

Original Article



Efficacy and Safety of Novel Botulinum Toxin Type A (Protoxin) in the Treatment of Moderate to Severe Glabellar Lines: A Multicenter, Randomized, Double-Blind, Active-Controlled Phase III Study

Hyung Seok Son ¹, Min Kyung Shin ², Jong Hun Lee ³, Moon Bum Kim ⁴, Kwang Ho Yoo ⁵, Sun Young Choi ⁵, Hye Sung Han ⁵, Joon Seok ⁶, Beom Joon Kim ⁶, Yang Won Lee ¹

¹Department of Dermatology, Konkuk University School of Medicine, Seoul, Korea

²Department of Dermatology, Kyung Hee University College of Medicine, Seoul, Korea

³Department of Plastic and Reconstructive Surgery, Inje University Sanggye Paik Hospital, School of Medicine, Inje University, Seoul, Korea

⁴Department of Dermatology, Pusan National University School of Medicine, Busan, Korea

⁵Department of Dermatology, Chung-Ang University Gwangmyeong Hospital, Chung-Ang University College of Medicine, Gwangmyeong, Korea

⁶Department of Dermatology, Chung-Ang University Hospital, Chung-Ang University College of Medicine, Seoul, Korea

OPEN ACCESS

Received: Jul 16, 2025

Revised: Sep 7, 2025

Accepted: Sep 9, 2025

Published online: Oct 29, 2025

Corresponding Author:

Yang Won Lee

Department of Dermatology, Konkuk University School of Medicine, 120-1 Neungdong-ro, Gwangjin-gu, Seoul 05030, Korea.

Email: 20050078@kuh.ac.kr

© 2026 The Korean Dermatological Association and The Korean Society for Investigative Dermatology
This is an Open Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (<https://creativecommons.org/licenses/by-nc/4.0/>) which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

ABSTRACT

Background: A novel botulinum toxin type A (Protoxin; Protox Inc.) has been developed.

Objective: To evaluate the efficacy and safety of the newly developed Protoxin compared to the approved drug onabotulinumtoxinA (OBoNT) in moderate to severe glabellar lines.

Methods: Adults with a glabellar line Facial Wrinkle Scale (FWS) score of 2 (moderate) or 3 (severe) were enrolled in the study. Subjects were randomized in a 1:1 ratio to receive either Protoxin or OBoNT. A total of 20 units of botulinum toxin was injected at five sites in the glabellar region (4 units at each site). FWS scores were assessed at baseline and at weeks 4, 8, 12, and 16 post-injection. The primary endpoint was the proportion of subjects at week 4 who had a reduction of 2 or more points in FWS and a final score of 0 (none) or 1 (mild).

Results: A total of 274 subjects were randomized, of whom 78.1% were female. At week 4 post-treatment, the improvement rate of glabellar lines was 62.22% in the Protoxin group and 62.96% in the OBoNT group. The lower limit of the two-sided 95% confidence interval (-12.24%) exceeded the -15% margin, confirming the non-inferiority of the new drug. Safety profiles were comparable between the two groups.

Conclusion: Protoxin demonstrated efficacy and safety profiles comparable to those of OBoNT in the treatment of moderate to severe glabellar lines.

Trial Registration: ClinicalTrials.gov Identifier: [NCT05364580](https://clinicaltrials.gov/ct2/show/study/NCT05364580)

Keywords: Aging; Botulinum toxins; Neurotoxins

INTRODUCTION

Glabella lines are primarily formed by the repeated contraction of the centrally located procerus muscle and the bilateral corrugator supercilii muscles¹. These lines typically begin to appear noticeable in individuals during their 30s to 40s, indicating an association with the aging process. Additionally, prominent glabella lines may convey an impression of anger or irritation, potentially influencing social perception.

Botulinum toxin type A, a neurotoxin produced by *Clostridium botulinum*, blocks the release of acetylcholine at motor nerve terminals and temporarily induces muscle relaxation, thereby reducing dynamic facial wrinkles². Since the initial observation in 1987—when botulinum toxin used to treat blepharospasm was also found to smooth the facial wrinkles³—research into its cosmetic applications has expanded significantly.

To date, efficacy of the various botulinum toxins for improvement of glabella lines has been demonstrated in numerous randomized controlled studies⁴⁻⁸. Furthermore, several studies have reported that patient satisfaction following treatment of glabella lines with botulinum toxin is associated with reduced psychological burden, and improvement of depressive symptoms^{9,11}.

Protoxin (Protox Inc., Hwaseong, Korea) is a botulinum toxin type A product derived from a master cell bank of *C. botulinum* type A (strain ATCC-3502). During manufacturing, the active ingredient undergoes purification and formulation processes. While many commercially available botulinum toxin type A products are manufactured using the conventional lyophilization (freeze-drying) method, Protoxin is distinguished by its use of a vacuum drying process. This process removes moisture through direct evaporation under reduced pressure, thereby eliminating the freezing step that could result in protein destabilization and potential loss of biological activity¹². By avoiding this step, vacuum drying may help better preserve the molecular integrity and biological potency of the toxin. Additionally, the vacuum drying process significantly reduces manufacturing time, which could improve production efficiency. These advantages may contribute to high product quality and enhanced cost-effectiveness.

OnabotulinumtoxinA (OBoNT), the first botulinum toxin product approved for the temporary improvement of glabella lines, was used as the active comparator in this study. The objective of this study was to evaluate whether Protoxin demonstrates efficacy and safety profiles comparable to those of OBoNT at a 1:1 dosage ratio in subjects with moderate to severe glabella lines.

MATERIALS AND METHODS

Study design

This clinical trial is a multicenter, randomized, double-blind,

active-controlled, phase III clinical trial targeting adult subjects with moderate to severe glabella lines (NCT05364580). Five medical centers in South Korea participated in the trial, and subjects were recruited from March 2023 to October 2023. The study protocol was approved by Institutional Review Board at Konkuk University Medical Center (IRB protocol approval number KUMC 2022-05-035). After obtaining written informed consent from the subjects, screening tests were conducted.

This clinical trial was conducted ethically and scientifically in accordance with Korea Good Clinical Practice and all relevant regulations. Furthermore, the trial respected the dignity and rights of participants and was conducted in accordance with the Declaration of Helsinki, ensuring that subjects were not subjected to any undue harm.

Subject selection

Subjects were adult men and women aged 19 to 65 years who had a moderate to severe glabella lines based on 4-point Facial Wrinkle Scale (FWS) (Table 1). The exclusion criteria were as follows: a history of neuromuscular junction disorders; a history of facial nerve palsy or ptosis; skin abnormalities at the injection site; current use of medications with muscle relaxing effects, a history of taking aspirin or non-steroidal anti-inflammatory drugs; a history of any facial procedures or surgery that may influence the study results; a known hypersensitivity to the components of injection (e.g., botulinum toxin, albumin); a history of alcohol or drug abuse, or other significant mental disorders; pregnancy or lactation; individuals of reproductive potential unwilling to use adequate contraception; and any individual deemed unsuitable for participation as judged by the investigator.

Drug administration

Eligible subjects who met the inclusion and exclusion criteria were randomly assigned to either the Protoxin group or OBoNT group. Randomization was performed by an independent statistician using a stratified block randomization method, with glabella lines severity as a stratification factor. The randomization sequence was generated using SAS® Version 9.4 (SAS Institute, Cary, NC, USA). One vial (100 U) of Protoxin or OBoNT was diluted with 2.5 ml of 0.9% sterile saline solution (resulting in a concentration of 4 U/0.1 ml). The diluted solution was then injected—0.1 ml at each of 5 sites. Four injection sites targeted the bilateral corrugator supercilii muscles and one site targeted the procerus muscle (Fig. 1). To reduce the risk of blepharoptosis, injections into the corrugator supercilii muscle were performed at least 1 cm above the bony supraorbital margin to avoid diffusion to the levator palpebrae superioris muscle.

Blinding was maintained for both the investigator and the subjects to minimize the potential bias in evaluation of treatment

Table 1. Description of 4-point FWS and 7-point subjects' self-assessment scale

Scale	Score	Severity	Definition
FWS (At maximal frown)	3	Severe	Facial lines are prominent, and the base of the deepest wrinkle is not visible from the surface
	2	Moderate	Facial lines are prominent, and the base of the deepest wrinkle is visible from the surface
	1	Mild	Some facial lines are visible
	0	None	No lines are visible
FWS (At rest)	3	Severe	Lines are very clearly visible
	2	Moderate	Lines are clearly visible
	1	Mild	Some lines are visible
	0	None	No lines are visible
Subjects' Self-Assessment Scale	+4		Almost 100% improved
	+3		75% improved
	+2		50% improved
	+1		25% improved
	0		Neither improved nor worsened
	-1		25% worsened
	-2		50% worsened
	-3		75% worsened
	-4		Almost 100% worsened

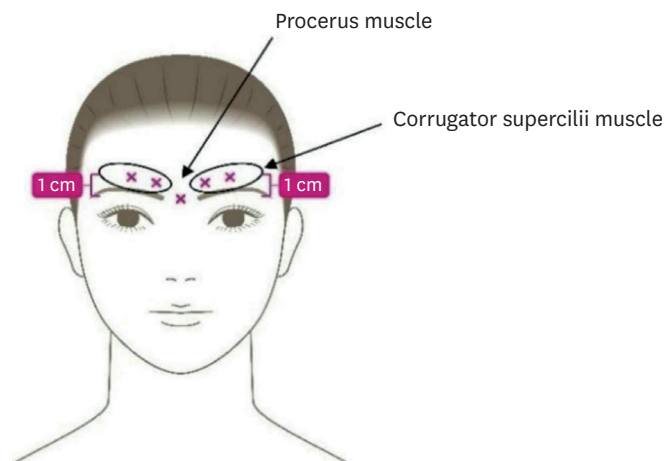


Fig. 1. Injection sites of the botulinum toxin.

effects and adverse reactions. Each subject was identified only by the assigned code during randomization, and allocation details remained undisclosed until the end of the clinical trial. To address the difference in drug vial appearance, dilution was performed by trained and designated research personnel who were not involved in subject assessment or injection. Identical syringes were prepared and delivered to the investigator who were responsible for the injection, thereby maintaining blinding.

Assessments

The glabellar lines evaluation by investigators was conducted at screening visits, at 4, 8, 12, and 16-weeks post-administration. Glabellar lines, both at maximum frown and at rest, were evaluated using the FWS. In addition to on-site evaluations, an independent investigator assessed glabellar lines using the FWS by reviewing photographs of the subjects. At maximum frown, "improvement"

was defined as FWS score of 0 or 1 and a decrease of two or more grades from the baseline. At rest, "improvement" was defined as FWS score 0 or 1.

Subjects self-assessed the improvement of their glabellar lines at 4, 8, 12, and 16 weeks post-administration using a 7-point scale (Table 1). An improvement was defined as a score increase of 2 or more points. Subject satisfaction with the appearance of their glabellar lines was also assessed at the same time points. They were asked to rate their satisfaction with the improvement due to the treatment, using a 7-point scale ranging from grade 1 (very dissatisfied) to grade 7 (very satisfied). Scores of grade 6 or 7 were considered indicative of satisfaction.

The primary efficacy endpoint was the improvement rate of glabellar lines at maximum frown, as evaluated by the on-site investigator at 4 weeks post-administration. Secondary efficacy endpoints included improvement rates of glabellar lines under the following conditions:

- 1) Maximum frown, evaluated by the on-site investigator at weeks 8, 12, and 16.
- 2) Maximum frown, evaluated by the independent investigator at weeks 4, 8, 12, and 16.
- 3) At rest, evaluated by the on-site investigator at weeks 4, 8, 12, and 16.
- 4) At rest, evaluated by the independent investigator at weeks 4, 8, 12, and 16.
- 5) Maximum frown, evaluated by subjects at weeks 4, 8, 12, and 16.
- 6) At rest, evaluated by subjects at weeks 4, 8, 12, and 16.

Subjects' overall satisfaction rates at weeks 4, 8, 12, and 16 were also included as secondary endpoints.

Adverse events were categorized and assessed as either localized (related to the forehead area) or systemic reactions. In case of adverse events, details such as symptoms, onset and resolution dates, severity, course, management, and causality were comprehensively recorded in the case report form. All adverse events were classified according to the Medical Dictionary for Regulatory Activities version 26.0, based on System Organ Class and Preferred Term.

Statistical methods

For efficacy evaluation, the primary analysis was conducted on the Per-Protocol Set (PPS), and supplementary analysis was performed on the Full Analysis Set (FAS). The safety analysis was conducted on the Safety Set (SS), which included all subjects who received at least one dose of the drug. Demographic characteristics, baseline medical history, and prior medication use were analyzed based on the Intention-To-Treat principle, including all randomly assigned subjects.

For analysis of primary efficacy endpoint, a 95% two-sided confidence interval (CI) was calculated using the Mantel-Haenszel method. Non-inferiority of the investigational drug compared to the active comparator was established if the lower limit of the 95% CI exceeded -15%. For the analysis of secondary efficacy endpoints, the frequency and percentage for each group were presented, and difference between groups were analyzed using the Cochran-Mantel-Haenszel test. All statistical tests were two-sided

and performed at a significance level of 5%. Statistical analyses were performed using SAS® Version 9.4 (SAS Institute).

RESULTS

Demographics

A total of 274 subjects were randomly assigned to two groups (137 in Protocoxin group and 137 in OBoNT group). Of 274 subjects, five subjects dropped out, and 269 subjects completed the clinical trial (**Fig. 2**).

The demographic characteristics of the subjects are presented in **Table 2**. The mean age (mean $\pm$ standard deviation) was 46.61 ± 9.26 years, and 78.1% were female. There was no significant difference between the two groups in terms of the age, gender, height, history of previous botulinum toxin injection. There was a statistically significant difference in weight distribution between the groups ($p=0.0461$). The distribution of FWS scores at maximum frown at baseline was not significantly different between the Protocoxin and OBoNT group. The proportion of subjects with a score of 2 (moderate) was 67.15% in Protocoxin group and 66.42% in OBoNT group.

The drug was administered at least once in all randomized 274 subjects, and they were included in the SS. In FAS, one subject was excluded due to the omission of evaluation after baseline. In PPS, which was used for primary efficacy analysis, four subjects

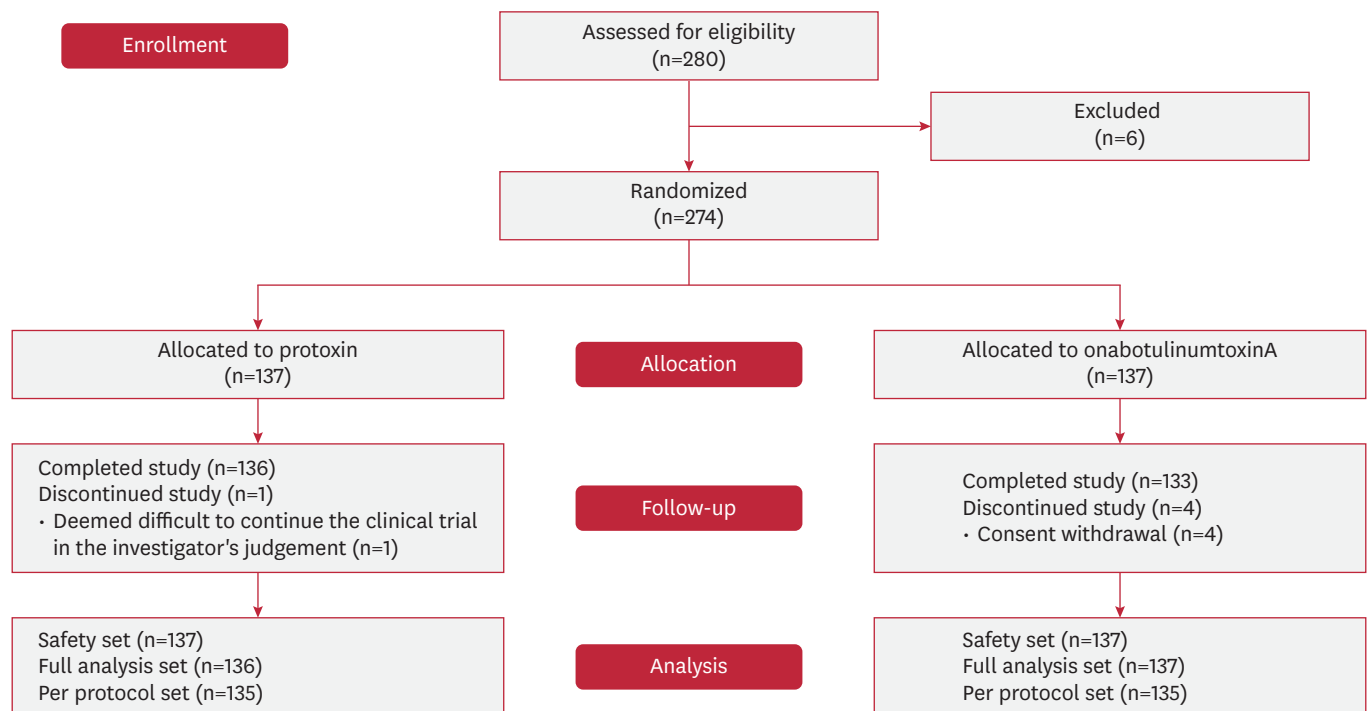


Fig. 2. Flow diagram of the study participants.

Table 2. Demographic characteristics of all randomized subjects

Characteristics	OBoNT (n=137)	Protoxin (n=137)	Total (n=274)	p-value*
Age (yr)				0.359
Number of subjects	137	137	274	
Mean ± SD	47.12±9.28	46.09±9.26	46.6±9.26	
Median (min–max)	47.0 (24.0–65.0)	45.0 (24.0–64.0)	46.0 (24.0–65.0)	
Age group				0.921
Number of subjects	137	137	274	
18–19 yr	0	0	0	
20–29 yr	7 (5.11)	7 (5.11)	14 (5.11)	
30–39 yr	18 (13.14)	22 (16.06)	40 (14.60)	
40–49 yr	62 (45.26)	63 (45.99)	125 (45.62)	
50–59 yr	32 (23.36)	31 (22.63)	63 (22.99)	
60–65 yr	18 (13.14)	14 (10.22)	32 (11.68)	
Sex				0.080
Number of subjects	137	137	274	
Male	24 (17.52)	36 (26.28)	60 (21.90)	
Female	113 (82.48)	101 (73.72)	214 (78.10)	
Previous botulinum toxin injection				0.715
Number of subjects	137	137	274	
Yes	78 (56.93)	75 (54.74)	153 (55.84)	
No	59 (43.07)	62 (45.26)	121 (44.16)	
Height (cm)				0.067
Number of subjects	137	137	274	
Mean ± SD	163.04±7.60	164.79±8.18	163.92±7.93	
Median (min–max)	162.1 (148.3–185.7)	163.1 (145.5–187.6)	162.5 (145.5–187.6)	
Weight (kg)				0.046
Number of subjects	137	137	274	
Mean ± SD	60.72±11.29	63.67±13.04	62.20±12.26	
Median (min–max)	59.2 (41.6–98.5)	60.0 (42.7–112.2)	59.5 (41.6–112.2)	
Baseline severity of glabellar lines (at maximum frown)				0.898
Number of subjects	137	137	274	
2 points (moderate)	92 (67.15)	91 (66.42)	183 (66.79)	
3 points (severe)	45 (32.85)	46 (33.58)	91 (33.21)	

Values are presented as number (%).

The text in bold indicates $p < 0.05$.

OBoNT: onabotulinumtoxinA, SD: standard deviation.

*Two sample t-test, Pearson's χ^2 test or Fisher's exact test. Statistically significant at a two-sided 5% significance level.

were excluded due to protocol violation. Thus, a total of 270 subjects were included in the PPS (**Fig. 2**).

Primary endpoint

The results of the primary endpoint are shown in **Table 3**. In the analysis of the PPS, the improvement rate of glabellar lines at maximum frown at 4 weeks after administration according to the

investigator's on-site evaluation, was 62.22% (84/135 subjects) in the test group and 62.96% (85/135 subjects) in the control group. The difference in improvement rates between the test and control groups was -0.88% (95% two-sided CI, -12.24% , 10.49%). The lower limit of the CI (-12.24%) exceeded -15% , demonstrating the non-inferiority of the test group compared to the control group. The FAS analysis results showed similar results to the PPS analysis.

Table 3. Comparison of the improvement rates at maximum frown, as evaluated by the on-site investigator at week 4

	Per Protocol Set		Full Analysis Set	
	OBoNT (n=135)	Protoxin (n=135)	OBoNT (n=137)	Protoxin (n=136)
Number of subjects	135	135	137	136
Improvement	85 (62.96)	84 (62.22)	87 (63.50)	85 (62.50)
No improvement	50 (37.04)	51 (37.78)	50 (36.50)	51 (37.50)
Treatment difference (%)*		-0.88		-1.18
95% confidence interval†		-12.24 , 10.49		-12.47 , 10.11
Non-inferiority		Yes		Yes

Values are presented as number (%).

OBoNT: onabotulinumtoxinA.

*Treatment difference (%): Protoxin – OBoNT.

†Mantel-Haenszel (adjusted for facial wrinkle scale score as a stratification covariate).

Secondary endpoints

1) Evaluation by on-site investigator

At weeks 8, 12, and 16, at maximum frown, the improvement rates were 39.55%, 22.90%, 19.23% in the Protoxin group, and 34.81%, 22.96%, 19.23% in the OBoNT group (**Supplementary Table 1**). The differences were not statistically significant at any time point (all $p > 0.05$). At weeks 4, 8, 12, and 16, at rest, the improvement rates were 87.41%, 85.82%, 85.50%, and 80.77% in the Protoxin group, and 85.93%, 81.48%, 80.00%, and 78.52% in the OBoNT group (**Supplementary Table 2**). No statistically significant differences were observed between the groups at any time point (all $p > 0.05$).

2) Evaluation by independent investigator

At weeks 4, 8, 12, and 16, at maximum frown, the improvement rates were 82.22%, 68.42%, 50.38%, and 43.08% in the Protoxin group, and 91.11%, 69.63%, 51.85%, and 42.96% in the OBoNT group (**Supplementary Table 3**). At week 4, the improvement rate was significantly higher in the control group compared to the test group (91.11% vs. 82.22%, $p = 0.033$). Except for week 4, no statistically significant differences were observed between the groups (all $p > 0.05$). At weeks 4, 8, 12, 16 weeks, at rest, the improvement rate was 74.07%, 74.44%, 74.81%, and 73.08% in the Protoxin group, and 74.07%, 76.30%, 74.07%, and 74.07% in the OBoNT group (**Supplementary Table 4**). The differences were not statistically significant at any time points (all $p > 0.05$).

3) Evaluation by subjects

At weeks 4, 8, 12, and 16, at maximum frown, the improvement rates were 95.56%, 91.04%, 76.34%, and 65.38% in the Protoxin group and 94.81%, 91.11%, 77.78%, and 62.22% in the OBoNT group (**Supplementary Table 5**). The differences were not statistically significant at all time points (all $p > 0.05$). At weeks 4, 8, 12, and 16, at rest, the improvement rates were 91.11%, 89.55%, 74.81%, and 63.85% in the Protoxin group and 93.33%, 91.85%, 81.48%, and 67.41% in the OBoNT group (**Supplementary Table 6**). No statistically significant differences were observed between the groups at any time point (all $p > 0.05$).

4) Subjects' overall satisfaction rates

Satisfaction was defined as subjects reporting scores of 6 (satisfied) or 7 (very satisfied). At weeks 4, 8, 12, and 16, the satisfaction rates were 91.11%, 85.82%, 77.10%, and 73.08% in the Protoxin group, and 85.93%, 82.22%, 73.33%, and 65.93% in the OBoNT group (**Fig. 3**). There were no statistically significant differences between the groups at any time point (all $p > 0.05$).

Safety evaluation

The overall incidence of treatment-emergent adverse events after

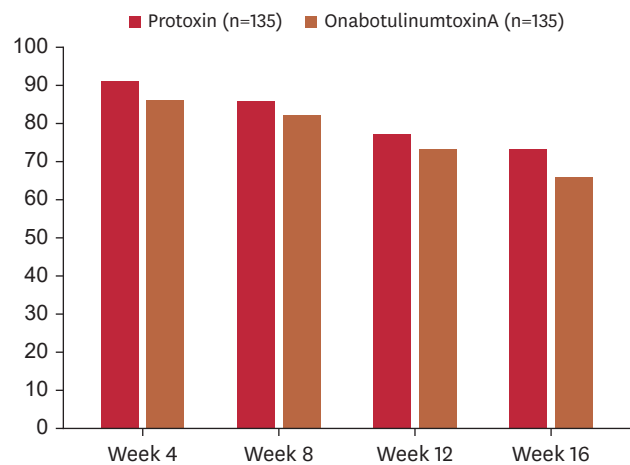


Fig. 3. Subjects' overall satisfaction rates at weeks 4, 8, 12, and 16.

drug administration was reported as 10.22% (14/137 persons, 16 events) in the test group and 8.03% (11/137 persons, 13 events) in the control group. Among these, adverse drug reactions with a potential causal relationship to the investigational drug were observed in 2.19% of the test group (3/137 subjects). These reactions were headache, injection site pruritus, and injection site reaction. In the control group, such reactions were observed in 1.46% (2/137 subjects), which were headache and injection site paresthesia. All drug-related adverse reactions were mild. Additionally, no acute adverse reactions were observed within 30 minutes after administration of the investigational drug.

No deaths or severe adverse reactions were reported after the administration of the drug. There were no clinically significant changes observed in vital signs, hematological tests, blood chemistry test, or urinalysis.

DISCUSSION

The difference in improvement rates at maximum frown between the groups at week 4, which was our primary endpoint, was -0.88% , confirming the non-inferiority of Protoxin. Up to week 16, there were no significant differences in improvement rate between the two groups. Both groups exhibited a decreasing trend in improvement rates over time, and the proportion of subjects with improvement at week 16 was under 20% in both groups. This can be attributed to gradual functional recovery of cholinergic synapses after blockade by botulinum toxin. In a meta-analysis of OBoNT in the treatment of glabellar lines¹³, the median duration effect was 120 days, which was consistent with our study results.

The improvement rate for the primary outcome observed in this study was slightly lower than that reported in previous studies evaluating the same outcome—defined as an improvement of

≥2 points from baseline and an FWS score of 0 or 1. In the two phase III placebo-controlled studies of prabotulinumtoxinA¹⁴, the improvement rate at week 4 was 77.5% and 82.5%, and in the phase III placebo-controlled study of letibotulinumtoxinA¹⁵, the week 4 improvement rate was 78.6%. Similarly, the phase III placebo-controlled study of incobotulinumtoxinA⁷ reported a week 4 improvement rate of 76.6%. In contrast, the improvement rate in this study was 62.22% in the Protoxin group and 62.96% in the OBoNT group. This discrepancy may be attributable to differences in the sex distribution of the study populations. While the proportion of male subjects in the three aforementioned studies ranged from 6.8% to 12%, nearly 20% of subjects in the present study were male. Several reports suggest that male subjects require higher doses of botulinum toxin than female subjects, as men generally have greater muscle mass¹⁶⁻¹⁸. As a fixed dose (20U) was administered, the relatively higher proportion of male participants may have contributed to the lower responder rates observed in this study. Importantly, subgroup analysis identified sex as a statistically significant predictor of treatment outcome in this cohort. After controlling for confounding factors, female subjects had 2.22 times higher odds of experiencing clinical improvement at week 4 (primary endpoint) compared to male subjects (reference group; 95% CI, 1.21, 4.07, $p=0.0099$), supporting the hypothesis that the higher proportion of males in our study population likely contributed to the reduced overall improvement rate.

Regarding secondary endpoints, the improvement rate recorded by independent investigators was not significantly different between the two groups, except for the photographic evaluation at week 4 (82.22% in the Protoxin group vs. 91.11% in the OBoNT group). No differences were observed at weeks 8, 12, and 16. The subjects' self-evaluation also showed no significant differences between the two groups at any time point. Although the independent photographic evaluation at week 4 showed a statistically higher improvement rate with OBoNT compared to Protoxin, this isolated difference should be interpreted with caution. Possible explanations include 1) slight variation in the timing of maximal clinical effect between the formulations, 2) a chance finding due to inter-individual variability and random variation—especially given multiple assessments at different timepoints, and 3) inherent differences between 2-dimensional photographic evaluations versus 3-dimensional on-site evaluations of wrinkle severity. Notably, the difference was not maintained at subsequent visits, and no difference was observed in on-site investigator evaluation or subject self-assessment at all timepoints, suggesting that the week-4 finding does not represent a consistent or clinically meaningful inferiority of Protoxin.

In procedures or surgical interventions conducted for cosmetic purposes, meeting the expectations of the subjects is also an important criterion for assessing efficacy^{19,20}. Up to the 16-week

time point, the subject-reported overall satisfaction rate exceeded 70% in both the test and control groups, with slightly higher satisfaction observed in the test group. Botulinum toxin injection is generally well tolerated, and produces immediate effects following the procedure, with minimal pain.

From a safety perspective, the new drug demonstrated a safety profile comparable to that of the control drug. There were no statistically significant differences between the groups in the incidence rates of local adverse reactions at the injection site, overall adverse reactions, adverse drug reactions. All adverse events were considered mild, and no serious adverse events were reported.

Historically, botulinum toxin type A formulations have progressively improved in purity, stability, and reduction of immunogenicity by employing advanced manufacturing techniques. Protoxin utilizes a relatively newer vacuum drying method, which mitigates several limitations associated with conventional lyophilization, including protein damage and extended manufacturing times. This process can contribute to enhanced product stability, production efficiency and cost-effectiveness. These manufacturing improvements support Protoxin's clinical value as a reliable alternative to existing neurotoxins.

This clinical trial has several limitations. First, study population consisted entirely of Koreans, which may limit the generalizability of the study findings. Second, anatomic variability of the target muscles was not considered. Various glabellar contraction patterns have been reported in previous studies²¹⁻²³, and classification by subtype along with subgroup analysis may help determine the optimal injection method, thereby maximizing efficacy and minimizing unwanted adverse events. Lastly, the data were limited to 16 week-period following injection; the long-term duration of drug effectiveness was not investigated.

In conclusion, based on its demonstrated equivalence to OBoNT, Protoxin may be considered a satisfying treatment option for adult subjects with moderate to severe glabellar lines.

ORCID iDs

Hyung Seok Son 
<https://orcid.org/0000-0002-0607-7530>
 Min Kyung Shin 
<https://orcid.org/0000-0001-9834-7931>
 Jong Hun Lee 
<https://orcid.org/0000-0001-6417-6986>
 Moon Bum Kim 
<https://orcid.org/0000-0003-4837-0214>
 Kwang Ho Yoo 
<https://orcid.org/0000-0002-0137-6849>
 Sun Young Choi 
<https://orcid.org/0000-0003-0248-7708>
 Hye Sung Han 
<https://orcid.org/0000-0002-3556-0740>

Joon Seok <https://orcid.org/0000-0002-8016-8385>Beom Joon Kim <https://orcid.org/0000-0003-2320-7621>Yang Won Lee <https://orcid.org/0000-0003-1268-1248>**Trial Registration**ClinicalTrials.gov Identifier: [NCT05364580](#)**FUNDING SOURCE**

This study was sponsored by Protox Inc.

CONFLICTS OF INTEREST

The authors have nothing to disclose.

DATA SHARING STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

SUPPLEMENTARY MATERIALS**Supplementary Table 1**

Comparison of the improvement rates at maximum frown, as evaluated by the on-site investigator at week 8, 12, and 16

Supplementary Table 2

Comparison of the improvement rates at rest, as evaluated by the on-site investigator at week 4, 8, 12, and 16

Supplementary Table 3

Comparison of the improvement rates at maximum frown, as evaluated by the independent investigator at week 4, 8, 12, and 16

Supplementary Table 4

Comparison of the improvement rates at rest, as evaluated by the independent investigator at week 4, 8, 12, and 16

Supplementary Table 5

Comparison of the improvement rates at maximum frown, as evaluated by the subjects at week 4, 8, 12, and 16

Supplementary Table 6

Comparison of the improvement rates at rest, as evaluated by the subjects at week 4, 8, 12, and 16

REFERENCES

- Wan J, Yi KH. Why do vertical glabellar lines form? A guideline on botulinum neurotoxin injection. *J Cosmet Dermatol* 2024;23:3878-3882. [PUBMED](#) | [CROSSREF](#)
- Choudhury S, Baker MR, Chatterjee S, Kumar H. Botulinum toxin: an update on pharmacology and newer products in development. *Toxins (Basel)* 2021;13:58. [PUBMED](#) | [CROSSREF](#)
- Carruthers A. History of the clinical use of botulinum toxin A and B. *Clin Dermatol* 2003;21:469-472. [PUBMED](#) | [CROSSREF](#)
- Won CH, Lee HM, Lee WS, Kang H, Kim BJ, Kim WS, et al. Efficacy and safety of a novel botulinum toxin type A product for the treatment of moderate to severe glabellar lines: a randomized, double-blind, active-controlled multicenter study. *Dermatol Surg* 2013;39:171-178. [PUBMED](#) | [CROSSREF](#)
- Kim BJ, Kwon HH, Park SY, Min SU, Yoon JY, Park YM, et al. Double-blind, randomized non-inferiority trial of a novel botulinum toxin A processed from the strain CBFC26, compared with onabotulinumtoxin A in the treatment of glabellar lines. *J Eur Acad Dermatol Venereol* 2014;28:1761-1767. [PUBMED](#) | [CROSSREF](#)
- Wu Y, Fang F, Lai W, Li C, Li L, Liu Q, et al. Efficacy and safety of abobotulinumtoxinA for the treatment of glabellar lines in Chinese patients: a pivotal, phase 3, randomized, double-blind and open-label phase study. *Aesthetic Plast Surg* 2023;47:351-364. [PUBMED](#) | [CROSSREF](#)
- Carruthers A, Carruthers J, Coleman WP 3rd, Donofrio L, Flynn T, Gold M, et al. Multicenter, randomized, phase III study of a single dose of incobotulinumtoxinA, free from complexing proteins, in the treatment of glabellar frown lines. *Dermatol Surg* 2013;39:551-558. [PUBMED](#) | [CROSSREF](#)
- Carruthers JD, Lowe NJ, Menter MA, Gibson J, Eadie N; Botox Glabellar Lines II Study Group. Double-blind, placebo-controlled study of the safety and efficacy of botulinum toxin type A for patients with glabellar lines. *Plast Reconstr Surg* 2003;112:1089-1098. [PUBMED](#) | [CROSSREF](#)
- Hexsel D, Brum C, Siega C, Schilling-Souza J, Dal'Forno T, Heckmann M, et al. Evaluation of self-esteem and depression symptoms in depressed and nondepressed subjects treated with onabotulinumtoxinA for glabellar lines. *Dermatol Surg* 2013;39:1088-1096. [PUBMED](#) | [CROSSREF](#)
- Hoffman L, Fabi S. Look better, feel better, live better? The impact of minimally invasive aesthetic procedures on satisfaction with appearance and psychosocial wellbeing. *J Clin Aesthet Dermatol* 2022;15:47-58. [PUBMED](#)
- Cox SE, Kaufman-Janette J, Cohen JL, Gold M, Joseph J, Nestor MS, et al. LetibotulinumtoxinA attenuates the psychological burden of glabellar lines and is associated with high subject satisfaction in phase 3 clinical trials. *Dermatol Surg* 2024;50:535-541. [PUBMED](#) | [CROSSREF](#)
- Arsiccio A, Giorsetto P, Marengo L, Pisano R. Considerations on protein stability during freezing and its impact on the freeze-drying cycle: a design space approach. *J Pharm Sci* 2020;109:464-475. [PUBMED](#) | [CROSSREF](#)
- Glogau R, Kane M, Beddingfield F, Somogyi C, Lei X, Caulkins C, et al. OnabotulinumtoxinA: a meta-analysis of duration of effect in the treatment of glabellar lines. *Dermatol Surg* 2012;38:1794-1803. [PUBMED](#) | [CROSSREF](#)
- Beer KR, Shamban AT, Avelar RL, Gross JE, Jonker A. Efficacy and safety of prabotulinumtoxinA for the treatment of glabellar lines in adult subjects: results from 2 identical phase III studies. *Dermatol Surg* 2019;45:1381-1393. [PUBMED](#) | [CROSSREF](#)
- Mueller DS, Prinz V, Adelglass J, Cox SE, Gold MH, Kaufman-Janette J, et al. Efficacy and safety of letibotulinumtoxin A in the treatment of glabellar lines: a randomized, double-blind, multicenter, placebo-controlled phase 3 study. *Aesthet Surg J* 2022;42:677-688. [PUBMED](#) | [CROSSREF](#)

16. Kane MAC, Brandt F, Rohrich RJ, Narins RS, Monheit GD, Huber MB, et al. Evaluation of variable-dose treatment with a new U.S. botulinum toxin type A (Dysport) for correction of moderate to severe glabellar lines: results from a phase III, randomized, double-blind, placebo-controlled study. *Plast Reconstr Surg* 2009;124:1619-1629. [PUBMED](#) | [CROSSREF](#)
17. Carruthers A, Carruthers J. Prospective, double-blind, randomized, parallel-group, dose-ranging study of botulinum toxin type A in men with glabellar rhytids. *Dermatol Surg* 2005;31:1297-1303. [PUBMED](#) | [CROSSREF](#)
18. Brandt F, Swanson N, Baumann L, Huber B. Randomized, placebo-controlled study of a new botulinum toxin type a for treatment of glabellar lines: efficacy and safety. *Dermatol Surg* 2009;35:1893-1901. [PUBMED](#) | [CROSSREF](#)
19. Fagien S, Carruthers JDA. A comprehensive review of patient-reported satisfaction with botulinum toxin type a for aesthetic procedures. *Plast Reconstr Surg* 2008;122:1915-1925. [PUBMED](#) | [CROSSREF](#)
20. Wang J, Rieder EA. A systematic review of patient-reported outcomes for cosmetic indications of botulinum toxin treatment. *Dermatol Surg* 2019;45:668-688. [PUBMED](#) | [CROSSREF](#)
21. Jiang H, Zhou J, Chen S. Different glabellar contraction patterns in Chinese and efficacy of botulinum toxin type A for treating glabellar lines: a pilot study. *Dermatol Surg* 2017;43:692-697. [PUBMED](#) | [CROSSREF](#)
22. de Almeida AR, da Costa Marques ER, Banegas R, Kadunc BV. Glabellar contraction patterns: a tool to optimize botulinum toxin treatment. *Dermatol Surg* 2012;38:1506-1515. [PUBMED](#) | [CROSSREF](#)
23. Rams DJ, Koziej M, Green JB, Biesman BS, Szczepanek E, Popiela TJ, et al. The relationship between glabellar contraction patterns and glabellar muscle anatomy: a magnetic resonance imaging-based study. *Aesthet Surg J* 2024;45:NP8-NP15. [PUBMED](#) | [CROSSREF](#)