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REVIEW PAPER

Use of botulinum toxin in aesthetic medicine and gynaecology: current approaches, controversies, and future directions

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Abstract

This review looks at the use of botulinum toxin in the gynaecological field with the aim of determining what needs to be further investigated to achieve a standardized application. Numerous studies have been conducted to explore how botulinum toxins (BoNT) can be applied, and it is becoming popular for treating various disorders such as chronic pelvic pain, vestibulodynia, and vaginism. However, the exact dosage and ideal location for injections still need to be clarified. The objective of this study is to point out which aspects need to be more carefully studied to ensure a consistent use of BoNT in gynaecology.

Key words: botulinum, vagina, vulva, vulvodynia, dyspareunia, Botox.

Introduction

Botulinum toxin is a neurotoxin produced by the anaerobic, gram-positive bacterium, *Clostridium botulinum* [1]. To date, 7 distinct types (serotypes A, B, C1, D, E, F, and G) of the toxin have been identified [2]. Each serotype is composed of a single polypeptide chain with a molecular mass of about 150 kDa. This chain is associated with non-toxic proteins, forming complexes of between 300 and 900 kDa. Splitting the 150 kDa polypeptide chain into 2 parts (100 and 50 kDa, respectively) linked by a disulphide bond and non-covalent interactions, maximizes the biological activity of the molecule. Not all forms are active on the human nervous system, and of the 7, only serotypes A and B are available for medical use. These are available as 4 different subtypes for clinical use with 3 forms for serotype A and one form for serotype B [3–6]. All forms are approved by the Food and Drug Administration (FDA) for the treatment of spasmodic torticollis, focal eye dystonia, chronic migraine, hyperhidrosis, urinary incontinence, and aesthetics [7, 8]. The neurotoxin onabotulinumtoxin A has the widest use in cosmetics [9]. Recently, there has been increased interest in the use of botulinum in gynaecology, with different solutions, routes of administration, and indications being presented in the literature and in practical medicine. This review aims to provide an overview of the use of botulinum in aesthetic medicine and gynaecology to bridge these 2 disciplines.

Material and methods

A literature search was conducted to identify studies exploring the use of botulinum toxins (BoNT) in treating vaginal and vulvar disorders. The search was performed in MEDLINE, EMBASE, Global Health, The Cochrane Library (Cochrane Database of Systematic Reviews, Cochrane Central Register of Controlled Trials, Cochrane Methodology Register), the Health Technology Assessment Database, and Web of Science, as well as research registers such as www.cliniclatrials.gov. The search terms were "botulinum" or "BoNT" and "vaginal", "vulvar" or "vulvovaginal", and the search was limited to articles published in English from the start of each database until 1 September 2022.

Two authors screened titles and abstracts from the search results and additional sources to identify potentially eligible studies. The full text of these ar-

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ticles was retrieved and independently assessed by 2 other review team members, with any disagreements resolved via discussion with an external collaborator. Data from selected articles were then extracted by 2 authors independently, with any discrepancies resolved via discussion (with an external collaborator if necessary). A narrative synthesis was used to summarize the findings.

Results

Mechanism of action

Botulinum toxin serotypes have a highly specific mechanism of action that targets cholinergic nerves [10, 11]. This leads to a temporary denervation of peripheral areas and the subsequent relaxation of muscle fibres. The light chain of BoNT binds to the SNARE complex, a transport protein responsible for releasing acetylcholine, while the heavy chain binds to glycoprotein structures found on cholinergic synaptic terminals, thus providing the neurotoxin's selectivity for cholinergic sites [12]. Injection of BoNT into the target tissue results in these effects. Botulinum toxins are known to have distinct target proteins for each subtype. Botulinum toxin A specifically binds to synaptosome-associated protein 25 kDa, while BoNT-B binds to vesicle-associated membrane protein, also known as synaptobrevin II [13]. These proteins are split by the toxin's light chain, leading to the prevention of the acetylcholine vesicle's attachment to the cell membrane, blocking its fusion. The full mechanism of action of BoNT on the nerve terminals can be broken down into 5 steps. First, it binds to cholinergic nerve terminals with high affinity and specificity. It is then internalized, which is followed by translocation of its light chain across the vesicle membrane. The light chain is then released, and the disulphide bond is disassociated, leading to the cleavage of the SNARE proteins. This ultimately results in the blockade of neurotransmitter release and neuroparalysis [14, 15].

The effects of BoNT on synaptic exocytosis can inhibit the release of acetylcholine at the neuromuscular junction. The results of this cholinergic block may differ depending on the type of organ affected. If the target organ is skeletal muscle, chemical denervation may lead to paresis, while if the target organ is an exocrine gland, the secretion of the gland can be blocked. The volume of the denervation field induced by BoNT is dependent on the dose and amount of solution injected. The turnover of the SNARE protein complex can be restored, which can reverse the inhibition of acetylcholine exocytosis caused by BoNT [16, 17].

The effects deriving from the administration of botulinum toxin can be schematically described as direct and indirect. The first one (direct) includes blocking the neuromuscular or autonomic cholinergic innervation. Indirect effects, on the other hand, occur at the level of the central nervous system, resulting in reflex inhibition, intracortical inhibition, normalization of reciprocal inhibition, and evocation of somatosensory potentials.

Available doses for application in clinical practice

Currently, there are 4 commonly used preparations of BoNT, all characterized by similar therapeutic effect but differentiated by chemical structure, associated proteins, production, and purification processes [18]: onabotulinumtoxinA (Botox; Botox Cosmetic, Allergan, Irvine, CA), abobotulinumtoxinA (Dysport; Ipsen, Ltd., Berkshire, UK), incobotulinumtoxinA (Xeomin; Merz Pharmaceuticals, Frankfurt, Germany), and rimabotulinumtoxinB (Myobloc; Solstice Neurosciences, San Francisco, CA).

The recommended dilutions are, respectively, 100 U of powder in 1–8 ml of normal saline solution for onabotulinumtoxin A and 300 U of powder in 0.6–2.5 ml of normal saline solution for abotulinumtoxinA (BoNT-A).

Experts in BoNT therapy have suggested a conversion ratio for facial aesthetic use, in which one unit of onabotulinumtoxinA 5 is equivalent to one unit of incobotulinumtoxinA 5 and 3 units of abobotulinumtoxinA [19–22]. However, due to a lack of experience, no such equivalence exists between BoNT-A and rinotulinumtoxinB. Moreover, an incorrect conversion ratio can result in an overdose, increasing the risk of adverse events. After injection, it may take up to 2 weeks for the full clinical effects to become noticeable [23].

The packaging and distribution of pharmaceutical formulations of BoNT as a powder must be done with caution due to the potential of incorrect dilution leading to its spread to unwanted regions and adverse effects, such as eyelid ptosis [24]. The Food and Drug Administration (FDA) approved dose for aesthetic glabellar lines are abobotulinumtoxinA 50 U, onabotulinumtoxinA cosmetic 20 U, and incobotulinumtoxinA 20 U. The FDA label advises against performing potency conversions. To ensure the most effective results, there are several variables that must be taken into consideration, such as the location, volume, and concentration of the targeted muscle, the intrinsic properties of the available toxins, the muscle selection, and injection technique. All these factors should be considered before deciding on the most suitable toxin, dilution, and injection site.

Aesthetic use

The effects of BoNT have been extensively studied, and the results are now widely used by many specialists in the field of cosmetic treatments.

Since 2002, BoNT has been used for aesthetic purposes after its approval. Numerous studies have been conducted to explore all the potential applications of this toxin in the beauty industry. The effects of BoNT have been thoroughly analysed, and their results are now frequently applied by cosmetic specialists.

The aging process of the face involves changes at all levels, from the epidermis to the subcutis and even the underlying bone. Stem cells are responsible for regenerating and balancing the degeneration of these tissues, but as people age, their regenerative properties diminish and a net loss of tissue can occur. This can result in deflation of the subcutaneous fat, a decrease in total collagen content, and the degeneration of dermal collagen and elastin. These changes can cause a variety of outward signs, such as skin laxity, folds, rhytides, roughness, and xerosis [25].

The use of BoNT for aesthetic purposes has become increasingly popular due to the broad range of improvements it can bring [26]. Patients have reported high levels of satisfaction with the effects of treatment, which involves selectively weakening facial expression muscles. This helps to reduce the appearance of dynamic wrinkles caused by muscle activity, while also creating a softer, more natural look. While BoNT can be used to reduce dynamic wrinkles, they cannot replace procedures such as skin resurfacing for treating static wrinkles.

In many countries, the use of botulinum toxin type A has been approved for treating glabellar lines. Furthermore, it is also used to address lateral canthal lines, facial hyperkinetic lines, transverse forehead rhytides, the glabellar region, brow lift, and the lateral orbital region in some countries [27–30].

It has been suggested that transverse forehead wrinkles can be improved with off-label use of BoNT-A. A patient's wrinkles must be evaluated to determine whether they are static or dynamic in nature. When treating dynamic rhytides, BoNT-A is applied to soften the wrinkles. On the other hand, static wrinkles may be treated with dermal filler or skin resurfacing. When administering BoNT-A to the frontalis muscle, care should be taken to stay at least 1–2 cm above the supraorbital rim to avoid brow or eyelid ptosis [31].

The first FDA-approved aesthetic use of BoNT-A was for the treatment of vertical glabellar rhytides in 2002, and it continues to be its most common indication. To evaluate the muscle location, strength, and size, the patient can be asked to frown as much as possible. Asymmetries in muscle strength or contraction must be carefully taken into account before administering the injection. Generally, BoNT-A is injected in a 5-point V pattern, with 2 injections in each corrugator and one in the central procerus. The injections should be kept at least 1 cm above the orbital rim to prevent diffusion into the elevator palpebrae superioris muscle, which could lead to iatrogenic ptosis. It is recommended that a maximum of 20 U be used among the 5 injection sites; however, it can vary slightly in terms of dosage and number of injections in clinical practice [32].

The treatment of the lateral brow is usually incorporated into the overall treatment of the glabellar complex and the frontalis, creating a pleasing brow contour. Injections of BoNT-A into the medial brow depressors can subtly raise the medial and central brow, with an additional lifting effect on the lateral brow [33]. Crow's feet, or lateral canthal lines, are a common sign of aging due to hyperactivity of the orbicularis oculi muscles. The use of BoNT-A injections to reduce the appearance of these wrinkles has been documented. To avoid any unwanted diffusion to nearby muscles, it is recommended to inject at least 1 cm away from the orbital rim, targeting the orbital partition of the orbicularis oculi muscle [34]. Extra caution should be taken when injecting medially to prevent any disruption of proper lid function in patients with lax lower lids.

As it pertains to the middle and lower face, aging is associated with tissue volume loss and descent, making botulinum toxin therapy a less commonly used option, while dermal fillers or surgical tissue resuspension are usually more effective. Furthermore, transverse nasalis muscle hyperactivity can give rise to oblique rhytides, often referred to as bunny lines, which are generally disliked by patients [35]. In the upper lip, dynamic vertical wrinkles are caused by the orbicularis muscle sphincter action, which is often a source of complaint for women. In this area, BoNT-A is rarely used alone, but dermal fillers are commonly employed to improve contour, eversion, and fullness. However, judicious use of BTA in the upper lip has been shown to produce positive results [36].

Treatment of aging-related changes in the midface and lower face and neck generally involve restoring volume and resuspending descending tissues. This can be done through various sites including the depressor anguli oris muscle, which when contracted can pull down the corners of the mouth [37]. An enlarged masseter can give the lower third of the face a squared appearance and give the face a heavy look [38]. The platysma muscle can become lipodystrophic with age and cannot be treated with BoNT-A. However, vertical platysmal bands and horizontal rhizomes can be improved with chemodenervation [39].

The Global Aesthetics Consensus Group has suggested that the middle and lower parts of the face are most amenable to a combined treatment of fillers and botulinum toxin. It has been noted that synergy can be achieved through carefully planned individual treatments. Not only is this approach applicable to the lower face, but also to the upper face and some areas of the midface. The recommended approach is one of individualization, tailored specifically to meet the needs of the patient [40].

Gynaecological use

Since the year 2000, researchers have been exploring the effects and potential therapeutic uses of BoNT in gynaecological pathologies. Several studies have been conducted which focus on the effectiveness of using BoNT to treat chronic pelvic pain, vaginism, dyspareunia, and vulvodynia.

Chronic pelvic pain

Chronic pelvic pain syndrome lasting more than 6 months can be caused by a spasm of the pelvic muscles, especially the elevator ani [41, 42]. This type of pain is conceptualized either as a syndrome of somatic functional pain or as a regional pain syndrome [41].

The visual analogue scale was used to measure the severity of the symptoms (dysmenorrhoea, dyspareunia, and pelvic pain of non-menstrual origin) in women. AbotulinumtoxinA injections into the elevator ani muscle have been found to provide pain relief and improve symptoms in most cases [43]. Additionally, the injection of BoNT-A into the internal obturator muscle may provide beneficial results with fewer side effects, such as motor weakness and bowel or bladder dysfunction, compared to local anaesthetics like bupivacaine [44].

Evidence suggests that BoNT-A may be effective in reducing pelvic floor hypertonia, which may lead to a decrease in some types of pelvic pain. Studies suggest that myelinated and unmyelinated fibres, which are known as nociceptors, are present in muscles and can be sensitized by a variety of substances, including bradykinins, prostaglandin E, substance P, calcitonin gene-related peptide, and adenosine triphosphate. It is believed that BoNT-A can inhibit the release of these substances and reduce the activation of the spinal cord neurons responsible for pain transmission. Additionally, BoNT-A may be able to inhibit spinal motor neurons α and γ, a mechanism that could potentially stop pelvic muscle spasm [45–49].

Vaginism

Vaginismus is a condition characterized by involuntary, recurrent, or persistent contractions of the perineal muscles surrounding the outer third of the vagina [42]. This disorder was first described by Lamont in 1978, and it is divided into 4 degrees [50]. Penetration can be impossible in the most extreme cases of vaginismus, and it is experienced during sexual intercourse and/or when trying to insert a tampon or gynaecological speculum [51]. In 1977, the first reported case of treating vaginismus with BoNT-A was reported, and since then, multiple studies have been conducted to assess the efficacy of this treatment. The results achieved have been encouraging, with improved sexual function and satisfaction observed in affected people [52]. Moreover, electromyographic studies have revealed that vaginismus is linked to decreased rest and poor inhibition when exercising the levator ani muscle [52].

Patients have reported an involuntary spastic contraction of the puborectalis and pubococcygeus muscles when attempting to insert a penis or in response to the mere thought of penetration. This spasm may also be accompanied by tensing of the thigh abductors, rectus abdominis, and gluteus muscles. Botulinum toxin works by blocking the release of acetylcholine at the neuromuscular junction, resulting in denervation and muscle paralysis. Thus, its use in cases of moderate and severe vaginismus can lead to muscle relaxation and reduced pain.

Vestibulodynia

Vulvodynia, a pain disorder affecting premenopausal women, is most commonly manifested by provoked vestibulodynia. This condition is characterized by persistent discomfort and pain around the vaginal opening, which is known as the vestibule. Symptoms can occur when the area is touched, when a tampon is used, during sexual activity, during a pelvic exam, when wearing tight clothing, or even when sitting for extended periods of time. Additionally, inflammation may also be present. The cause of this condition is not well understood, but it appears to involve neurogenic inflammation and sensitization of the vestibular area, hyperactivity of the pelvic floor muscles, and changes in pain regulation [53].

The bulbocavernosus muscle is an ideal site for intravaginal injection due to its proximity to the levator ani. This treatment has been associated with positive effects, which could be explained by 2 theories: decreased hypertonicity of the pelvic floor muscles and reduced pain, as well as the blocking of nociceptive receptors for pain control. Women report minimal discomfort with tampon use, and during and after sexual intercourse, and evidence suggests that the combination of BoNT-A and physical therapy can have a beneficial effect on muscle response [42].

Discussion

Botulinum toxin A application in the aesthetic use has been widely investigated for more than 20 years, and there is strong evidence that it reduces wrinkles within 4 weeks of treatment, even if it probably increases risk of ptosis [54].

In the gynaecological field, on the other hand, there is still much potential for research. As with hyaluronic acid, extensive research has been done on its safe use

for the vulvovaginal atrophy [55, 56], or in pregnancy [57] or during the Sars-CoV-2 pandemic [58], botulinum toxin may also be research cues.

Some studies have been conducted for the treatment of symptomatic pelvic floor myofascial pain with the botulinum toxin obtaining significant reduction in patient-reported pain scores [59]. Other studies have investigated the BoNT effect on vulvodynia and vaginismus. with promising results. The effects of botulinum toxin on gynaecological disorders could be attributed to 2 theories. The first hypothesizes that it reduces pelvic muscle tension and pain by temporarily paralyzing the muscles. The second suggests that it blocks the transmission of pain signals at the nociceptive receptors in the submucosal layers of the vestibule [60]. Randomized trials on the matter are scarce, and additional research on appropriate dosages, injection sites, and periodicity could shed further light on this issue.

Conclusions

On the one hand, the summary of the available evidence that we collected in this overview suggests that the use of Botulinum toxin in gynaecology needs further investigations to clarify indications, feasibility, safety, and cost-effectiveness compared to other available approaches. On the other hand, the robust experience gained in the field of aesthetic medicine may be partially translated in the gynaecological field and pave the way for proper and evidence-based trials. In the current scenario, we take the opportunity to solicit a close collaboration between experts in aesthetic medicine and gynaecology, to adopt clear indications, reproducible clinical application, and implement the best quality management for our women.

Disclosure

The authors report no conflict of interest.

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