8 Medytoxin / Neuronox®

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Summary and Key Features

- Neuronox® is a botulinum toxin type A (BoNT-A) product approved in 23 countries
- Neuronox® shows similar microbiological, physicochemical, and biochemical features to Botox®
- Neuronox® has proven its non-inferiority to Botox® in terms of efficacy and safety in various clinical studies in 1:1 dose ratio
- The recommended dosage of botulinum toxin type A for wrinkle treatment in Asians is usually lower than that in Caucasians
- The combination technique of multiple intradermal injection of botulinum toxin type A with conventional intramuscular injection is widely utilized in Asia for the purpose of facial rejuvenation
- The use of botulinum toxin type A to reduce the volume of muscles such as masseter, temporalis, calf, and deltoid is becoming more popular and promise to have increasingly broader applications

Introduction

Neuronox® (Medytox Inc., Ochang, South Korea) is a botulinum toxin type A (BoNT-A) product first approved in 2006 by the Korean Food and Drug Administration. Since then, it has been approved under different brand names as Botulift®, Siax®, Cunox®, and Meditoxin® in 23 countries world wide (Fig. 8.1). Among currently available BoNT-A products, Neuronox® seems to be one of the viable alternatives to Botox® in terms of microbiological, physicochemical, and biochemical features, and clinical interchangeability in 1:1 dose ratio.

Microbiological, physicochemical, and biochemical profiles

Considering that botulinum toxin is a biological agent produced in living cells, one ought to first analyze the bacterial strain that produced the toxin itself. In the late 1970s, Dr Kyu-Hwan Yang, a microbiologist brought *Clostridium botulinum* to the Korean Advanced Institute of Science and Technology (KAIST) with him after several years of research at the University of Wisconsin. While the C. *botulinum* produced at the University of Wisconsin became the source of Oculinum® – the first licensed botulinum toxin product, later renamed Botox®, the exact same strain at KAIST became the foundation of botulinum toxin research in Korea as well as the source of Neuronox®, which was formulated by a group of scientists from KAIST, who later founded Medytox Inc. As a result the C. *botulinum* type A Hall strain used by Medytox Inc. (GenBank accession number DQ409059) is exactly the same bacterial strain as that used by Allergan (AF488749, 488748, 488747, AF488746, AF488745), and there is no difference between the two deduced amino acid sequences derived from the DNA sequence of the type A toxin complex.

However, differences in manufacturing processes, such as purification and filtration, can result in differences in the bulk toxin even amongst those from the same culture solution. One important physicochemical feature potentially affected by such processes would be the molecular weight. The size exclusion high-performance liquid chromatography (SE-HPLC) analysis of Neuronox® yields a single peak in the chromatogram with an average molecular weight of 904 ± 7 kDa. This is highly comparable with that of Allergan: 880kDa by SE-HPLC analysis and 925 ± 45 kDa by light-scattering analysis.

In addition, the botulinum toxin is formulated in different ways in the final stages. Neuronox® is formulated using a freeze-drying method, whereas Botox® is formulated as vacuum-dried powder. Nevertheless, Neuronox® ultimately has a similar formulation to Botox®, with one vial containing 100 mouse units of the purified toxin complex, 0.9 mg of sodium chloride, and 0.5 mg of human serum albumin.

It is still unclear whether the origin of the bacterial strain, molecular weight, or product formulation is truly clinically significant. But such specifications supported by clinical data could provide outline guidelines to practicing physicians.

Clinical studies for Neuronox®

The safety and efficacy of Neuronox® were compared with Botox® in two randomized, double-blind studies, at

Figure 8.1 Neuronox® is marketed under different brand names such as Botulift®, Siax®, Cunox®, and Meditoxin®.

a 1:1 dose ratio, for the treatment of essential blepharospasm and spasticity in children with cerebral palsy (Kim et al and Yoon et al). Neuronox® was proven to be non-inferior to Botox® in both studies, and there was no significant difference in the safety profiles. Although published studies on Neuronox® are limited to the therapeutic area owing to its recent introduction to the market, several clinical studies using Neuronox® in aesthetic treatment have already been performed and their results are in the process of publication.

The safety and efficacy of Neuronox® in aesthetic applications were presented at the 22nd World Congress of Dermatology in 2011. A total of 49 women with symmetric crow's feet were enrolled in a double-blind, randomized, intraindividual controlled study. Patients' crow's feet were treated with 12 units of Botox® on one side and with same dose of Neuronox® on the other side. Clinical improvement was assessed by blinded investigators based on photographs taken at 1, 4, and 12 weeks post-treatment on a four point grading scale from poor to excellent; at all timepoints, there was no significant difference between two treatments ($p>0.05$). At 4 weeks post-treatment, 95.7% of the sides treated with Botox® and 100% of those treated with Neuronox® demonstrated a good or excellent result, with 42.6% of either group having an 'excellent' result. Also, both products were well tolerated without any clinically significant treatment-related adverse events.

In another clinical trial, Neuronox® was also directly compared with Botox® at 1:1 dose ratio for the treatment of glabellar lines in a double-blind, randomized Phase III study. A total of 314 patients aged 20 to 65 with moderate to severe glabellar lines were randomly treated with 20 units of Neuronox® or Botox® and assessed by blinded investigators. Neuronox® was again proven to be non-inferior to Botox® without any significant difference in any of the efficacy end-points, including live assessment at maximum frown and at rest, photographic assessment, and the subject's self-assessment of improvement and satisfaction for up to 16 weeks. Both products were well tolerated (publication in progress). Interestingly, the response rates at maximum frown at 4 weeks were higher in this study (93.8% in the Neuronox® group and 94.6% in the Botox® group) than those conducted in the USA (76.7% and 83.7% with Botox®) and comparable with those from China and Japan (88.6%, 95.1%, and 94.1% with Botox®). All these studies had the same timepoint, method of assessment, and the definition of 'responder', but different demographics (Asian versus Caucasian).

Based on these clinical data, we could conclude that the efficacy and safety of Neuronox® are comparable with those of Botox® not only in the treatment of blepharospasm and muscle spasticity in cerebral palsy, but also in the treatment of wrinkles such as glabellar lines and crow's feet.

Reconstitution and handling

Neuronox® is supplied as a single-use vial containing 50, 100, or 200 units of BoNT-A complex. The package insert for Neuronox® recommends reconstituting it with non-preserved 0.9% sodium chloride injection. In a 2009 survey of 500 Korean dermatologists and plastic surgeons, 49.8% of responders dilute a 100 unit vial with 2.5 mL, yielding 4 units per 0.1mL, and 20.6% of responders dilute a 100 units vial with 2 mL yielding 5 units per 0.1 mL. Unreconstituted Neuronox® must be stored in a refrigerator at 2–8°C or in a freezer at below –5°C. Once reconstituted, the product should be stored at 2–8°C for later use. The package insert recommends using the reconstituted product within 24 hours.

The use of Neuronox® in Asians

Although many clinicians in Asia use Neuronox® and Botox® interchangeably, their practice does not necessarily coincide with the published guidelines for Botox®. We

identified the following cases with regard to the aesthetic use of Neuronox® in Asians, which is thought to be more due to ethnic differences than to a difference between the products themselves. This might also be valid for other botulinum toxin products.

First, the optimal dose and injection sites for treating wrinkles are different from the guidelines advocated in North America or Europe. For example, when treating forehead wrinkles, parts of the frontalis lateral to the mid-pupillary lines are areas not recommended for Caucasians, but in Asians are routinely treated in conjunction with the medial parts in order to avoid the so called 'samurai eyebrow', referring to the exaggerated elevation of the lateral part of the eyebrows following injection of BoNT-A into the forehead. Because Asians have a wider and flatter face compared with Caucasians, the 'samurai eyebrow' becomes more evident and results in an malevolent appearance in Asians.

Another concern is the optimal dose of BoNT-A for the treatment of wrinkles. Although more recently published recommendations by Carruthers et al in 2004 and 2007 modified the original techniques and decreased dosage of BoNT-A, the optimal initial doses for wrinkle reduction in Asians seems to be still lower than that of Caucasians (Table 8.1). From the above-mentioned data of clinical studies on the treatment of glabellar wrinkles using BoNT-A, we found differences in the dosages of BoNT-A between Asians and Caucasians. Intrinsic ethnic differences in the muscle mass as well as cultural factors, such as Asians being less facially expressive, could be the reason why lower doses are more appropriate.

Secondly, the practice of using multiple intradermal injections of BoNT-A has been widely adopted in Asia, under various names such as 'mesobotox', 'dermotoxin', or 'Botox lift'. Asian physicians have incorporated the technique of multiple intradermal injections of BoNT-A as an additional solution to achieve subtle changes while retaining a natural expression. This is applied with the expectation of not only the reduction of dynamic facial wrinkles but also the reduction of static wrinkles and pore size, as well as creating the so-called perceived 'lifted effect' – even though the actual lifting effects have not been objectively verified.

Table 8.1 Recommended dose of BoNT-A for the reduction of dynamic wrinkles in Asians

Wrinkles	Muscles	Initial dose (in units)
Forehead wrinkles	Frontalis	3–10
Glabellar frown lines	Corrugator and procerus	10–14
Crow's feet	Orbicularis oculi	6–8
Nasal bunny lines	Nasalis	6
Perioral wrinkles	Orbicularis oris	2–4
Cobbled chin	Mentalis	6–8
Neck platysmal bands	Platysma	20–30

The hypothesized effects of multiple intradermal injections can be classified into three categories. The first is the same dynamic wrinkle reduction as delivered by conventional intramuscular injection of BoNT-A. The second is the improvement of static wrinkles and tightening of pores, which imparts shine and a tighter look to the skin. The last is the improvement of facial contours resulting in a lifted and younger appearance via the elevation of the lateral part of eyebrow, sharper definition to the chin line by weakening mentalis and platysma muscle and reducing the volume of the masseter muscle.

The exact mechanism by which the improvement in skin texture, including static wrinkles and pores is attained, has not yet been elucidated. Our hypothesis involves dermal edema resulting from the transient and mild lymphatic insufficiency induced by underlying muscular paralysis and the smooth muscle relaxation of the dermal vasculature. Dermal edema might improve the fine static wrinkles and dilated pores. In our recent study on skin changes post intradermal injections of Neuronox®, its efficacy in static wrinkle depth was demonstrated in a randomized double-blinded, placebo-controlled, split-face clinical study design (publication in progress). Other parameters related to skin changes, such as elasticity, hydration, sebum expression, skin pore size, and skin thickness, failed to show any significant changes. However, we note that this study treated only a limited area of the lateral cheek, and thus is not appropriate for evaluating sebum production and pore size. Further studies with a modified regimen are required for capturing those variables.

Combining multiple intradermal injections with the conventional intramuscular injection seems more promising than using either method in isolation. Conventional intramuscular injections can be used for supplementation in areas where more targeted paralysis is needed (Fig. 8.2). This could tailor a more comprehensive approach to using BoNT for facial rejuvenation.

The third striking feature of BoNT-A treatment in Asians is its expanding role in facial and body contouring. Since Dr Smyth et al first reported the use of BoNT-A to treat masseteric hypertrophy with favorable results, BoNT-A has proven reduce muscle volume through disuse atrophy. This novel method has become a great success among Asians, whose faces tend to be wider or flatter compared with Caucasians and who aspire to having a more oval and slimmer face. Before BoNT-A became widely used for masseter reduction, mandibular angle resection surgery was the mainstay of treatment. Considering the downtime, risks, and costs, BoNT-A treatment for masseteric hypertrophy is far more attractive than surgery. In patients where muscles contribute more to

their lower facial contours than bone or fat, BoNT-A treatment can change their appearance dramatically (Fig. 8.3). Indeed, it is highly suitable for Asians, considering that even normal Asians have masseteric muscles 0.8 cm thick per side.

Usually 10–40 units per side are required, depending on masseteric volume (Table 8.2). In our clinical study using three-dimensional computed tomography, the overall masseter muscle volume had decreased by 27.5% on average at 12 weeks' post-treatment with 20–40 units of Neuronox®. Volume reduction of the lower part of masseter, lying below an imaginary line connecting the anterior tragus and the corner of the mouth, attributed more to the contouring of the lower face, by up to 35.1% (Fig. 8.4) (publication in progress). Other published studies yielded comparable results. The duration of effect varied from 3 to more than 12 months depending on the individual's dietary habits, bruxism, and so forth. Although this procedure is generally considered simple and safe, the possibility of adverse effects should be anticipated before proceeding with treatment and our suggestions considered.

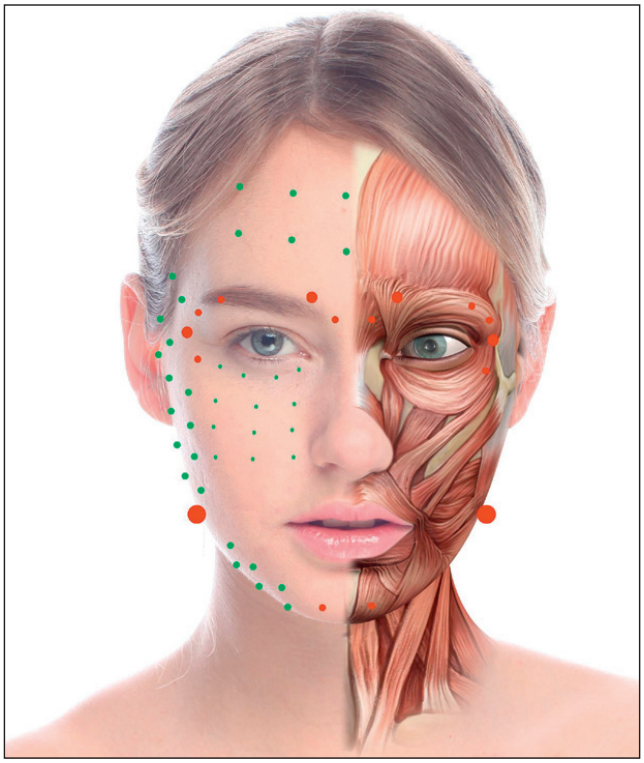

Figure 8.2 A combination of multiple intradermal and conventional intramuscular injections of botulinum toxin type A. The green dots indicate intradermal injection sites (large green dots, 0.4 units; small green dots, 0.2 units) whereas red dots indicate intramuscular injection (large red dots, 5–10 units; medium red dots, 3 units; small red dots, 2 units).

Pearl 1

An unnatural or asymmetric smile after treatment of masseter hypertrophy with BoNT-A is the most embarrassing side effect of this treatment; it seems to occur mainly due to the inadvertent diffusion of BoNT-A into the risorius muscle overlying the masseter muscle. This can be prevented if the toxin is injected into the masseter muscle in sufficient depth by inserting the needle perpendicularly to the skin and advancing it to touch the mandibular bone. One should choose the right needle length, at least half an inch (1.25 cm), for this purpose. Another important tip is to screen out high risk patients using this simple test: those patients whose ears move when they smile fully and pull their lips outward more than others tend to have more developed risorius muscle, and thus are more at risk. We advise trying with a reduced dose of less than 10 units per side in those patients.

Pearl 2

Sunken cheeks are another potential side effect of treating masseter hypertrophy with BoNT-A. This can be aesthetically undesirable because it can impart a gaunt, aged, and fatigued look. However, this condition is due rather to the surfacing of a pre-existing condition that was previously masked by the volume of masseter muscle. Therefore it is important for physicians to mention such pre-existing condition to susceptible patients and to inform them of the need for a subsequent fat graft or filler injection before treatment with BoNT-A.

Table 8.2 Recommended dosage of BoNT-A for the reduction of muscle volume.

Muscle	Dose of botulinum toxin type A per side (in units)
Masseter	10–40
Temporalis	20–40
Gastrocnemius	70–150

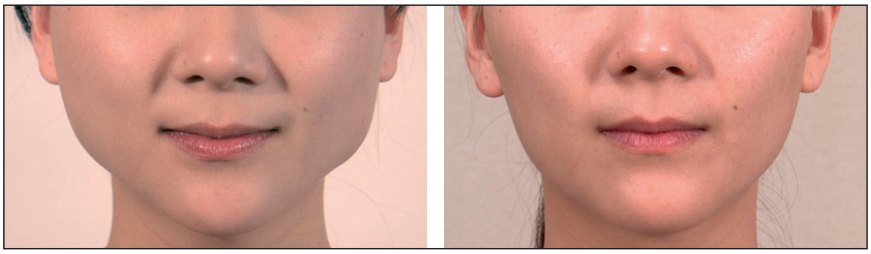

Figure 8.3 Botulinum toxin type A for masseteric hypertrophy dramatically changes lower facial contours: left, pretreatment and right, 3 months post-treatment.

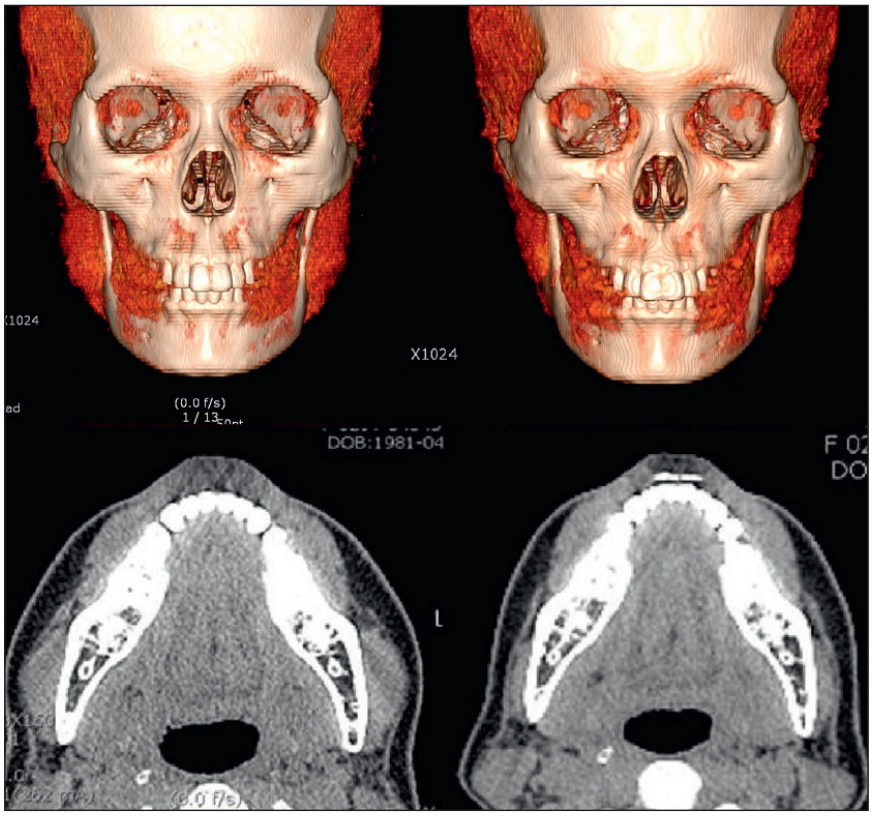

Figure 8.4 Reduction in masseteric volume post-injection with Neuronox® demonstrated on 3-dimensional computed tomography (upper) and computed tomography scans of transverse section (lower). The right masseter muscle was treated with 20 units and the left masseter muscle was treated with 30 units of Neuronox®. In both sides a third of the total muscle volume was reduced from baseline: left, pretreatment, and right, 3 months post-treatment.

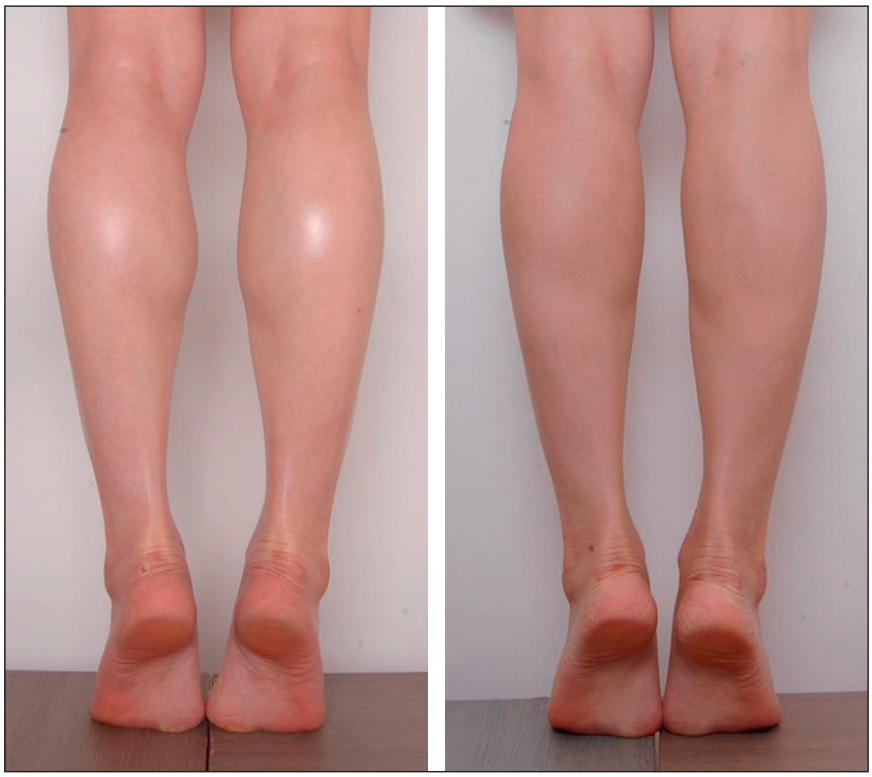

Figure 8.5 Bulging of medial gastrocnemius was improved with 100 unit of BoNT-A injections per side: left, pretreatment; right, 3 months' post-treatment.

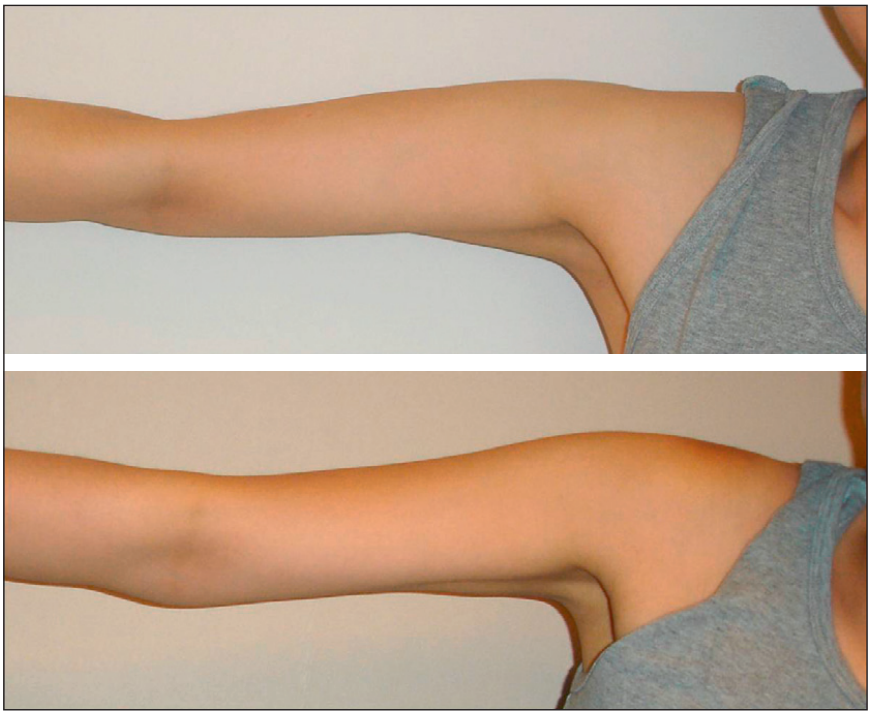

Figure 8.6 Upper arm became slender after injection of BoNT-A (50 unit per side) into deltoid muscle: above, pretreatment; below, 2 months post-treatment.

Pearl 3

Unnatural bulging of focal muscle upon mastication may also occur after treatment of masseter hypertrophy with BoNT-A. This is caused by the imbalance of muscle activity between the muscle fibers affected and those unaffected by BoNT-A. This symptom can be a serious concern because it looks weird to others. This side effect usually begins at 1–3 days after treatment and resolves spontaneously within a month, but an additional injection into the upper or deeper part of the masseter muscle unaffected by the initial treatment can help correct this embarrassing adverse effect more quickly.

Disuse muscular atrophy induced by BoNT-A could be applied to other areas such as the temple, calves and upper arms (see Table 8.2). Hypertrophied temporalis muscle may result in a less attractive bulging appearance, and BoNT-A can create smooth and neat contours by volume reduction of temporalis muscle. Calf muscle reduction has rapidly become popular in Asia, as a close second to masseter reduction in terms of muscle volume reduction by BoNT-A. Patients are more keen on reducing the muscular bulge during walking or in tip-toe position than in the diameter itself. Reducing muscle bulge makes the lower legs look slimmer (Fig. 8.5). Shaping the upper arms by reducing deltoid muscle is another novel indication of BoNT-A utilizing disuse atrophy. With 50 Unit per side, it can deliver impressive results and patient satisfaction (Fig. 8.6).

The rapidly increasing demand for aesthetic procedures has fueled novel techniques and has defined new trends in BoNT-A treatment in Asians. However, to further refine the techniques and clarify any safety issues, the need to establish concrete scientific evidence for these novel indications is paramount.

Further reading

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