subdermal wells to cause unwanted paralysis in surrounding areas of the body. This problem is exacerbated when the area to be treated is large and many injections of toxin are required to treat the area. Moreover, because the injected toxin complexes contain non-toxin proteins and albumin that stabilize the botulinum toxin and increase the molecular weight of the toxin complex, the toxin complexes have a long half-life in the body and may cause an undesirable antigenic response in the patient. For example, some patients will, over time, develop an allergy to the albumin used as a stabilizer in current commercial formulations. Also, the toxin complexes may induce the immune system of the patient to form neutralizing antibodies, so that larger amounts of toxin are required in subsequent administrations to achieve the same effect. When this happens, subsequent injections must be carefully placed so that they do not release a large amount of toxin into the bloodstream of the patient, which could lead to fatal systemic poisoning, especially since the non-toxin proteins and albumin stabilize the botulinum toxin in blood.

[0014] In view of the drawbacks associated with current botulinum toxin formulations, it would be highly desirable to have an injectable botulinum toxin formulation that is efficacious and stable, but exhibits reduced antigenicity and a lower tendency to diffuse locally after injection. It would also be desirable to use such a botulinum toxin formulation for various therapeutic, aesthetic and/or cosmetic purposes.

SUMMARY

[0015] In one of its examples, this disclosure provides injectable compositions comprising botulinum toxin non-covalently associated with a positively charged carrier molecule. In preferred examples, the compositions of the disclosure possess one or more advantages over conventional commercial botulinum toxin formulations, such as BOTOX® or MYOBLOC®. For instance, in certain examples, the compositions may exhibit one or more advantages over conventional injectable botulinum formulations, including reduced antigenicity, a reduced tendency to undergo diffusion into surrounding tissue following injection, increased duration of clinical efficacy or enhanced potency relative to conventional botulinum toxin formulations, faster onset of clinical efficacy, and/or improved stability.

[0016] Another example of this disclosure is the recognition that certain non-native molecules (i.e., molecules not found in botulinum toxin complexes obtained from *Clostridium*

botulinum bacteria) can be added to botulinum toxin, botulinum toxin complexes, and in particular reduced botulinum toxin complexes (as defined herein), to improve toxin diffusion through tissues. The non-native molecules associate non-covalently with the toxin and act as penetration enhancers that improve the ability of the toxin to reach target structures after injection. Furthermore, the non-native molecules may increase the stability of the toxin prior to and after injection. By way of example, the penetration enhancers may be positively charged carriers, such as cationic peptides, which have no inherent botulinum-toxin-like activity and which also contain one or more protein transduction domains as described herein.

[0017] Another example of this disclosure is to provide a composition comprising botulinum toxin, a botulinum toxin complex (or a reduced protein botulinum toxin complex including just the 150 kD neurotoxin itself, or the neurotoxin with some, but not all, of the native complex proteins) and a positively charged carrier.

[0018] In another of its examples, the disclosure further relates to a method for producing a biologic effect by injecting an effective amount, preferably a therapeutically effective amount, of the compositions of this disclosure to a subject or patient in need of such treatment. The biologic effect may include, for example, muscle paralysis, reduction of hypersecretion or sweating, treatment of neurologic pain or migraine headache, management of rhinitis or sinusitis, treatment of hyperactive bladder, reduction of muscle spasms, prevention or reduction of acne, reduction or enhancement of an immune response, reduction of wrinkles, fine lines, or glabellar lines, also known as frown lines, particularly in the face, or prevention or treatment of various other disorders.

[0019] In another example, the disclosure provides effective doses and amounts of the compositions of this disclosure that afford a long-lasting, sustained efficacy e.g., a response rate of long duration, following administration by injection to a subject or patient in need of treatment. Such doses and amounts are preferably therapeutically or cosmetically effective doses and amounts that produce or result in a desired therapeutic or cosmetic effect in a subject to whom the doses and amounts are administered. In an example, the effect of reducing wrinkles and lines of the face, particularly, glabellar lines, provided by a dose of a composition of the disclosure administered by injection as a single treatment to the subject or patient in need, lasted several weeks to several months compared with conventional treatment, such as BOTOX®

Cosmetic injection. In particular examples, and as described in the Examples herein, a single treatment of a subject or patient with a composition of the disclosure comprising a botulinum toxin, such as botulinum toxin A, and a positively charged carrier, as described herein, in therapeutically effective amounts of 20 U to 60 U, afforded a response rate of wrinkle and facial line reduction for at least 20 weeks, at least 24 weeks, or about 6 to 10 months, or even longer, compared with conventional BOTOX® injection. Moreover, the compositions of the disclosure provide an attribute of reduced diffusion or spread from the injection site following injection, thereby localizing the toxin and its effect where desired and decreasing nonspecific or unwanted effects of the toxin at sites or locations distant from the site of injection for treatment.

[0020] The duration of effect provided by compositions of the disclosure, e.g., RT002 of Example 5, as well as by the described treatment methods and uses, affords significant advantages compared to the art. By way of example, subjects undergoing treatment, such as aesthetic treatment, with compositions containing botulinum toxin consider that duration of effect following treatment is of high importance to them. Such a long, sustained duration of effect, which is achieved by even a single treatment with an effective dose of a product of the disclosure, for example, RT002, permits fewer injections per treatment course for a subject, which is extremely important for the subject. A prolonged duration effect from a single treatment with a product which has clear efficacy and safety, as provided by the inventive compositions and methods described herein, offer less discomfort, less cost and more convenience to subjects undergoing a course of treatment. Furthermore, a product that affords significant and sustained effects, which are maintained for at least a 20 or 24 week period, or for at least a 6-month period, or for greater than a 6-month period, following the single injectable treatment of the product to a subject, provides a solution to an unmet need in the art for both practitioners and patients alike. Thus, the compositions and methods of the disclosure provide a solution to the problem of too frequent treatments and improve patients' overall well-being. Such prolonged duration of action provides for fewer treatments over an entire treatment course.

[0021] In another example, the disclosure provides a method of administering botulinum toxin to achieve an extended duration therapeutic or cosmetic effect in an individual, in which the method comprises administering by injection a dose of a sterile injectable composition into an area of the individual in need of treatment to achieve the therapeutic or cosmetic effect following a first treatment with the composition; wherein the composition comprises a botulinum

toxin, a botulinum toxin complex, or a reduced botulinum toxin complex component and a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of (gly)p-RGRDDRRQRRR-(gly)q, (gly)p-YGRKKRRQRRR-(gly)q or (gly)p-RKKRRQRRR-(gly)q, wherein the subscripts p and q are each independently an integer of from 0 to 20; wherein the botulinum toxin, botulinum toxin complex, or reduced botulinum toxin complex component is administered to the individual in a treatment dose of 20 U to 60 U; wherein the positively charged carrier is non-covalently associated with the botulinum toxin, botulinum toxin complex, or reduced botulinum toxin complex component; and a pharmaceutically acceptable diluent suitable for injection; and wherein the first treatment dose of the composition administered by injection to the individual achieves the extended duration therapeutic or cosmetic effect having at least about a 6 month to about a 10 month duration of effect, optionally, before a second or subsequent treatment dose is administered.

[0021a] In one aspect, the disclosure provides a method of administering botulinum toxin to achieve an extended duration therapeutic or cosmetic effect in an individual, the method comprising: administering by injection a first treatment dose of a sterile injectable composition into an area of the individual in need of treatment to achieve the therapeutic or cosmetic effect following a first treatment with the composition; wherein the composition comprises a pharmaceutically acceptable diluent suitable for injection; and a botulinum toxin component selected from the group consisting of a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex; and a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20; wherein the botulinum toxin component is administered to the individual in a treatment dose of 20 U to 60 U; wherein the positively charged carrier is non-covalently associated with the botulinum toxin component; and wherein the first treatment dose of the composition administered by injection to the individual achieves the extended duration therapeutic or cosmetic effect having at least about a 6 month to about a 10 month duration of effect, optionally, before a second or subsequent treatment dose is administered.

[0021b] In another aspect, the disclosure provides use of botulinum toxin in the manufacture of a sterile injectable composition for achieving an extended duration therapeutic or cosmetic effect in an individual, wherein a first treatment dose of the sterile injectable composition is to be administered by injection into an area of the individual in need of treatment to achieve the therapeutic or cosmetic effect following a first treatment with the composition; wherein the composition comprises a pharmaceutically acceptable diluent suitable for injection; and a botulinum toxin component selected from the group consisting of a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex; and a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20; wherein the botulinum toxin component is to be administered to the individual in a treatment dose of 20 U to 60 U; wherein the positively charged carrier is non-covalently associated with the botulinum toxin component; and wherein the first treatment dose of the composition administered by injection to the individual achieves the extended duration therapeutic or cosmetic effect having at least about a 6 month to about a 10 month duration of effect, optionally, before a second or subsequent treatment dose is to be administered.

[0022] In another example, the disclosure provides a method of reducing wrinkles, lines, or furrows in an individual in need thereof, the method comprising administering to the individual as a single dose injection a composition comprising: a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex and a positively charged carrier comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of (gly)p-RGRDDRRQRRR-(gly)q, (gly)p-YGRKKRRQRRR-(gly)q or (gly)p-RKKRRQRRR-(gly)q, wherein the subscripts p and q are each independently an integer of from 0 to 20; and a pharmaceutically acceptable diluent for injection; wherein the botulinum toxin is administered to the individual in an amount of 20 U to 60 U; wherein the positively charged carrier is non-covalently associated with the botulinum toxin, botulinum toxin complex, or reduced botulinum toxin complex component; and wherein the single dose injection of the composition provides a single treatment having at least about a six month to about a 10 month duration of effect in reducing the wrinkles, lines, or furrows in the individual, thereby extending treatment interval duration for the individual. In an example, the composition reduces, or reduces the severity of, glabellar lines in the face of the individual.

[0022a] In one aspect, the disclosure provides a method of reducing wrinkles, lines, or furrows in an individual in need thereof, the method comprising: administering to the individual as a single dose injection a composition comprising: a pharmaceutically acceptable diluent for injection; a botulinum toxin componenst selected from the group consisting of a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex; and a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20; wherein the botulinum toxin is administered to the individual in an amount of 20 U to 60 U; wherein the positively charged carrier is non-covalently associated with the botulinum component; and wherein the single dose injection of the composition provides a single treatment having at least about a six month to about a 10 month duration of effect in reducing the wrinkles, lines, or furrows in the individual, thereby extending treatment interval duration for the individual.

[0022b] In another aspect, the disclosure provides a pharmaceutical composition in a sterile injectable formulation for use in administering botulinum toxin to achieve an extended duration therapeutic or cosmetic effect in an individual, said composition comprising a pharmaceutically acceptable diluent suitable for injection; a botulinum toxin component in a treatment dose of 20 U to 60 U, wherein said botulinum toxin component is selected from the group consisting of a botulinum toxin complex, a reduced botulinum toxin complex, or a botulinum toxin; and a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20; wherein the positively charged carrier is non-covalently associated with the botulinum toxin component; and wherein said treatment dose of the composition achieves the extended duration therapeutic or cosmetic effect having at least about a 6 month to about a 10 month duration of effect in the individual administered said formulation by injection.

[0022c] In another aspect, the disclosure provides a pharmaceutical composition in a sterile injectable formulation for use in reducing wrinkles, lines, or furrows in an individual in need thereof, said composition comprising: a botulinum toxin component in a dose of 20 U to 60 U, said botulinum toxin component selected from the group consisting of a botulinum toxin complex, a reduced botulinum toxin complex, or a botulinum toxin, a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20; and a pharmaceutically acceptable diluent for injection; wherein the positively charged carrier is non-covalently associated with the botulinum toxin component; and wherein said dose of the composition provides a single treatment having at least about a six month to about a 10 month duration of effect in reducing the wrinkles, lines, or furrows in the individual, thereby extending treatment interval duration for the individual.

[0023] In another example, the disclosure provides a sterile injectable composition comprising a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex

in a dosage amount selected from 20 U, 40 U, or 60 U; and a positively charged carrier comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of (gly)p-RGRDDRRQRRR-(gly)q, (gly)p-YGRKKRRQRRR-(gly)q or (gly)p-RKKRRQRRR-(gly)q, wherein the subscripts p and q are each independently an integer of from 0 to 20; and a pharmaceutically acceptable diluent for injection; wherein the positively charged carrier is non-covalently associated with the botulinum toxin, botulinum toxin complex, or reduced botulinum toxin complex component; and wherein the composition provides a cosmetic or therapeutic effect which endures for at least 20 to 24 weeks, or for at least 6 months, or greater than 6 months, e.g., about 6 months to about 10 months, following a treatment of an individual with an effective dose of the injectable composition.

[0023a] In one aspect, the disclosure provides a sterile injectable composition comprising: a botulinum toxin component selected from the group consisting of a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex, in a dosage amount selected from 20 U, 40 U, or 60 U; and a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20; and a pharmaceutically acceptable diluent for injection; wherein the positively charged carrier is non-covalently associated with the botulinum toxin component; and wherein the composition provides a cosmetic or therapeutic effect which endures for at least 20 to 24 weeks following a single treatment of an individual with the injectable composition.

[0024] In some examples of these above methods and composition, the composition comprises botulinum toxin of serotype A, preferably a serotype A botulinum toxin having a molecular weight of 150 kDa. In an example, the positively charged carrier has the amino acid sequence RKKRRQRRRG-$(K)_{15}$-GRKKRRQRRR. In an example, the botulinum toxin is present in the composition in a dosage amount of 20 U. In an example, the botulinum toxin is present in the composition in a dosage amount of 40 U. In an example, the botulinum toxin is present in the composition in a dosage amount of 60 U. In an example, the composition reduces the severity of glabellar lines in an individual who has undergone a single treatment by injection of the composition. In certain examples, the duration of the treatment effect comprises greater than 6 months; greater than 7 months; greater than 8 months; greater than 9 months; or at least 6 months through 10 months.

[0025] In another of its examples, the disclosure provides a method of treating an individual who is in need of treatment with injectable botulinum toxin, in which the method of treatment comprises a treatment course having multiple treatment intervals with prolonged duration of effect and duration time between each treatment interval, the treatment course comprising: administering by injection an initial treatment dose of a sterile injectable composition into an area of the individual in need of treatment to achieve a therapeutic or a cosmetic effect following the initial treatment with the composition; wherein the composition comprises a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex component and a positively charged carrier component comprising a positively charged polylysine backbone having

covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of (gly)p-RGRDDRRQRRR-(gly)q, (gly)p-YGRKKRRQRRR-(gly)q or (gly)p-RKKRRQRRR-(gly)q, wherein the subscripts p and q are each independently an integer of from 0 to 20; and a pharmaceutically acceptable diluent suitable for injection; wherein the botulinum toxin, botulinum toxin complex, or reduced botulinum toxin complex component is administered to the individual in a treatment dose of 20 U to 60 U; wherein the positively charged carrier is non-covalently associated with the botulinum toxin, botulinum toxin complex, or reduced botulinum toxin complex component; wherein the initial treatment dose of the composition administered by injection to the individual provides a therapeutic or cosmetic duration of effect lasting through at least about 10 months; and administering subsequent treatment doses of the composition by injection to the individual at treatment intervals comprising a duration of greater than or equal to 3 months to at least about 10 months following the initial treatment dose and between each subsequent treatment dose.

[0025a] In one aspect, the disclosure provides a method of treating an individual in need of treatment with injectable botulinum toxin, wherein the method of treatment comprises a treatment course having multiple treatment intervals with prolonged duration of effect and duration time between each treatment interval, the treatment course comprising: administering by injection an initial treatment dose of a sterile injectable composition into an area of the individual in need of treatment to achieve a therapeutic or a cosmetic effect following the initial treatment with the composition; wherein the composition comprises a pharmaceutically acceptable diluent suitable for injection; a botulinum toxin component selected from the group consisting of a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex; and a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20; and wherein the botulinum toxin component is administered to the individual in a treatment dose of 20 U to 60 U; wherein the positively charged carrier is non-covalently associated with the botulinum toxin component; wherein the initial treatment dose of the composition administered by injection to the individual provides a therapeutic or cosmetic duration of effect lasting through at least about 10 months; and administering subsequent treatment doses of the composition by injection to the individual at treatment intervals comprising a duration of greater than or equal to 3 months to at least about 10 months following the initial treatment dose and between each subsequent treatment dose.

[0025b] In another aspect, the disclosure provides use of an injectable botulinum toxin in the manufacture of a medicament for treating an individual in need of treatment with injectable botulinum toxin, wherein the treatment comprises a treatment course having multiple treatment intervals with prolonged duration of effect and duration time between each treatment interval, the treatment course comprising: an initial treatment dose of a sterile injectable composition to be administered by injection into an area of the individual in need of treatment to achieve a therapeutic or a cosmetic effect following the initial treatment with the composition; wherein the composition comprises a pharmaceutically acceptable diluent suitable for injection; a botulinum toxin component selected from the group consisting of a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex; and a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20; and wherein the botulinum toxin component is to be administered to the individual in a treatment dose of 20 U to 60 U; wherein the positively charged carrier is non-covalently associated with the botulinum toxin component; wherein the initial treatment dose of the composition administered by injection to the individual provides a therapeutic or cosmetic duration of effect lasting through at least about 10 months; and subsequent treatment doses of the composition to be administered by injection to the individual at treatment intervals comprising a duration of greater than or equal to 3 months to at least about 10 months following the initial treatment dose and between each subsequent treatment dose.

[0026] In examples of the above-described treatment method, the therapeutic or cosmetic effect is treatment or reduction of wrinkles, lines, or furrows. In an example, the therapeutic or cosmetic effect is reduction of glabellar lines in the face of the individual. In an example, the composition comprises botulinum toxin of serotype A, preferably, botulinum toxin of serotype A having a molecular weight of 150 kDa. In an example, the positively charged carrier is a positively charged peptide having the amino acid sequence RKKRRQRRRG-(K)15-GRKKRRQRRR. In an example, the composition does not locally diffuse from the site of injection following injection. In specific examples, the botulinum toxin is administered to the individual in an amount of 20 U, or in an amount of 40 U, or in an amount of 60 U. In an example, the therapeutic effect is reduction of a symptom associated with a disorder selected from the group consisting of hemifacial spasm, adult onset spasmodic torticollis, anal fissure, blepharospasm, cerebral palsy, headaches, strabismus, temporomandibular joint disorder, neurologic pain, overactive bladder, rhinitis, sinusitis, acne, dystonia, dystonic contractions, hyperhidrosis, and hypersecretion of a gland controlled by the cholinergic nervous system. In certain examples, the duration of the treatment interval comprises greater than 3 months; greater than 4 months; greater than 5 months; greater than 6 months; greater than 7 months; greater than 8 months; greater than 9 months; or at least 6 months through 10 months.

[0027] This disclosure also provides kits for preparing formulations or compositions containing a botulinum toxin, a botulinum toxin complex, or a reduced protein botulinum toxin complex and positively charged carrier, or a premix that may in turn be used to produce such a formulation or composition. Also provided are kits that contain means for sequentially administering a botulinum toxin complex (or a reduced botulinum toxin complex including just the 150 KD neurotoxin itself or the neurotoxin with some native complex proteins) and a positively charged carrier.

DESCRIPTION OF THE FIGURES

[0028] **Figure 1** presents a bar graph showing the required time to return to the baseline digit abduction score (DAS) value (0.4) following repeated administration of either RT003 or BOTOX®.

[0029] **Figures 2A and 2B: Figure 2A** shows the hind leg of a mouse injected with a dark dye to indicate the portion of a mouse's gastrocnemius muscle that is affected by lateral-to-midline injection. **Figure 2B** shows the hind leg of a mouse injected with a dark dye to indicate the portion of a mouse's gastrocnemius muscle that is affected by midline injection.

[0030] **Figure 3** presents digital abduction scores (DAS) measured as a function of time following injection of RT003, RTT150, or BOTOX® into either the lateral-to-midline or midline portion of the gastrocnemius muscle of a mouse.

[0031] **Figures 4A and 4B** present Kaplan-Meier Curves showing the duration of response in various treatment groups from the clinical study described in Example 5 herein. The Kaplan-Meier Curves represent results from primary efficacy analyses of the clinical study and demonstrate duration of response for at least a 1-point improvement from baseline for Investigator Global Assessment-Facial Wrinkle Severity (IGA-FWS) Assessment in the indicated treatment groups. **Figure 4A** presents Kaplan-Meier Curves for the Placebo treatment group, the VISTABEL®/BOTOX® 20 U treatment group and the RT002 40 U treatment group. **Figure 4B** presents Kaplan-Meier Curves for the Placebo treatment group, the VISTABEL®/BOTOX® 20 U treatment group, the RT002 20 U treatment group, the RT002 40 U treatment group and the RT002 60 U treatment group.

Claims:

1. A method of administering botulinum toxin to achieve an extended duration therapeutic or cosmetic effect in an individual, the method comprising:

administering by injection a first treatment dose of a sterile injectable composition into an area of the individual in need of treatment to achieve the therapeutic or cosmetic effect following a first treatment with the composition;

wherein the composition comprises a pharmaceutically acceptable diluent suitable for injection; and

a botulinum toxin component selected from the group consisting of a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex; and

a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20;

wherein the botulinum toxin component is administered to the individual in a treatment dose of 20 U to 60 U;

wherein the positively charged carrier is non-covalently associated with the botulinum toxin component; and

wherein the first treatment dose of the composition administered by injection to the individual achieves the extended duration therapeutic or cosmetic effect having at least about a 6 month to about a 10 month duration of effect, optionally, before a second or subsequent treatment dose is administered.

2. Use of botulinum toxin in the manufacture of a sterile injectable composition for achieving an extended duration therapeutic or cosmetic effect in an individual, wherein a first treatment dose of the sterile injectable composition is to be administered by injection into an area of the individual in need of treatment to achieve the therapeutic or cosmetic effect following a first treatment with the composition;

wherein the composition comprises a pharmaceutically acceptable diluent suitable for injection; and

a botulinum toxin component selected from the group consisting of a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex; and

a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20;

wherein the botulinum toxin component is to be administered to the individual in a treatment dose of 20 U to 60 U;

wherein the positively charged carrier is non-covalently associated with the botulinum toxin component; and

wherein the first treatment dose of the composition administered by injection to the individual achieves the extended duration therapeutic or cosmetic effect having at least about a 6 month to about a 10 month duration of effect, optionally, before a second or subsequent treatment dose is to be administered.

3. A method of reducing wrinkles, lines, or furrows in an individual in need thereof, the method comprising:

administering to the individual as a single dose injection a composition comprising:

a pharmaceutically acceptable diluent for injection;

a botulinum toxin componenst selected from the group consisting of a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex; and

a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20;

wherein the botulinum toxin is administered to the individual in an amount of 20 U to 60 U;

wherein the positively charged carrier is non-covalently associated with the botulinum component; and

wherein the single dose injection of the composition provides a single treatment having at least about a six month to about a 10 month duration of effect in reducing the wrinkles, lines, or furrows in the individual, thereby extending treatment interval duration for the individual.

4. A pharmaceutical composition in a sterile injectable formulation for use in administering botulinum toxin to achieve an extended duration therapeutic or cosmetic effect in an individual,

said composition comprising a pharmaceutically acceptable diluent suitable for injection;

a botulinum toxin component in a treatment dose of 20 U to 60 U, wherein said botulinum toxin component is selected from the group consisting of a botulinum toxin complex, a reduced botulinum toxin complex, or a botulinum toxin; and

a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20;

wherein the positively charged carrier is non-covalently associated with the botulinum toxin component; and

wherein said treatment dose of the composition achieves the extended duration therapeutic or cosmetic effect having at least about a 6 month to about a 10 month duration of effect in the individual administered said formulation by injection.

5. A pharmaceutical composition in a sterile injectable formulation for use in reducing wrinkles, lines, or furrows in an individual in need thereof, said composition comprising:

a botulinum toxin component in a dose of 20 U to 60 U, said botulinum toxin component selected from the group consisting of a botulinum toxin complex, a reduced botulinum toxin complex, or a botulinum toxin,

a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20; and

a pharmaceutically acceptable diluent for injection;

wherein the positively charged carrier is non-covalently associated with the botulinum toxin component; and

wherein said dose of the composition provides a single treatment having at least about a six month to about a 10 month duration of effect in reducing the wrinkles, lines, or furrows in the individual, thereby extending treatment interval duration for the individual.

6. The method according to claim 1 or claim 3, the use according to claim 2, or the pharmaceutical composition for use according to claim 4 or claim 5, wherein the composition reduces glabellar lines in the face of the individual.

7. The method, use or pharmaceutical composition for use according to claim 6, wherein the composition comprises botulinum toxin of serotype A.

8. The method, use or pharmaceutical composition for use according to claim 7, wherein the composition comprises botulinum toxin of serotype A having a molecular weight of 150 kDa.

9. The method, use or pharmaceutical composition for use according to any one of claims 1 to 8, wherein the positively charged polylysine backbone has covalently attached thereto one or more positively charged efficiency groups having the amino acid sequence $(gly)_p$-

RGRDDRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20.

10. The method, use or pharmaceutical composition for use according to any one of claims 1 to 8, wherein the positively charged polylysine backbone has covalently attached thereto one or more positively charged efficiency groups having the amino acid sequence $(gly)_p$-YGRKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20.

11. The method, use or pharmaceutical composition for use according to any one of claims 1 to 8, wherein the positively charged polylysine backbone has covalently attached thereto one or more positively charged efficiency groups having the amino acid sequence $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20.

12. The method, use or pharmaceutical composition for use according to any one of claims 1 to 11, wherein (i) the subscripts p and q are each independently an integer of from 0 to 8; or (ii) are each independently an integer of from 2 to 5.

13. The method, use or pharmaceutical composition for use according to any one of claims 1 to 12, wherein the one or more positively charged efficiency groups are attached to either end, or both ends, of the positively charged polylysine backbone of the positively charged carrier.

14. The method, use or pharmaceutical composition for use according to claim 13, wherein the positively charged carrier has the amino acid sequence RKKRRQRRRG-$(K)_{15}$-GRKKRRQRRR.

15. The method, use or pharmaceutical composition for use according to any one of claims 1 to 14, wherein the composition does not locally diffuse from the site of injection following injection.

16. The method, use or pharmaceutical composition for use according to any one of claims 1 to 15, wherein the botulinum toxin is administered to the individual in an amount of 20 U.

17. The method, use or pharmaceutical composition for use according to any one of claims 1 to 15, wherein the botulinum toxin is administered to the individual in an amount of 40 U.

18. The method, use or pharmaceutical composition for use according to any one of claims 1 to 15, wherein the botulinum toxin is administered to the individual in an amount of 60 U.

19. The method according to claim 1, the use according to claim 2, or the pharmaceutical composition for use according to claim 4, wherein the therapeutic effect is reduction of a symptom associated with a disorder selected from the group consisting of hemifacial spasm, adult onset spasmodic torticollis, anal fissure, blepharospasm, cerebral palsy, headaches, strabismus, temporomandibular joint disorder, neurologic pain, overactive bladder, rhinitis, sinusitis, acne, dystonia, dystonic contractions, hyperhidrosis, and hypersecretion of a gland controlled by the cholinergic nervous system.

20. The method according to claim 1, the use according to claim 2, or the pharmaceutical composition for use according to claim 4, wherein the therapeutic or cosmetic effect is treatment or reduction of wrinkles, lines, or furrows in the individual.

21. The method, use or pharmaceutical composition for use according to any one of claims 1 to 20, wherein the duration of treatment effect comprises greater than 3 months.

22. The method, use or pharmaceutical composition for use according to any one of claims 1 to 20, wherein the duration of treatment effect comprises greater than 4 months.

23. The method, use or pharmaceutical composition for use according to any one of claims 1 to 20, wherein the duration of treatment effect comprises greater than 5 months.

24. The method, use or pharmaceutical composition for use according to any one of claims 1 to 20, wherein the duration of treatment effect comprises greater than 6 months.

25. The method, use or pharmaceutical composition for use according to any one of claims 1 to 20, wherein the duration of treatment effect comprises greater than 7 months.

26. The method, use or pharmaceutical composition for use according to any one of claims 1 to 20, wherein the duration of treatment effect comprises greater than 8 months.

27. The method, use or pharmaceutical composition for use according to any one of claims 1 to 20, wherein the duration of treatment effect comprises greater than 9 months.

28. The method, use or pharmaceutical composition for use according to any one of claims 1 to 20, wherein the duration of treatment effect comprises at least 6 months through 10 months.

29. A sterile injectable composition comprising:

a botulinum toxin component selected from the group consisting of a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex, in a dosage amount selected from 20 U, 40 U, or 60 U; and

a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20; and

a pharmaceutically acceptable diluent for injection;

wherein the positively charged carrier is non-covalently associated with the botulinum toxin component; and

wherein the composition provides a cosmetic or therapeutic effect which endures for at least 20 to 24 weeks following a single treatment of an individual with the injectable composition.

30. The composition according to claim 29, wherein the positively charged carrier has the amino acid sequence RKKRRQRRRG-$(K)_{15}$-GRKKRRQRRR.

31. The composition according to claim 29 or claim 30, wherein the composition comprises botulinum toxin of serotype A.

32. The composition according to claim 31, wherein the composition comprises botulinum toxin of serotype A having a molecular weight of 150 kDa.

33. The composition according to any one of claims 29 to 32, wherein the botulinum toxin is to be administered to the individual in a dosage amount of 20 U.

34. The composition according to any one of claims 29 to 32, wherein the botulinum toxin is to be administered to the individual in a dosage amount of 40 U.

35. The composition according to any one of claims 29 to 32, wherein the botulinum toxin is to be administered to the individual in a dosage amount of 60 U.

36. A method of treating an individual in need of treatment with injectable botulinum toxin, wherein the method of treatment comprises a treatment course having multiple treatment intervals with prolonged duration of effect and duration time between each treatment interval, the treatment course comprising:

administering by injection an initial treatment dose of a sterile injectable composition into an area of the individual in need of treatment to achieve a therapeutic or a cosmetic effect following the initial treatment with the composition;

wherein the composition comprises a pharmaceutically acceptable diluent suitable for injection;

a botulinum toxin component selected from the group consisting of a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex; and

a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20; and

wherein the botulinum toxin component is administered to the individual in a treatment dose of 20 U to 60 U;

wherein the positively charged carrier is non-covalently associated with the botulinum toxin component;

wherein the initial treatment dose of the composition administered by injection to the individual provides a therapeutic or cosmetic duration of effect lasting through at least about 10 months; and

administering subsequent treatment doses of the composition by injection to the individual at treatment intervals comprising a duration of greater than or equal to 3

months to at least about 10 months following the initial treatment dose and between each subsequent treatment dose.

37. Use of an injectable botulinum toxin in the manufacture of a medicament for treating an individual in need of treatment with injectable botulinum toxin, wherein the treatment comprises a treatment course having multiple treatment intervals with prolonged duration of effect and duration time between each treatment interval, the treatment course comprising:

an initial treatment dose of a sterile injectable composition to be administered by injection into an area of the individual in need of treatment to achieve a therapeutic or a cosmetic effect following the initial treatment with the composition;

wherein the composition comprises a pharmaceutically acceptable diluent suitable for injection;

a botulinum toxin component selected from the group consisting of a botulinum toxin, a botulinum toxin complex, or a reduced botulinum toxin complex; and

a positively charged carrier component comprising a positively charged polylysine backbone having covalently attached thereto one or more positively charged efficiency groups having an amino acid sequence of $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, $(gly)_p$-YGRKKRRQRRR-$(gly)_q$ or $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20; and

wherein the botulinum toxin component is to be administered to the individual in a treatment dose of 20 U to 60 U;

wherein the positively charged carrier is non-covalently associated with the botulinum toxin component;

wherein the initial treatment dose of the composition administered by injection to the individual provides a therapeutic or cosmetic duration of effect lasting through at least about 10 months; and

subsequent treatment doses of the composition to be administered by injection to the individual at treatment intervals comprising a duration of greater than or equal to 3 months to at least about 10 months following the initial treatment dose and between each subsequent treatment dose.

38. The method according to claim 36 or use according to claim 37, wherein the therapeutic or cosmetic effect is treatment or reduction of wrinkles, lines, or furrows.

39. The method or use according to claim 38, wherein the therapeutic or cosmetic effect is reduction of glabellar lines in the face of the individual.

40. The method or use according to any one of claims 36 to claim 39, wherein the composition comprises botulinum toxin of serotype A.

41. The method or use according to claim 40, wherein the composition comprises botulinum toxin of serotype A having a molecular weight of 150 kDa.

42. The method or use according to any one of claims 36 to 41, wherein the positively charged polylysine backbone has covalently attached thereto one or more positively charged efficiency groups having the amino acid sequence $(gly)_p$-RGRDDRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20.

43. The method or use according to any one of claims 36 to 41, wherein the positively charged polylysine backbone has covalently attached thereto one or more positively charged efficiency groups having the amino acid sequence $(gly)_p$-YGRKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20.

44. The method or use according to any one of claims 36 to 41, wherein the positively charged polylysine backbone has covalently attached thereto one or more positively charged efficiency groups having the amino acid sequence $(gly)_p$-RKKRRQRRR-$(gly)_q$, wherein the subscripts p and q are each independently an integer of from 0 to 20.

45. The method or use according to any one of claims 36 to 44, wherein (i) the subscripts p and q are each independently an integer of from 0 to 8; or (ii) are each independently an integer of from 2 to 5.

46. The method or use according to any one of claims 36 to 45, wherein the one or more positively charged efficiency groups are attached to either end, or both ends, of the positively charged polylysine backbone of the positively charged carrier.

47. The method or use according to any one of claims 36 to 41, wherein the positively charged carrier has the amino acid sequence RKKRRQRRRG-$(K)_{15}$-GRKKRRQRRR.

48. The method or use according to any one of claims 36 to 47, wherein the composition does not locally diffuse from the site of injection following injection.

49. The method or use according to any one of claims 36 to 48, wherein the botulinum toxin is administered to the individual in an amount of 20 U.

50. The method or use according to any one of claims 36 to 48, wherein the botulinum toxin is administered to the individual in an amount of 40 U.

51. The method or use according to any one of claims 36 to 48, wherein the botulinum toxin is administered to the individual in an amount of 60 U.

52. The method according to claim 36 or use according to claim 37, wherein the therapeutic effect is reduction of a symptom associated with a disorder selected from the group consisting of hemifacial spasm, adult onset spasmodic torticollis, anal fissure, blepharospasm, cerebral palsy, headaches, strabismus, temporomandibular joint disorder, neurologic pain, overactive bladder, rhinitis, sinusitis, acne, dystonia, dystonic contractions, hyperhidrosis, and hypersecretion of a gland controlled by the cholinergic nervous system.

53. The method or use according to any one of claims 36 to 52, wherein the duration of the treatment interval comprises greater than 3 months.

54. The method or use according to any one of claims 36 to 52, wherein the duration of the treatment interval comprises greater than 4 months.

55. The method or use according to any one of claims 36 to 52, wherein the duration of the treatment interval comprises greater than 5 months.

56. The method or use according to any one of claims 36 to 52, wherein the duration of the treatment interval comprises greater than 6 months.

57. The method or use according to any one of claims 36 to 52, wherein the duration of the treatment interval comprises greater than 7 months.

58. The method or use according to any one of claims 36 to 52, wherein the duration of the treatment interval comprises greater than 8 months.

59. The method or use according to any one of claims 36 to 52, wherein the duration of the treatment interval comprises greater than 9 months.

60. The method or use according to any one of claims 36 to 52, wherein the duration of the treatment interval comprises at least 6 months through 10 months.