We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,300

Open access books available

172,000

International authors and editors

190M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities

WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com

How Can We Clinically Apply Ultrasound-Guided BoNT-A Injection Technology for Muscle Spasticity in Stroke Patients?

Li Jiang and Zulin Dou

Additional information is available at the end of the chapter

<http://dx.doi.org/10.5772/66697>

Abstract

In this chapter, the primary focus is towards four topics related to the ultrasound (US)-guided injection: (1) the advantages of various guided injection techniques including US-guided injection, (2) a brief review of recently published studies on the US-guided botulinum toxin type A (BoNT-A) injection in stroke patients, (3) standardized operational procedures for the US-guided injection and (4) a description of the skills necessary to properly locate the probe and limb during the US-guided injection operation. Illustrations will be presented in the chapter to assist the readers in gaining a better understanding of the US-guided BoNT-A injection technique.

Keywords: botulinum toxin type A, spasticity, post-stroke, ultrasound

1. Introduction

In the post-stroke rehabilitation setting, botulinum toxin type A (BoNT-A) represents a first-line treatment for focal spasticity. Botulinum toxin type A is regarded as an effective treatment agent, and the efficacy and safety of BoNT-A injected in post-stroke patients with lower limb spasticity have been suggested in a few limited-scale randomized controlled trials [1, 2], as well as a meta-analysis study [3]. A successful and safe therapy using BoNT-A requires an anatomically accurate administration of BoNT-A into the muscles of the belly. One should be aware that BoNT-A may induce undue weakness to adjacent unaffected muscles. Knowing the location of the needle can help clinicians more accurately inject BoNT-A into the target muscle. To date, manual needle placement (MNP), electromyography (EMG), electrical stimulation (ES) and ultrasound (US) guidance have all been applied during BoNT-A injection [4].

Moreover, there are less frequently used localization techniques that exist to include fluoroscopy, computed tomography (CT) and endoscopic guidance. As a clinician, one should be knowledgeable of the characteristics regarding the localized injection technique and the disadvantages and advantage of each injection technique. Then, the clinician can determine the appropriate localization technique that is specifically designed for the patient under examination. The issues related to injection techniques will be discussed in the following chapter.

2. Comparisons of different guided injection techniques for BoNT-A

2.1. Manual needle placement

As presented above, there are four types of localization techniques that are traditionally used in clinical practice. Manual needle placement, also referred to as anatomical localization, is the simplest type of localization technique. This technique does not require the use of any equipment, unlike the other three above-mentioned localization techniques. When using MNP for injection, the clinician should be knowledgeable of the positions of the bones in the procedural area of the body and use palpation to identify the target muscles. To correctly inject patients, the physician should have a thorough understanding of the anatomical position of the target muscle as well as the surrounding muscles. Texts provide electromyographers with consistent electrode insertion sites for each muscle [5]. Following these same insertion sites of electrography, physicians can locate the target muscles. Sometimes, in order to verify the location better, the physician should use some manoeuvres and indirect signs emitted from passive palpation spastic muscles prior to injection.

There are several advantages of MNP localization. The most apparent advantage of this technique is that no equipment is required. A small-gauge needle is used to complete the injection instead of the larger-gauge needles that are required with EMG or ES injection. Moreover, the use of a smaller needle may decrease the level of discomfort. In addition, MNP localization is a relatively quick method that can be utilized to reduce the injection duration. Several disadvantages surface when MNP localization is used alone. The large, superficial muscles may be easily distinguishable with anatomic localization alone; however, other small, deeper muscles may not be as easily identified. Further, the spastic muscles can atrophy or twist, and these conditions can potentially alter the anatomical location of the muscle itself. In some cases, patients are unable to have a standard position that is appropriate for injection as demonstrated by the electrography tests. These conditions result in MNP localization as the sub-optimal localization [6]. Henzel et al. [7] demonstrated this concept to determine whether US localization is equivalent to anatomical localization in order to identify BoNT-A injection targets. The investigators were able to locate the forearm spastic muscle using two separate localization techniques. The study results showed that significant differences were observed between MNP localization and US localization for several flexor muscles. This group believed that the landmark measurement was based on cadaveric studies and that it is difficult to place patients with spasticity in a standard supination and extension position. Furthermore, in the case of in-patients with spasticity, the typical three-dimensional structure of the forearm may be distorted due to severe muscle atrophy [8].

2.2. Electromyography-guided injection

The EMG-guided injection technique is familiar amongst many physicians who treat spasticity using BoNT-A. This injection technique has the ability to precisely identify spastic muscles and requires an EMG machine and a hollow insulated monopolar needle electrode [6]. To conduct this technique, the physician first ensures that the target muscles align with the anatomical localization and then administers the EMG-guided injection. Once the EMG needle electrode is inserted into spastic muscle, the physician should hear the involuntary motor unit action potentials (MUAPs), which may initially have a dull muffled sound, but will become sharper as the needle is advanced closer to the end plate. In order to make sure that the spastic muscle is correctly targeted, the physician may ask the patient to move that muscle, and the physician will then listen for increased signalling of MUAPs. In the event that the patient is unable to voluntarily contract the muscle using active motions, the physician may conduct a passive range of motion (PROM) to stretch the target muscle in the patient [9].

There are several advantages that are associated with the use of EMG-guided injection. An auditory EMG device is inexpensive and provides a mechanism for more precisely localizing the spastic target muscles. In addition, the EMG-guided injection is helpful for the delivery of the BoNT-A near the motor endplate of the spastic muscle or in the location of a high concentration of active MUAPs [10, 11]. There are a few disadvantages associated with EMG-guided technique. At first, this technique does not guarantee that the monopolar needle is inserted in the target muscle. Voluntary activation and PROM are used to decrease this risk; however, for deeper or overlapping muscles, there would be a greater probability for misplacement of the needle. In some instances, the physician may inject into a spastic muscle that may not be the target muscle. To circumvent these issues, the clinician may increase the frequency and intensity of the muscle stimulus to identify spastic muscle, which may extend the time duration of the procedure and intensify the level of pain as compared with anatomic guidance. Other disadvantages of the EMG-guided injection technique are the costs of the insulated EMG needle and the costs of the EMG machine compare with anatomic guidance [6].

2.3. Electrical stimulation-guided injection

The electrical stimulation-guided injection is a popular technique for muscle localization. In this method, like EMG-guided injection, a hollow insulated monopolar needle is connected to a portable ES machine, and the target muscle is located by anatomical landmarks. Once the needle electrode is located in the target muscle, electric current is delivered to the muscle through the needle electrode. Normally, a 5-mA stimulus at 1-Hz intervals is applied to contract the muscle. Once the level of muscle contraction is identified as appropriate, the clinician may attempt to maintain a robust contraction while slowly decreasing the intensity of the muscle stimulation or incrementally modify the needle electrode, as needed. If the clinician is able to maintain the muscle contraction using a low-intensity stimulus, such as a 1 mA, the clinician can be relatively certain that the tip of the needle is proximal to the motor endplate [6].

One advantage of the ES-guided injection technique is the accuracy of the targeted muscle localization. The visual feedback from the spastic muscle contraction ensures that the needle is properly inserted in the target muscle, especially in the event that muscle contraction occurs

under a low-intensity stimulus. The disadvantage of using the ES-guided technique to locate the target muscle is that this technique may result in increased time consumption and may require additional training compared to the other techniques mentioned herein. Moreover, this technique may cause the patient to experience a higher level of discomfort than the other techniques. In addition to the disadvantages of the ES-guided technique, for patients with severe spasticity and limited range of motion, it may be difficult to assess individual muscle contraction. Lastly, the costs of both the needle electrode and the ES device must be considered in clinical practice when conducting this method.

2.4. Ultrasound-guided injection

Ultrasonography is well established as a reliable and reproducible imaging method that is used to identify the anatomy of the muscle [12]. Ultrasound machines consist of several components: the transducer, the computer processor unit and the monitor. Transducers are available in high and low frequencies, while the higher-frequency transducers are used for more superficial structures at a high resolution and the lower-frequency transducers can be used to assess deeper structures. The clinician who is experienced with the use of US-guided injection is able to recognize the cross-sectional anatomy that is displayed on the monitor and is able to visualize the needle tip once it is injected into the target muscle.

There are many advantages of the US-guided injection with BoNT-A. This technique allows real-time visualization of the needle into structures including the target muscles and adjunct tissues. This method not only permits the clinician to more precisely identify the target muscles but also permits the avoidance of needle penetration in some other bodily structures, to include the blood vessels and nerves. Other potential benefits of this technique is that US-guided technique procedure is relatively more efficient with respect to the time needed to conduct this technique, and the patients experience less pain compared with the ES-guided and EMG-guided injections. The lowered pain indication can be attributed to the use of a smaller gauge needle [7]. In addition, the use of US-guided injection may help to reduce side effects of the injected medicine, such as the dissemination of BoNT-A into nontargeted areas. Moreover, the clinician is able to visualize the volume of the injected BoNT-A solution in real time and administer the appropriate amount of solution into the targeted muscle. This feature permits the physician to relocate the needle tip to a different area within the same target muscle in order to complete the injection and minimize the spread of the BoNT-A solution into the adjacent off-target muscles.

Although the US-guided injection technique for BoNT-A is regarded as the most accurate method for locating the target muscles, shortcomings exist for this technique. At first, operation of the US transducer and the syringe simultaneously may require the presence of an assistant. This is particularly necessary for the clinicians who are novel to this technique. It is advisable that clinicians who are interested in using the US guidance to localize the target muscle should seek the appropriate training in great sufficiency prior to administering this technique.

Which type of guided injection technique is most appropriate to eliminate toxins? Other investigators have reviewed and compared these guided injection techniques in details as reported in recent publications [4, 6, 13]. First, we believe that the clinicians who will perform

the BoNT-A injections for spasticity management should be knowledgeable of the advantages and disadvantages of each of the injection techniques as previously discussed. Second, the clinicians should undergo combined training experiences in order to adequately explain each technique in sufficient detail to the patient and ensure that the appropriate technique aligns with the needs of the patient. Finally, one should consider additional factors, such as equipment cost, spastic muscles location when considering the most appropriate guided injection technique. Although it seems that each of these localization techniques is superior to the use of anatomical localization alone, we believe that the anatomical localization should be the established standard when considering the use of an instrument-guided injection technique. Further, more studies are needed to determine which combination of localization techniques can produce the best clinical outcomes.

3. A review of the studies that administered US-guided BoNT-A injection into the upper and lower limb muscles of stroke patients

The use of US is a well-established reliable and reproducible imaging method for defining muscle anatomy. An ultrasound system with a 7.5-MHz linear transducer can provide sufficient resolution for both superficial and deep-seated muscles [14]. As an alternative to the electrophysiological techniques, US offers a visually controlled method of injection of BoNT-A [15, 16]. Schiano et al. were the first to report on the US-guided BoNT-A injection for the treatment of achalasia [17]. Since that time, the advantages of the US-guided BoNT-A injection have been recognized, and the use of this technique is becoming more widespread in the present years. In the previous years, Berweck and colleagues utilized US-guided injections for spastic muscles in children. From the years 2000 to 2003, these investigators administered over 6000 injections into 70 different muscles in a total of 350 children. Berweck et al. recommended the use of US to conduct anatomically precise injection of BoNT-A due to the advantageous features of sonography, which is easy, quick, painless and available in most hospitals [15, 16]. Until now, the use of US was mostly popular amongst pediatrics, and it seemed that the use of US was the obvious choice for spastic muscle identification and injection control [4].

Several studies suggested that the various guided BoNT-A injection techniques such as ES guided, EMG guided and US guided showed a greater spasticity reduction in conjunction with improved clinical outcomes when compared with MNP localization. In a clinical study conducted by Yang et al. [18], MNP into the gastrocnemius muscle (GCM) for BoNT-A injection in children with spastic cerebral palsy (CP) was investigated to analyze its accuracy and effectiveness. The accuracy of MNP by one researcher using anatomic landmarks alone was assessed by another researcher who used US visualization prior to BoNT-A injection. These researchers found that the MNP was accurately placed into the GCM in 78.7% of cases, with the greatest accuracy being with the needle insertion into the medial GCM (92.6%) and the lowest accuracy was observed in the lateral GCM (64.7%). These investigators reported that the lateral portion of the GCM was thinner than the medial GCM, and this resulted in a greater rate of misplaced needle. Injection of the BoNT-A into GCMs using an anatomical landmark was an acceptable approach for injection into the medial GCM; however, this approach was

not effective when administered in the lateral GCM. Py et al. [19] evaluated the effectiveness of injecting BoNT-A into the lower limbs of children with CP using the needle placement technique (MNP or US guided). Thirty of the children received US-guided injections, and the remainder received injections using anatomical localization. These investigators evaluated the gait and the spasticity (Tardieu scale), a functional evaluation (the Gross Motor Function Measure) 4 weeks after the injection. Clinical effectiveness was noted in the children receiving US-guided injections compared with MNP alone, and the “functional effectiveness” was also improved in the children with the use of US. The results of the aforementioned studies support the concept that the US-guided injection is more effective than the MNP localization technique administered in the spastic muscles of patients with CP. In contrast to the study conducted by Py et al., Kwon et al. [20] evaluated the clinical outcomes in 32 children with CP following BoNT-A injections into the GCMs. These investigators compared the efficacy of ES guided and US guided for BoNT-A injections into the GCMs in the children. These researchers found no significant differences between the groups according to the scores on the modified Ashworth Scale scores (MAS) and the Modified Tardieu Scale (MTS) assessments; however, the subscales of the Physician’s Rating Scale significantly improved in children who received BoNT-A injections when guided by US. Although similar results showed that spasticity levels between the US-guided injection and ES-guided injection were decreased, the investigators concluded that the visual feedback provided by the US-guided injection may improve the accuracy of the administration of BoNT-A into the GCMs in the children.

To date, a few studies have compared the precision of the MNP and US localization in efforts to identify the forearm flexor muscles that will undergo BoNT-A injections in subjects with arm spasticity. In an observational study of 18 adult patients with upper extremity spasticity, Henzel et al. [7] explored the accuracy of the MNP to locally administer the investigational agent in the forearm flexor muscles. After the measurements of the surface, marks were obtained, and US was used to determine the optimal injection site, which is described as the portion of the target muscle with the largest cross-sectional area. There was a statistically significant difference between the proposed injection sites as per the methods of MNP and the actual optimal injections sites as determined via the US-guided injections for the flexor pollicis longus, pronator teres and flexor digitorum superficialis (FDS) to digit 3, with trends towards significance in the flexor carpi radialis and FDS to digits 2 and 4. Based on these findings, the investigators recommend that the US-guided technique should be considered for muscle localization in patients with upper extremity spasticity. Picelli et al. [21] completed a single-blinded, randomized controlled study comparing the outcomes of 60 chronic stroke patients with clenched fist or flexed wrist who underwent BoNT-A injections with MNP or ES-guided or US-guided injections. Each subject underwent pre-injection and postinjection evaluations that included MAS, Tardieu angle and PROM. These investigators found enhanced improvement in all of the measurements when comparing the ES group with the MNP group. Likewise, the subjects in the US-guided BoNT-A injection group demonstrated greater improvement in all of the measurements when compared with the MNP group. There was no difference noted in the clinical outcomes when comparing patients who received injections when guided by ES versus the injections that were guided by US. These authors concluded that the use of ES- and US-guided BoNT-A injections decreases spasticity and results in vast improvements in the range of motion when compared to MNP techniques alone.

Santamato et al. [22] compared the reduction of spasticity and the related finger position at rest improvement in post-stroke patients treated with BoNT-A in the wrist and finger flexor limb muscles using US-guided injection and MNP localization. In the randomized clinical trial, two groups of 15 stroke patients were treated with BoNT-A injections in the wrist and finger flexor muscles of the affected upper limb using US-guided injections or MNP localization. The MAS and the finger position at rest were measured before and 4 weeks after administration of the injections. The results showed the MAS and finger position at rest significantly improved in both of the treatment groups, although these clinical outcomes were more effective in patients who received the US-guided BoNT-A injection compared to patients injected with BoNT-A using MNP localization. The investigators concluded that the US-guided BoNT-A injections were capable of improving the clinical outcome more effectively than MNP in post-stroke patients with spasticity.

In a study conducted by our team [23], we injected BoNT-A while being guided by US in patients with post-stroke wrist and finger flexor muscle spasticity and assessed the clinical outcomes following the injection and rehabilitation intervention. The results showed significant decreases in the MAS scores of both the finger flexor muscle and wrist flexor muscle at all time points after BoNT-A injections in comparison with the baseline scores. Compared with the baseline, the PROM of the wrist and finger extensions and the FMA scores of the wrist and hand significantly increased ($p < 0.001$) at 2, 4 and 12 weeks after the injections. We concluded that the US-guided injection of BoNT-A combined with rehabilitation exercise decreased the spasticity of the wrist and finger flexor muscles and improved the motor function in stroke patients up to 12 weeks following the BoNT-A injection.

Based on these aforementioned studies, all of the researchers have suggested that the US-guided injection technique is more effective in improving the accuracy of toxin placement in patients with limb spasticity. Most importantly, the US-guided injection is visually controlled. In viewing the entire process, the operator gains a better understanding of the anatomy of the individual under observation. This enables a more effective and safer technique selection and assists in the reduction of side effects due to the unintentional spread of medicine. Despite the fact that US cannot measure the muscular hyperactivity and detect motor endplate regions, the ultrasound images can provide information about muscle size and fibrosis, which are all factors that can be important in the decision-making process.

4. A description of the procedures for US-guided BoNT-A injection in clinical practices

In order to make US-guided injections feasible and effective in clinical practice, in accordance with the practice experience, the authors have developed a set of standardized operational procedures that include target muscle identification, the selected probe for upper and muscles images, the relationship between the limb and probe positioning, the proper needle types used for different muscles, the distance between the injecting needle and probe and needle tilting angle and the coordination between the operators and others involved in the injection process.

Figure 1. Sonographic image of the upper limb (Bic = bicep, H = humerus).

1. Probe selection

The 7.5-MHz linear transducer can provide sufficient resolution for superficial muscles but is also able to detect deep-seated muscles.

2. Probe viewing mode

The transverse viewing mode is arranged such that the medial part of the limb is seen on the right and the lateral part of the limb (the right side) is seen on the left side of the monitor screen.

3. Muscle imaging

The musculature appears poorly echogenic, while the perimysia and fascicular tissue between the muscle bellies are apparently echogenic. The two principles of muscle identification are (1) recognizing the characteristic pattern of the individual muscles. The transverse sonogram corresponds to the transverse anatomic sections. (2) Each muscle has a characteristic contour line. These muscles exhibit specific patterns and allow prompt (within a few seconds) identification of the individual muscles. An example for the upper or lower limb is shown in **Figures 1 and 2**.

4. Imaging of neighbouring structures

Visualization of the neighbouring structures, such as the bones and vessels, helps to accurately determine the injection site in target muscles. An example is shown in **Figure 3**.

5. Identification of target muscles

The suggested manner to identify target muscles includes the following: (1) The skin surface location for the target muscle was identified on the basis of the use of the specific anatomic landmarks from Delagi et al. [5] and was subsequently marked. (2) The transducer was located

Figure 2. Sonographic image of the lower limb (SOL, soleus; FDL, flexor hallucis longus; TP, tibialis posterior; T, tibia; F, fibula).

at the marker and positioned perpendicularly to the skin surface to obtain a transverse view of the target muscle. (3) By adjusting the parameters such as view depth, focus and gain, a clear image of the target muscle and other muscles could be displayed (**Figures 4–6**).

Figure 3. Sonographic image of the forearm, including the FDL and surrounding structures (Bra, brachioradialis; FPL, flexor pollicis longus; FDS, flexor digitorum superficialis; FDP, flexor digitorum profundus; A, artery; R, radius; U, ulna).

Note 1: Passive movements at small amplitudes are visible as concurrent oscillations of the intramuscular echo. Passive movement of the corresponding part of the body may help to visualize the dynamic contraction of the target muscle.

6. US-guided injection

To conduct the US-guided injection, (1) in the transverse (axial) view, the target muscle was scanned from the proximal to distal direction or vice versa until the largest cross-sectional area was identified. (2) The needle was inserted and was closely aligned to the longitudinal axis of the transducer (**Figure 7**). Slight movements of the needle along the transducer longitudinal axis are helpful to obtain a satisfactory image. (3) As the needle penetrates the skin, its path through

Figure 4. Probe location at different target muscles (a, pronator teres; b, pronator quadratus).

the tissue to the target site for injection is continually monitored on the screen. (4) To confirm the surface injection site, in some cases, the second injection site of each target muscle was identified by distally moving the transducer approximately 1.5–2.0 cm. (5) Pre-injection sonographic images and the images during the injection are able to be stored for future references.

Upon injection, the solution spreads out in the muscle, usually as an echogenic cloud, sometimes with echo obliteration (**Figures 8 and 9**).

Note 2: Slight movements of the needle along its longitudinal axis can help to obtain a satisfactory image.

Figure 5. Sonographic image of the upper third of the anterior forearm (PT, pronator teres; Bra, brachioradialis; A, artery).

Figure 6. Sonographic image of the lower third portion of the anterior forearm (PQ, pronator quadratus; FDS, flexor digitorum superficialis; FDP, flexor digitorum profundus; R, radius; U, ulna).

Figure 7. Sonographic image indicating the position of the inserted needle (arrow head points to the hyperecho line).

Figure 8. Needle in the target muscle before injection (arrow head points to the hyperecho point).

Figure 9. The solution disseminates into the target muscle (arrow head points to the hyperecho area).

5. A summary of the necessary skills to effectively administer the US-guided BoNT-A injection

1. Familiarity with the regional anatomy of the limb

It is well known that due to the size of the ultrasound probe, the pictures that are displayed on the screen of the ultrasound machine are specific only to the regional anatomy of the tissue under examination. Therefore, various probe locations result in different sonographic images. In order to achieve accurate injections, the clinician should undergo intense practice sessions in administering a variety of injections which will allow the clinician to become more familiar with the characteristics of the muscle and the surrounding structures as viewed through the sonographic machine. These practices are useful for precise injection into the target muscles and also ensure a safe and effective injection.

2. Basic operation of the ultrasound machine

The operators of the US machine should be competent in the use of this machine. A good understanding of the panel operation is helpful for adjusting the parameter settings in order to obtain the best image quality, which in turn would project the best injection site.

3. Spastic limb placement

In preparation of the injection, the injection site of the affected limb should be fully exposed. If patients with the higher spasticity levels are unable to position the limb appropriately, the clinician should ask for an assistant to help correctly position the limb.

4. The probe placement

The probe should be perpendicular to the plane of the proposed limb for injection. Proper probe placement is one of the basic requirements to obtain an excellent quality of ultrasound images. If the angle from the probe and body is too large or too small, the probe will receive sound reduction. Furthermore, the probe will produce unclear images of anatomical structure, and the accuracy of the injection will undergo severe alterations.

5. Relationship between the needle and probe (**Figures 10 and 11**)

In the in-plane position, the needle should be parallel to the probe. Next, the needle is inserted along the longitudinal axis of the probe. In this orientation, the needle is easy to visualize and the tip can be observed in its entirety during the injection.

In the out-of-plane position, the needle is inserted across the short axis of the probe at an angle to the skin. To properly conduct this technique, it is necessary to acquire the skills through repetitive practices and obtain a sufficient level of experience. The tip or shaft of the needle was seen as a hyper-echoic dot using the method.

The most common choice is the in-plane method in which the needle process can be observed throughout the muscle to ensure the safety and accuracy of the injection. In the out-of-plane position, the accuracy of injection may be affected due to the fact that only a portion of the needle is visible in the sonographic picture.

6. Ambidextrous coordination

As one hand holds the probe, the clinician should pay careful attention to the manner in which the probe is handled such that the probe is well positioned on the surface of the

Figure 10. In plane (needle is parallel to probe).

Figure 11. Out of plane (needle inserted across the short axis of probe).

skin. Using the other hand, the clinician is capable of the injection using the US-guided technique. For beginners, this ambidextrous coordination may seem challenging. With repetitive practices and increased exposure to this technique, the clinician will be able to master the skills.

7. Other skills

The operators involved with the injection procedure should practise sterile techniques.

Author details

Li Jiang* and Zulin Dou

*Address all correspondence to: 908455721@qq.com

Department of Rehabilitation Medicine, The Third Affiliated Hospital, Sun Yat-sen University, Guangzhou, China

References

- [1] Farina, S., Migliorini, C., Gandolfi, M., Bertolasi, L., Casarotto, M., Manganotti, P., et al. Combined effects of botulinum toxin and casting treatments on lower limb spasticity after stroke. *Funct Neurol.* 2008, 23 (2): 87–91.

- [2] Mancini, F., Sandrini, G., Moglia, A., Nappi, G., Pacchetti, C. A randomised, double-blind, dose-ranging study to evaluate efficacy and safety of three doses of botulinum toxin type A (Botox) for the treatment of spastic foot. *Neurol Sci.* 2005, 26 (1): 26–31.
- [3] Rosales, R.L., Chua-Yap, A.S. Evidence-based systematic review on the efficacy and safety of botulinum toxin-A therapy in post-stroke spasticity. *J Neural Transm.* 2008, 115 (4): 617–623. doi: 10.1007/s00702-007-0869-3.
- [4] Schroeder, A.S., Berweck, S., Lee, S.H., Heinen, F. Botulinum toxin treatment of children with cerebral palsy—a short review of different injection techniques. *Neurotox Res.* 2006, 9 (2–3): 189–196.
- [5] Delagi, E.F., Perotto, A., Iazetti, J., Morrison, D. *Anatomic Guide for the Electromyographer: The Limbs and Trunk.* 4th ed. Springfield, IL: Charles C Thomas. Publisher, Ltd; 2005.
- [6] Walker, H.W., Lee, M.Y., Bahroo, L.B., Hedera, P., Charles, D. Botulinum toxin injection techniques for the management of adult spasticity. *PM R.* 2014, 7 (4): 417–427. doi: 10.1016/j.pmrj.2014.09.021.
- [7] Henzel, M.K., Munin M.C., Niyonkuru C., Skidmore E.R., Weber D.J., Zafonte R.D. Comparison of surface and ultrasound localization to identify forearm flexor muscles for botulinum toxin injections. *PM R.* 2010, 2 (7): 642–646. doi: 10.1016/j.pmrj.2010.05.002.
- [8] Pillen, S., Arts, I.M., Zwarts, M.J. Muscle ultrasound in neuromuscular disorders. *Muscle Nerve.* 2008, 37 (6): 679–693. doi: 10.1002/mus.21015.
- [9] O'Brien, C.F. Injection techniques for botulinum toxin using electromyography and electrical stimulation. *Muscle Nerve Suppl.* 1997, 6: S176–S180.
- [10] Lapatki, B.G., van Dijk, J.P., van de Warrenburg, B.P., Zwarts, M.J. Botulinum toxin has an increased effect when targeted toward the muscle's endplate zone: a high-density surface EMG guided study. *Clin Neurophysiol.* 2011, 122 (8): 1611–1616. doi: 10.1016
- [11] Gracies, J.M., Lugassy, M., Weisz, D.J., Vecchio, M., Flanagan, S., Simpson, D.M. Botulinum toxin dilution and endplate targeting in spasticity: a double-blind controlled study. *Arch Phys Med Rehabil.* 2009, 90 (1): 9–16.e2. doi: 10.1016.
- [12] Fischer, A.Q., Carpenter, D.W., Hartlage, P.L., Carroll, J.E., Stephens, S. Muscle imaging in neuromuscular disease using computerized real-time sonography. *Muscle Nerve.* 1988, 11 (3): 270–275.
- [13] Lim, E.C., Quek, A.M., Seet, R.C. Accurate targeting of botulinum toxin injections: how to and why. *Parkinsonism Relat Disord.* 2011, 17 (Suppl1): S34–S39. doi: 10.1016
- [14] Willenborg, M.J., Shilt, J.S., Smith, B.P., Estrada, R.L., Castle, J.A., Koman, L.A. Technique for iliopsoas ultrasound-guided active electromyography-directed botulinum a toxin injection in cerebral palsy. *J Pediatr Orthop.* 2002, 22 (2): 165–168.
- [15] Berweck, S., Feldkamp, A., Francke, A., Nehles, J., Schwerin, A., Heinen, F. Sonography-guided injection of botulinum toxin A in children with cerebral palsy. *Neuropediatrics.* 2002, 33 (4): 221–223.

- [16] Berweck, S., Schroeder, A.S., Heinen, F. Sonography-guided injection of botulinum toxin in children with cerebral palsy. *Lancet*. 2004, 363 (9404): 249–250.
- [17] Schiano, T.D., Fisher, R.S., Parkman, H.P., Cohen, S., Dabiezies, M., Miller, L.S. Use of high-resolution endoscopic ultrasonography to assess esophageal wall damage after pneumatic dilation and botulinum toxin injection to treat achalasia. *Gastrointest Endosc*. 1996, 44 (2): 151–157.
- [18] Yang, E.J., Rha, D.W., Yoo, J.K., Park, E.S. Accuracy of manual needle placement for gastrocnemius muscle in children with cerebral palsy checked against ultrasonography. *Arch Phys Med Rehabil*. 2009, 90 (5): 741–744. doi: 10.1016
- [19] Py, A.G., Zein Addeen, G., Perrier, Y., Carlier, R.Y., Picard, A. Evaluation of the effectiveness of botulinum toxin injections in the lower limb muscles of children with cerebral palsy. Preliminary prospective study of the advantages of ultrasound guidance. *Ann Phys Rehabil Med*. 2009, 52 (3): 215–223. doi: 10.1016/j.rehab.2009.02.005.
- [20] Kwon, J.Y., Hwang, J.H., 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, 89 (4): 279–286. doi: 10.1097/PHM.0b013e3181ca24ac.
- [21] Picelli, A., Lobba, D., Midiri, A., 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*. 2014, 28 (3): 232–242. doi: 10.1177/0269215513497735.
- [22] Santamato, A., Micello, M.F., Panza, F., Fortunato, F., Baricich, A., Cisari, C., Pilotto, A., Logroscino, G., Fiore, P., Ranieri, M. Can botulinum toxin type A injection technique influence the clinical outcome of patients with post-stroke upper limb spasticity? A randomized controlled trial comparing manual needle placement and ultrasound-guided injection techniques. *J Neurol Sci*. 2014, 15, 347 (1–2): 39–43. doi: 10.1016/j.jns.2014.09.016.
- [23] Jiang, L., Dou, Z.L., Wang, Q., Wang, Q.Y., Dai, M., Wang, Z., Wei, X.M., Chen, Y.B. Evaluation of clinical outcomes of patients with post-stroke wrist and finger spasticity after ultrasonography-guided BTX-A injection and rehabilitation training. *Front Hum Neurosci*. 2015 Sep 2, 9: 485. doi: 10.3389/fnhum.2015.00485.