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ORIGINAL ARTICLE

Using translational medicine to understand clinical differences between botulinum toxin formulations

K. R. Aoki[a], D. Ranoux[b] and J. Wissel[c]

[a]*Department of Biological Sciences, Neurotoxin Research Program, Allergan Inc., Irvine, CA, USA;* [b]*Service de Neurologie, Centre Hospitalier Sainte-Anne, Paris Cedex 14, France; and* [c]*Neurological Rehabilitation Clinic, Berlin and Beelitz, Germany*

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When using botulinum toxin-based products, the physician must decide the optimal location and dose required to alleviate symptoms and improve the patient's quality of life. To deliver effective treatment, the physician needs to understand the importance of accurate target muscle selection and localization and the implications of each product's migration properties when diluted in different volumes. Pre-clinical mouse models of efficacy and safety have been utilized to compare local and distal muscle relaxation effects following defined intramuscular administration. Data from the model allow the products to be ranked based on their propensity for local efficacy versus their distal migration properties. Using standardized dilutions, the non-parallel dose–response curves for the various formulations demonstrate that they have different efficacy profiles. Distal effects were also noted at different treatment doses, which are reflected in the different safety and/or therapeutic margins. Based on these pre-clinical data, the safety and therapeutic margin rankings are ordered, largest to smallest, as BOTOX®, Dysport® and Myobloc®. The results of subsequent clinical trials are variable and dose comparisons are inconclusive, thus supporting the regulatory position that the dose units of the individual preparations are unique and cannot be simply converted between products.

Introduction

Botulinum toxin (BoNT) has been used for approximately 30 years in a variety of indications, initially in ophthalmological conditions such as strabismus and blepharospasm, and more recently in indications of spasticity, hyperhidrosis and migraine headaches. Its use is supported by a wealth of pre-clinical and clinical trial data, resulting in a high degree of confidence in its current clinical application.

There are currently three commercially available preparations of BoNT type-A (BoNT-A) in Europe – BOTOX® (manufactured by Allergan Inc., Irvine, California, USA), Dysport® (manufactured by Ipsen Limited, Berkshire, UK) and Xeomin® (manufactured by Merz Pharmaceuticals GmbH, Frankfurt am Main, Germany, and currently only available in Germany). The only approved BoNT type-B (BoNT-B) formulation is known as Myobloc® in the United States and as Neurobloc® outside of the United States. Although they are all BoNT formulations, there are differences between the products that the treating physician must understand to ensure that each is used safely and effectively. For more than a decade, there has been ongoing debate concerning the comparative effectiveness of these commercially available BoNT-A formulations and whether a conversion factor or dose ratio can be established between the products. The reasons for wanting such a conversion factor are twofold. Firstly, patients can request to switch to another formulation based on advertising of one particular product, because of side effects or lack of efficacy of their existing treatment. In these cases, a simple dose conversion would be beneficial instead of having to re-establish the effective dose (ED_{50}) in the patient with the new formulation. Secondly, there are commercial implications for fixed ratios, as a dose ratio of 4:1 means the costs of BOTOX treatment would be lower than the costs of Dysport treatment, but a dose ratio of 3:1 would make the cost of BOTOX treatment higher than that of Dysport.

Data that have been used to try to determine a dose equivalence ratio between BOTOX and Dysport have come from both pre-clinical and clinical sources. However, to date, there have been no randomized, controlled studies in which doses are titrated to the same effect and safety to establish a suitable dose ratio.

Given the current availability of three formulations in Europe and the increasing pre-clinical and clinical data in this area, it seemed an opportune time to re-evaluate the characteristics of each formulation, thus

Correspondence: Dr K. R. Aoki, Department of Biological Sciences, Neurotoxin Research Program, Allergan Inc., 2525 Dupont Drive, Irvine, CA 92612-1599, USA (tel.: +1 714 2465413; fax: +1 714 246 4374; e-mail: aoki_roger@allergan.com).

enabling the clinician to make educated treatment decisions and optimize safety and efficacy on an up-to-date knowledge base.

Properties of botulinum toxin

Botulinum toxin is very safe and effective, primarily because it is a highly specific and selective treatment administered locally in very small doses. The specificity of the BoNT treatment is conferred by its injection at a localized target site, thereby minimizing systemic exposure [1]. The selectivity of BoNT treatment is due to the unique mode of action of the toxin, which binds with high affinity to cell ecto-acceptors, which then allow it to enter the target nerve cells [2–8]. This means that the toxin only acts on cells expressing the appropriate ecto-acceptor on their surface [8–10]. The combination of these characteristics means that low doses of BoNT can be administered locally at the required site of action and that the toxin will only act on the appropriate nerve cells within that tissue. As a result, BoNT has an extensive history of safe use in a number of clinical indications and over extended time-periods (for review of Botox safety see Ref. [11]).

The characteristics of the clinically approved BoNT formulations differ in terms of physical form and the diluents and vehicles used, as summarized in Table 1. BOTOX (manufactured by Allergan) was approved by the US Food and Drug Administration for human use in 1989 and is manufactured with a 900-kDa complex of toxin serotype A (BoNT-A). The commercial BOTOX preparation is supplied as a vacuum-dried powder at approximately 5 ng/vial. Dysport (manufactured by Ipsen) was first licensed in 1991 in the United Kingdom and also comprises a complex of toxin serotype A, but has a molecular mass ranging from 500 to 900 kDa. Xeomin (manufactured by Merz and licensed since 2005 in Germany) also consists of toxin serotype A, but in the form of the naked toxin (150 kDa). The three commercial preparations of BoNT-A are supplied in freeze-dried or lyophilized form, which rapidly equilibrates to form a solution of neutral pH on reconstitution. The only approved toxin serotype B (BoNT-B) formulation is known as Myobloc in the United States and as Neurobloc outside of the United States, and consists of a 700-kDa complex, supplied as a solution maintained at pH 5.6 to preserve activity. Only BOTOX and Myobloc are licensed for use in the United States, in contrast to Europe where BOTOX, Dysport, Xeomin and Neurobloc are all available (Xeomin is currently only available in Germany). In addition, another toxin serotype A called BTX-A is produced in China and is being used in an increasing number of countries.

Characteristics of different botulinum toxin formulations

As the different BoNT products are manufactured using different purification methods and formulations, they are unlikely to be clinically equivalent. Translational medicine aims to assess the physical characteristics of the various BoNT formulations and to develop hypotheses as to the pharmacological, biochemical and physiological basis of efficacy and safety differences between products. In addition, pre-clinical studies are used to consider the characteristics of different BoNT formulations in terms of dose–response profiles and to assess how the dose administered correlates with local treatment efficacy and the occurrence of distal effects.

Botulinum toxin complex size

The size of the BoNT complex present in the different serotype A formulations is more likely to account for some of the pre-clinical and clinical differences seen when, for example, comparing BOTOX and Dysport. The production of BOTOX uses the crystallization method of Schantz [12] and results in a product that exclusively comprises a 900-kDa complex. The large size of the 900-kDa complex limits fluid-based diffusion of the toxin within the target muscle. The molecular

Table 1 The characteristics of commercially available botulinum toxin formulations

Formulation (license date)	BOTOX (1989)	Dysport (1991)	Myobloc (2000)	Xeomin (2005)
Serotype	A	A	B	A
Molecular mass of complex (kDa)	900	> 500	700 (500–900)	150
Package size (units)	100	500	2500 5000 10 000	100
Amount of neurotoxin (ng/vial)	~5	12.5	25 50 100	0.6
Form	Vacuum-dried	Lyophilized	Solution	Lyophilized
pH	~7	~7	5.6	~7

mass of isolated BoNT-A is 150 kDa; however, in its native form, the toxin associates with accessory proteins to produce various sized complexes ranging from 300 to 900 kDa [13,14]. These accessory proteins comprise non-toxic haemagglutinins that stabilize biological activity *in vivo* and most probably allows the complex to adhere to the muscle tissue, thus acting as a depot-like substance (opinion expressed by E. Johnson in the review [15]). Using fluid-based diffusion theories, the active toxin molecule is then thought to diffuse from the complex to interact with target receptors on the surface of neuronal cells. The migration of radio-labelled 900-kDa BOTOX complex within the muscle has been compared experimentally with that of the isolated 150-kDa neurotoxin. Radio-iodinated toxins were injected into the gastrocnemius muscle of rats and migration of the toxin from the injection site was monitored at various time points [16]. The findings showed that a greater proportion of the radiolabelled 900-kDa BOTOX complex persisted in the target muscle over the 24 h following injection compared with the 150-kDa toxin form. As the majority of the radio-label was found on the accessory proteins, this suggests that the accessory haemagglutinins might confer an advantage in staying power over the naked neurotoxin.

Botulinum toxin type-A complex in the Dysport formulation is purified using column chromatography and the size of the resulting product has been shown to be heterogeneous in composition [14,17]. The Dysport preparation comprises a small proportion of the 900-kDa complex in addition to toxin moieties ranging from 500 to 700 kDa. As a significant proportion of the Dysport toxin comprises material of lower molecular mass, it will migrate further from the injection site as a result of fluid-based diffusion. The resulting larger distribution zone for Dysport means that active toxin is more probably to migrate over greater distances within the target muscle and subsequently reach adjacent tissues or the systemic circulation.

Pre-clinical dose–response studies

The efficacies of different BoNT preparations have been assessed pre-clinically using the mouse digit abduction score model [18]. In this model, mice were injected with BoNT in the head of one gastrocnemius muscle. Localized muscle weakness was scored by considering the toe-spreading reflex in the injected foot in comparison with the untreated foot. This involves briefly lifting each animal by the tail to elicit a startle response in which the animal extends its hind limbs and abducts the hind digits. By assessing groups of mice with different doses but using identical injection volumes of the various BoNT formulations, dose–response curves could be produced and used to calculate the intramuscular ED_{50}.

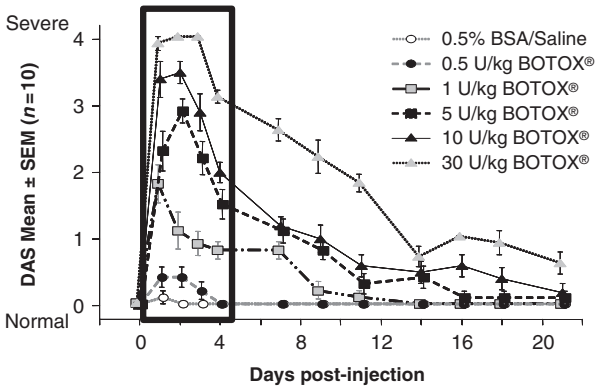

Figure 1 The muscle-weakening efficacy of different BOTOX® doses using the mouse digital abduction assay.

Response curves following BOTOX treatment, determined using the mouse digit abduction score model, are presented in Fig. 1. At the lowest dose of BOTOX (0.5 U/kg), there was a small increase in the mean digital abduction score, which peaked approximately 2 days after injection. With increasing doses of BOTOX, there was an increase in the mean digital abduction score, indicative of greater muscle weakness, and the duration of muscle weakness was also prolonged [19]. Similar experiments have shown other BoNT preparations produce comparable digital abduction score efficacy curves. The data considering dose versus peak digital abduction score (the boxed area in Fig. 1) can be used to construct dose–response curves for the individual BoNT formulations. Figure 2 shows log dose–response curves for BOTOX, BTX-A, Dysport and Myobloc, with each formulation showing the characteristic sigmoid-shaped curves [20]. However, the dose–response curves are not parallel for the different formulations indicative of different efficacy and safety ratios. Comparison of the dose–response curves

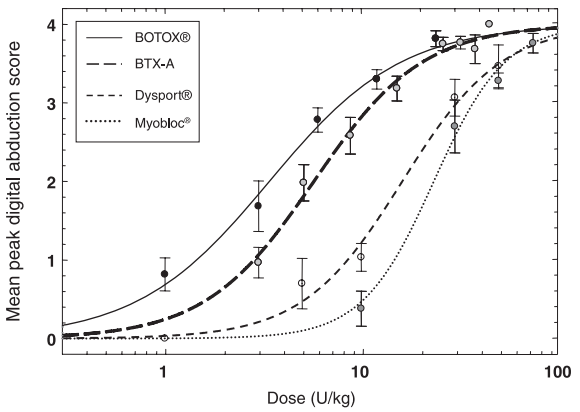

Figure 2 Dose–response curves for different botulinum toxin formulations, assessed using the mouse digital abduction assay.

allows the different products to be ranked in order of potency. Ranking shows BOTOX to be of highest potency, followed by BTX-A, Dysport and Myobloc. The dramatic difference in potency seen when comparing BOTOX and Myobloc is even greater in human subjects and this is thought to reflect an increased murine sensitivity to BoNT-B that may not be reflected in the clinical setting. These dose–response curves can also be used to determine the intramuscular ED_{50} values for the different formulations, taken as the dose required to effect a mean digital abduction score of 2. This methodology results in mean intramuscular ED_{50} values of: 3.4 ± 0.3 U/kg for BOTOX, 5.7 ± 0.3 U/kg for BTX-A, 16.2 ± 1.1 U/kg for Dysport and 23.4 ± 1.4 U/kg for Myobloc.

Having established the degree of local efficacy at the injection site using identical injection volumes, the overall performance of BoNT formulations can be considered in terms of systemic toxicity. This is used to establish an overall safety margin for each preparation and compares the intramuscular dose at which systemic leakage leads to the occurrence of side effects (LD_{50}) with the dose required to achieve local efficacy (ED_{50}). The resulting safety margin dose ratio for BOTOX, of 15.1, was higher than those for Dysport (6.1) or Myobloc (4.8). The rank order of these values is comparable with the rank order of potency and indicates that rapid migration of Myobloc from the injection site is responsible for systemic treatment effects.

However, in clinical terms the appearance of systemic side effects is not an appropriate outcome measure and so pre-clinical efficacy is also assessed in terms of the therapeutic margin. The intramuscular therapeutic margin compares the intramuscular threshold dose (TD) that shows localized toxin spread, reflected by atrophy of the quadriceps muscle, with the dose required to achieve local efficacy (ED_{50}). The resulting therapeutic margin dose ratio for BOTOX, of 6.8, was higher than those for Dysport (1.5) or Myobloc (1.0). The rank order of these values is comparable with the rank order of potency and indicates rapid migration of Dysport and Myobloc from the injection site into neighbouring muscles.

Figure 3 shows the dose–response curves for BOTOX, Dysport and Myobloc, indicating graphically the magnitude of the safety and therapeutic margins for each formulation [21]. When considering BOTOX, the ED_{50} dose is well separated from the doses where local (TD) and systemic spread (LD_{50}) become apparent, resulting in broad therapeutic and safety margins. It is apparent that systemic spread of BOTOX only starts to occur at the top of the dose–response curve (e.g. near maximal local efficacy). With Dysport the area of the graph where systemic effects occur is considerably larger, starting approximately half way up the dose–response curve, and the therapeutic margin is much narrower than that seen with BOTOX. This means that systemic side effects can potentially occur before maximal toxin efficacy is achieved. In addition, the loss of toxin by local and systemic migration may contribute to the reduced potency of Dysport in the murine digital abduction model. With Myobloc (BoNT-B) the toxin appears to migrate from the target muscle very rapidly, resulting in a negligible therapeutic margin. This also results in systemic spread being associated with a broad area of the dose–response curve. These data consider the properties of the different toxin formulations solely in the mouse gastrocnemius muscle when using equal injection volumes of each toxin; however, the findings are likely to vary depending on the size of the target muscle and the injection volume.

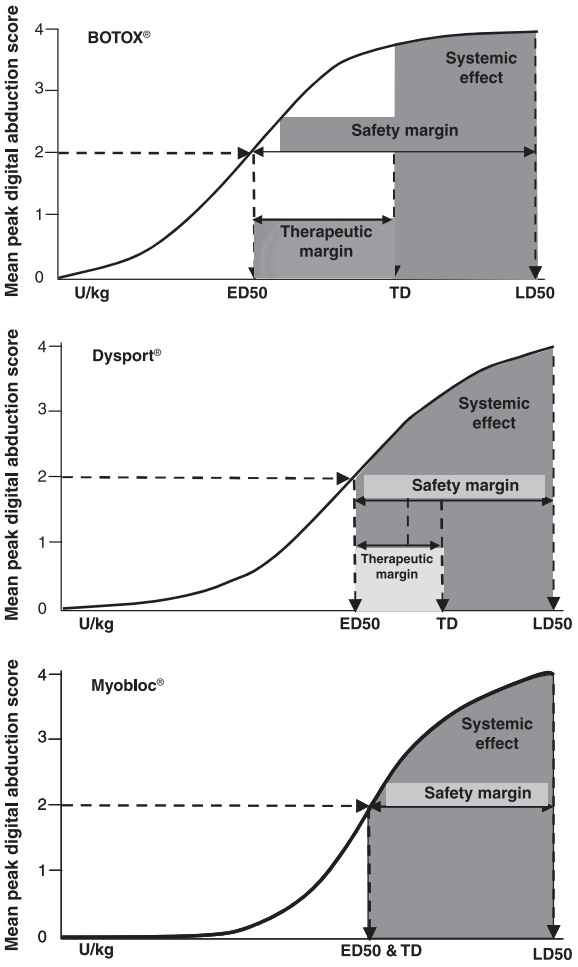

Figure 3 Different clinical profiles for botulinum toxin formulations based on pre-clinical studies using murine digital abduction scores.

Overall, these findings show that when considering the two BoNT-A and one BoNT-B formulations, distinct characteristics are seen in terms of efficacy and

local and systemic toxin migration. In addition, the non-parallel nature of the dose–response curves for the different preparations indicate different pharmacological properties. In conjunction, these findings indicate that the different BoNT formulations are not comparable or interchangeable in the clinical setting [19,22]. These findings are acknowledged in BoNT summary of product characteristics, which state that simple dose conversion factors are not applicable clinically.

Toxin serotype

Some of the differences in the dose–response characteristics seen when comparing Myobloc with the other BoNT formulations are likely to be the result of the different ecto-acceptors required to mediate toxin binding to target tissues [23–26]. The distribution of ecto-acceptors may contribute to the different therapeutic profiles seen in terms of efficacy, by affecting the amount of toxin taken up at a particular nerve terminal. The intracellular levels of toxin that accumulate will also influence the duration of any therapeutic effect seen. Toxin that does not bind to its ecto-acceptor and is not internalized into the nerve cell will be able to migrate out of the muscle to distal tissues [27]. Therefore, the presence and number of ecto-acceptors in the target muscle will influence not just efficacy parameters but also the profile of local and systemic side effects.

Clinical differences between botulinum toxin formulations

Differences are apparent when considering the clinical application and adverse event profiles of the different toxin formulations.

1. The doses administered for the different clinical indications vary greatly between BoNT formulations.

2. The injection pattern (dose, volume and muscle pattern) differs between preparations and this depends on the migration characteristics of the individual products. Following injection, BoNT migrates from this treatment depot to varying degrees, depending on the formulation and the dilution injection volume used. This migration can represent focal dispersal within the target tissue, movement into adjacent or distal muscles, or systemic spread as a result of toxin reaching the circulation or lymphatic systems. Widespread sub-clinical effects on neuromuscular transmission can be detected in muscles distant from the injection site using single fibre electromyography [28,29]. The migration profile of the different BoNT formulations is of major interest, as it influences the profile of side effects seen following treatment. However, careful consideration of the formulation, dilution volume and dosing options can minimize migration to non-target tissues.

3. The amount of toxin that can be administered during each treatment session varies, with this primarily influenced by the incidence of neutralizing antibody development seen with the different preparations. This is of greatest concern with Myobloc, where the large quantities of neurotoxin that needs to be administered to achieve clinical benefit increases the likelihood of antibody formation [27,30]. The impact of these factors on the clinical use of the different BoNT formulations has been assessed in both pre-clinical models and in comparative clinical trials.

Comparison between BOTOX and Dysport

A summary of published studies that have either tested predetermined ratios or that have used both products in a clinical trial is provided in Table 2. In addition, various recommendations have been made over the years

Table 2 Summary of studies comparing different ratios of BOTOX to Dysport

Authors	BOTOX to Dysport ratio	Indication or outcome measure
Studies testing predetermined ratios		
Marion *et al.* [47]	1:3	Blepharospasm or hemifacial spasm
Durif [48]	1:5 to 1:6	Cervical dystonia
Nüssgens and Roggenkamper [37]	1:4	Blepharospasm
Sampaio *et al.* [38]	1:4	Blepharospasm
Odergren *et al.* [40]	1:3	Cervical dystonia
Vanden Bergh and Lison [49]	1:2.5	Hemifacial spasm or cervical dystonia
Annese *et al.* [50]	1:2.5	Achalasia
Ranoux *et al.* [41]	1:3 and 1:4	Cervical dystonia
Bihari [36]	1:4 to 1:5	Blepharospasm, cervical dystonia or hemifacial spasm
Clinical studies using both products		
Bhakta *et al.* [51]	1:4 to 1:5	Spasticity
Bhaumik and Behari [52]	1:4.6	Cervical dystonia
Hesse *et al.* [53]	1:5	Spasticity
Marchetti *et al.* [54]	1:2 to 1:11	Blepharospasm and/or cervical dystonia

by clinical experts in the field of BoNT therapy, with Clarke recommending a dose ratio of 1:3 to 1:6 [31], Brin recommending a dose ratio of 1:4 to 1:5 [32] and Lowe, Rasmussen and the Royal College of Physicians recommending a dose ratio of 1:4 [33–35]. As the information suggests, there is no consensus for a set dose ratio amongst the experts.

One of the most recent studies looking at the efficacy and safety of different dose ratios was carried out by Bihari in 2005 [36]. This was a single-arm, crossover, comparative study in 48 patients with blepharospasm ($n = 27$), cervical dystonia/torticollis ($n = 12$) or hemifacial spasm ($n = 9$). Each patient received a Dysport injection and was assessed over a 12-week period, followed by a BOTOX injection and then a second 12-week assessment period. Both products were diluted so that the injection volumes were the same in each session and the same injection sites were used. In patients with blepharospasm 120 U Dysport and 30 U BOTOX were used (1:4 ratio), in cervical dystonia patients 654 U Dysport and 130 U BOTOX were used (1:5 ratio) and for hemifacial spasm 78 U Dysport and 16 U BOTOX were used (1:5 ratio). Assessments were carried out at baseline and at 3 and 12 weeks using the Jankovic Rating Scale and the Toronto Western Spasmodic Torticollis Rating Scale (TWSTRS) and patients also kept a diary to record when efficacy wore off and also the side effects they experienced. The results are summarized in Fig. 4 and show that BOTOX affected significantly greater symptom improvement and had a longer duration of effect compared with Dysport ($P < 0.05$). In addition, no side effects were recorded in the BOTOX group, whereas 40% of patients (19/48) reported side effects whilst receiving Dysport treatment.

However, although these results show a better response rate for BOTOX based on a 1:4 dose ratio for blepharospasm and a 1:5 ratio for cervical dystonia and hemifacial spasm, the study was openlabel and the order of injection was fixed (first injection with Dysport and then 12 weeks later a second injection with BOTOX), and as such could have been subject to bias. Therefore, it is more appropriate to consider the controlled studies that have been carried out that allow direct comparison of different dose ratios between BOTOX and Dysport. To date this comprises four studies, with the results of each of these studies summarized briefly below.

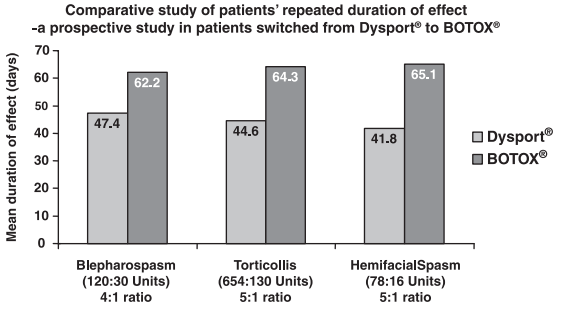

Figure 4 Summary of results comparing different ratios of BOTOX® to Dysport® in the treatment of blepharospasm, torticollis and hemifacial spasm.

Nüssgens and Roggenkämper. Comparison of two botulinum-toxin preparations in the treatment of essential blepharospasm [37]

This was a double-blind study carried out in 212 patients with blepharospasm. A dose ratio of BOTOX to Dysport of 1:4 was used, with each patient receiving one injection session with BOTOX and one injection session with Dysport in a randomized order. All patients had received a mean of 15.3 previous BoNT-A injections, characterizing a well-trained study population able to describe and detect efficacy and safety parameters.

The results of this study showed no significant difference in duration and efficacy between the two groups, with patients treated with Dysport having a duration of effect of 8.03 weeks compared with 7.98 weeks for patients treated with BOTOX.

The incidence of adverse events in this study was significantly ($P < 0.05$) higher in the Dysport group (24%) compared with the BOTOX group (17%), mainly because of a higher incidence of ptosis in the Dysport group (6.6%) compared with the BOTOX group (1.4%).

Sampaio et al. *A single-blind, randomized parallel study to determine whether any differences can be detected in the efficacy and tolerability of two formulations of BoNT-A (Dysport and BOTOX) assuming a ratio of 4:1 [38]*

This was a single-blind, parallel group study in 91 patients with blepharospasm or hemifacial spasm. A dose ratio of BOTOX to Dysport of 1:4 was used, but different dilution volumes were used: 2.5 ml for a vial of 500 U Dysport and 4 ml for a vial of 100 U BOTOX. None of the patients had received previous treatment with any BoNT and efficacy was assessed using the Blepharospasm Rating Scale [39].

The results of this study showed no significant difference in efficacy and duration of effect between the two groups, with patients treated with Dysport having a duration of effect of 12.8 weeks compared with 13.1 weeks for patients treated with BOTOX. In this study, patients were allowed to have a booster injection if they were not satisfied with the effect of the initial injections. A larger number of patients in the Dysport

group (24%) required booster injections compared with the BOTOX group (12%), indicating that the Dysport-treated patients may have been under-dosed.

The incidence of adverse events in this study was comparable between the two groups (50% in the Dysport group and 47% in the BOTOX group), but this probably reflects the under-dosing in the Dysport group.

Odergren et al. *A double blind, randomized, parallel group study to investigate the dose equivalence of Dysport and BOTOX in the treatment of cervical dystonia [40]*

This was a double-blind, parallel group study carried out in 73 patients with rotational cervical dystonia. All patients had received four previous treatments with BOTOX prior to entering the study. A dose ratio of BOTOX to Dysport of 1:3 was used, with 35 patients being treated with BOTOX and 38 patients being treated with Dysport. No fixed injection sites per muscle were used, although a fixed volume was administered. In every patient, an identical muscle pattern was injected.

The results of this study showed no significant difference between the two treatment groups based on the reduction in Tsui score (Dysport 4.8 and BOTOX 5.0) and duration of effect (Dysport 83.9 days and BOTOX 80.7 days). The incidence of adverse events was also similar between the two groups (58% in the Dysport group and 69% in the BOTOX group). Therefore, this study concluded overall that no difference was seen between the two treatments using a dose ratio of 1:3. However, it should be noted that the Tsui score is not very sensitive to small changes and as such a small difference between the groups might not have been detected.

Ranoux et al. *Respective potencies of Dysport and BOTOX: a double blind, randomized, crossover study in cervical dystonia [41]*

This was a double-blind, crossover, three-period study carried out in 54 patients with cervical dystonia. All patients had received two previous treatments with BOTOX prior to entering the study. Dose ratios of BOTOX to Dysport of 1:4 and 1:3 were used, with each patient receiving in a randomized order: BOTOX as per their prior treatment regimen, Dysport at a dose of 1:3 and Dysport at a dose of 1:4. The site of injection, the muscle injected and the volume used were fixed for all treatment groups.

The results of this study showed statistically significant better clinical results for both Dysport doses compared with BOTOX based on the reduction in Tsui score (Dysport 1:3 ratio = 4.32, Dysport 1:4 ratio = 4.89, BOTOX = 3.22, $P < 0.05$) and pain scale reduction (Dysport 1:3 ratio = 4.41, Dysport 1:4 ratio = 5.37, BOTOX = 2.59, $P < 0.05$). The mean duration of effect was also reported to be longer by 25 days in the Dysport 1:4 group compared with the BOTOX group ($P = 0.02$).

The incidence of adverse events was higher in the Dysport group (36% in the 1:4 group and 33% in the 1:3 group, compared with 18% in the BOTOX group). This difference was mainly due to higher incidences of dysphagia, asthenia and dysphonia in the Dysport groups (Table 3).

Thus, these four controlled studies show concordant results. However, it should be noted that the studies have some methodological limitations, such as the research tools used in these studies. The Jankovic Scale, Tsui Score and TWSTRS are not sensitive enough to detect small differences in efficacy between active formulations, particularly as the studies to date have generally been underpowered. For assessments of efficacy duration, the current estimates based on patient interviews are too imprecise, relying on patient memory, whereas use of a daily patient diary would produce more accurate results.

Other published studies have also shown divergent results, primarily because of open-label methodology and the number of study variables, such as dilution factors, injection volumes, site of injection and muscles injected. Thus, it is recommended to ensure that the outcomes obtained are exclusively because of the BoNT-A formulation, only patients with stable disease conditions should be included in the studies and standardized injection techniques and patterns should be used. In particular, identical muscles and locations should be used, as fibre density and type differ between muscles can cause variation in the behaviour of the toxin. In addition, muscle size and the number of cholinergic synapses involved in the different clinical conditions vary (e.g. disease duration) and so the dose of toxin required to achieve an effective response will also vary. As distribution of the toxin also depends on the volume of injection, the dilution factor should remain constant.

Table 3 Summary of adverse events in Ranoux study (Ranoux *et al.* [42])

	BOTOX	Dysport 1:3	Dysport 1:4
Mean dose (range)	105 U (70–180)	315 U (210–540)	420 U (280–720)
All adverse events	18%	33%*	36%**
Dysphagia	4%	16%	17%
Asthenia	4%	4%	13%
Dysphonia	0%	6%	6%

*BOTOX versus Dysport ($P = 0.06$); **BOTOX versus Dysport ($P = 0.03$).

Comparison between BOTOX and Xeomin

Two clinical studies have been carried to date in patients with blepharospasm and cervical dystonia comparing BOTOX and Xeomin. They showed non-inferiority in efficacy and no significant differences in side effects when using a 1:1 dose ratio of Xeomin compared with BOTOX [42,43].

However, the 1:1 dose ratio used for conversion in both the trials was based on a small neurophysiological study in 14 healthy volunteers, where the paralytic efficacy of 4 U Xeomin was compared with 4 U BOTOX in the extensor digitorum brevis [44]. As such the justification for the 1:1 ratio tested is limited and as the trials were not designed to establish a fixed dose conversion ratio, the 1:1 ratio therefore cannot be regarded as being substantiated in this clinical setting.

Pharmacovigilance data

The lower incidence of adverse events seen with BOTOX in comparative clinical studies is supported by pharmacovigilance data. The incidence of adverse events spontaneously reported to the Agência Nacional de Vigilância Sanitária in Brazil showed that a higher incidence of adverse events was associated with Dysport treatment (http://www.anvisa.gov.br) [45]. In the period from January 2003 to June 2006, a total of 124 813 vials of BOTOX and 70 810 vials of Dysport were sold in Brazil. Treatment was associated with 113 reported adverse events for BOTOX and 414 adverse events for Dysport, equating to one event per 1000 vials and six events per 1000 vials, respectively. This compared with an adverse event rate of four per 1000 vials for the Chinese BoNT formulation (Prosigne Lanzhou Institute of Biological Products, China). The World Health Organization database also showed a higher adverse event rate associated with Dysport treatment. Using data acquired over 5 years from France, Germany, Italy, Spain and the United Kingdom, the reporting rates associated with all toxin indications were in excess of 7×10^{-5} events per vial of Dysport sold and 3×10^{-5} events per vial of BOTOX sold (http://www.who.int) [46].

The incidences of specific adverse events following treatment of cervical dystonia, as cited in the summary of product characteristics for different countries, are summarized in Table 4. Dysphagia is the most commonly reported adverse event and is seen with a higher incidence following treatment with Dysport. Other events include dizziness, disturbed vision, muscle weakness and dry mouth, all of which are more frequently reported following Dysport treatment compared with BOTOX treatment.

Conclusions

Translational medicine has shown that different BoNT preparations differ in terms of their serotype and physical composition. It is proposed that the homogeneous nature of the high molecular weight toxin complex in BOTOX affects tissue distribution by minimizing fluid-based migration from the injection site compared with Dysport. This is seen in terms of greater retention of radiolabelled toxin in the target muscle and higher therapeutic and safety margins for BOTOX in animal model systems. The reduced local and systemic migration seen with BOTOX is consistent with the lower incidence of adverse events apparent in the clinical setting.

Dysport's greater mobility is sometimes presented as an advantage, e.g. in large muscles where fewer injection sites are required to achieve clinical efficacy. However, the tendency for greater migration means that not only is the Dysport dose more likely to disperse throughout the target muscle, but it may also enter adjacent muscles or the systemic circulation via capillary absorption and/or lymphatic drainage. Migration into adjacent muscles is likely to cause unwanted local muscle weakness and systemic spread increases the likelihood of adverse events characteristic of BoNT poisoning or antibody formation.

Table 4 Incidence of adverse events as cited in the summary of product characteristics for treatment of cervical dystonia

Country	Event	Dysport	BOTOX
Australia, Finland, Denmark, France, Ireland and Germany	Dysphagia	26% for 250 U 29% for 500 U 39% for 1000 U	Australia: quoted as 12.2% usually mild to moderate, occasionally more severe 13%
Argentina, Brazil, Mexico and New Zealand	Dysphagia	29% with 500 U, often following injection of stemocleidomastoid muscle	
Germany and Greece	Dry mouth	> 10% for 500, 1000 and 1500 U	< 10%
Germany	Disturbed vision	> 10% at 1000 U	< 10%
Germany	Muscle weakness	> 10% at 1000 U	< 10%
Germany	Dizziness	> 10% at 1000 U	< 10%

Doses are not interchangeable between the different preparations of BoNT-A, as stated in all data sheets. This can lead to confusion and in some cases misinformation, which can lead to dangerous consequences as use of an incorrect fixed dose ratio can result in under- or over-dosing, leading to a sub-therapeutic effect or clinically significant safety issues.

In well-designed clinical studies, it must be remembered that the results are only valid for the treated muscles and disease studied in the trial. Until well-designed multicentre studies are carried out, which take efficacy and safety aspects of both low- and high-dose BoNT-A regimens into account, the establishment of a fixed dose is currently not achievable. Therefore, in clinical practice, clinicians should be guided by the dosing instructions specific to each product and based on previous patient response and clinical experience.

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Declaration of interest

Dr K. Roger Aoki is an employee of Allergan Inc., which manufactures and sells BOTOX®.

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