Botulinum Toxin Convergence on Skills for Movement Disorders and Spasticity (ABCs-MDS)

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Botulinum Toxin: History, Pharmacology, Dilution Techniques and Immunogenicity

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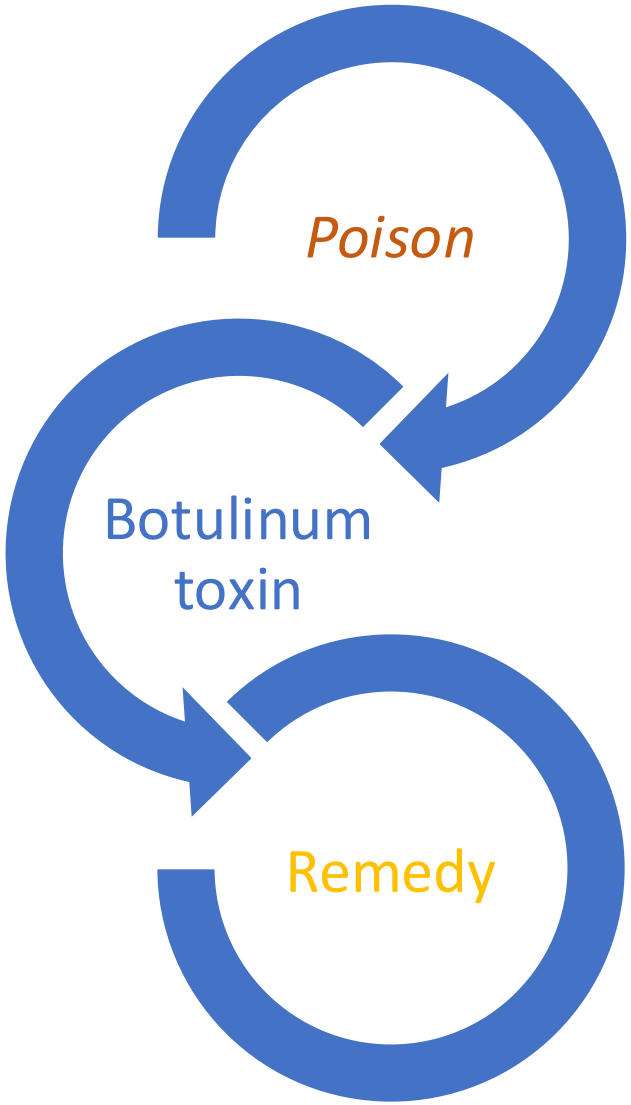
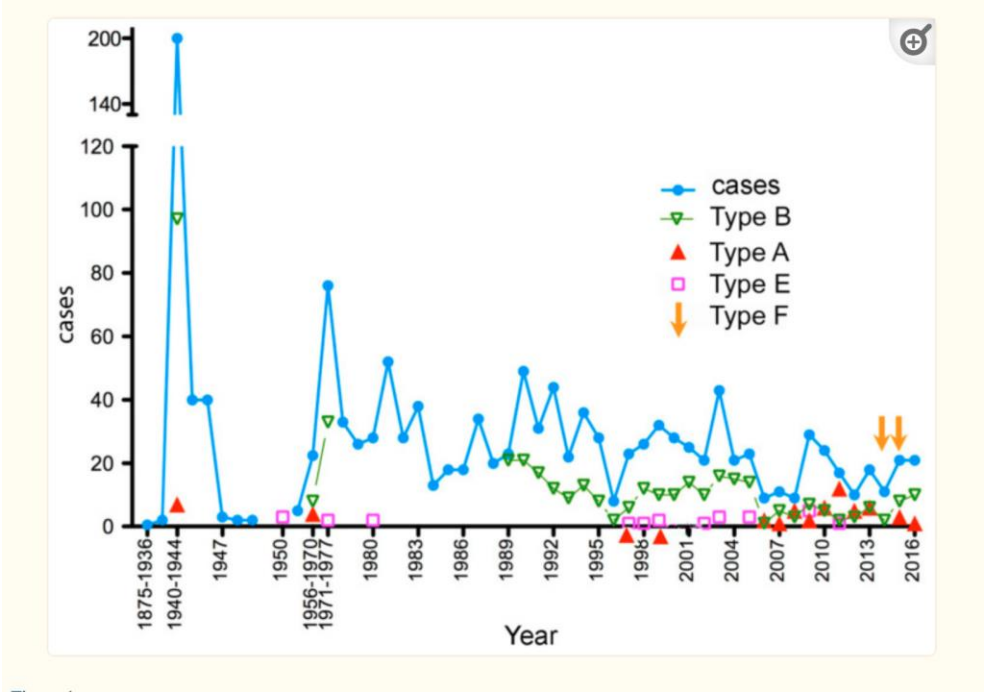
History



From poison to remedy: botulinum toxin

Botulism outbreaks in the 18th and 19th century

- At the end of the 18th century, food poisoning manifested after consumption of *meat and blood sausages* caused many deaths throughout the Kingdom of Wurttemberg.
- Due to the combination of *mydriasis and progressive muscle paralysis in the victims, some physicians suspected an atropine intoxication*, others related the disease to the consumption of meat.



Justinus Kerner's systematic publications: 'Sausage poisoning'

- The German physician and romantic poet Justinus Kerner (1786–1862)
- As a medical officer, the 29-year-old Kerner reported a case of lethal food poisoning in 1817
- Kerner published a first monograph in 1820 on ''sausage poisoning''
- He summarised the case histories of 76 patients and gave a complete clinical description of botulism.

The discovery of ‘Bacillus botulinus’ in Belgium

- In December 1895, there was an extraordinary outbreak of botulism in the small Belgian village of Ellezelles after a funeral meal
- The 34 guests displayed symptoms of botulism
- Three of them died and 10 nearly died.
- The microbiologist Emile Pierre Marie van Ermengem of the University of Ghent conducted the medical investigation
- He correlated ‘‘sausage poisoning’’ with an anaerobic microorganism

Progress in botulinum toxin research

- In 1904, when an outbreak of botulism in Germany was caused by canned white beans
- Two serological subtypes were noted: types A and B
- Type C- 1922
- Type D- 1928
- Type E- 1936
- Type F- 1960
- Type G- 1970

In 1949, Burgen and his colleagues (Burgen et al., 1949) in London discovered that botulinum toxin blocked the release of acetylcholine at neuromuscular junctions.

The potential use of botulinum toxin as a weapon

- World war I and II
- Hooper Foundation, University of California, San Francisco in the 1920s
- With the outbreak of World War II, the United States government began intensive research into biological weapons, including botulinum toxin, especially in the laboratory at Camp Detrick (later named Fort Detrick) in Maryland.
- The methodology was subsequently used by Edward J. Schantz to produce the first batch of toxin which was the basis for the later clinical product

Botulinum toxin in therapy

- After the end of World War II, Schantz worked at Fort Detrick as a civilian.
- In 1972, when Fort Detrick was closed, Schantz continued his research on the use of botulinum toxin at the University of Wisconsin
- Around 1968, Schantz was contacted by Alan Scott, an ophthalmologic surgeon

Development of modern botulinum toxin therapy

- Scott injected into the ocular muscles of monkeys various anesthetics, alcohol, enzymes, enzyme blockers, snake neurotoxins.
- Eventually, motivated by Drachmann's studies, he also tried botulinum toxin.
- With botulinum toxin, he observed a remarkable effect.
- Scott recalled: '*An injection of a few picograms would induce paralysis confined to the target muscles, long in duration, and with no side effects whatsoever'*
- *Human experimentation began first in healthy volunteers and strabismus patients in 1977*

Alan Scott

Hannover, Germany, May, 2018

Scott named the drug Oculinum and formed a company of the same name in 1978.

Oculinum

- Successful studies on the therapeutic value of botulinum toxin in blepharospasm and strabismus led to a first approval of the toxin batch 79-11 (prepared in November 1979) by the US Food and Drug Administration (FDA) as an orphan drug called ‘‘Oculinum’’ in 1989.
- The Allergan company, which had acquired the rights to distribute Oculinum, received FDA approval to change the therapeutic toxin’s name from Oculinum to Botox

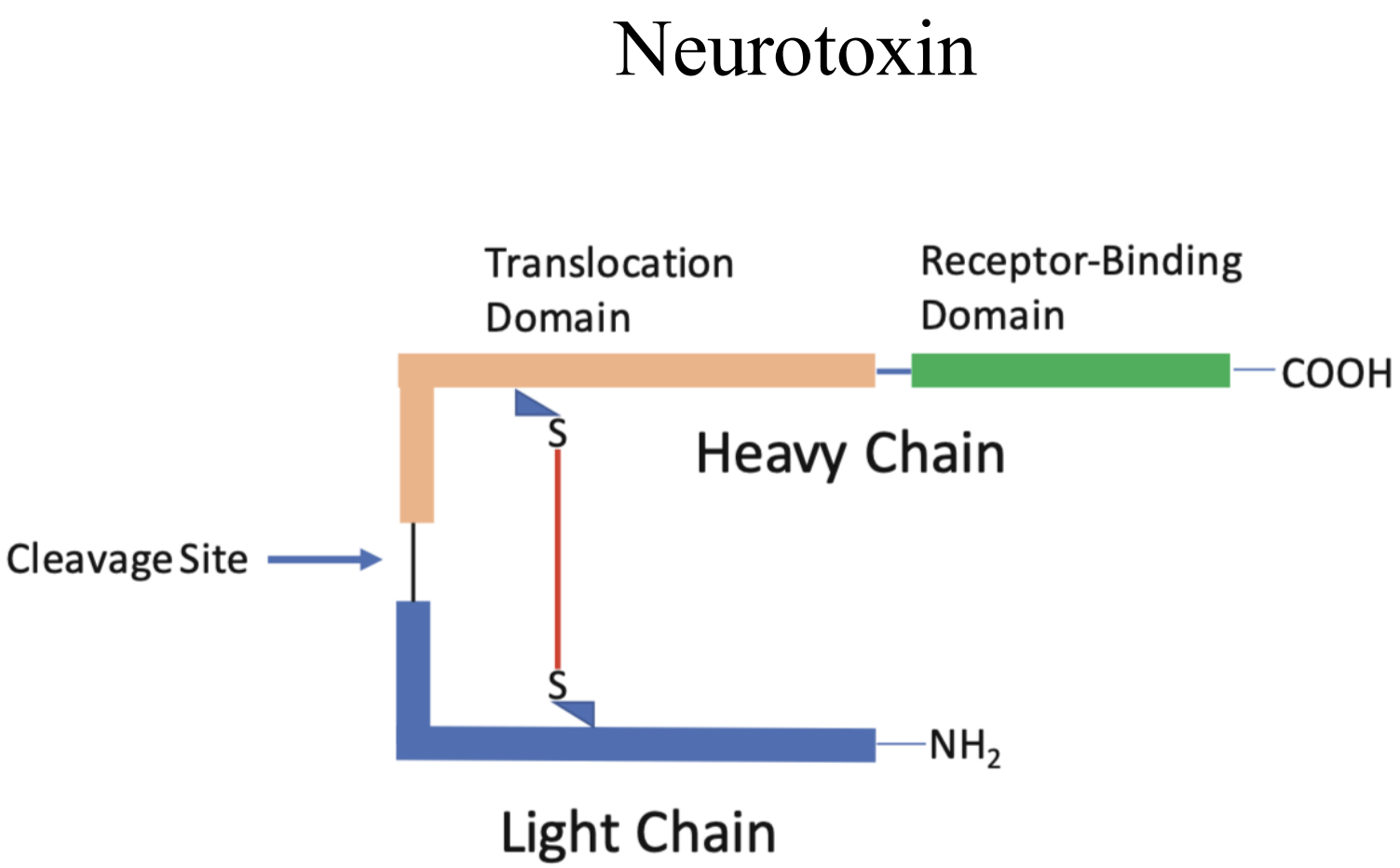
Similar products

- In 1984 Ispen pharma from Europe came with another type A toxin ‘Dysport’
- Indications expanded to to other forms of contractions of smooth and skeletal muscles, as well as to conditions of glandular hypersecretion and pain
- The third officially approved toxin was a type B toxin formulation named MyoblocTM in the United States and Neurobloc+ in Europe
- In Germany, in 2005 another type A preparation (Xeomin+ manufactured by Merz company) that is free of complex non-toxic proteins was approved for treating blepharospasm and cervical dystonia

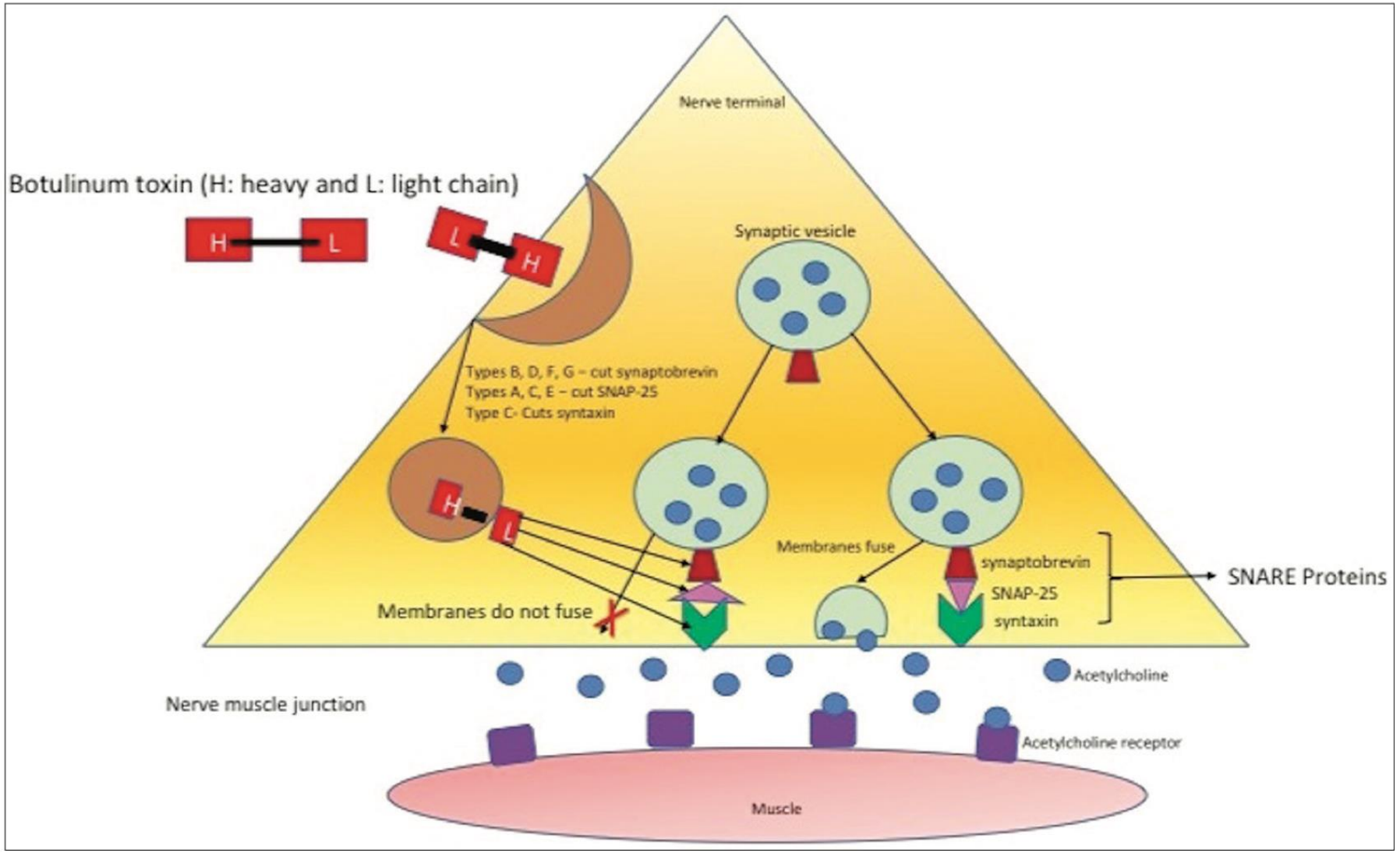
Pharmacology

150 kD core neurotoxins

900 kD complexing proteins



How Does Botulinum Toxin Work?



Tater P, Pandey S. Botulinum toxin in movement disorders. *Neurol India*. 2018;66(Supplement):S79-S89. doi:10.4103/0028-3886.226441

Botulinum toxin works in four areas

- The neuromuscular junction,
- Autonomic ganglia,
- Postganglionic parasympathetic nerve endings,
- Sympathetic nerve endings

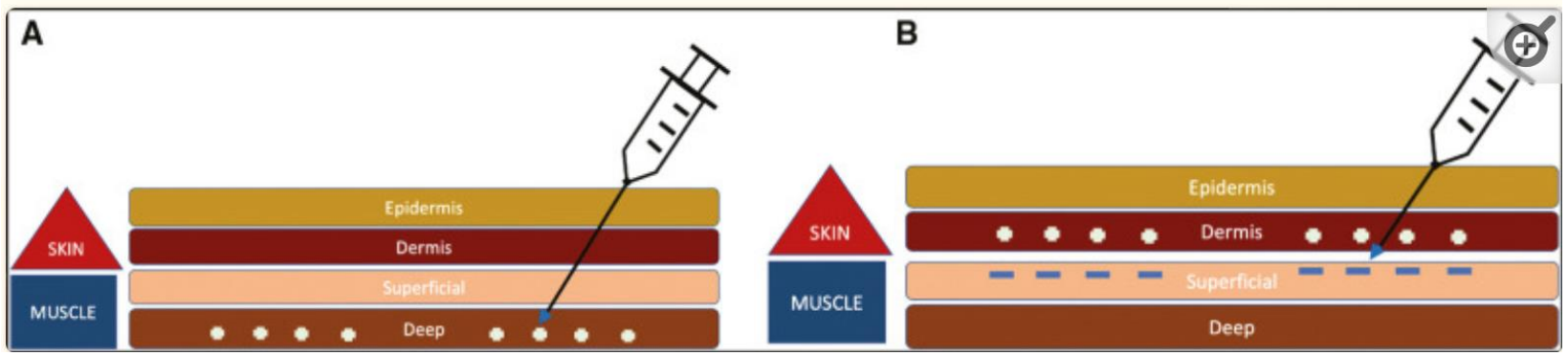
How Does Botulinum Toxin Work? Sensory pathway

Kaji R, Bhatia K, Graybiel AM. Pathogenesis of dystonia: is it of cerebellar or basal ganglia origin? J Neurol Neurosurg Psychiatry. 2018 May;89(5):488-492.

Botulinum toxin (brand name)	Abbreviated indication[a]	FDA approval (year)
OnabotulinumtoxinA (Botox and Botox cosmetic)	Strabismus	1989
	Blepharospasm	1989
	Cervical dystonia	2000
	Glabellar lines	2002
	Axillary hyperhidrosis	2004
	Adult upper limb spasticity	2010
	Chronic migraine	2010
	Urinary incontinence due to detrusor overactivity	2011
	Overactive bladder	2013
	Lateral canthal lines	2013
	Adult lower limb spasticity	2016
	Forehead lines	2017
	Pediatric upper limb spasticity	2019
AbobotulinumtoxinA (Dysport)	Cervical dystonia	2009
	Glabellar lines	2009
	Adult upper limb spasticity	2015
	Pediatric lower limb spasticity	2016
	Adult lower limb spasticity	2017
IncobotulinumtoxinA (Xeomin)	Cervical dystonia	2010
	Blepharospasm	2010
	Glabellar lines	2011
	Adult upper limb spasticity	2015
	Sialorrhea	2018
RimabotulinumtoxinB (Myobloc)	Cervical dystonia	2000

daxibotulinumtoxinA-lanm (Daxxify), approved by the FDA in September 2022 for moderate to severe frown lines

Microdroplet Botulinum Toxin



Traditional technique of administration of botulinum toxin into the deeper fibers of the muscle.

Microdroplet technique showing administration of neurotoxin in intradermal plane, into the superficial muscle fibers thus sparing the deeper muscle fibers.

Aesthetics
Hyperhydrosis

Kandhari et al. Microdroplet Botulinum Toxin: A Review. J Cutan Aesthet Surg. 2022 Apr-Jun;15(2):101-107.

Non-proprietary name: BoNT type	Onabotulinumtoxin A	Abobotulinumtoxin A	Incobotulinumtoxin A	Rimabotulinumtoxin B
Company	Allergan, Inc., Irvine, CA, USA	Ipsen Biopharm Ltd., Wrexham, UK	Merz Pharmaceuticals GmbH, Frankfurt, Germany	Europe/US WorldMeds, Louisville, KY, USA
Trade name	Botox	Dysport	Xeomin	NeuroBloc/Myobloc
Mechanism of action	Cleaves SNAP 25	Cleaves SNAP 25	Cleaves SNAP 25	Cleaves VAMP
Molecular weight, kD	900	500–900	150	700
Dosage form	Spray-dried powder	Freeze-dried powder	Freeze-dried powder	Sterile solution
Shelf life, mo	36	24	36	24
Storage temperature, °C	< 8	< 8	< 25	< 8
pH value after reconstitution	7.4	7.4	7.4	5.6
Excipients	500 μg HSA and 0.9 mg NaCl in 100-U vial	125 μg HSA and 2.5 mg lactose in 500-U vial	1000 μg HSA and 4.7 mg sucrose in 100-U vial	0.5 mg/cc HSA; 0.01 M sodium succinate; 0.1 M NaCl; and SWI in 2,500-U, 5,000-U, and 10,000-U vials
Units per vial	50, 100, 200	300, 500	50, 100	2,500; 5,000; 10,000
Recommended volume of reconstitution	Maximum, 10 mL	Maximum, 1 mM	Maximum, 8 mL	0.5 mL; 1 mL; 2 mL
Total protein, ng/vial	~5	~5	~0.6	~50
Antigenic protein load, ng/vial	~0.8	Unknown	~0.6	~10.7
Biologic activity	100 MU-A/vial	500 MU-I/vial	100 MU-M/vial	1.0/2.5/10.0 kMU-E/vial
Specific activity, U/ng	20	40	167	75–125

Ramirez-Castaneda et al. Diffusion, spread, and migration of botulinum toxin. Mov Disord. 2013 Nov;28(13):1775-83.

Dilution

- Onabotulinumtoxin A and incobotulinum A toxins are known to have a conversion ratio of 1:1 .
- A conversion ratio of 1:2-3 was reported for onabotulinumtoxin A/incobotulinum A and abobotulinumtoxin A
- In addition, a conversion ratio of 1:50 was reported for onabotulinumtoxin A/incobotulinum A and rimabotulinumtoxin B

Dilution

100 units [Ona and Inco] 300 units [Abo]

1 ml----------2ml----------2.5 ml----------4 ml

Little diffusion Larger diffusion

Smaller muscle Larger muscle

Hyperhidrosis

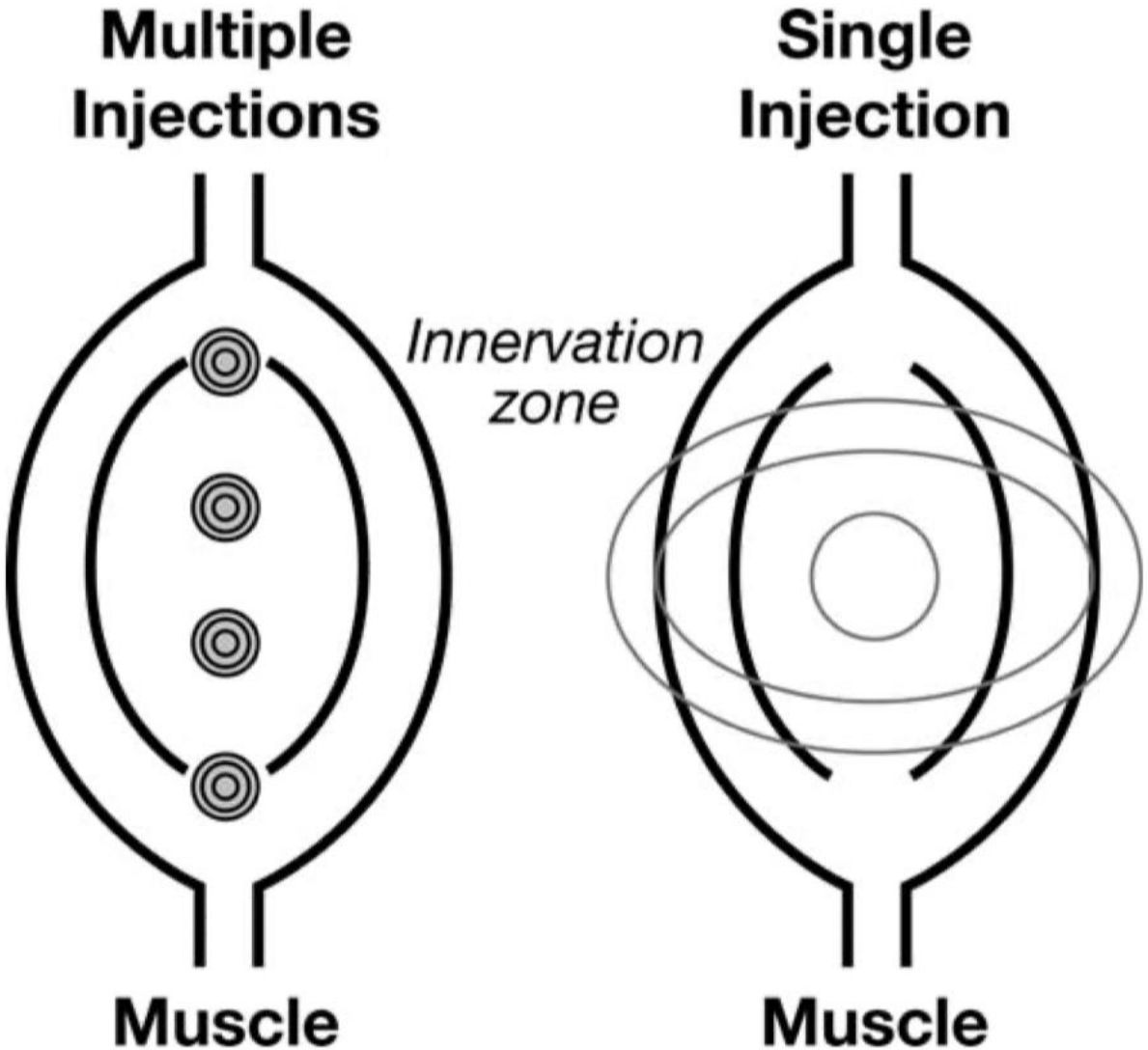
Choose a Syringe, So That 1 Dash Equals 1 Unit

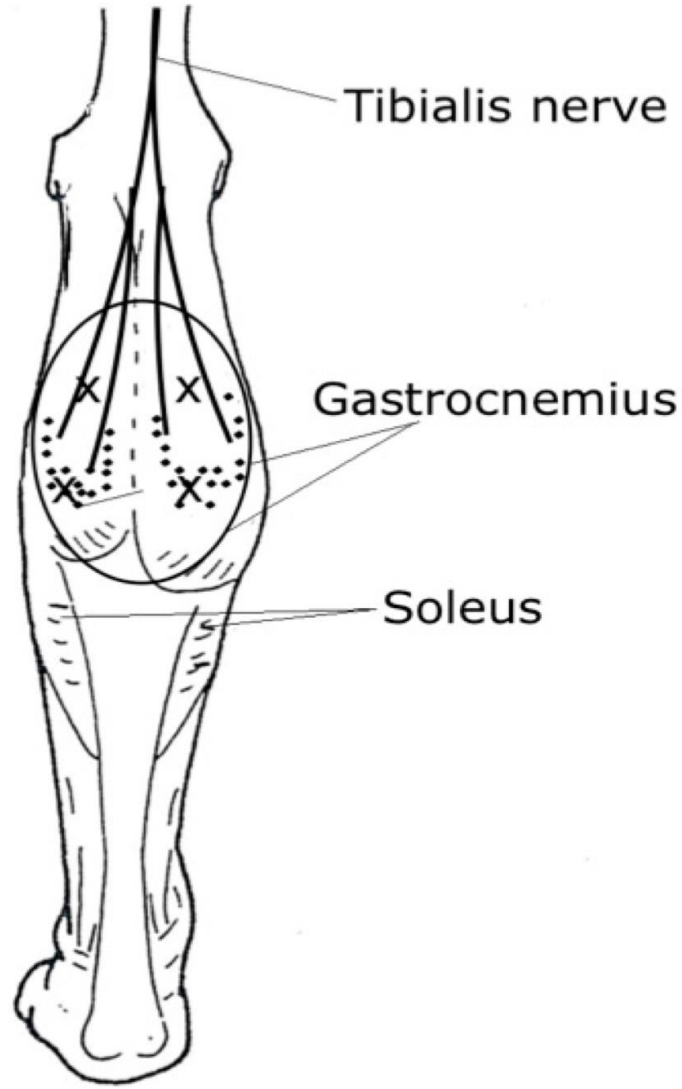
Ona and Incobotulinum toxin

1 ml Dilution	2 ml Dilution	2.5 ml Dilution	4 ml Dilution
Each 0.1 ml = 10 u	Each 0.1 ml = 5 u	Each 0.1 ml = 4 u	Each 0.1 ml = 2.5 u
0.25 ml = 25 u	0.5 ml = 25 u	0.5 ml = 20 u	1 ml = 25 u
0.5 ml = 50 u	1.0 ml = 50 u	1.0 ml = 40 u	2 ml = 50 u
0.75 ml = 75 u	1.5 ml = 75 u	1.5 ml = 60 u	3 ml = 75 u
1.0 ml = 100 u	2.0 ml = 100 u	2.0 ml = 80 u	4 ml = 100 u
		2.5 ml = 100 u	

Abobotulinum toxin

1 ml Dilution	2 ml Dilution	2.5 ml Dilution	4 ml Dilution
0.25 ml = 75 u	0.5 ml = 75 u	0.5 ml = 60 u	1 ml = 75 u
0.5 ml = 150 u	1.0 ml = 150 u	1.0 ml = 120 u	2 ml = 150 u
0.75 ml = 225 u	1.5 ml = 225 u	1.5 ml = 180 u	3 ml = 225 u
1.0 ml = 300 u	2.0 ml = 300 u	2.0 ml = 240 u	4 ml = 300 u
		2.5 ml = 300 u	





Ramirez-Castaneda et al. Diffusion, spread, and migration of botulinum toxin. Mov Disord. 2013 Nov;28(13):1775-83.

The base for dose calculation and dose modifiers

Base for Dose Calculation:
Total units per treatment session
Total units per kg body weight per session
Units per muscle
Units per injection site
Units per kg body weight per muscle (U/kg/muscle)

Dose Modifiers:
Weight of the patient
Number of muscles needing treatment
Size and activity of the muscle
Knowledge of the muscle configuration and MEP distribution
Severity of spasticity/dystonia
The goal of the treatment
Comorbidities (e.g., dysphagia, aspiration, and breathing problems)
Dynamic vs fibrotic muscle
Experience from previous injections

Sätilä H. Over 25 Years of Pediatric Botulinum Toxin Treatments: What Have We Learned from Injection Techniques, Doses, Dilutions, and Recovery of Repeated Injections? Toxins (Basel). 2020 Jul 6;12(7):440.

Immunogenicity

	Complexing proteins
150 kD core neurotoxins	Hemagglutinin (HA)
	Non-hemagglutinin (NTNHA) proteins
900 kD complexing proteins	three HA proteins are HA1 (=HA-33), HA2 (=HA-17), and HA3 (=HA-70), and HA3 is divided into HA3a and HA3b
	HA-1 (HA-33), which is the largest component of the complexing proteins, is known to be the most important protein for the immunogenicity of botulinum toxin

Complexing protein

- Unlike onabotulinumtoxin and abobotulinumtoxin, incobotulinumtoxin is composed of only neurotoxins without complexing proteins
- complexing proteins are related to the interleukin (IL)-6 pathway and stimulate an immune response by inducing the secretion of inflammatory cytokines
- Increases the antigenic protein load and is a risk factor for the formulation of neutralizing antibodies, which leads to treatment failure

Treatment failure

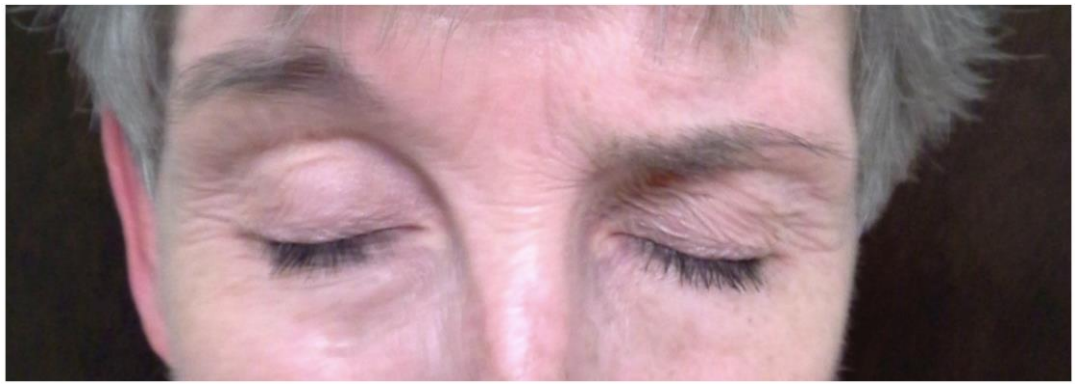
- Treatment failure (non-response) reflects the patient's and/or the physician's lack of satisfaction with the outcome of treatment, implying that the treatment is partially or completely ineffective
- Primary and secondary
- Primary failure is defined as less than a 25% response despite an increase in dose or 2-3 trials of injections
- Primary failure may be due to low sensitivity to botulinum toxin, misdiagnosis, an insufficient dose, and incorrect injection into other muscles

Secondary treatment failure

- Secondary failure is observed when the toxin is effective at the first injection but has no effect afterward
- This may be due to deterioration of the disease, or presence of neutralizing antibodies and immunogenic proteins
- Presence of neutralizing antibodies and immunogenicity may be determined by the injection session dose, injection interval, cumulative dose, number of injections, previous injection history, and botulinum toxin formulation

Tools to detect neutralizing antibodies

- Pharmacological: bioassays such as the mouse protection assay (MPA) and mouse hemidiaphragm assay (MHDA) are used to identify neutralizing antibodies in animals
- MPA has low sensitivity and MHDA has low specificity
- Clinical detection is easier to implement: injected into the right medial eyebrow: onabotulinumtoxinA; incobotulinumtoxinA [20 U], or abobotulinumtoxinA [50 U], or rimabotulinumtoxinB [1000 U].



After 15 days

Bellows S, Jankovic J. Immunogenicity associated with botulinum toxin treatment. Toxins (Basel) 2019;11:491.

Steps to prevent and overcome treatment failure

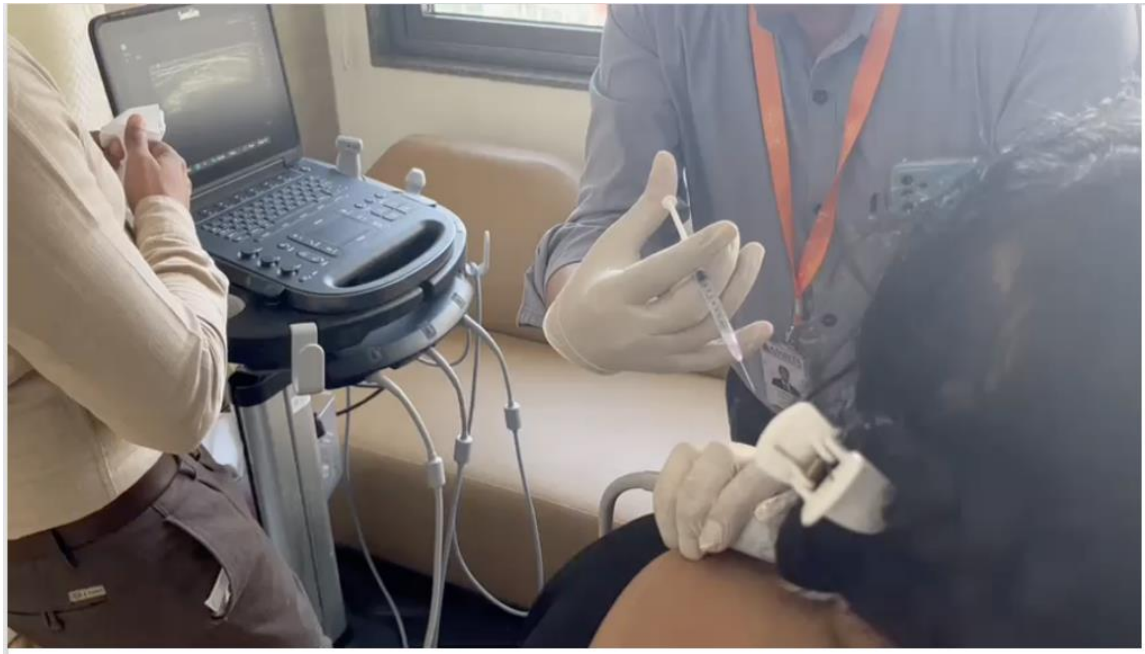
- Therefore, to reduce antigen occurrence, a product with a low risk of antigenicity should be selected.
- to use the product at the minimum dose, avoid additional booster injections, reduce the single injection dose, and perform injections at intervals of at least 3 months

Injection-techniques

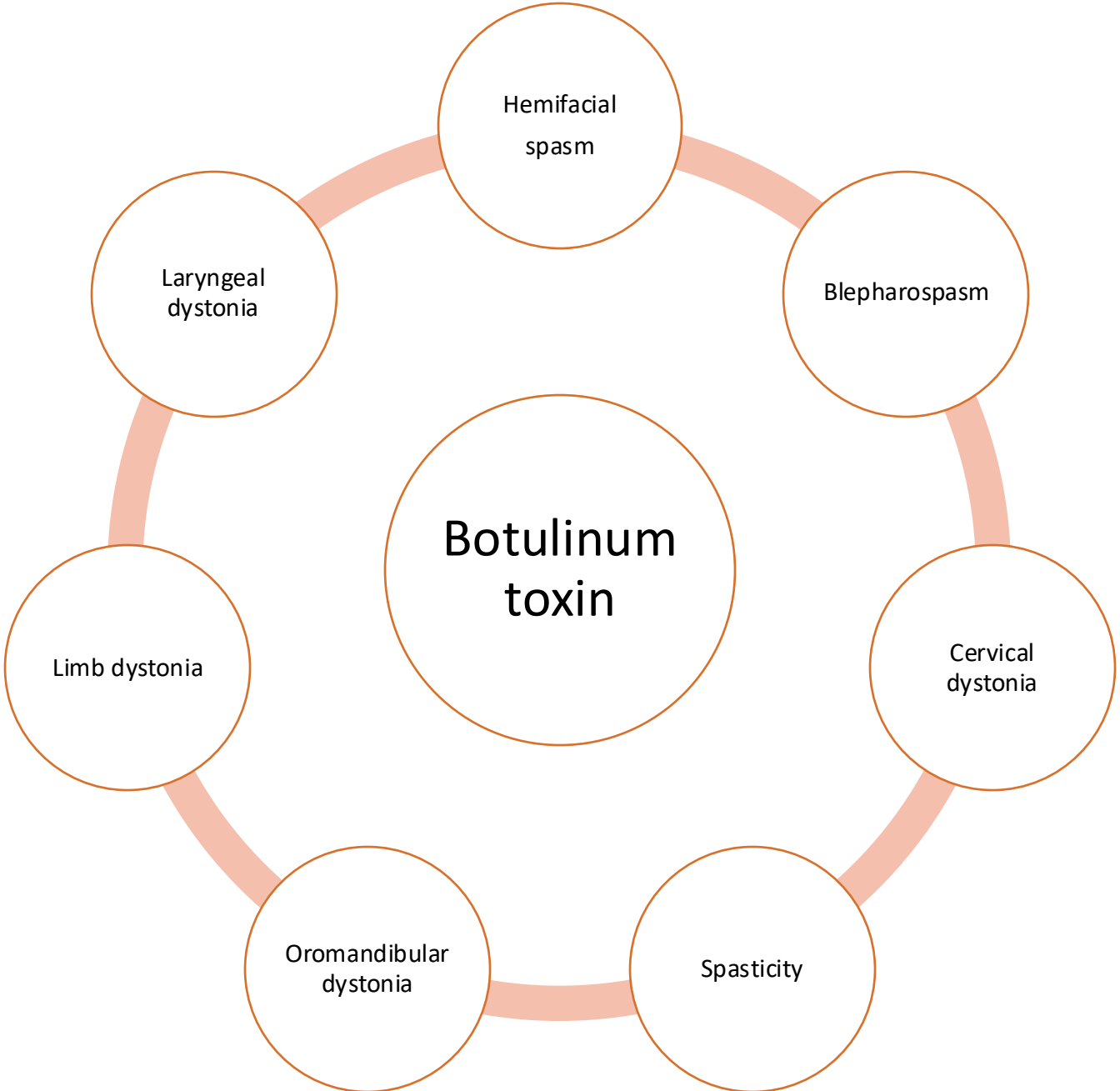
- Anatomical guidance
- Electromyography (EMG)
- Electrical Stimulation (E-Stim)
- Ultrasound

EMG + ES

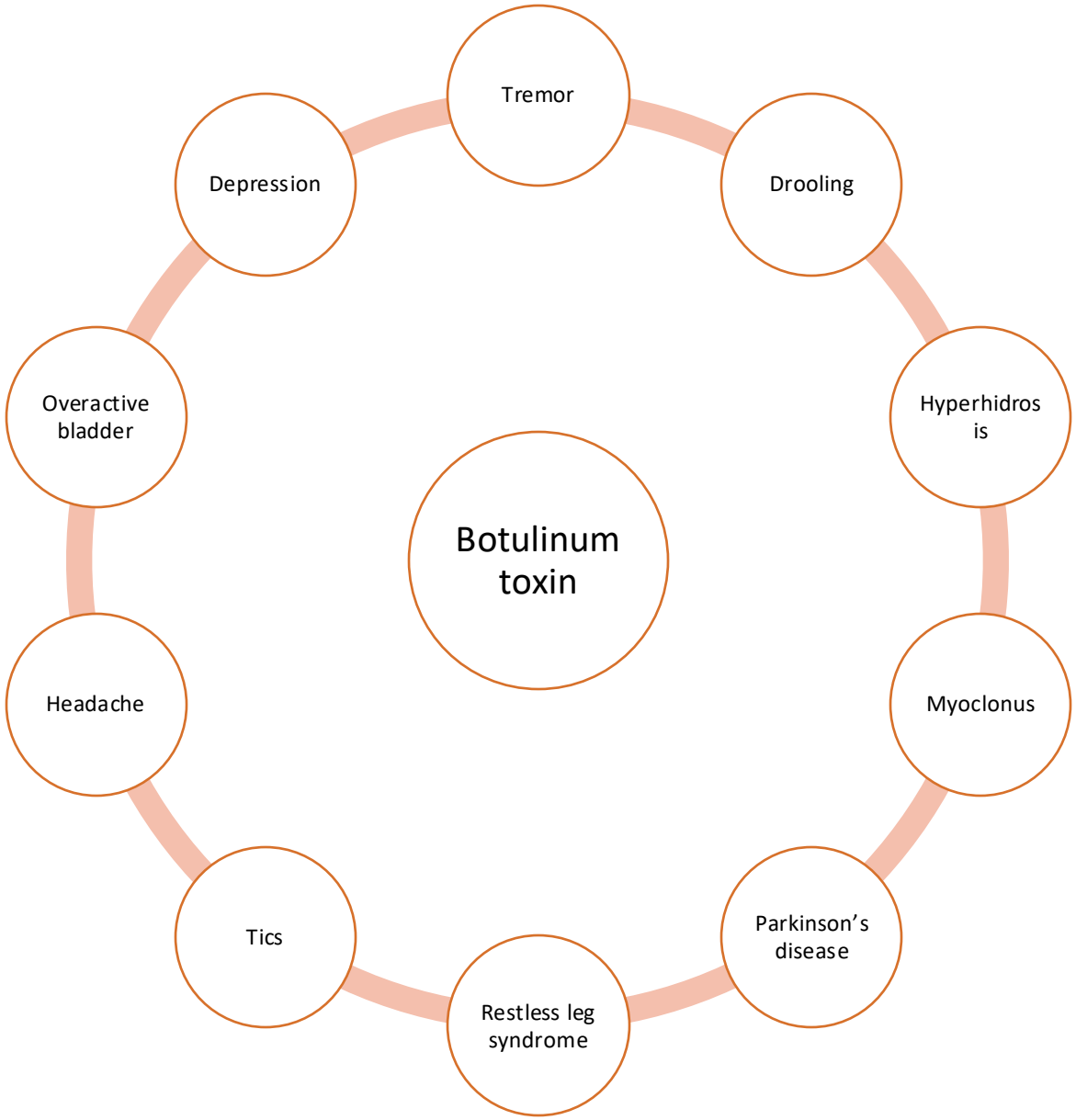
Ultrasonography



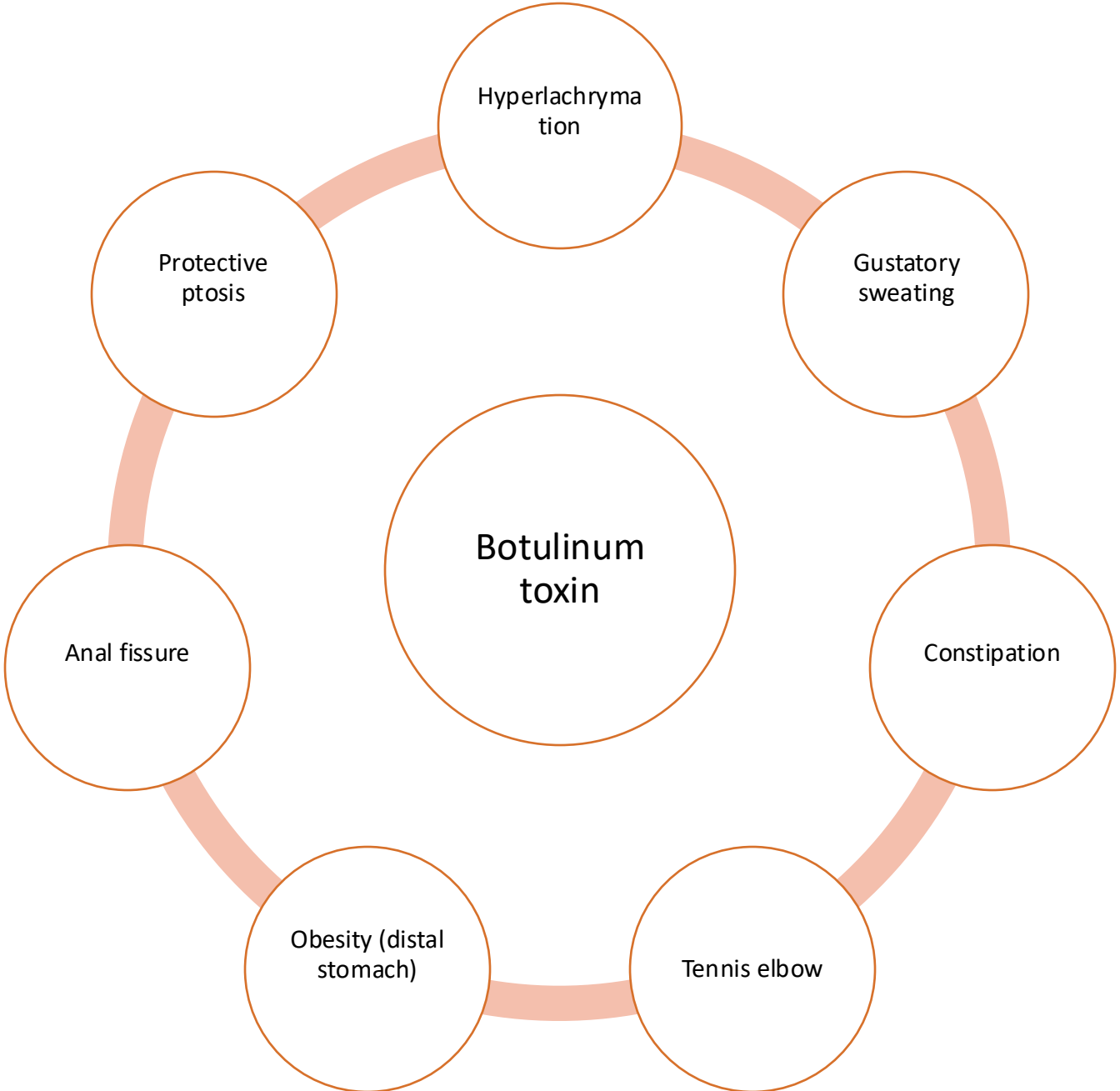
Botulinum toxin in movement disorders: Common indications



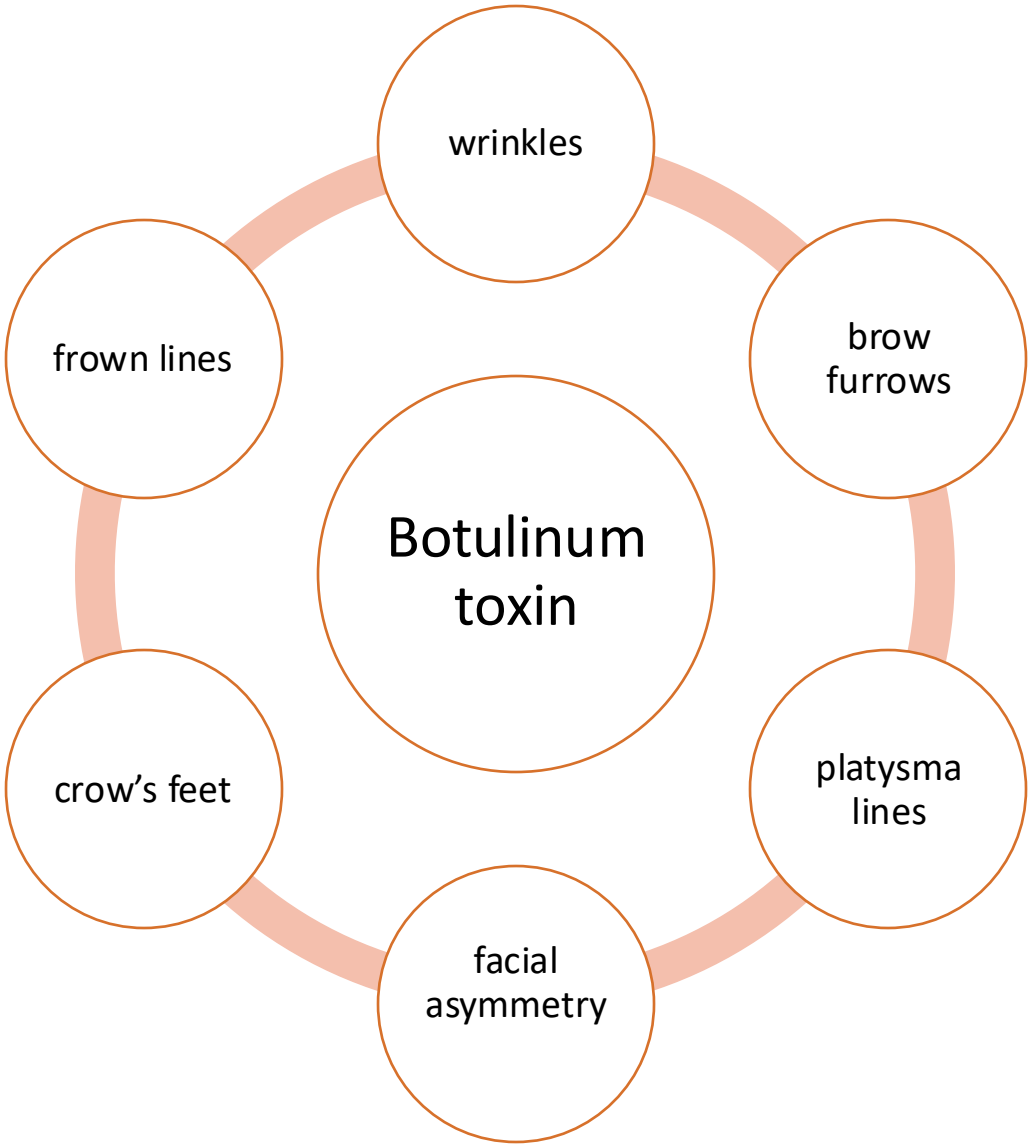
Botulinum toxin: Rare neurological indications



Botulinum toxin: Other Indications

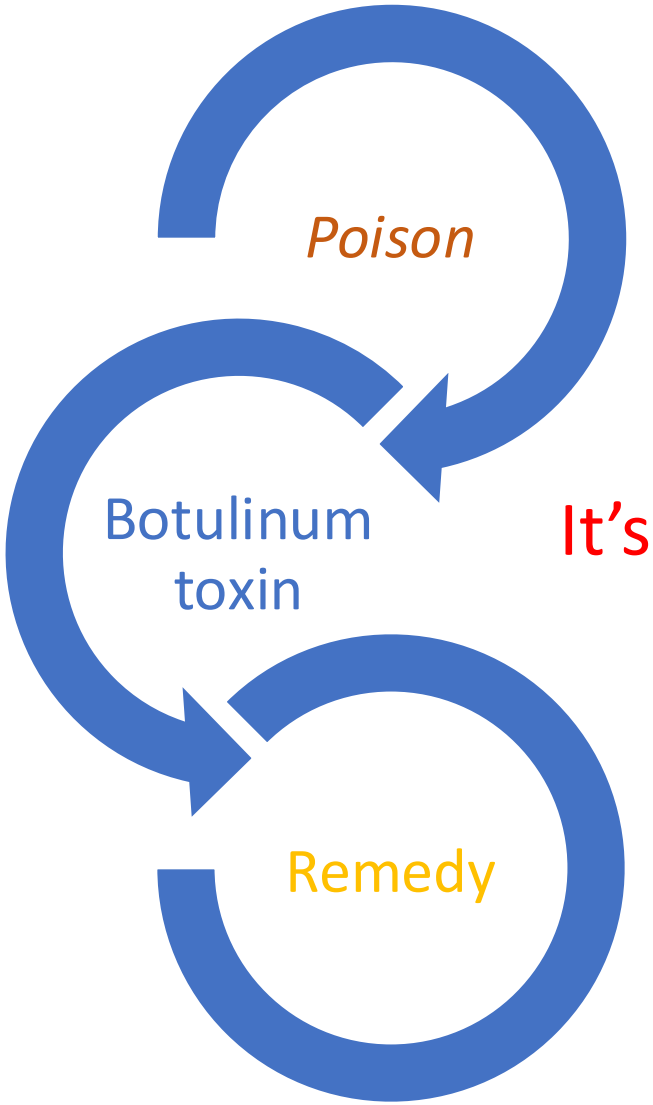


Botulinum toxin: Cosmetic indications



The Drug That's Treating Everything

Thanks



It’s a remarkable arc for a drug with so many indications

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DYSTONIA COALITION

Collis-Caput-Collaboration

Mark Hallett

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सत्यमेव जयते

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