking techniques consist of anatomy palpation, electrostimulation, electromyography, and, more recently, ultrasound. Palpating the patient to guide injection is not reliable, particularly when the deeper muscles are concerned (*e.g.,* only 12% successfully-positioned injections in the tibialis posterior and flexor carpi ulnaris), but also for more superficial muscles (22% failure rate for the gastrocnemius) [2]. Injection guiding *via* electromyogram (EMG) is not always appropriate, when there is difficulty obtaining active or passive muscle activation, thus preventing differentiation between muscular activation of a specific muscle and that of surrounding muscles [3]. In addition, with this technique, there is no correlation between the extent of spasticity and muscular activity [3].

Tracking *via* ultrasound is widely used in other indications, such as infiltrations in the locomotor system (particularly the tendons and joints) [4] or anesthetic nerve blocks [5]. The primary advantages of ultrasound-guided botulinum toxin injection are that tracking is painless [3], fast [6], more precise [3], and thus safer, avoiding complications associated with subcutaneous, intravascular, or too-deep injections [7].

A comparative study has assessed the efficacy of ultrasound-based tracking with an electrostimulation-based technique [8]. The authors evaluated 32 children presenting with cerebral palsy sequelae, who were divided into two groups. All received botulinum toxin injection into the gastrocnemius, which was guided by either ultrasound or electrostimulation depending on the group. The techniques were evaluated based on three different scales: the Ashworth, Tardieu, selective motor control (SMC), and Physician Rating scales. The authors observed a non-significant improvement in spasticity, assessed by the Ashworth and Tardieu scales, at 3 months post-injection, in the group treated with ultrasound-guided injections. In contrast, the electrostimulation-guided group exhibited non-significant improvement in motor control of the antagonistic muscles. The only significant differences revealed were improvements in walking pattern and foot-to-ground contact in the ultrasound-guided group. Nonetheless, the numerous controversial methodological choices made by the authors limited the relevance of these results.

There have also been two comparative studies evaluating the efficacy of ultrasound-guiding with that of techniques using electrostimulation or anatomy palpation [9-10].

The first [9], conducted in 2012, compared these three injection-guiding techniques in the lower limbs. The trial involved 49 patients presenting with lower-limb spasticity following stroke, who were randomized into three groups, the first receiving injections guided by anatomy palpation, the second by electrostimulation, and the last by ultrasound. All received a botulinum toxin injection into the gastrocnemius, administered by the same physician. The investigator, who was blinded to the injection type, evaluated each patient on inclusion and at 1 month. The patients were forbidden from undergoing any form of physical therapy within the 3 months preceding the study and during its entirety. Ashworth and Tardieu scale results were assessed for all, along with passive dorsiflexion of the foot. The authors reported significantly improved passive dorsiflexion of the foot in the ultrasound-

guided injection group compared to the electrostimulation-guided group. Moreover, the Ashworth scale results were significantly improved 1 month following botulinum toxin injection in the ultrasound-guided group compared to the group where anatomy palpation was used.

The second study [10] was conducted in 2013 and assessed upper-limb spasticity in 60 patients who had suffered from strokes. As in the above-described study, these patients were randomized into three groups of 20 each in order to compare the three injection-guiding techniques: ultrasound, electrostimulation, and anatomy palpation. Two injections were administered in at least two of the following muscles: flexor carpi ulnaris, flexor carpi radialis, flexor digitorum superficialis, and the flexor digitorum profundus. The same physician, experienced with using botulinum toxin under ultrasound-guiding, administered all the injections. An investigator who was blinded to the injection type assessed each patient at the beginning and 4 weeks into the study. The patients were forbidden from undergoing any type of physical therapy in the 3 months preceding the study and during its entirety. The Ashworth and Tardieu scale results were assessed, along with passive dorsiflexion of the wrist and fingers. One month following injection, the modified Ashworth scale scores significantly improved in the group having undergone ultrasound-guided injection compared to the group tracked using anatomy palpation, as did the Tardieu scale scores and passive mobilizations. In contrast, the authors found no significant differences between ultrasound- and electrostimulation-guiding for the different evaluations.

In both of these studies, the authors described limitations consisting of the absence of functional evaluation of the upper or lower limbs, owing to the short follow-up rendering this assessment difficult to implement, as well as of the injections being administered by only one physician experienced with ultrasound-guided injection. The authors also indicated that body

mass index (BMI) was not taken into account in their studies, despite obesity potentially constituting a limitation to the accurate assessment of anatomical landmarks.

In a literature review [11], all four guiding techniques (anatomy palpation, electromyography [EMG], electrostimulation, and ultrasound) were compared, with advantages and disadvantages outlined for each. The authors retrieved and analyzed 15 articles, concluding that injection guided by anatomy palpation required no equipment and only a small-sized needle. Yet deep or slighter muscles were more difficult to access. In addition, while EMG enabled the toxin to be injected closest to the motor end-plate, this technique could, however, not guarantee that the needle was actually in the target muscle. As for the electrostimulation-guided technique, its primary advantage appeared to be its precise localization capacity. Despite this, it can take a long time to perform and require more training than the EMG and anatomy palpation techniques. Finally, ultrasound was found to enable the real-time visualization of the needle's progression while avoiding certain structures like blood vessels or nerves, among other advantages. In addition, the needle used in this technique was finer and thus less painful. On the other hand, this technique was highly dependent on the operator's skill, potentially requiring the presence of an assistant for beginners.

All in all, guiding injections by anatomy palpation thus appears to be the least precise technique. The other guiding techniques appear to offer superiority, in terms of precision and thus efficacy, although further studies must be conducted in order to determine which technique achieves the best clinical results.

Another literature review [12] evaluated the impact of the different injection-guiding techniques on the efficacy of botulinum toxin when treating not only spasticity but also dystonia. This review covered 10 studies, seven of which were randomized. The authors reported a high level of evidence (Grade A) that instrument-based guiding, *i.e.,* ultrasound,

electrostimulation or electromyography, was more effective than manual guiding in the treatment of upper limb spasticity, spastic equinus following stroke in adults, and cerebral palsy in children. The review's conclusions were that no instrument-based guiding technique proved superior to another. At the present time, no recommendation can be made in terms of choosing the optimal guiding technique, although ultrasound nevertheless appears to be more effective than electrostimulation in spastic equinus treatment following stroke in adults (passive mobilization of the ankle) [10].

No study has as yet successfully proven the benefits of ultrasound-guided botulinum toxin injections in terms of efficacy and patient comfort compared to other guiding techniques.

2) METHODS/DESIGN

Objective

Our main objective is to compare the efficiency of botulinum toxin injections in terms of guiding technique: ultrasound *vs.* electrical stimulation.

The secondary objective is to demonstrate that ultrasound-guidance is a less painful localization technique.

Study design

This prospective, randomized, single-center, single-blind, crossover study will be conducted in chronic stroke patients presenting with triceps surae spasticity. The patients will

receive two injections; each administered using a different guiding technique. Randomization will determine which technique will be used in the first and second instances. The patients will be selected among those who attend for consultation the Physical Medicine and Rehabilitation Department of the Clermont-Ferrand University Hospital. The botulinum toxin injections and assessments will take place in the same department. The study will last 5 months for each patient. This study does not present a major risk for the subjects. The main potential disadvantages to the treatment consist of injection pain or side-effects from the botulinum toxin (increased motor deficit or dysphagia).

The study design is presented in Figure 1.

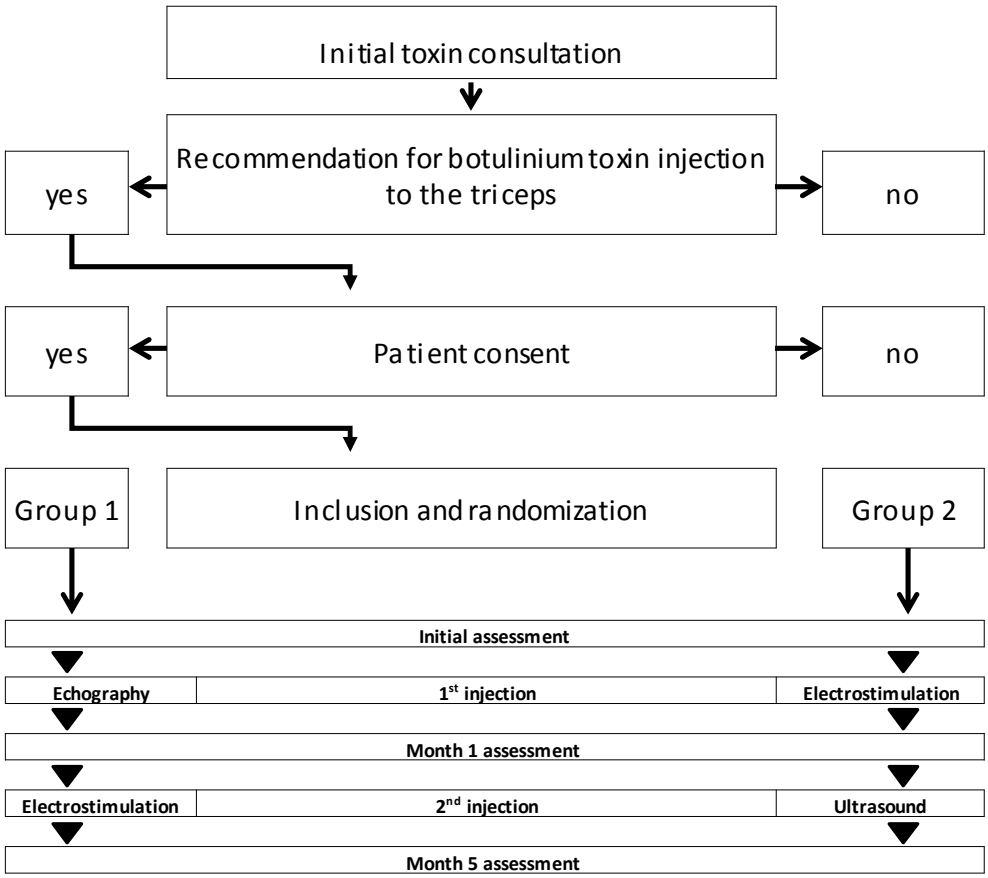

Figure 1: Flow diagram showing the different stages of the protocol

Randomization

The patients will be randomly assigned to one of the above-described groups by means of a Latin square design in order to balance out the group numbers.

Study description

The patients pre-selected during consultation at the Physical Medicine and Rehabilitation Department of the Clermont-Ferrand University Hospital will be handed a letter containing information on the study protocol. They will then have 1 month to grant their consent, should they wish, and be included at their next consultation.

The following data will then be collected for each patient: age, gender, time since stroke, side affected by the cerebral lesion, current treatments and dosages (for managing spasticity and pain), date of first botulinum toxin injection, as well as severity of deficit (functional walking scale).

The initial assessment of the patients included in the study will be performed just prior to the first injection. This evaluation will be both clinical (assessment of the triceps surae spasticity based on the Tardieu and modified Ashworth scales) and instrument-based (walking speed using GAITRite, CIR Systems Inc. Sparta, New Jersey, the USA).

The first injection will be administered in the outpatient clinic, guided using ultrasound or electrostimulation, depending on the group. The clinical investigator will randomize the patients then administer the injection according to the guiding method assigned. In total, 500 units of A Dysport® botulinum toxin will be injected into four separate areas of the triceps surae. Further injections will be administered into other muscle groups, if necessary. The total

dose for this injection will be 1,500 Dysport® units. Any pain experienced during the injection will be assessed by means of vertical visual analogue scales, and the time required for tracking and administering the injection will be recorded.

The second injection will be administered 4 months after the first, also in the outpatient clinic. The tracking method used on this occasion will differ from that used for the first. A total of 500 units of A Dysport® botulinum toxin will be injected into four different areas of the triceps surae. Further injections will be administered into other muscle groups, if necessary. The total dose administered for this injection will be 1,500 Dysport® units. Any pain experienced during the injection will be assessed using vertical visual analogue scales, and the time required for tracking and administering the injection will be recorded.

The two follow-up visits will take place 1 month after each botulinum toxin injection. Each patient will be asked to attend the clinic for consultation so as to allow the efficacy of the injection to be assessed. This assessment will be both clinical (assessment of the spasticity of the triceps surae by means of the Tardieu and modified Ashworth scales) and instrument-based (walking speed using GAITRite). Each follow-up visit will be performed by an investigator (physiotherapist) blinded to the tracking technique.

This study is actually ongoing and the investigators are currently still collecting data. The contents of the manuscript have not been submitted or published elsewhere.

Patients

The patients will be recruited among those attending the physical medicine and rehabilitation (PM+R) hospital consultations. The inclusion and exclusion criteria are described in Table 1. The patients will be randomized on inclusion into two groups, each due to receive two botulinum toxin injections spaced 4 months apart. The order of guiding

techniques used for each injection will differ between the groups: ultrasound then electrostimulation for one group and electrostimulation then ultrasound for the other. The indication for botulinum toxin injection to the upper limb will not constitute a non-inclusion criterion for this study.

Inclusion Criteria	Age 18 to 80 years
	Hemiplegic sequelae of stroke
	Triceps surae spasticity evaluated at 1 + / 4 on the modified Ashworth scale
	Ability to provide written consent
Exclusion Criteria	Injection of botulinum toxin dating from over 3 months
	Previous ultrasound-guided injection of botulinum toxin
	Swallowing impairment
	Ongoing anti-vitamin K (AVK) anticoagulation treatment with international normalized ratio (INR) >3 during one week before randomization
	Ongoing aminoglycoside treatment
	General anesthesia with planned curare injection during study participation
	Implanted with a pacemaker

	History of ankle arthrodesis
	Other contra-indication for botulinum toxin injection: myasthenia gravis, pregnancy, or breast feeding

Evaluation

The primary endpoint is variation in passive ankle dorsiflexion range of motion at slow and high speeds (Tardieu scale) while keeping the knee straight.

The procedure consists of assessing the angle at which resistance manifests, as well as the intensity of this resistance to mobilization at slow and fast speeds [13]. The ankle dorsal flexion angle will thus be measured by means of a goniometer during passive manipulation of the ankle with the knee being kept straight, before and after treatment.

This straight-knee assessment is relevant for simultaneously obtaining measurement of the soleus and gastrocnemius muscle spasticity, which are bi-articular.

The Tardieu scale is more sensitive than the commonly-used modified Ashworth scale. The latter only consists of five stages, which does not always allow for treatment efficacy to be evaluated. Furthermore, this scale does not take into account the velocity factor during spasticity [14]. Nevertheless, validation studies pertaining to the Tardieu scale and involving the adult population are scarce in the scientific literature.

Assessing the difference in the range of motion between slow and fast speeds is relevant because this takes into account not only the spastic component but also any potential tendon retraction.

The principal evaluation criterion will be measured on the day of injection and at the Month 1 assessment.

The secondary endpoints are:

- other components of the "Tardieu scale": quality of muscle reaction (X) at slow and fast speeds, as well as angle of apparition of the muscle reaction (Y) at slow and fast speeds;
- assessment of the triceps surae spasticity on the modified Ashworth scale;
- walking speed;
- extent of pain at the injection site using a visual analogue scale;
- duration of tracking and injection.

Statistical considerations :

To date, only one comparative study focused on the protocol's topic has been published [15]. Therefore, if scientific literature data provides information on the statistical variability of ankle dorsiflexion range of motion at slow and high speeds obtained using the Tardieu scale for patients having suffered from stroke [15], exhibiting a standard deviation around 8.5°, proposing an expected difference between the two randomized groups proves challenging. In addition, in order to highlight the efficiency of botulinum toxin injections in terms of guiding technique, namely ultrasound vs. electrical stimulation, sample size estimation was based on statistical power simulations in relation to recruitment capacity. To demonstrate a minimum difference of 7.12, with an effect size of 0.8, 15 patients per group (ultrasound stimulation then electrical vs. electrical stimulation then ultrasound) will be needed for a two-sided Type I error at 5%, a statistical power of 90%, and a correlation coefficient equal to 0, owing due to

the cross-over design. For a more favorable correlation coefficient (for example 0.5), the difference expected with 30 subjects will be near to 5° (effect size of 0.6). Finally, if there would be an interaction effect "order processing x group", only the results of the first period could be considered. Under the previous assumptions, notably 15 subjects per group, the expected difference between the two groups would be 10°. Statistical power estimations will be performed a posteriori on other components of the Tardieu scale: quality of muscle reaction (X) at slow and fast speeds, as well as angle of apparition of the muscle reaction (Y) at slow and fast speeds.

Statistical analysis will be performed on an intention-to-treat basis using the Stata software (Version 13, StataCorp, College Station, US) for a two-sided Type I error at α=5%. The patient characteristics will be described by numbers and associated percentages for categorical data. For quantitative parameters, mean (standard-deviation) or median (interquartile range) values will be calculated and presented according to statistical distribution. The assumption of normality will be studied by Shapiro-Wilk test. The primary endpoint, namely change in the ankle dorsiflexion range of motion at slow and high speeds obtained using the Tardieu scale, will be compared between the groups using a repeated analysis of variance (ANOVA) for cross-over designs while taking into account the following effects: treatment group (ultrasound vs. electrical stimulation), order processing, sequence, subject (as random-effect), and carry-over. A particular focus will be given to the interaction "order processing x group". If this test proves significant, the statistical analysis will only cover the first period of this cross-over study. The normality of residuals will be studied, as described previously. When endpoints do not assume the normality assumption, a non-parametric paired test like the Wilcoxon will be proposed. Analyses concerning the secondary endpoints (quality of muscle reaction [X] at slow and fast speeds, the angle of the muscle reaction apparition [Y] at slow

and fast speeds, assessment of the triceps surae spasticity on the modified Ashworth scale, walking speed, and extent of pain at the injection site using a visual analogue scale, along with the duration of tracking and injection will be studied in a similar way as the primary endpoint. For categorical parameters, Stuart-Maxwell test for paired data or generalized linear mixed model taking into account the above-mentioned effects will be applied. Concerning non-crossover comparisons, usual statistical tests will be performed: Student t-test or Mann-Whitney test if the conditions of t-test are no met (normality or homoscedasticity verified using Fisher-Snedecor test) for quantitative parameters and Chi-squared test or Fischer's exact test for categorical variables, if appropriate. As discussed by Feise [16], adjustment of Type I error (α) will not be proposed systematically, but on a case-by-case basis in the light of clinical considerations rather than statistical ones only.

3) DISCUSSION

The various comparative studies currently available [8-10] have demonstrated that instrument-guided procedures, such as electrostimulation and ultrasound, improve the efficacy of botulinum toxin injections compared to that obtained by means of simple anatomy palpation, in line with current recommendations for good clinical practices.

Ultrasound enables us to visualize in real-time the needle's progress, resulting in a precise localization of the target muscle, while avoiding certain structures like blood vessels and nerves. In addition, this technique allows for a passive manipulation of the limb part under study in order to distinguish the muscular body of the target muscle from that of other adjacent muscular structures [10].

Ultrasound-guided botulinum toxin injection can be subject to the same limitations inherent to ultrasound itself. The technique is highly dependent on the skills of the operator, who needs to be experienced, thus requiring further investment in terms of training and equipment. Additionally, the structural evolution of spastic muscles, and in particular fibrous involution, alter the ultrasound features of the muscle, rendering it at times difficult to distinguish from the different adjacent muscles [16].

The substantial cost of ultrasound equipment no longer appears to represent an obstacle to using this guiding technique. It is now possible to directly employ different ultrasound waves with a digital tablet, thus considerably reducing the equipment costs.

With regard to the guiding speed of the different techniques, the literature currently provides contradictory views. The Berweck team [6] demonstrated that the mean time of muscle localization and injection was only 5 seconds for superficial muscles and 30 seconds for deeper ones when using ultrasound. On the other hand, the 2010 Henzel study [16] reported an average increase of 5 to 10 minutes in procedure time when adding ultrasound-guiding to usual guiding techniques. If ultrasound-guided injection was concretely proven to be faster than other methods, this could represent a particular advantage for children and poorly compliant adults displaying low tolerance for procedures involving prolonged immobilization [10].

In this study, we hypothesize that botulinum toxin injections guided by ultrasound are more efficacious than those using electrostimulation, with the triceps surae spasticity as primary evaluation criterion. In addition, we also seek to prove that ultrasound-guided

botulinum toxin injections are less painful than those administered using electrostimulation, and that the time needed for localizing and injecting is shorter for the former.

The expected benefit for the patient is thus a more efficacious injection and consequently reduced spasticity of the triceps surae. The benefits of ultrasound-guided injection compared to that of electrostimulation-guided consist of reduced tracking and injection times, in addition to reduced pain on injection.

This study's objective is to improve the techniques pertaining to guiding and injection. When injecting botulinum toxin, it is, in fact, all the more crucial to be as precise as possible in order to ensure the best efficacy in the target muscles while avoiding any unwanted-effects that could arise in relation with toxin diffusion or intravascular injection. For this reason, it is highly-desirable to use the most reliable guiding method possible. Furthermore, toxin injection can be a painful procedure, particularly for certain patients suffering from hyperesthesia or cognitive disorders, meaning a guiding technique that enables the highest tolerance is all the more crucial.

4) FOOTNOTES

Authors' contributors

BP: data analysis, critical revision, and final approval of the manuscript. CM: manuscript writing, critical revision, and final approval of the manuscript. EC: conception and design, analysis and interpretation, manuscript writing, critical revision, and final approval of the manuscript. HI: conception and design, data collection, analysis and interpretation,

manuscript writing, critical revision, and final approval of the manuscript. NA: study setup, critical revision, and approval of the final manuscript.

Funding statement:

This work was supported by CHU Clermont Ferrand and IPSEN provided funding for the ultrasound.

Competing interest:

None

Ethics approval

The study protocol, patient information, and patient consent form, along with the case report form, have been submitted to the ethics committee of the Rhône-Alpes region (*Comité de Protection des Personnes Rhône-Alpes*, *CPP Sud-Est I*). The CPP's favorable assessment was transmitted to the study sponsor and ANSM (*l'Agence nationale de sécurité des medicaments et des produits de santé* – the French national agency of medicine and health products safety) on November 29, 2012.

5) ABBREVIATIONS

ANSM: *Agence nationale de sécurité du médicament et des produits de santé* - the French national agency of medicine and health products safety

CPP: *Comité de protection des personnes* – ethics committee

EMG: Electromyogram

PM+R: Physical medecine and rehabilitation

6) AUTHORS' INFORMATION

1 Service de Médecine Physique et de Réadaptation; CHU Clermont-Ferrand, 58 rue Montalembert, 63 000 Clermont-Ferrand, France

2 Université Clermont Auvergne, Clermont-Ferrand, France

3 Centre médical Etienne Clémentel, 63530 Enval, France

4 Biostatistics Unit, Délégation Recherche Clinique & Innovation (DRCI), CHU Clermont-Ferrand, 63 000 Clermont-Ferrand F-63003;

5 INRA, Unité de Nutrition Humaine (UNH, UMR 1019), CRNH Auvergne, Clermont-Ferrand, France

7) REFERENCES

1. Yelnik AP, Simon O, Bensmail D, Chaleat-Valayer E, Decq P, Dehail P, et al. Drug treatments for spasticity. Ann PhysRehabil Med. 2009 Dec;52(10):746–56.

2. Chin TYP, Nattrass GR, Selber P, Graham HK. Accuracy of intramuscular injection of botulinum toxin A in juvenile cerebral palsy: a comparison between manual needle placement and placement guided by electrical stimulation. J PediatrOrthop. 2005 Jun;25(3):286–91.

3. Schroeder AS, Berweck S, Lee SH, Heinen F. Botulinum toxin treatment of children with cerebral palsy - a short review of different injection techniques. Neurotox Res. 2006 Apr;9(2-3):189–96.

4. Adler RS, Sofka CM. Percutaneous ultrasound-guided injections in the musculoskeletal system. Ultrasound Q. 2003 Mar;19(1):3–12.

5. Walker KJ, McGrattan K, Aas-Eng K, Smith AF. Ultrasound guidance for peripheral nerve blockade. Cochrane Database Syst Rev. 2009;(4):CD006459.

6. Berweck S, Schroeder AS, Fietzek UM, Heinen F. Sonography-guided injection of botulinum toxin in children with cerebral palsy. Lancet. 2004 Jan 17;363(9404):249–50.

7. Berweck S, Heinen F. Use of botulinum toxin in pediatric spasticity (cerebral palsy). MovDisord. 2004 Mar;19Suppl 8:S162–7.

8. Kwon J-Y, Hwang JH, Kim J-S. Botulinum toxin a injection into calf muscles for treatment of spastic equinus in cerebral palsy: a controlled trial comparing sonography and electric stimulation-guided injection techniques: a preliminary report. Am J Phys

Med Rehabil. 2010 Apr;89(4):279–86.

9. Picelli A, Lobba D, Midiri A, Prandi P, Melotti C, Baldessarelli S, et al. Botulinum toxin injection into the forearm muscles for wrist and fingers spastic overactivity in adults with chronic stroke: a randomized controlled trial comparing three injection techniques. ClinRehabil. 2014 Mar;28(3):232–42.

10. Picelli A, Tamburin S, Bonetti P, Fontana C, Barausse M, Dambruoso F, et al. Botulinum toxin type A injection into the gastrocnemius muscle for spastic equinus in adults with stroke: a randomized controlled trial comparing manual needle placement, electrical stimulation and ultrasonography-guided injection techniques. Am J Phys Med Rehabil. 2012 Nov;91(11):957–64.

11. Walker HW, Lee MY, Bahroo LB, Hedera P, Charles D. Botulinum Toxin Injection Techniques for the Management of Adult Spasticity. PM R. 2015 Apr;7(4):417–27.

12. Grigoriu A-I, Dinomais M, Rémy-Néris O, Brochard S. Impact of Injection-Guiding Techniques on the Effectiveness of Botulinum Toxin for the Treatment of Focal Spasticity and Dystonia: A Systematic Review. Arch Phys Med Rehabil. 2015 May 14;

13. Ben Smaïl D, Kiefer C, Bussel B. [Clinical evaluation of spasticity]. Neurochirurgie. 2003 May;49(2-3 Pt 2):190–8.

14. Haugh AB, Pandyan AD, Johnson GR. A systematic review of the Tardieu Scale for the measurement of spasticity.DisabilRehabil. 2006 Aug 15;28(15):899–907.

15. Singh P, Joshua AM, Ganeshan S, Suresh S. Intra-rater reliability of the modified Tardieu scale to quantify spasticity in elbow flexors and ankle plantar flexors in adult stroke subjects. Ann Indian Acad Neurol. 2011 Jan;14(1):23–6.

16. Henzel MK, Munin MC, Niyonkuru C, Skidmore ER, Weber DJ, Zafonte RD. Comparison of surface and ultrasound localization to identify forearm flexor muscles for botulinum toxin injections. PM R. 2010 Jul;2(7):642–6.

APPENDIX 1:

Modified Ashworth Scale

Modified Ashworth Scale for Grading Spasticity	
Grade	**Description**
0	No increase in muscle tone
1	Slight increase in muscle tone, manifested by a catch and release or by minimal resistance at the end of the ROM when the affected part(s) is moved in flexion or extension
1+	Slight increase in muscle tone, manifested by a catch, followed by minimal resistance throughout the remainder (less than half) of the ROM
2	More marked increase in muscle tone through most of the ROM, but affected part(s) easily moved
3	Considerable increase in muscle tone, passive movement difficult
4	Affected part(s) rigid in flexion or extension

APPENDIX 2:

Tardieu Scale

Muscle reaction to stretch recorded for specific velocities:

- V1: as slow as possible
- V3: as fast as possible

Quality of muscle reaction (X)	
0	No resistance throughout the course of the passive movement
1	Slight resistance throughout the course of the passive movement with no clear catch at a precise angle
2	Clear catch at a precise angle interrupting the passive movement, followed by a release
3	Fatiguable clonus (<10s when maintaining the pressure) appearing at a precise angle
4	Unfatiguable clonus (>10s when maintaining the pressure) appearing at a precise angle
Angle of muscle reaction (Y)	
	Measure relative to the position of minimal stretch of the muscle (corresponding at angle 0), with the exception of the hip, for which the measure is relative to the resting anatomical position

570

571

572

Inclusion Criteria	Age 18 to 80 years
	Hemiplegic sequelae of stroke
	Triceps surae spasticity evaluated at 1 + / 4 on the modified Ashworth scale
	Ability to provide written consent
Exclusion Criteria	Injection of botulinum toxin dating from over 3 months
	Previous ultrasound-guided injection of botulinum toxin
	Swallowing impairment
	Ongoing anti-vitamin K (AVK) anticoagulation treatment with international normalized ratio (INR) >3 during one week before randomization
	Ongoing aminoglycoside treatment
	General anesthesia with planned curare injection during study participation
	Implanted with a pacemaker
	History of ankle arthrodesis
	Other contra-indication for botulinum toxin injection: myasthenia gravis, pregnancy, or breast feeding

Table 1: Inclusion and exclusion criteria

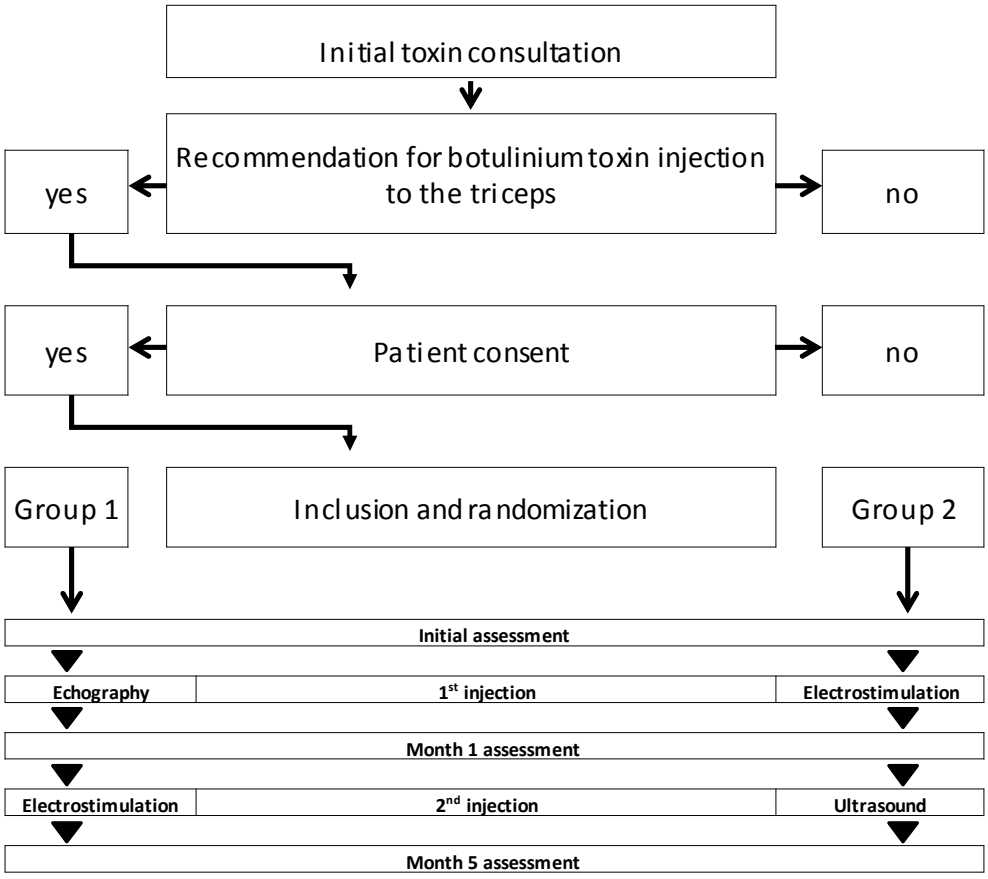

Figure 1: Flow diagram showing the different stages of the protocol

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	3
	2b	All items from the World Health Organization Trial Registration Data Set	X
Protocol version	3	Date and version identifier	X
Funding	4	Sources and types of financial, material, and other support	20
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	21
	5b	Name and contact information for the trial sponsor	21
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	20
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	NA

Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	5
	6b	Explanation for choice of comparators	X
Objectives	7	Specific objectives or hypotheses	9
Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)	11
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	9
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	13
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	10
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	X
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	X
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	X
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	14
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	10

Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	15
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	12
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	15
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	X
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	11
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	12
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	X
Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	X
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	X

Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	X
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	15
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	15
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	15
Methods: Monitoring			
Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	X
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	X
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	X
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	X
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	20
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	X

Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	20
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	NA
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	X
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	20
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	X
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	X
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	X
	31b	Authorship eligibility guidelines and any intended use of professional writers	X
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	X
Appendices			
Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	X
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	NA

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.

BMJ Open

Efficacy of two injection-site localization techniques for botulinum toxin injections: a single-blind, crossover randomized trial protocol among adults with hemiplegia due to stroke.

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2016-011751.R1
Article Type:	Protocol
Date Submitted by the Author:	08-Jul-2016
Complete List of Authors:	MOREL, Claire; CHU Gabriel Montpied, Centre Hospitalier Universitaire de Clermont Ferrand, service Médecine Physique et de Réadaptation HAURET, Isabelle; Centre Médical Etienne Clementel ANDANT, Nicolas; Biostatistics Unit, Délégation Recherche Clinique & Innovation, CHU Clermont Ferrand Pereira, Bruno; University Hospital CHU Clermont-Ferrand, Bonnin, Armand; CHU Gabriel Montpied, Centre Hospitalier Universitaire de Clermont Ferrand, service Médecine Physique et de Réadaptation; Centre Hospitalier Universitaire de Clermont-Ferrand, Médecine Physique et de Réadaptation Coudeyre, Emmanuel; Centre Hospitalier Universitaire de Clermont-Ferrand, Médecine Physique et de Réadaptation
Primary Subject Heading:	Rehabilitation medicine
Secondary Subject Heading:	Neurology
Keywords:	echography, electrostimulation, soleus, gastrocnemius

Efficacy of two injection-site localization techniques for botulinum toxin injections: a single-blind, crossover randomized trial protocol among adults with hemiplegia due to stroke.

Claire Morel[1,2], Isabelle Hauret[3], Nicolas Andant[4], Bruno Pereira[4] , Armand Bonnin[1,2] and Emmanuel Coudeyre[1,2,5]

1 Service de Médecine Physique et de Réadaptation; CHU Clermont-Ferrand, 58 rue Montalembert, 63 000 Clermont-Ferrand, France

2 Université Clermont Auvergne, Clermont-Ferrand, France

3 Centre médical Etienne Clémentel, 63530 Enval, France

4 Biostatistics Unit, Délégation Recherche Clinique & Innovation (DRCI), CHU Clermont-Ferrand, 63 000 Clermont-Ferrand F-63003;

5 INRA, Unité de Nutrition Humaine (UNH, UMR 1019), CRNH Auvergne, Clermont-Ferrand, France

Corresponding author:

Emmanuel COUDEYRE, Service de Médecine Physique et Réadaptation, CHU Clermont-Ferrand, Hôpital Nord,

Route de Chateaugay, F-63118 Cébazat, France

ecoudeyre@chu-clermontferrand.fr

Key words: echography, electrostimulation, soleus, gastrocnemius

Word count: 3694 words

ABSTRACT

Introduction

Botulinum toxin injections are an effective treatment for limb spasticity following stroke. Different tracking techniques are used for this purpose: palpation, electrostimulation, electromyography and ultrasound. Yet very few studies have compared these different techniques, and none has successfully proven the superior efficacy of ultrasound-guided injections compared to another tracking method. The primary objective of our study was therefore to compare the efficacy of botulinum toxin injections depending on the tracking technique used: ultrasound *versus* electrostimulation.

Methods and analysis

This is a clinical, single-center, prospective, interventional, single-blind, crossover randomized trial. In total, 30 patients aged between 18 and 80 years old presenting with triceps surae spasticity (evaluated >1 on the modified Ashworth scale) associated with hemiplegia sequelae due to stroke will be included. The patients will be selected among those who attend for consultation the Physical Medicine and Rehabilitation Department of the Clermont-Ferrand University Hospital. One group will receive the abobotulinumtoxinA (BoNT-A) injection guided by electrostimulation then ultrasound, the second group's botulinum toxin injections will be guided by ultrasound then electrostimulation. For each patient, the duration of study participation is 5 months. The primary endpoint is variation in passive ankle dorsiflexion range of motion at slow and high speeds (Tardieu scale) with the knee straight.

Ethics and dissemination

This study has ethical approval form the CPP of Rhônes-Alpes region. Results will be published in a peer-reviewed journal.

Trial registration

ClinicalTrials.gov Identifier, NCT01935544

Strengths and limitations of this study:

The management of muscle spasticity proves to be a major challenge in hemiplegia following a stroke, with botulinum toxin injections constituting the first-line treatment for local or loco-regional spasticity.

Concerning the limitations, it is a prospective study with inherent risks due to this type of studies such as lost to follow up bias.

1) INTRODUCTION

The management of muscle spasticity is a major challenge in hemiplegia following a stroke, with botulinum toxin injections constituting the first-line treatment for local or loco-regional spasticity (1).

Yet there is a range of techniques involving different methods for both injection and tracking. The most commonly-used tracking techniques are, anatomy palpation, electrostimulation, electromyography, and, more recently, ultrasound. Palpating children to guide injection is not reliable, particularly when the deeper muscles are concerned (*e.g.,* only 12% successfully-positioned injections in the tibialis posterior and flexor carpi ulnaris), but also for more superficial muscles (22% failure rate for the gastrocnemius) (2). For children, injection guided *via* electromyogram (EMG) is not always appropriate, when there is difficulty obtaining active or passive muscle activation, to differentiate muscular activation of a specific muscle from surrounding muscles (3). In addition, with this technique, there is no correlation between the extent of spasticity and muscular activity (3). One article (4) shows that neither manual needle placement nor electrical stimulation is wholly accurate to inject gastrocnemius muscle of adults with spasticity.

Tracking *via* ultrasound is widely used in other indications, such as infiltrations in the locomotor system (particularly the tendons and joints) (5) or anesthetic nerve blocks (6). The primary advantages of ultrasound-guided botulinum toxin injection are that tracking is painless (3), fast (7), more precise (3), and thus safer, avoiding complications associated with subcutaneous, intravascular, or too-deep injections (8).

A comparative study has assessed the efficacy of ultrasound-based tracking with an electrostimulation-based technique (9). The authors evaluated 32 children presenting with cerebral palsy sequelae, who were divided into two groups. All received botulinum toxin injection into the gastrocnemius, which was guided by either ultrasound or electrostimulation depending on the group. The techniques were evaluated based on three different scales: the Ashworth (Appendix 1), Tardieu (Appendix 2) (10), selective motor control (SMC), and Physician Rating scales. The authors observed a non-significant improvement in spasticity, assessed by the Ashworth and Tardieu scales, at 3 months post-injection, in the group treated with ultrasound-guided injections. In contrast, the electrostimulation-guided group showed non-significant improvement in motor control of the antagonistic muscles. The only significant differences revealed were improvements in walking pattern and foot-to-ground contact in the ultrasound-guided group. Nonetheless, the numerous controversial methodological choices made by the authors limited the relevance of these results.

There have also been two comparative studies evaluating the efficacy of ultrasound-guiding with that of techniques using electrostimulation or anatomy palpation (11) (12).

The first (11), conducted in 2012, compared these three injection-guiding techniques in the lower limbs. The trial involved 49 patients presenting with lower-limb spasticity following stroke, who were randomized into three groups, the first receiving injections guided by anatomy palpation, the second by electrostimulation, and the last by ultrasound. All received a botulinum toxin injection into the gastrocnemius, administered by the same physician. The investigator, who was blinded to the injection type, evaluated each patient on inclusion and at 1 month. The patients were forbidden from undergoing any form of physical therapy within the 3 months preceding the study and during its entirety. Ashworth and Tardieu scale results were assessed for all, along with passive dorsiflexion of the foot. The authors reported significantly improved passive dorsiflexion of the foot in the ultrasound-

guided injection group compared to the electrostimulation-guided group. Moreover, the Ashworth scale results were significantly improved 1 month following botulinum toxin injection in the ultrasound-guided group compared to the group where anatomy palpation was used.

The second study (12) was conducted in 2013 and assessed upper-limb spasticity in 60 patients who had suffered from strokes. As in the above-described study, these patients were randomized into three groups of 20 each in order to compare the three injection-guiding techniques: ultrasound, electrostimulation, and anatomy palpation. Two injections were administered in at least two of the following muscles: flexor carpi ulnaris, flexor carpi radialis, flexor digitorum superficialis, and the flexor digitorum profundus. The same physician, experienced with using botulinum toxin under ultrasound-guiding, administered all the injections. An investigator who was blinded to the injection type assessed each patient at the beginning and 4 weeks into the study. The patients were forbidden from undergoing any type of physical therapy in the 3 months preceding the study and during its entirety. The Ashworth and Tardieu scale results were assessed, along with passive dorsiflexion of the wrist and fingers. One month following injection, the modified Ashworth scale scores significantly improved in the group having undergone ultrasound-guided injection compared to the group tracked using anatomy palpation, as did the Tardieu scale scores and passive mobilizations. In contrast, the authors found no significant differences between ultrasound- and electrostimulation-guiding for the different evaluations.

In both of these studies, the authors described limitations consisting of the absence of functional evaluation of the upper or lower limbs, owing to the short follow-up rendering this assessment difficult to implement, as well as of the injections being administered by only one physician experienced with ultrasound-guided injection. The authors also indicated that body

mass index (BMI) was not taken into account in their studies, despite obesity potentially constituting a limitation to the accurate assessment of anatomical landmarks.

In a literature review (13), all four guiding techniques (anatomy palpation, electromyography [EMG], electrostimulation, and ultrasound) were compared, with advantages and disadvantages outlined for each. The authors retrieved and analyzed 15 articles, concluding that injection guided by anatomy palpation required no equipment and only a small-sized needle. Yet deep or slighter muscles were more difficult to access. In addition, while EMG enabled the toxin to be injected closest to the motor end-plate, this technique could, however, not guarantee that the needle was actually in the target muscle. As for the electrostimulation-guided technique, its primary advantage appeared to be its precise localization capacity. Despite this, it can take a long time to perform and require more training than the EMG and anatomy palpation techniques. Finally, ultrasound was found to enable the real-time visualization of the needle's progression while avoiding certain structures like blood vessels or nerves, among other advantages. In addition, the needle used in this technique was finer and thus less painful. On the other hand, this technique was highly dependent on the operator's skill, potentially requiring the presence of an assistant for beginners.

All in all, guiding injections by anatomy palpation thus appears to be the least precise technique. The other guiding techniques appear to offer superiority, in terms of precision and thus efficacy, although further studies must be conducted in order to determine which technique achieves the best clinical results.

Another literature review (14) evaluated the impact of the different injection-guiding techniques on the efficacy of botulinum toxin when treating not only spasticity but also dystonia. This review covered 10 studies, seven of which were randomized. The authors reported a high level of evidence (Grade A) that instrument-based guiding, *i.e.,* ultrasound,

electrostimulation or electromyography, was more effective than manual guiding in the treatment of upper limb spasticity, spastic equinus following stroke in adults, and cerebral palsy in children. The review's conclusions were that no instrument-based guiding technique proved superior to another. At the present time, no recommendation can be made in terms of choosing the optimal guiding technique, although ultrasound nevertheless appears to be more effective than electrostimulation in spastic equinus treatment following stroke in adults (passive mobilization of the ankle) (11).

No study has as definitely yet successfully proven the benefits of ultrasound-guided botulinum toxin injections in terms of efficacy and patient comfort compared to other guiding techniques.

2) METHODS/DESIGN

Objective

Our main objective is to compare the efficacy of botulinum toxin injections in terms of guiding technique: ultrasound *vs.* electrical stimulation in patients with hemiplegia due to stroke.

The secondary objective is to demonstrate that ultrasound-guidance is a less painful localization technique.

Study design

This prospective, randomized, single-center, single-blind, crossover study will be conducted in chronic stroke patients (>6 months) presenting with triceps surae spasticity. Severity of the ambulation deficit was considered by using the Functional Ambulation Classification modified and stratification was made on ambulation. The patients will receive two injections; each administered using a different guiding technique. Randomization will determine which technique will be used in the first and second instances. The patients will be selected among those who attend for consultation at the Physical Medicine and Rehabilitation Department of the Clermont-Ferrand University Hospital. The botulinum toxin injections and assessments will take place in the same department. The study will last 5 months for each patient. This study does not present a major risk for the subjects. The main potential disadvantages to the treatment are injection pain or side-effects from the botulinum toxin (increased motor deficit or dysphagia).

The study design is presented in Figure 1.

Randomization

The patients will be randomly assigned to one of the above-described groups by means of a Latin square design in order to balance out the group numbers.

Study description

The patients pre-selected during consultation at the Physical Medicine and Rehabilitation Department of the Clermont-Ferrand University Hospital will be handed a

letter containing information on the study protocol. They will then have 1 month to grant their consent, should they wish, and be included at their next consultation.

The following data will then be collected for each patient: age, gender, time since stroke, side affected by the cerebral lesion, current treatments and dosages (for managing spasticity and pain), date of first botulinum toxin injection, as well as severity of deficit (functional walking scale).

The initial assessment of the patients included in the study will be performed just prior to the first injection. This evaluation will be both clinical (assessment of the triceps surae spasticity based on the Tardieu and modified Ashworth scales) and instrument-based (walking speed using GAITRite, CIR Systems Inc. Sparta, New Jersey, the USA).

The first injection will be administered in the outpatient clinic by a therapist with injection experience of over 3 years, guided using ultrasound or electrostimulation, depending on the group. We use Dantec Clavis® for electrostimulation injection and Sonosite Edge® with a 6-13 MHz probe for ultrasound injection. The clinical investigator will randomize the patients then administer the injection according to the guiding method assigned. In total, 500 units of BoNT-A (Dysport®) will be injected into four separate areas of the triceps surae to have a good reproductibility. Further injections will be administered into other muscle groups, if necessary. The total dose for this injection will be minus 1,500 BoNT-A units. Any pain experienced during the injection will be assessed by means of vertical visual analogue scales, and the time required for tracking and administering the injection will be recorded.

The second injection will be administered 4 months after the first, also in the outpatient clinic. The procedure will be identical to the first injection apart from the tracking method used on this occasion which will differ from that used for the first.

The two follow-up visits will take place 1 month after each botulinum toxin injection. Each patient will be asked to attend the clinic for consultation so as to allow the efficacy of the injection to be assessed. This assessment will be both clinical (assessment of the spasticity of the triceps surae by means of the Tardieu and modified Ashworth scales) and instrument-based (walking speed using GAITRite®). Each follow-up visit will be performed by an investigator (physiotherapist) blinded to the tracking technique. The patients are also told to hide the kind of injection they received. After each injection patients were told to continue their regular physiotherapy.

This study is actually ongoing and the investigators are currently still collecting data. The contents of the manuscript have not been submitted or published elsewhere.

Patients

The inclusion criteria are: age 18 to 80 years, hemiplegic sequelae of stroke, triceps surae spasticity evaluated >1 on the modified Ashworth scale and ability to provide written consent. The exclusion criteria are: injection of botulinum toxin dating from over 3 months, previous ultrasound-guided injection of botulinum toxin, swallowing impairment, ongoing anti-vitamin K (AVK) anticoagulation treatment with international normalized ration (INR) > 3 during one week before randomization, ongoing aminoglycoside treatment, general anesthesia with planned curare injection during study participation, implant with a pacemaker, history of ankle arthrodesis, other contra-indication for botulinum toxin injection: myasthenia gravis, pregnancy or breast feeding and patient included in other trials. The indication for botulinum toxin injection to the upper limb will not constitute a non-inclusion criterion for this study.

Evaluation

The primary endpoint is variation in passive ankle dorsiflexion range of motion at slow and high speeds (Tardieu scale) while keeping the knee straight.

The procedure consists of assessing the angle at which resistance manifests, as well as the intensity of this resistance to mobilization at slow and fast speeds (15). The ankle dorsal flexion angle will thus be measured by means of a goniometer during passive manipulation of the ankle with the knee being kept straight, before and after treatment. For the Tardieu Scale the minimally clinical important change differs according to studies (16) The effect size calculation is based on an improvement of 7 angular degrees which is quite important considering regular ankle range of motion from 0 to 50°(17). Concerning the gait analysis, an improvement of 0.2 meters/second of the gait speed is considered as a minimally clinical important change (18).

This straight-knee assessment is relevant for simultaneously obtaining measurement of gastrocnemius muscles, which are bi-articular, and the soleus spasticity.

The Tardieu scale is more sensitive than the commonly-used modified Ashworth scale. The latter only consists of five stages, which does not always allow for treatment efficacy to be evaluated. Furthermore, this scale does not take into account the velocity factor during spasticity (16). Nevertheless, validation studies pertaining to the Tardieu scale and involving the adult population are scarce in the scientific literature. Moreover the Tardieu scale is reliable for assessing spasticity in lower limb muscles of adults with chronic neurologic injuries(19).

Assessing the difference in the range of motion between slow and fast speeds is relevant because this takes into account not only the spastic component but also any potential tendon retraction.

The principal evaluation criterion will be measured on the day of injection and at the Month 1 assessment.

The secondary endpoints are:

- other components of the "Tardieu scale": quality of muscle reaction (X) at slow and fast speeds, as well as angle of apparition of the muscle reaction (Y) at slow and fast speeds;
- assessment of the triceps surae spasticity on the modified Ashworth scale;
- walking speed;
- extent of pain at the injection site using a visual analogue scale;
- duration of tracking and injection.

Statistical considerations :

To date, only one comparative study focused on the protocol's topic has been published (20). Therefore, if scientific literature data provides information on the statistical variability of ankle dorsiflexion range of motion at slow and high speeds obtained using the Tardieu scale for patients having suffered from stroke (20), exhibiting a standard deviation around 8.5°, proposing an expected difference between the two treatments (ultrasound vs. electrical stimulation) proves challenging. In addition, in order to highlight the efficacy of botulinum toxin injections in terms of guiding technique, namely ultrasound vs. electrical stimulation, sample size estimation was based on statistical power simulations in relation to recruitment capacity. To demonstrate a minimum difference of 7.12, with an effect size of 0.8, 15 patients per sequence (ultrasound stimulation then electrical vs. electrical stimulation then ultrasound) will be needed for a two-sided Type I error at 5%, a statistical power of 90%, and a correlation coefficient equal to 0, owing due to the cross-over design. For a more favorable

correlation coefficient (for example 0.5), the difference expected with 30 subjects (15 patients per sequence) will be near to 5° (effect size of 0.6).

Statistical analysis will be performed on an intention-to-treat basis using the Stata software (Version 13, StataCorp, College Station, US) for a two-sided Type I error at α=5%. The patient characteristics will be described by numbers and associated percentages for categorical data. For quantitative parameters, mean (standard-deviation) or median (interquartile range) values will be calculated and presented according to statistical distribution. The assumption of normality will be studied by Shapiro-Wilk test. The primary endpoint, namely change in the ankle dorsiflexion range of motion at slow and high speeds obtained using the Tardieu scale, will be compared between the groups using a repeated analysis of variance (anova) for cross-over designs while taking into account the following effects: treatment group (ultrasound vs. electrical stimulation), sequence, subject (as random-effect), and carry-over. A particular focus will be given to the interaction "sequence x treatment (ultrasound vs. electrical stimulation)". A sensitivity analysis will be proposed to determine the nature of missing data and to apply the more appropriate imputation approach as multiple imputation. If this test proves significant, the statistical analysis will only cover the first period of this cross-over study. The normality of residuals will be studied, as described previously. When endpoints do not assume the normality assumption, a non-parametric paired test like the Wilcoxon will be proposed. Analyses concerning the secondary endpoints (quality of muscle reaction [X] at slow and fast speeds, the angle of the muscle reaction apparition [Y] at slow and fast speeds, assessment of the triceps surae spasticity on the modified Ashworth scale, walking speed, and extent of pain at the injection site using a visual analogue scale, along with the duration of tracking and injection will be studied in a similar way as the primary endpoint. For categorical parameters, Stuart-Maxwell test for paired data or generalized linear mixed model taking into account the above-mentioned effects will be applied. Concerning non-crossover comparisons,

usual statistical tests will be performed: Student t-test or Mann-Whitney test if the conditions of t-test are not met (normality or homoscedasticity verified using Fisher-Snedecor test) for quantitative parameters and Chi-squared test or Fischer's exact test for categorical variables, if appropriate. As discussed by Feise (21), adjustment of Type I error (α) will not be proposed systematically, but on a case-by-case basis in the light of clinical considerations rather than statistical ones only.

3) DISCUSSION

The various comparative studies currently available (9), (11), (12) have demonstrated that instrument-guided procedures, such as electrostimulation and ultrasound, improve the efficacy of botulinum toxin injections compared to that obtained by means of simple anatomy palpation, in line with current recommendations for good clinical practices.

Ultrasound enables us to visualize in real-time the needle's progress, resulting in a precise localization of the target muscle, while avoiding certain structures like blood vessels and nerves. In addition, this technique allows a passive manipulation of the limb part under study in order to distinguish the muscular body of the target muscle from that of other adjacent muscular structures (11).

Ultrasound-guided botulinum toxin injection can be subject to the same limitations inherent to ultrasound itself. The technique is highly dependent on the skills of the operator, who needs to be experienced, thus requiring further investment in terms of training and equipment. Additionally, the structural evolution of spastic muscles, as fatty infiltration and, in particular, fibrous involution, alter the ultrasound features of the muscle, rendering it at times difficult to distinguish from the different adjacent muscles (22), (23).

The substantial cost of ultrasound equipment no longer appears to represent an obstacle to using this guiding technique. It is now possible to directly employ different ultrasound waves with a digital tablet, thus considerably reducing the equipment costs.

With regard to the guiding speed of the different techniques, the literature currently provides contradictory views. The Berweck team (7) demonstrated that the mean time of muscle localization and injection was only 5 seconds for superficial muscles and 30 seconds for deeper ones when using ultrasound. On the other hand, the 2010 Henzel study (22) reported an average increase of 5 to 10 minutes in procedure time when adding ultrasound-guiding to usual guiding techniques. If ultrasound-guided injection was concretely proven to be faster than other methods, this could represent a particular advantage for children and poorly compliant adults displaying low tolerance for procedures involving prolonged immobilization (11).

In this study, we hypothesize that botulinum toxin injections guided by ultrasound are more efficacious than those using electrostimulation, with the triceps surae spasticity as primary evaluation criterion. In addition, we also seek to prove that ultrasound-guided botulinum toxin injections are less painful than those administered using electrostimulation, and that the time needed for localizing and injecting is shorter for the former.

The expected benefit for the patient is thus a more efficacious injection and consequently reduced spasticity of the triceps surae. The benefits of ultrasound-guided injection compared to that of electrostimulation-guided consist of reduced tracking and injection times, in addition to reduced pain on injection.

This study's objective is to improve the techniques pertaining to guiding and injection. When injecting botulinum toxin, it is, in fact, all the more crucial to be as precise as possible in order to ensure the best efficacy in the target muscles while avoiding any unwanted-effects that could arise in relation with toxin diffusion or intravascular injection. For this reason, it is highly-desirable to use the most reliable guiding method possible. Furthermore, toxin injection can be a painful procedure, particularly for certain patients suffering from hyperesthesia or cognitive disorders, meaning a guiding technique that enables the highest tolerance is all the more crucial.

4) FOOTNOTES

Authors' contributors

BP: data analysis, critical revision, and final approval of the manuscript. CM: manuscript writing, critical revision, and final approval of the manuscript. EC: conception and design, analysis and interpretation, manuscript writing, critical revision, and final approval of the manuscript. HI: conception and design, data collection, analysis and interpretation, manuscript writing, critical revision, and final approval of the manuscript. NA: study setup, critical revision, and approval of the final manuscript. AB: english reviewing and critical revision.

Funding statement:

This work was supported by CHU Clermont Ferrand and IPSEN provided funding for the ultrasound.

Competing interest:

None

Ethics approval

The study protocol, patient information, and patient consent form, along with the case report form, have been submitted to the ethics committee of the Rhône-Alpes region (*Comité de Protection des Personnes Rhône-Alpes*, *CPP Sud-Est I*). The CPP's favorable assessment was transmitted to the study sponsor and ANSM (*l'Agence nationale de sécurité des medicaments et des produits de santé* – the French national agency of medicine and health products safety) on November 29, 2012.

5) ABBREVIATIONS

ANSM: *Agence nationale de sécurité du médicament et des produits de santé* - the French national agency of medicine and health products safety

CPP: *Comité de protection des personnes* – ethics committee

EMG: Electromyogram

PM+R: Physical medecine and rehabilitation

6) AUTHORS' INFORMATION

1 Service de Médecine Physique et de Réadaptation; CHU Clermont-Ferrand, 58 rue Montalembert, 63 000 Clermont-Ferrand, France

2 Université Clermont Auvergne, Clermont-Ferrand, France

3 Centre médical Etienne Clémentel, 63530 Enval, France

4 Biostatistics Unit, Délégation Recherche Clinique & Innovation (DRCI), CHU Clermont-Ferrand, 63 000 Clermont-Ferrand F-63003;

5 INRA, Unité de Nutrition Humaine (UNH, UMR 1019), CRNH Auvergne, Clermont-Ferrand, France

7) REFERENCES

1. Yelnik AP, Simon O, Bensmail D, Chaleat-Valayer E, Decq P, Dehail P, et al. Drug treatments for spasticity. Ann Phys Rehabil Med. déc 2009;52(10):746‑56.

2. Chin TYP, Nattrass GR, Selber P, Graham HK. Accuracy of intramuscular injection of botulinum toxin A in juvenile cerebral palsy: a comparison between manual needle placement and placement guided by electrical stimulation. J Pediatr Orthop. juin 2005;25(3):286‑91.

3. Schroeder AS, Berweck S, Lee SH, Heinen F. Botulinum toxin treatment of children with cerebral palsy - a short review of different injection techniques. Neurotox Res. avr 2006;9(2‑3):189‑96.

4. Picelli A, Bonetti P, Fontana C, Barausse M, Dambruoso F, Gajofatto F, et al. Accuracy of botulinum toxin type A injection into the gastrocnemius muscle of adults with spastic equinus: manual needle placement and electrical stimulation guidance compared using ultrasonography. J Rehabil Med. mai 2012;44(5):450‑2.

5. Adler RS, Sofka CM. Percutaneous ultrasound-guided injections in the musculoskeletal system. Ultrasound Q. mars 2003;19(1):3‑12.

6. Walker KJ, McGrattan K, Aas-Eng K, Smith AF. Ultrasound guidance for peripheral nerve blockade. Cochrane Database Syst Rev. 2009;(4):CD006459.

7. Berweck S, Schroeder AS, Fietzek UM, Heinen F. Sonography-guided injection of botulinum toxin in children with cerebral palsy. Lancet. 17 janv 2004;363(9404):249‑50.

8. Berweck S, Heinen F. Use of botulinum toxin in pediatric spasticity (cerebral palsy). Mov Disord Off J Mov Disord Soc. mars 2004;19 Suppl 8:S162-167.

9. Kwon J-Y, Hwang JH, Kim J-S. Botulinum toxin a injection into calf muscles for treatment of spastic equinus in cerebral palsy: a controlled trial comparing sonography and electric stimulation-guided injection techniques: a preliminary report. Am J Phys Med Rehabil Assoc Acad Physiatr. avr 2010;89(4):279‑86.

10. Gracies J-M, Burke K, Clegg NJ, Browne R, Rushing C, Fehlings D, et al. Reliability of the Tardieu Scale for assessing spasticity in children with cerebral palsy. Arch Phys Med Rehabil. mars 2010;91(3):421‑8.

11. Picelli A, Tamburin S, Bonetti P, Fontana C, Barausse M, Dambruoso F, et al. Botulinum toxin type A injection into the gastrocnemius muscle for spastic equinus in adults with stroke: a randomized controlled trial comparing manual needle placement, electrical stimulation and ultrasonography-guided injection techniques. Am J Phys Med Rehabil Assoc Acad Physiatr. nov 2012;91(11):957‑64.

12. Picelli A, Lobba D, Midiri A, Prandi P, Melotti C, Baldessarelli S, et al. Botulinum toxin injection into the forearm muscles for wrist and fingers spastic overactivity in adults with chronic stroke: a randomized controlled trial comparing three injection techniques. Clin Rehabil. mars 2014;28(3):232‑42.

13. Walker HW, Lee MY, Bahroo LB, Hedera P, Charles D. Botulinum Toxin Injection Techniques for the Management of Adult Spasticity. PM R. avr 2015;7(4):417‑27.

14. Grigoriu A-I, Dinomais M, Rémy-Néris O, Brochard S. Impact of Injection-Guiding Techniques on the Effectiveness of Botulinum Toxin for the Treatment of Focal Spasticity and Dystonia: A Systematic Review. Arch Phys Med Rehabil. 14 mai 2015;

15. Ben Smaïl D, Kiefer C, Bussel B. [Clinical evaluation of spasticity]. Neurochirurgie. mai 2003;49(2‑3 Pt 2):190‑8.

16. Haugh AB, Pandyan AD, Johnson GR. A systematic review of the Tardieu Scale for the measurement of spasticity. Disabil Rehabil. 15 août 2006;28(15):899‑907.

17. Singh P, Joshua AM, Ganeshan S, Suresh S. Intra-rater reliability of the modified Tardieu scale to quantify spasticity in elbow flexors and ankle plantar flexors in adult stroke subjects. Ann Indian Acad Neurol. 2011;14(1):23‑6.

18. Lewek MD, Randall EP. Reliability of spatiotemporal asymmetry during overground walking for individuals following chronic stroke. J Neurol Phys Ther JNPT. sept 2011;35(3):116‑21.

19. Ben-Shabat E, Palit M, Fini NA, Brooks CT, Winter A, Holland AE. Intra- and interrater reliability of the Modified Tardieu Scale for the assessment of lower limb spasticity in adults with neurologic injuries. Arch Phys Med Rehabil. déc 2013;94(12):2494‑501.

20. Singh P, Joshua AM, Ganeshan S, Suresh S. Intra-rater reliability of the modified Tardieu scale to quantify spasticity in elbow flexors and ankle plantar flexors in adult stroke subjects. Ann Indian Acad Neurol. janv 2011;14(1):23‑6.

21. Feise RJ. Do multiple outcome measures require p-value adjustment? BMC Med Res Methodol. 17 juin 2002;2:8.

22. Henzel MK, Munin MC, Niyonkuru C, Skidmore ER, Weber DJ, Zafonte RD. Comparison of surface and ultrasound localization to identify forearm flexor muscles for botulinum toxin injections. PM R. juill 2010;2(7):642‑6.

23. Picelli A, Bonetti P, Fontana C, Barausse M, Dambruoso F, Gajofatto F, et al. Is spastic muscle echo intensity related to the response to botulinum toxin type A in patients with stroke? A cohort study. Arch Phys Med Rehabil. juill 2012;93(7):1253‑8.

568

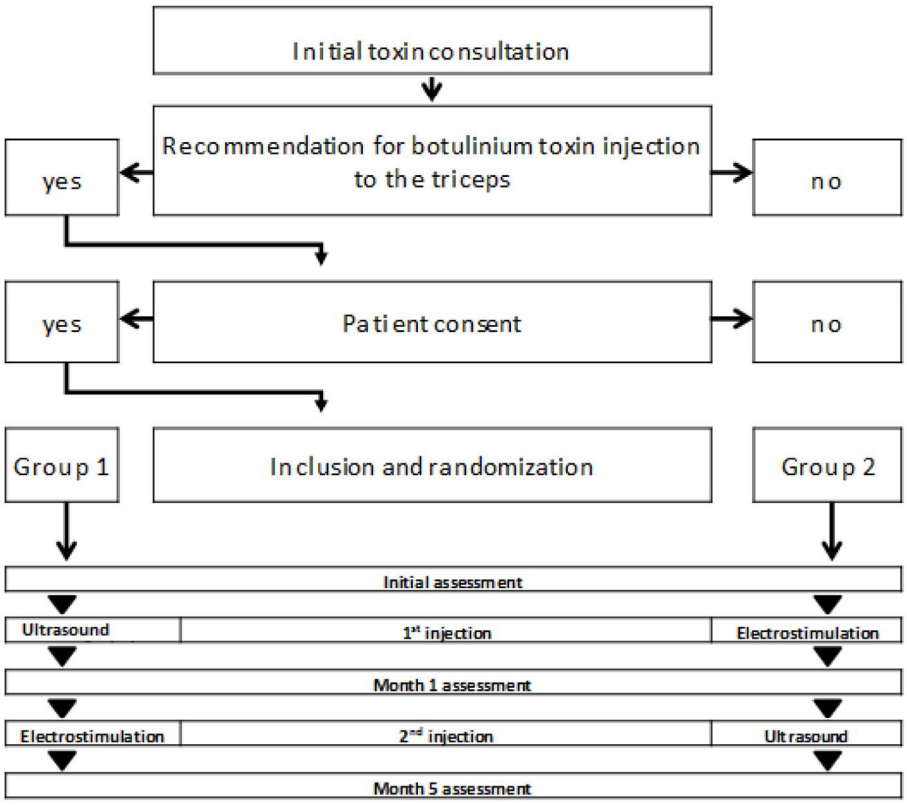

Figure 1: Flow diagram showing the different stages of the protocol

154x127mm (300 x 300 DPI)

APPENDIX 1:

Modified Ashworth Scale

Modified Ashworth Scale for Grading Spasticity	
Grade	**Description**
0	No increase in muscle tone
1	Slight increase in muscle tone, manifested by a catch and release or by minimal resistance at the end of the ROM when the affected part(s) is moved in flexion or extension
1+	Slight increase in muscle tone, manifested by a catch, followed by minimal resistance throughout the remainder (less than half) of the ROM
2	More marked increase in muscle tone through most of the ROM, but affected part(s) easily moved
3	Considerable increase in muscle tone, passive movement difficult
4	Affected part(s) rigid in flexion or extension

APPENDIX 2:

Tardieu Scale: Principles

Grading always performed:

- On a muscle at rest before the stretch maneuver
- At a reproducible velocity of stretch. Once the fast velocity is selected for a muscle, it remains the same for all subsequent tests.
- At the same time of the day
- In a constant body position for a given limb
- Other joints, particularly the neck, must also remain in a constant position throughout the assessment and for all other assessments.

Velocity of Stretch

- SLOW = V1: As slow as possible (slower than the rate of natural drop of the limb segment under gravity)
- FAST = Either V2 or V3
  - V2: Speed of the limb segment falling under gravity
  - V3: As fast as possible (faster than the rate of natural drop of the limb segment under gravity)

Tardieu Scale: Grading

X = Spasticity Angle (Threshold)

Angle of arrest at slow speed X_{V1} *minus* Angle of catch at fast speed X_{V3}

Y = Spasticity Grade (Gain)

0. No resistance throughout passive movement
1. Slight resistance throughout passive movement
2. Clear catch at precise angle, interrupting passive movement, followed by release
3. Fatigable clonus (<10s when maintaining pressure) occurring at a precise angle, followed by release
4. Unfatigable clonus (>10s when maintaining pressure) occurring at a precise angle
- Catch without release: graded 0 if $X_{V1}=X_{V3}$; "unratable" spasticity otherwise
- Catch with "minimal" release: graded 2 if X_{V3} is consistent and consistently less than X_{V1}
- Angle 0° = position of minimal stretch of the tested muscle
- For grades 0 and 1, spasticity angle X=0° by definition

19

20

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	3
	2b	All items from the World Health Organization Trial Registration Data Set	X
Protocol version	3	Date and version identifier	X
Funding	4	Sources and types of financial, material, and other support	20
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	21
	5b	Name and contact information for the trial sponsor	21
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	20
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	NA

Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	5
	6b	Explanation for choice of comparators	X
Objectives	7	Specific objectives or hypotheses	9
Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)	11
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	9
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	13
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	10
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	X
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	X
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	X
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	14
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	10

Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	15
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	12
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	15
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	X
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	11
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	12
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	X
Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	X
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	X

Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	X
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	15
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	15
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	15
Methods: Monitoring			
Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	X
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	X
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	X
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	X
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	20
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	X

Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	20
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	NA
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	X
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	20
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	X
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	X
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	X
	31b	Authorship eligibility guidelines and any intended use of professional writers	X
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	X
Appendices			
Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	X
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	NA

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.

BMJ Open

Efficacy of two injection-site localization techniques for botulinum toxin injections: a single-blind, crossover randomized trial protocol among adults with hemiplegia due to stroke.

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2016-011751.R2
Article Type:	Protocol
Date Submitted by the Author:	02-Sep-2016
Complete List of Authors:	MOREL, Claire; CHU Gabriel Montpied, Centre Hospitalier Universitaire de Clermont Ferrand, service Médecine Physique et de Réadaptation HAURET, Isabelle; Centre Médical Etienne Clementel ANDANT, Nicolas; Biostatistics Unit, Délégation Recherche Clinique & Innovation, CHU Clermont Ferrand Pereira, Bruno; University Hospital CHU Clermont-Ferrand, Bonnin, Armand; Centre Hospitalier Universitaire de Clermont-Ferrand, Médecine Physique et de Réadaptation Coudeyre, Emmanuel; Centre Hospitalier Universitaire de Clermont-Ferrand, Médecine Physique et de Réadaptation
Primary Subject Heading:	Rehabilitation medicine
Secondary Subject Heading:	Neurology
Keywords:	echography, electrostimulation, soleus, gastrocnemius

Efficacy of two injection-site localization techniques for botulinum toxin injections: a single-blind, crossover randomized trial protocol among adults with hemiplegia due to stroke.

Claire Morel[1,2], Isabelle Hauret[3], Nicolas Andant[4], Armand Bonnin[1,2], Bruno Pereira[4] and Emmanuel Coudeyre[1,2,5]

1 Service de Médecine Physique et de Réadaptation; CHU Clermont-Ferrand, 58 rue Montalembert, 63 000 Clermont-Ferrand, France

2 Université Clermont Auvergne, Clermont-Ferrand, France

3 Centre médical Etienne Clémentel, 63530 Enval, France

4 Biostatistics Unit, Délégation Recherche Clinique & Innovation (DRCI), CHU Clermont-Ferrand, 63 000 Clermont-Ferrand F-63003;

5 INRA, Unité de Nutrition Humaine (UNH, UMR 1019), CRNH Auvergne, Clermont-Ferrand, France

Corresponding author:

Emmanuel COUDEYRE, Service de Médecine Physique et Réadaptation, CHU Clermont-Ferrand, Hôpital Nord,

Route de Chateaugay, F-63118 Cébazat, France

ecoudeyre@chu-clermontferrand.fr

Key words: echography, electrostimulation, soleus, gastrocnemius

Word count: 4002 words

ABSTRACT

Introduction

Botulinum toxin injections are an effective treatment for limb spasticity following stroke. Different tracking techniques are used for this purpose: palpation, electrostimulation, electromyography and ultrasound. Yet very few studies have compared these different techniques, and none has successfully proven the superior efficacy of ultrasound-guided injections compared to another tracking method. The primary objective of our study was therefore to compare the efficacy of botulinum toxin injections depending on the tracking technique used: ultrasound *versus* electrostimulation.

Methods and analysis

This is a clinical, single-center, prospective, interventional, single-blind, crossover randomized trial. In total, 30 patients aged between 18 and 80 years old presenting with triceps surae spasticity (evaluated >1 on the modified Ashworth scale) associated with hemiplegia sequelae due to stroke will be included. The patients will be selected among those who attend for consultation the Physical Medicine and Rehabilitation Department of the Clermont-Ferrand University Hospital. One group will receive the abobotulinumtoxinA (BoNT-A) injection guided by electrostimulation then ultrasound, the second group's botulinum toxin injections will be guided by ultrasound then electrostimulation. For each patient, the duration of study participation is 5 months. The primary endpoint is variation in passive ankle dorsiflexion range of motion at slow and high speeds (Tardieu scale) with the knee straight.

Ethics and dissemination

This study has ethical approval form the CPP of Rhônes-Alpes region. Results will be published in a peer-reviewed journal.

Trial registration

ClinicalTrials.gov Identifier, NCT01935544

Strengths and limitations of this study:

The management of muscle spasticity proves to be a major challenge in hemiplegia following a stroke, with botulinum toxin injections constituting the first-line treatment for local or loco-regional spasticity.

Concerning the limitations, it is a prospective study with inherent risks due to this type of studies such as lost to follow up bias.

1) INTRODUCTION

The management of muscle spasticity is a major challenge in hemiplegia following a stroke, with botulinum toxin injections constituting the first-line treatment for local or loco-regional spasticity (1).

Yet there is a range of techniques involving different methods for both injection and tracking. The most commonly-used tracking techniques are, anatomy palpation, electrostimulation, electromyography, and, more recently, ultrasound. Palpating children to guide injection is not reliable, particularly when the deeper muscles are concerned (*e.g.,* only 12% successfully-positioned injections in the tibialis posterior and flexor carpi ulnaris), but also for more superficial muscles (22% failure rate for the gastrocnemius) (2). For children, injection guided *via* electromyogram (EMG) is not always appropriate, when there is difficulty obtaining active or passive muscle activation, to differentiate muscular activation of a specific muscle from surrounding muscles (3). In addition, with this technique, there is no correlation between the extent of spasticity and muscular activity (3). One article (4) shows that neither manual needle placement nor electrical stimulation is wholly accurate to inject gastrocnemius muscle of adults with spasticity.

Tracking *via* ultrasound is widely used in other indications, such as infiltrations in the locomotor system (particularly the tendons and joints) (5) or anesthetic nerve blocks (6). The primary advantages of ultrasound-guided botulinum toxin injection are that tracking is painless (3), fast (7), more precise (3), and thus safer, avoiding complications associated with subcutaneous, intravascular, or too-deep injections (8).

A comparative study has assessed the efficacy of ultrasound-based tracking with an electrostimulation-based technique (9). The authors evaluated 32 children presenting with cerebral palsy sequelae, who were divided into two groups. All received botulinum toxin injection into the gastrocnemius, which was guided by either ultrasound or electrostimulation depending on the group. The techniques were evaluated based on three different scales: the Ashworth (Appendix 1), Tardieu (Appendix 2) (10), selective motor control (SMC), and Physician Rating scales. The authors observed a non-significant improvement in spasticity, assessed by the Ashworth and Tardieu scales, at 3 months post-injection, in the group treated with ultrasound-guided injections. In contrast, the electrostimulation-guided group showed non-significant improvement in motor control of the antagonistic muscles. The only significant differences revealed were improvements in walking pattern and foot-to-ground contact in the ultrasound-guided group. Nonetheless, the numerous controversial methodological choices made by the authors limited the relevance of these results.

There have also been two comparative studies evaluating the efficacy of ultrasound-guiding with that of techniques using electrostimulation or anatomy palpation (11) (12).

The first (11), conducted in 2012, compared these three injection-guiding techniques in the lower limbs. The trial involved 49 patients presenting with lower-limb spasticity following stroke, who were randomized into three groups, the first receiving injections guided by anatomy palpation, the second by electrostimulation, and the last by ultrasound. All received a botulinum toxin injection into the gastrocnemius, administered by the same physician. The investigator, who was blinded to the injection type, evaluated each patient on inclusion and at 1 month. The patients were forbidden from undergoing any form of physical therapy within the 3 months preceding the study and during its entirety. Ashworth and Tardieu scale results were assessed for all, along with passive dorsiflexion of the foot. The authors reported significantly improved passive dorsiflexion of the foot in the ultrasound-

guided injection group compared to the electrostimulation-guided group. Moreover, the Ashworth scale results were significantly improved 1 month following botulinum toxin injection in the ultrasound-guided group compared to the group where anatomy palpation was used.

The second study (12) was conducted in 2013 and assessed upper-limb spasticity in 60 patients who had suffered from strokes. As in the above-described study, these patients were randomized into three groups of 20 each in order to compare the three injection-guiding techniques: ultrasound, electrostimulation, and anatomy palpation. Two injections were administered in at least two of the following muscles: flexor carpi ulnaris, flexor carpi radialis, flexor digitorum superficialis, and the flexor digitorum profundus. The same physician, experienced with using botulinum toxin under ultrasound-guiding, administered all the injections. An investigator who was blinded to the injection type assessed each patient at the beginning and 4 weeks into the study. The patients were forbidden from undergoing any type of physical therapy in the 3 months preceding the study and during its entirety. The Ashworth and Tardieu scale results were assessed, along with passive dorsiflexion of the wrist and fingers. One month following injection, the modified Ashworth scale scores significantly improved in the group having undergone ultrasound-guided injection compared to the group tracked using anatomy palpation, as did the Tardieu scale scores and passive mobilizations. In contrast, the authors found no significant differences between ultrasound- and electrostimulation-guiding for the different evaluations.

In both of these studies, the authors described limitations consisting of the absence of functional evaluation of the upper or lower limbs, owing to the short follow-up rendering this assessment difficult to implement, as well as of the injections being administered by only one physician experienced with ultrasound-guided injection. The authors also indicated that body

mass index (BMI) was not taken into account in their studies, despite obesity potentially constituting a limitation to the accurate assessment of anatomical landmarks.

In a literature review (13), all four guiding techniques (anatomy palpation, electromyography [EMG], electrostimulation, and ultrasound) were compared, with advantages and disadvantages outlined for each. The authors retrieved and analyzed 15 articles, concluding that injection guided by anatomy palpation required no equipment and only a small-sized needle. Yet deep or slighter muscles were more difficult to access. In addition, while EMG enabled the toxin to be injected closest to the motor end-plate, this technique could, however, not guarantee that the needle was actually in the target muscle. As for the electrostimulation-guided technique, its primary advantage appeared to be its precise localization capacity. Despite this, it can take a long time to perform and require more training than the EMG and anatomy palpation techniques. Finally, ultrasound was found to enable the real-time visualization of the needle's progression while avoiding certain structures like blood vessels or nerves, among other advantages. In addition, the needle used in this technique was finer and thus less painful. On the other hand, this technique was highly dependent on the operator's skill, potentially requiring the presence of an assistant for beginners.

All in all, guiding injections by anatomy palpation thus appears to be the least precise technique. The other guiding techniques appear to offer superiority, in terms of precision and thus efficacy, although further studies must be conducted in order to determine which technique achieves the best clinical results.

Another literature review (14) evaluated the impact of the different injection-guiding techniques on the efficacy of botulinum toxin when treating not only spasticity but also dystonia. This review covered 10 studies, seven of which were randomized. The authors reported a high level of evidence (Grade A) that instrument-based guiding, *i.e.,* ultrasound,

electrostimulation or electromyography, was more effective than manual guiding in the treatment of upper limb spasticity, spastic equinus following stroke in adults, and cerebral palsy in children. The review's conclusions were that no instrument-based guiding technique proved superior to another. At the present time, no recommendation can be made in terms of choosing the optimal guiding technique, although ultrasound nevertheless appears to be more effective than electrostimulation in spastic equinus treatment following stroke in adults (passive mobilization of the ankle) (11).

No study has as definitely yet successfully proven the benefits of ultrasound-guided botulinum toxin injections in terms of efficacy and patient comfort compared to other guiding techniques.

2) METHODS/DESIGN

Objective

Our main objective is to compare the efficacy of botulinum toxin injections in terms of guiding technique: ultrasound *vs.* electrical stimulation in patients with hemiplegia due to stroke.

The secondary objective is to demonstrate that ultrasound-guidance is a less painful localization technique.

Study design

This prospective, randomized, single-center, single-blind, crossover study will be conducted in chronic stroke patients (>6 months) presenting with triceps surae spasticity. Severity of the ambulation deficit was considered by using the Functional Ambulation Classification modified and stratification was made on ambulation. The patients will receive two injections; each administered using a different guiding technique. Randomization will determine which technique will be used in the first and second instances. The patients will be selected among those who attend for consultation at the Physical Medicine and Rehabilitation Department of the Clermont-Ferrand University Hospital. The botulinum toxin injections and assessments will take place in the same department. The study will last 5 months for each patient. This study does not present a major risk for the subjects. The main potential disadvantages to the treatment are injection pain or side-effects from the botulinum toxin (increased motor deficit or dysphagia).

The study design is presented in Figure 1.

Randomization

The patients will be randomly assigned to one of the above-described groups by means of a Latin square design in order to balance out the group numbers.

Study description

The patients pre-selected during consultation at the Physical Medicine and Rehabilitation Department of the Clermont-Ferrand University Hospital will be handed a

letter containing information on the study protocol. They will then have 1 month to grant their consent, should they wish, and be included at their next consultation.

The following data will then be collected for each patient: age, gender, time since stroke, side affected by the cerebral lesion, current treatments and dosages (for managing spasticity and pain), date of first botulinum toxin injection, as well as severity of deficit (functional walking scale).

The initial assessment of the patients included in the study will be performed just prior to the first injection. This evaluation will be both clinical (assessment of the triceps surae spasticity based on the Tardieu and modified Ashworth scales) and instrument-based (walking speed using GAITRite, CIR Systems Inc. Sparta, New Jersey, the USA).

The first injection will be administered in the outpatient clinic by a therapist with injection experience of over 3 years, guided using ultrasound or electrostimulation, depending on the group. We use Dantec Clavis® for electrostimulation injection and Sonosite Edge® with a 6-13 MHz probe for ultrasound injection. The clinical investigator will randomize the patients then administer the injection according to the guiding method assigned. In total, 500 units of BoNT-A (Dysport®) will be injected into four separate areas of the triceps surae to have a good reproductibility. Further injections will be administered into other muscle groups, if necessary. The total dose for this injection will be minus 1,500 BoNT-A units. Any pain experienced during the injection will be assessed by means of vertical visual analogue scales, and the time required for tracking and administering the injection will be recorded.

The second injection will be administered 4 months after the first, also in the outpatient clinic. The procedure will be identical to the first injection apart from the tracking method used on this occasion which will differ from that used for the first.

The two follow-up visits will take place 1 month after each botulinum toxin injection. Each patient will be asked to attend the clinic for consultation so as to allow the efficacy of the injection to be assessed. This assessment will be both clinical (assessment of the spasticity of the triceps surae by means of the Tardieu and modified Ashworth scales) and instrument-based (walking speed using GAITRite®). Each follow-up visit will be performed by an investigator (physiotherapist) blinded to the tracking technique. The patients are also told to hide the kind of injection they received. After each injection patients were told to continue their regular physiotherapy.

This study is actually ongoing and the investigators are currently still collecting data. The contents of the manuscript have not been submitted or published elsewhere.

Patients

The inclusion criteria are: age 18 to 80 years, hemiplegic sequelae of stroke, triceps surae spasticity evaluated >1 on the modified Ashworth scale and ability to provide written consent. The exclusion criteria are: injection of botulinum toxin dating from over 3 months, previous ultrasound-guided injection of botulinum toxin, swallowing impairment, ongoing anti-vitamin K (AVK) anticoagulation treatment with international normalized ration (INR) > 3 during one week before randomization, ongoing aminoglycoside treatment, general anesthesia with planned curare injection during study participation, implant with a pacemaker, history of ankle arthrodesis, other contra-indication for botulinum toxin injection: myasthenia gravis, pregnancy or breast feeding and patient included in other trials. The indication for botulinum toxin injection to the upper limb will not constitute a non-inclusion criterion for this study.

Evaluation

The primary endpoint is variation in passive ankle dorsiflexion range of motion at slow and high speeds (Tardieu scale) while keeping the knee straight.

The procedure consists of assessing the angle at which resistance manifests, as well as the intensity of this resistance to mobilization at slow and fast speeds (15). The ankle dorsal flexion angle will thus be measured by means of a goniometer during passive manipulation of the ankle with the knee being kept straight, before and after treatment. For the Tardieu Scale the minimally clinical important change differs according to studies (16) The effect size calculation is based on an improvement of 7 angular degrees which is quite important considering regular ankle range of motion from 0 to 50°(17). Concerning the gait analysis, an improvement of 0.2 meters/second of the gait speed is considered as a minimally clinical important change (18).

This straight-knee assessment is relevant for simultaneously obtaining measurement of gastrocnemius muscles, which are bi-articular, and the soleus spasticity.

The Tardieu scale is more sensitive than the commonly-used modified Ashworth scale. The latter only consists of five stages, which does not always allow for treatment efficacy to be evaluated. Furthermore, this scale does not take into account the velocity factor during spasticity (16). Nevertheless, validation studies pertaining to the Tardieu scale and involving the adult population are scarce in the scientific literature. Moreover the Tardieu scale is reliable for assessing spasticity in lower limb muscles of adults with chronic neurologic injuries(19).

Assessing the difference in the range of motion between slow and fast speeds is relevant because this takes into account not only the spastic component but also any potential tendon retraction.

The principal evaluation criterion will be measured on the day of injection and at the Month 1 assessment.

The secondary endpoints are:

- other components of the "Tardieu scale": quality of muscle reaction (X) at slow and fast speeds, as well as angle of apparition of the muscle reaction (Y) at slow and fast speeds;
- assessment of the triceps surae spasticity on the modified Ashworth scale;
- walking speed;
- extent of pain at the injection site using a visual analogue scale;
- duration of tracking and injection.

Statistical considerations :

To date, only one comparative study focused on the protocol's topic has been published (20). Therefore, if scientific literature data provides information on the statistical variability of ankle dorsiflexion range of motion at slow and high speeds obtained using the Tardieu scale for patients having suffered from stroke (20), exhibiting a standard deviation around 8.5°, proposing an expected difference between the two treatments (ultrasound vs. electrical stimulation) proves challenging. In addition, in order to highlight the efficacy of botulinum toxin injections in terms of guiding technique, namely ultrasound vs. electrical stimulation, sample size estimation was based on statistical power simulations in relation to recruitment capacity. To demonstrate a minimum difference of 7.12 between ultrasound and electrical stimulations, with an effect size of 0.8 (so, an expected standard deviation of difference at 8.9°), 15 patients per sequence (ultrasound stimulation then electrical vs. electrical stimulation then ultrasound) will be needed for a two-sided Type I error at 5%, a statistical power of 80%,

an intra-individual correlation coefficient equals 0.5 (owing to the crossover design) and no carryover effect assumed.

Statistical analysis will be performed on an intention-to-treat basis using the Stata software (Version 13, StataCorp, College Station, US) for a two-sided Type I error at α=5%. The patient characteristics will be described by numbers and associated percentages for categorical data. For quantitative parameters, mean (standard-deviation) or median (interquartile range) values will be calculated and presented according to statistical distribution. The assumption of normality will be studied by Shapiro-Wilk test. The primary endpoint, namely change in the ankle dorsiflexion range of motion at slow and high speeds obtained using the Tardieu scale, will be compared between the groups using a repeated analysis of variance (anova) for crossover designs while taking into account the following effects: treatment group (ultrasound vs. electrical stimulation), sequence, subject (as random-effect), and carry-over. A particular focus will be given to the interaction "sequence x treatment (ultrasound vs. electrical stimulation)". A sensitivity analysis will be proposed to determine the nature of missing data and to apply the more appropriate imputation approach as multiple imputation. If this test proves significant, the statistical analysis will only cover the first period of this crossover study. The normality of residuals will be studied, as described previously. When endpoints do not assume the normality assumption, a non-parametric paired test like the Wilcoxon will be proposed. Analyses concerning the secondary endpoints (quality of muscle reaction [X] at slow and fast speeds, the angle of the muscle reaction apparition [Y] at slow and fast speeds, assessment of the triceps surae spasticity on the modified Ashworth scale, walking speed, and extent of pain at the injection site using a visual analogue scale, along with the duration of tracking and injection will be studied in a similar way as the primary endpoint. For categorical parameters, Stuart-Maxwell test for paired data or generalized linear mixed model taking into account the above-mentioned effects will be applied. Concerning non-crossover comparisons,

usual statistical tests will be performed: Student t-test or Mann-Whitney test if the conditions of t-test are not met (normality or homoscedasticity verified using Fisher-Snedecor test) for quantitative parameters and Chi-squared test or Fisher's exact test for categorical variables, if appropriate. As discussed by Feise (21), adjustment of Type I error (α) will not be proposed systematically, but on a case-by-case basis in the light of clinical considerations rather than statistical ones only.

3) DISCUSSION

The various comparative studies currently available (9), (11), (12) have demonstrated that instrument-guided procedures, such as electrostimulation and ultrasound, improve the efficacy of botulinum toxin injections compared to that obtained by means of simple anatomy palpation, in line with current recommendations for good clinical practices.

Ultrasound enables us to visualize in real-time the needle's progress, resulting in a precise localization of the target muscle, while avoiding certain structures like blood vessels and nerves. In addition, this technique allows a passive manipulation of the limb part under study in order to distinguish the muscular body of the target muscle from that of other adjacent muscular structures (11).

Ultrasound-guided botulinum toxin injection can be subject to the same limitations inherent to ultrasound itself. The technique is highly dependent on the skills of the operator, who needs to be experienced, thus requiring further investment in terms of training and equipment. Additionally, the structural evolution of spastic muscles, as fatty infiltration and, in particular, fibrous involution, alter the ultrasound features of the muscle, rendering it at times difficult to distinguish from the different adjacent muscles (22), (23).

The substantial cost of ultrasound equipment no longer appears to represent an obstacle to using this guiding technique. It is now possible to directly employ different ultrasound waves with a digital tablet, thus considerably reducing the equipment costs.

With regard to the guiding speed of the different techniques, the literature currently provides contradictory views. The Berweck team (7) demonstrated that the mean time of muscle localization and injection was only 5 seconds for superficial muscles and 30 seconds for deeper ones when using ultrasound. On the other hand, the 2010 Henzel study (22) reported an average increase of 5 to 10 minutes in procedure time when adding ultrasound-guiding to usual guiding techniques. If ultrasound-guided injection was concretely proven to be faster than other methods, this could represent a particular advantage for children and poorly compliant adults displaying low tolerance for procedures involving prolonged immobilization (11).

In this study, we hypothesize that botulinum toxin injections guided by ultrasound are more efficacious than those using electrostimulation, with the triceps surae spasticity as primary evaluation criterion. In addition, we also seek to prove that ultrasound-guided botulinum toxin injections are less painful than those administered using electrostimulation, and that the time needed for localizing and injecting is shorter for the former.

The expected benefit for the patient is thus a more efficacious injection and consequently reduced spasticity of the triceps surae. The benefits of ultrasound-guided injection compared to that of electrostimulation-guided consist of reduced tracking and injection times, in addition to reduced pain on injection.

This study's objective is to improve the techniques pertaining to guiding and injection. When injecting botulinum toxin, it is, in fact, all the more crucial to be as precise as possible in order to ensure the best efficacy in the target muscles while avoiding any unwanted-effects that could arise in relation with toxin diffusion or intravascular injection. For this reason, it is highly-desirable to use the most reliable guiding method possible. Furthermore, toxin injection can be a painful procedure, particularly for certain patients suffering from hyperesthesia or cognitive disorders, meaning a guiding technique that enables the highest tolerance is all the more crucial.

4) FOOTNOTES

Authors' contributors

BP: data analysis, critical revision, and final approval of the manuscript. CM: manuscript writing, critical revision, and final approval of the manuscript. EC: conception and design, analysis and interpretation, manuscript writing, critical revision, and final approval of the manuscript. HI: conception and design, data collection, analysis and interpretation, manuscript writing, critical revision, and final approval of the manuscript. NA: study setup, critical revision, and approval of the final manuscript. AB: english reviewing and critical revision.

Funding statement:

This work was supported by CHU Clermont Ferrand and IPSEN provided funding for the ultrasound.

Competing interest:

None

Ethics approval

The study protocol, patient information, and patient consent form, along with the case report form, have been submitted to the ethics committee of the Rhône-Alpes region (*Comité de Protection des Personnes Rhône-Alpes*, *CPP Sud-Est I*). The CPP's favorable assessment was transmitted to the study sponsor and ANSM (*l'Agence nationale de sécurité des medicaments et des produits de santé* – the French national agency of medicine and health products safety) on November 29, 2012.

5) ABBREVIATIONS

ANSM: *Agence nationale de sécurité du médicament et des produits de santé* - the French national agency of medicine and health products safety

CPP: *Comité de protection des personnes* – ethics committee

EMG: Electromyogram

PM+R: Physical medecine and rehabilitation

6) AUTHORS' INFORMATION

1 Service de Médecine Physique et de Réadaptation; CHU Clermont-Ferrand, 58 rue Montalembert, 63 000 Clermont-Ferrand, France

2 Université Clermont Auvergne, Clermont-Ferrand, France

3 Centre médical Etienne Clémentel, 63530 Enval, France

4 Biostatistics Unit, Délégation Recherche Clinique & Innovation (DRCI), CHU Clermont-Ferrand, 63 000 Clermont-Ferrand F-63003;

5 INRA, Unité de Nutrition Humaine (UNH, UMR 1019), CRNH Auvergne, Clermont-Ferrand, France

7) REFERENCES

1. Yelnik AP, Simon O, Bensmail D, Chaleat-Valayer E, Decq P, Dehail P, et al. Drug treatments for spasticity. Ann Phys Rehabil Med. déc 2009;52(10):746‑56.

2. Chin TYP, Nattrass GR, Selber P, Graham HK. Accuracy of intramuscular injection of botulinum toxin A in juvenile cerebral palsy: a comparison between manual needle placement and placement guided by electrical stimulation. J Pediatr Orthop. juin 2005;25(3):286‑91.

3. Schroeder AS, Berweck S, Lee SH, Heinen F. Botulinum toxin treatment of children with cerebral palsy - a short review of different injection techniques. Neurotox Res. avr 2006;9(2‑3):189‑96.

4. Picelli A, Bonetti P, Fontana C, Barausse M, Dambruoso F, Gajofatto F, et al. Accuracy of botulinum toxin type A injection into the gastrocnemius muscle of adults with spastic equinus: manual needle placement and electrical stimulation guidance compared using ultrasonography. J Rehabil Med. mai 2012;44(5):450‑2.

5. Adler RS, Sofka CM. Percutaneous ultrasound-guided injections in the musculoskeletal system. Ultrasound Q. mars 2003;19(1):3‑12.

6. Walker KJ, McGrattan K, Aas-Eng K, Smith AF. Ultrasound guidance for peripheral nerve blockade. Cochrane Database Syst Rev. 2009;(4):CD006459.

7. Berweck S, Schroeder AS, Fietzek UM, Heinen F. Sonography-guided injection of botulinum toxin in children with cerebral palsy. Lancet. 17 janv 2004;363(9404):249‑50.

8. Berweck S, Heinen F. Use of botulinum toxin in pediatric spasticity (cerebral palsy). Mov Disord Off J Mov Disord Soc. mars 2004;19 Suppl 8:S162-167.

9. Kwon J-Y, Hwang JH, Kim J-S. Botulinum toxin a injection into calf muscles for treatment of spastic equinus in cerebral palsy: a controlled trial comparing sonography and electric stimulation-guided injection techniques: a preliminary report. Am J Phys Med Rehabil Assoc Acad Physiatr. avr 2010;89(4):279‑86.

10. Gracies J-M, Burke K, Clegg NJ, Browne R, Rushing C, Fehlings D, et al. Reliability of the Tardieu Scale for assessing spasticity in children with cerebral palsy. Arch Phys Med Rehabil. mars 2010;91(3):421‑8.

11. Picelli A, Tamburin S, Bonetti P, Fontana C, Barausse M, Dambruoso F, et al. Botulinum toxin type A injection into the gastrocnemius muscle for spastic equinus in adults with stroke: a randomized controlled trial comparing manual needle placement, electrical stimulation and ultrasonography-guided injection techniques. Am J Phys Med Rehabil Assoc Acad Physiatr. nov 2012;91(11):957‑64.

12. Picelli A, Lobba D, Midiri A, Prandi P, Melotti C, Baldessarelli S, et al. Botulinum toxin injection into the forearm muscles for wrist and fingers spastic overactivity in adults with chronic stroke: a randomized controlled trial comparing three injection techniques. Clin Rehabil. mars 2014;28(3):232‑42.

13. Walker HW, Lee MY, Bahroo LB, Hedera P, Charles D. Botulinum Toxin Injection Techniques for the Management of Adult Spasticity. PM R. avr 2015;7(4):417‑27.

14. Grigoriu A-I, Dinomais M, Rémy-Néris O, Brochard S. Impact of Injection-Guiding Techniques on the Effectiveness of Botulinum Toxin for the Treatment of Focal Spasticity and Dystonia: A Systematic Review. Arch Phys Med Rehabil. 14 mai 2015;

15. Ben Smaïl D, Kiefer C, Bussel B. [Clinical evaluation of spasticity]. Neurochirurgie. mai 2003;49(2‑3 Pt 2):190‑8.

16. Haugh AB, Pandyan AD, Johnson GR. A systematic review of the Tardieu Scale for the measurement of spasticity. Disabil Rehabil. 15 août 2006;28(15):899‑907.

17. Singh P, Joshua AM, Ganeshan S, Suresh S. Intra-rater reliability of the modified Tardieu scale to quantify spasticity in elbow flexors and ankle plantar flexors in adult stroke subjects. Ann Indian Acad Neurol. 2011;14(1):23‑6.

18. Lewek MD, Randall EP. Reliability of spatiotemporal asymmetry during overground walking for individuals following chronic stroke. J Neurol Phys Ther JNPT. sept 2011;35(3):116‑21.

19. Ben-Shabat E, Palit M, Fini NA, Brooks CT, Winter A, Holland AE. Intra- and interrater reliability of the Modified Tardieu Scale for the assessment of lower limb spasticity in adults with neurologic injuries. Arch Phys Med Rehabil. déc 2013;94(12):2494‑501.

20. Singh P, Joshua AM, Ganeshan S, Suresh S. Intra-rater reliability of the modified Tardieu scale to quantify spasticity in elbow flexors and ankle plantar flexors in adult stroke subjects. Ann Indian Acad Neurol. janv 2011;14(1):23‑6.

21. Feise RJ. Do multiple outcome measures require p-value adjustment? BMC Med Res Methodol. 17 juin 2002;2:8.

22. Henzel MK, Munin MC, Niyonkuru C, Skidmore ER, Weber DJ, Zafonte RD. Comparison of surface and ultrasound localization to identify forearm flexor muscles for botulinum toxin injections. PM R. juill 2010;2(7):642‑6.

23. Picelli A, Bonetti P, Fontana C, Barausse M, Dambruoso F, Gajofatto F, et al. Is spastic muscle echo intensity related to the response to botulinum toxin type A in patients with stroke? A cohort study. Arch Phys Med Rehabil. juill 2012;93(7):1253‑8.

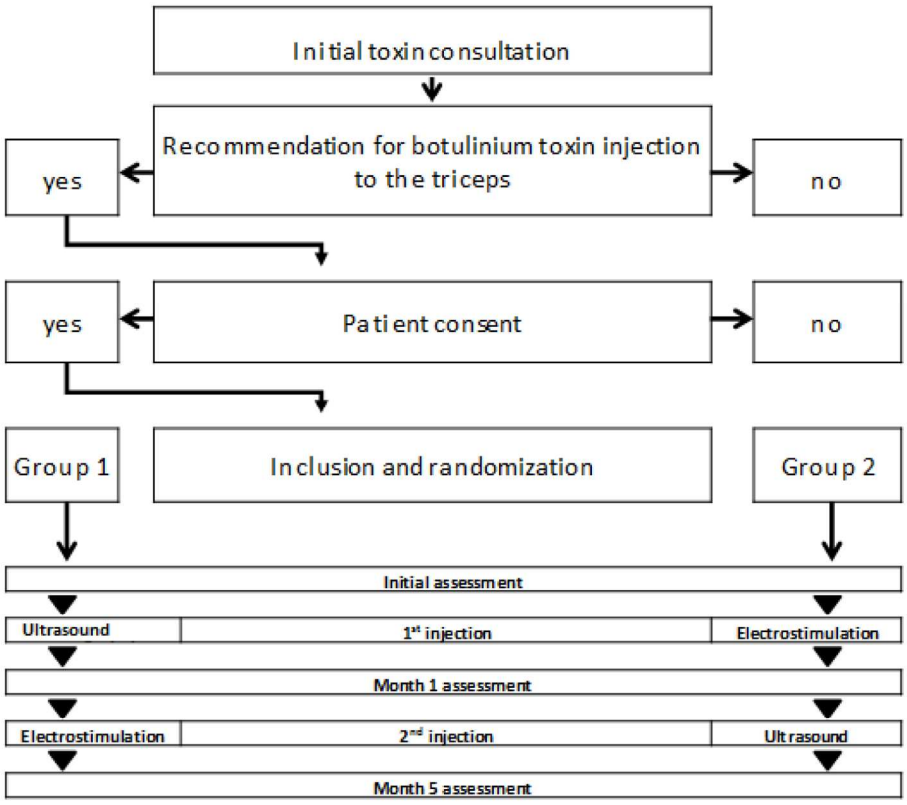

Figure 1: Flow diagram showing the different stages of the protocol

154x127mm (300 x 300 DPI)

APPENDIX 1:

Modified Ashworth Scale

Modified Ashworth Scale for Grading Spasticity	
Grade	**Description**
0	No increase in muscle tone
1	Slight increase in muscle tone, manifested by a catch and release or by minimal resistance at the end of the ROM when the affected part(s) is moved in flexion or extension
1+	Slight increase in muscle tone, manifested by a catch, followed by minimal resistance throughout the remainder (less than half) of the ROM
2	More marked increase in muscle tone through most of the ROM, but affected part(s) easily moved
3	Considerable increase in muscle tone, passive movement difficult
4	Affected part(s) rigid in flexion or extension

APPENDIX 2:

Tardieu Scale: Principles

Grading always performed:

- On a muscle at rest before the stretch maneuver
- At a reproducible velocity of stretch. Once the fast velocity is selected for a muscle, it remains the same for all subsequent tests.
- At the same time of the day
- In a constant body position for a given limb
- Other joints, particularly the neck, must also remain in a constant position throughout the assessment and for all other assessments.

Velocity of Stretch

- SLOW = V1: As slow as possible (slower than the rate of natural drop of the limb segment under gravity)
- FAST = Either V2 or V3
  - V2: Speed of the limb segment falling under gravity
  - V3: As fast as possible (faster than the rate of natural drop of the limb segment under gravity)

Tardieu Scale: Grading

X = Spasticity Angle (Threshold)

Angle of arrest at slow speed X_{V1} *minus* Angle of catch at fast speed X_{V3}

Y = Spasticity Grade (Gain)

0. No resistance throughout passive movement
1. Slight resistance throughout passive movement
2. Clear catch at precise angle, interrupting passive movement, followed by release
3. Fatigable clonus (<10s when maintaining pressure) occurring at a precise angle, followed by release
4. Unfatigable clonus (>10s when maintaining pressure) occurring at a precise angle
- Catch without release: graded 0 if $X_{V1}=X_{V3}$; "unratable" spasticity otherwise
- Catch with "minimal" release: graded 2 if X_{V3} is consistent and consistently less than X_{V1}
- Angle 0° = position of minimal stretch of the tested muscle
- For grades 0 and 1, spasticity angle X=0° by definition

19

20

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	3
	2b	All items from the World Health Organization Trial Registration Data Set	X
Protocol version	3	Date and version identifier	X
Funding	4	Sources and types of financial, material, and other support	20
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	21
	5b	Name and contact information for the trial sponsor	21
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	20
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	NA

Introduction

Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	5
	6b	Explanation for choice of comparators	X
Objectives	7	Specific objectives or hypotheses	9
Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)	11

Methods: Participants, interventions, and outcomes

Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	9
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	13
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	10
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	X
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	X
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	X
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	14
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	10

Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	15
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	12
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	15
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	X
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	11
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	12
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	X
Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	X
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	X

Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	_____X_____
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	_____15_____
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	_____15_____
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	_____15___
Methods: Monitoring			
Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	_____X_____
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	_____X_____
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	_____X____
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	_____X_____
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	_____20_____
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	_____X_____

Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	20
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	NA
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	X
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	20
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	X
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	X
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	X
	31b	Authorship eligibility guidelines and any intended use of professional writers	X
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	X
Appendices			
Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	X
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	NA

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.