3Dose™

White Paper

3Dose 1 ml Syringes

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Document

VMR2026-C178-White Paper-v1
1 Feb 2022

1. SUMMARY

The use of botulinum neurotoxin (toxin) is growing fast, and is used for various Aesthetic (Ax) treatments, because it is a pain-free treatment that makes the clients appearing youthful and healthy without the problems of invasive cosmetic surgery.
This is a cause for concern, since toxin injections are not without risks. For instance, the clinical pharmacology differs in formulation of the products, the formulations are poorly understood and besides that the low dosing volumes are very difficult to administer with the scale print on the barrel of traditional syringes.

Many factors may influence the result of the treatment, including the doses of toxin's. Even physicians with years of experience are facing difficulties with accurate, predictable and reproduceable dosing. That is why accurate dosing syringes enable better treatment results, satisfied patients, and significant costs savings.

For Therapeutic (Tx) treatments the use of botulinum toxin is used in a broad range of treatments like: spasms and migraine treatment.

"Accurate dose injection has become paramount to assure patients reproducibly excellent aesthetic results under the current treatment paradigm"

Dr. Michael S. Kaminer

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2. THE IMPORTANCE OF DOSING

2.1 Increase of the Botulinum Toxin Market

The range of clinical applications of botulinum toxin is growing fast according to Dr. Samizadeh, the founder of the Great British Academy of Aesthetic Medicine and the clinical director of Revivify London clinic, and Dr. De Boulle, who is specialized at dermatosurgery and dermatocosmetology (2018).
Despite the huge decline of non-covid related healthcare services and procedures, the global botulinum toxin market is expected to continue to increase worldwide in the coming years (Fortune Business Insights, 2020).

The increasing demand of minimally invasive cosmetic procedures, increasing R&D initiatives, expansion of Aesthetic (Ax), Therapeutic (Tx) indications and recent regulatory approvals are increasing the demand for toxins. In July 2020, an expanded use of Botox is approved for treatments of spasticity in pediatric for patients of two years and older. Furthermore, Fortune Business Insights noted down Xeomin is approved in Japan in June 2020 and Nabota and Nuceiva are approved in Europe in October 2019. North America contains the largest market of botulinum toxin with an expected revenue of $ 5,127 in 2027 (table 1).

Global Botulinum Toxin Market Revenue Breakdown (USD Mn, %) by Region, 2019-2027				
Region	2019		2027	
	USD	%	USD	%
North America	3.193	66.0%	5.127	66.5%
Europe	934	19.3%	1.457	18.9%
Asia Pacific	407	8.4%	708	9.2%
Latin America	169	3.5%	236	3.1%
Middle East	136	2.8%	179	2.3%
Total	4.839	100%	7.707	100%
CAGR (2020-2027) 7,5%				

Table 1: Source: Fortune Business Insights, 2020

Ophthalmologist Dr. Steven G. Yoelin (2020), board certified dermatologist Dr. Marilyn S. Kwolek and PhD adviser and consultant Dr. Jon Block argued “No other single medical product has revolutionized and quite literally defined the aesthetic medicine field like botulinum toxin injections”.

Toxins are used for novel therapies for a range of neuromuscular conditions (Atassi & Oshima, 1999; Mahant, Clouston & Lorentz, 2000; Glogau, 2000; Thant & Tan, 2003; Bell & Williams, 2003; Atassi, 2004; Benedetto, 2004; Cheng, Chen & Patel, 2006; Mahajan & Brubaker, 2007). Toxins are meeting the need of an easy, pain-free method of appearing youthful and healthy without the problems of invasive cosmetic surgery (Fortune Business Insights, 2020).

Chen (2012) advocates toxins are the most widely used therapeutic proteins, due to its high efficacy, longevity of action and satisfactory safety profile, it has been used empirically in a variety of ophthalmological, gastrointestinal, urological, orthopedic, dermatological, secretory, painful disorders and other treatments (table 1).

Botulinum toxins are often used by specialty, dermatology clinics, hospitals, and other clinics for aesthetic purposes (Ax 43,2%) as well as for therapeutic purposes (Tx 56,8%) (Fortune Business Insights, 2020).

In short, toxins are frequently used for various treatments (table 2).

Clinical Uses of Botulinum Toxin A					
Aesthetic (Ax)	Therapeutic (Tx)				
	Ophthalmology	Neurology	Otolaryngology	Pain	Autonomic Dysfunction
Hyperfunctional facial lines: glabellar frown lines, crown's feet, forehead lines, lateral oblique forehead lines, perioral lines, platysmal bands	Strabismus	Hemifacial spasm	Vocal tics	Migraine	Frey's syndrome
Brow ptosis, brow repositioning	Blepharospasm	Facial asymmetry	Stuttering	Tension headaches	Sialorrhea
Hyperhidrosis: palmar, axillary	Nystagmus	Oromandibular dystonia	Spasmodic dysphonia	Knee, shoulder, neuropathic pain	Rhinorrhea
Facial contouring	Concomitant and non-concomitant misalignment	Cervical dystonia	Oromandibular dystonia	Chronic lower back pain	
Reducing excessive columellar show	Primary or secondary esotropia or exotropia	Spasmodic torticollis	Cricopharyngeal achalasia	Lateral epicondylitis	
Excessive gingiva display	Duane's syndrome	Achalasia	Hemifacial spasm		

Table 2: Source: Samizadeh, De Boulle, 2018

Reversibility is one of the most important properties of toxin; muscles will function again upon clearance of toxin from the affected neuronal cells (Chen, 2012; Samizadeh & De Boulle, 2018). Samizadeh and De Boulle (2018) explain reversibility of toxins is caused by the temporary inhibition of neurotransmitter release, due to toxins cleave on one or two of the three core proteins of the neuroexocytosis apparatus at the peripheral cholinergic nerve terminals.

This process consists of five steps (Rossetto, Pirazzini & Montecucco, 2014; Pirazzini et al., 2017; Rummel, 2015): the toxin is bounded to the peripheral cholinergic nerve terminals, internalization of the toxin, translocation of the light chain, release of the light chain and dissociation of the disulfide bond and cleavage of the SNARE proteins.

Toxin can last for up to six months, which means frequent reapplications are not needed (Chen, 2012).

2.2 Incorrect Use of Botulinum Toxin

So far, toxins seems to be the ideal product, but the fact toxins are reversable does not mean it is without risks. Huang, Foster & Rogachefsky (2000) argue toxins are the most potent toxins known to mankind and can cause botulism, because the human nervous system is susceptible to Botulinum Toxin-A, B, C, E, F, and G. Various short-term effects can be the cause, which include pain, swelling or bruising at the injection site, headache or flu-like symptoms, droopy eyelid or cockeyed eyebrows, crooked smile or drooling, and eye dryness or excessive tearing (Fortune Business Insights, 2020).

According to Samizadeh and De Boulle (2018) the long-term effects include inhibition or blockage of the cholinergic neuromuscular or the cholinergic autonomic innervation of exocrine glands and smooth muscles, reflex inhibition, normalization of reciprocal inhibition, intracortical inhibition and somatosensory evoked potentials. Samizadeh and De Boulle advocate a correct and optimal treatment plan and procedure requires an in-depth knowledge of the product(s) used, anatomy and injection technique.
Samizadeh and De Boulle (2018) argue that increase in number of treatments with toxins are a cause for concern, because the clinical pharmacology and differences in formulation of commonly available products are poorly understood.

> *"small volumes with high doses distributed over a greater number of injection sites are more effective and superior to large injection volume with low dose."*
>
> *Samizadeh & De Boulle*

Brunger, et al. (2007) and Fisher and Montal (2007) confirm that a lot of questions remain regarding the mechanisms of action of toxins. Samizadeh and De Boulle (2018) argue that it is significantly important to proper understand each product, because the products are unique and vary in terms of units, chemical properties, biological activities, and weight, and are therefore not interchangeable. To perform a safe clinical practice and to achieve optimal results, they suppose clinical issues of potency, conversion ratio, toxin spread, and immunogenicity need to be understood by the practitioner. The complex construction of toxins and the complex applying make it hard to have a proper knowledge of the applying of toxins.

First, botulinum toxin is complex due to the consists of a complex mixture of proteins containing botulinum neurotoxin and several nontoxic proteins (Samizadeh & De Boulle, 2018). The single disulfide bridge and its integrity play an integral role in biological activity

of botulinum toxin, making it highly fragile to various environmental variations and influences (Dressler & Benecke, 2007).
The complexity of toxins is caused because they vary in terms, composition, amount of neurotoxin, units, toxin complex size, molecular weight, chemical properties, biological activity, pH, storage, risk of antigenicity, indication of use, geographic distribution (Huang, Foster & Rogachefsky, 2000; Boone, 2015; Matarasso, 2003). Different strains of Clostridium botulinum are what distinguishes the eight distinct toxin serotypes (A-G) from each other (Rossetto, Pirazzini & Monecucco, 2014; Scaglione, 2016). Especially serotypes A and B are widely used for clinical applications according to Samizadeh and De Boulle (2008), because of their long-lasting effects.
Recently, genes encoding for many new toxins are discovered, which are grouped within an existing serotype but with various amino acid sequences. The various structures of toxins indicates the importance of an accurate knowledge of practitioners.

> *"the doses are significantly important, because it affects the efficacy and duration of the effect."*
>
> *Carruthers & Carruthers*

Second, toxins are complex because clinicians have a significant contribution to the result of a treatment (Samizadeh & De Boulle, 2018). Rosales, Bigalke and Dressler (2006) argue that all possible contributing factors must be taken into consideration.
First, intrinsic properties, like the protein load of the available toxins must be examined. Thereafter the muscle must be selected correctly, by paying attention to the muscle activity pattern, muscle architecture and fascial planes.
Last, the injection technique must be correct, which includes a proper dilution, volume, and dose. Especially the doses are significantly important, because it is a stronger predictor of area of paralysis (Shaari & Sanders, 1993), poor response to the treatment (Nestor & Pickett, 2017), substantial paresis (Dressler, et al., 2005), more adverse events (Poewe, et al., 1998) and affect the efficacy and duration of the effect (Carruthers & Carruthers, 2005). When applied correctly and local, toxicity of toxins do spreads hardly or very slowly to the central neuron (Chen, 2012).

2.3 Accurate Dosing

The technique of applying toxins has evolved beyond a one size fits all approach toward the adoption of a tailored treatment protocol, resulting in a more aesthetically pleasing, natural and relaxed appearance (Yoelin, Kwolek & Block, 2020). Accurate dose injection has become paramount to assure patients reproducibly excellent aesthetic results under the current treatment paradigm (Kaminer, et al., 2020). Samizadeh and De Boulle (2018), Yoelin, Kwolek and Block (2020) and Chen (2012) all reported that the combination of small

volumes with high doses distributed over a greater number of injection sites are more effective and superior to large injection volume with low dose.
In this way, the toxin is localized (Huang, Foster & Rogachefsky, 2000; Borodic, et al., 1992), potential antibody resistance is minimized (Samizadeh & De Boulle, 2018) and a more natural appearance was achieved (Kaminer, et al., 2020; Anido, et al., 2017).

On the other hand, a large volume with low dose can weak the muscle and does not result in an overall smoothing effect with an associated risk of toxin spread to adjacent untargeted muscles (Garcia, Fulton, 1996; Edelstein, et al., 1998). Therefore, Bonati and Fabi (2017) argue a unique dosing strategy employing discretely placed, small volume doses are required.

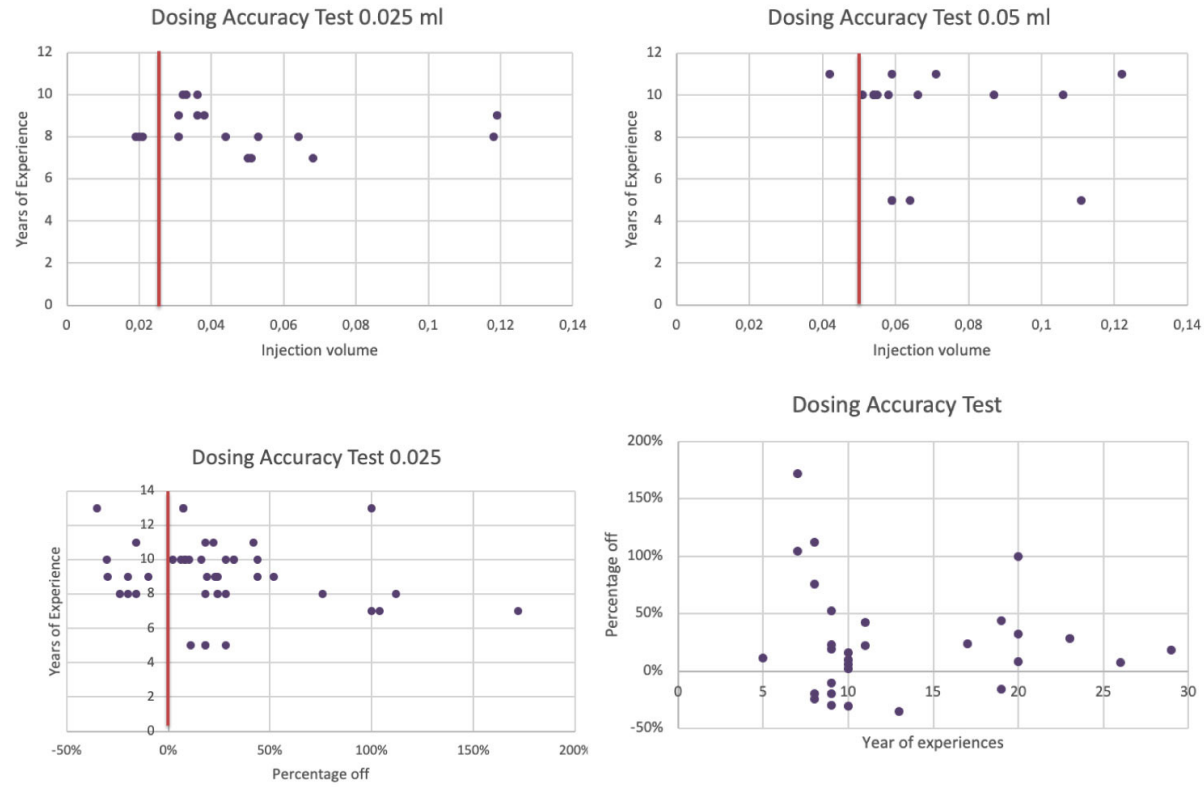
"The average deviation is 24% more than the intended injecting volume."

Study Dr. Dalvi Humzah

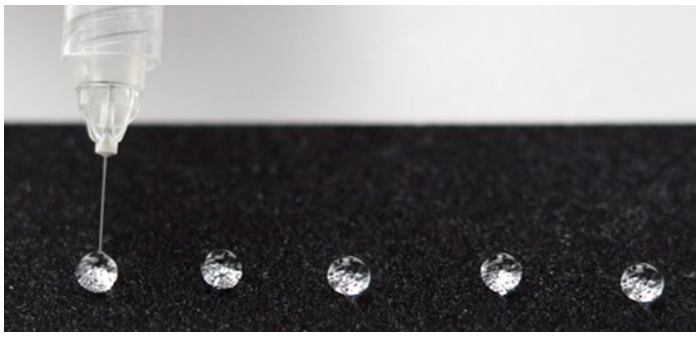
To perform this unique dosing strategy with small volumes, the doses need to be accurate, predictable and reproduceable. The research of Dr. Dalvi Humzah (Consultant Plastic, Reconstructive and Aesthetic Surgeon) commissioned by Vlow Medical (2017) has demonstrated the injection accuracy with the freehand method deviates significantly of the intended amount.

The average deviation is 24% more than the intended injecting volume. One out of ten respondents performing a percentage off, of more than 100% of the intended injecting volume, and all these physicians have at least seven years of experience. The study of Mr. Humzah clarifies even physicians with years of experience are facing difficulties with accurate dosing. Therefore, this product is relevant for all physicians, especially physicians who want to finetune their professionality, willing to pay more attention to the patient.

Kwolek and Block (2019) recently reported the average accuracy error associated with freehand injection of 40 consecutive single units (0.025 ml/unit) from a 1.0 ml disposable syringe was approximately ten percent for two highly experienced injectors. The injection accuracy of smaller doses deteriorates, while small doses are more favorable for optimal results (Yoelin, Kwolek and Block, 2020). That is why Yoelin, Kwolek and Block (2020) plead for a more delicate touch at current treatment paradigm by adopting sufficiently accurate injection techniques to achieve a consistently high quality of aesthetic outcomes "A new level of expertise is required to deliver consistently small-volume doses at varying tissue depths and to have the ability to precisely adjust on the fly to areas that require more and/or differing units of neurotoxin based on patient-specific characteristics".



Source: Dr. Dalvi Humzah, 2017



Source: Vlow Medical – Equal doses, 2017

Yoelin, Kwolek and Block (2020) notice a growing enthusiasm for injection assist devices as an instructional tool for new injectors as well as for experienced injectors to facilitate a broader scope of aesthetic applications (Ax). The 3Dose™ syringe of Vlow Medical is an accurate dosage device with audible and tactile feedback, dedicated for toxin injections. Each click is one dose, which makes it easy to use. “The syringe is easy to use and read number of units for different dilutions” (Samizadeh & De Boulle, 2018). On this way calculating errors in toxin units can be eliminated and it is easily to switch between botulinum toxin brands. The dosing options for the dilution amounts 0.63 ml and 1.25 ml are 0.0125 ml, 0.025 ml and 0.05 ml, the options for the the dilution amounts 1.0 ml and 2.0 ml are 0.01 ml, 0.02 ml and 0.04 ml. According to Yoelin, Kwolek and Block (2020) little or no learning curve is required to use the 3Dose™ syringe of Vlow Medical. Furthermore, the 3Dose™ Syringe in combination with a low deadspace needle reduce the loss of the product in

comparison with traditional needles with a dead space of 0.1 ml (Samizadeh & De Boulle, 2018). According to the Fortune Business Insights (2020), the costs of toxin have increased significantly in recent years. The 3Dose™ Syringe in combination with a TSK™ LDS Low Dead space hub needle results in 0.09 ml of toxin savings.

2.4 Ready-to-use (liquid) toxin combination with accurate dosing

Unlike all other Botulinum Toxin A products currently approved in Europe that come in powder form and require reconstitution with sodium chloride before use, Alluzience™ is a ready-to-use formulation. Galderma says this avoids any calculations and allows for more precision, meaning that the product has the potential to improve safety and dosing accuracy compared with powder- form toxin preparations.

"As the first ready-to-use neurotoxin in Europe, Alluzience™ is fit for the needs of today; it will allow to consistently optimize results and deliver patient satisfaction with more precision."

Dr. Joanna Czuwara

Ready-to-use toxin which has the dilution precision included in the product matches perfectly with 3Dose™ Syringe. The combination assures a guaranteed best delivery of a small doses of the right toxin dilution.

PREDICTABLE perfection®

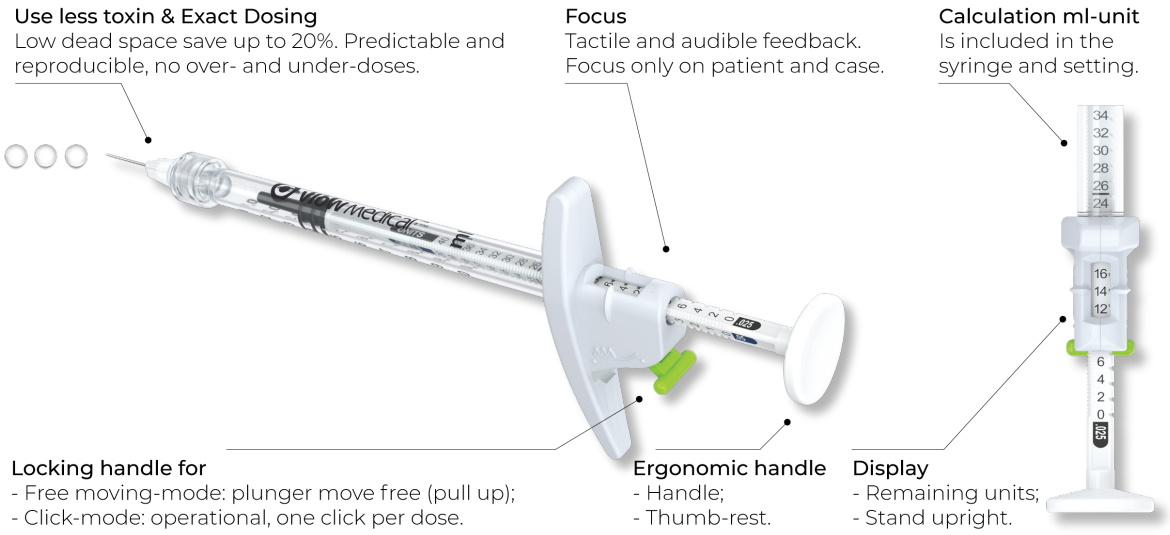
3 3DOSE™ INFORMATION

The use of an accurate dosage syringe is beneficial to both experienced doctors as well as to professionals who inject less frequently.

The 3Dose™ Syringe greatly improves the ease and efficiency of injections which results in improved user convenience and enhanced patient satisfaction. With the 3Dose™ Syringe, calculating units per ml is simplified and calculating errors in units can immediately be eliminated. The focus can now be on the patient where to inject rather than how much to inject.

3Dose 1 ml Syringe 125 Green - Doses: 0.0125 ml, 0.025 ml., 0.05 ml

3Dose 1 ml Syringe 100 Orange - Doses: 0.01 ml, 0.02 ml, 0.04 ml



Dilution Table

3Dose Syringe - green - orange	Vial [units]	Solvent Added Sodium Chloride [ml]	Reconstitution Solution [units/0.1 ml]	3Dose Plunger Setting - green - black - blue	Number of units per click **1 Click =**
Green 3DG125	50 units	1.25 ml	4 units / 0.1 ml	green: 0.0125	0.5 units
				black: 0.025	1 unit
	100 units	2.50 ml		blue: 0.05	2 units
	125 Speywood units	0.625 ml	20 Speywood units / 0.1 ml	black: 0.025	5 Speywood units
	300 Speywood units	1.5 ml		blue: 0.05	10 Speywood units
	500 Speywood units	2.5 ml			
	125 Speywood units	1.25 ml	10 Speywood units / 0.1 ml	blue: 0.05	5 Speywood units
Orange 3DO100	50 units	1.0 ml	5 units / 0.1 ml	green: 0.01	0.5 units
				black: 0.02	1 unit
	100 units	2.0 ml		blue: 0.04	2 units

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