

**An Evaluator Blinded, Split-Neck,
Randomized Placebo Controlled
Clinical Study Investigating the
Efficacy and Safety of Restylane
Refyne for Correction of Static
Horizontal Neck Rhytids Utilizing
Either a Cannula or Needle**

Robyn Siperstein M.D.

August 20th. 2020



An Evaluator Blinded, Split-Neck, Randomized Placebo Controlled Clinical Study Investigating the Efficacy and Safety of Restylane Refyne for Correction of Static Horizontal Neck Rhytids Utilizing Either a Cannula or Needle

Principal Investigator: Robyn Siperstein MD

Sub-Investigator: Lisa Grunebaum, MD

9897 Hagen Ranch Road

Boynton Beach Florida 33472

Phone: 954-494-1400

Fax: 888-650-7801

Email Address: Doctorsip@Sipderm.com

Study Coordinators:

Veronica German, CCRC, Brurva Pierre, CCRC & Stacy Stankiewicz

9897 Hagen Ranch Road

Boynton Beach Florida 33472

Phone: 561-364-7774

Fax: 888-650-7801

Email Addresses: Stacy@Sipderm.com

Sponsor: Siperstein Dermatology Group - Robyn Siperstein, MD

V6.082020

STATEMENT OF COMPLIANCE

The trial will be conducted in accordance with the ICH E6 and the Code of Federal Regulations on the Protection of Human Subjects (45 CFR Part 46). The Principal Investigator will assure that no deviation from, or changes to the protocol will take place without prior agreement from the sponsor and documented approval from the Institutional Review Board (IRB), except where necessary to eliminate an immediate hazard(s) to the trial participants. All personnel involved in the conduct of this study have completed Human Subjects Protection Training.

I agree to ensure that all staff members involved in the conduct of this study are informed about their obligations in meeting the above commitments.

Robyn Siperstein, MD

Title: *An Evaluator Blinded, Split Neck, Randomized Placebo Controlled Clinical Study Investigating the Efficacy and Safety of Restylane Refyne for the Correction of Static Horizontal Neck Rhytids Utilizing Either a Cannula or Needle*

Précis: Twenty Subjects with grades ranging from 1-3 on the Transverse Neckline Scale¹, will be randomized to receive up to 1cc of Restylane® Refyne on one side of their neck utilizing a cannula and up to 1cc of Restylane® Refyne on the other side utilizing a needle. In addition, there will be six subjects in the placebo group that will receive the same treatments with saline instead of Restylane® Refyne. On Day 30, the treatments with the same left-right assignment can be repeated if optimal correction on either side has not been achieved according to the treating investigator. Subjects will be given patient diaries for the first 30 days after each treatment and will return 24-48 hours after their first treatment, to fill out questionnaires, take pictures, and to be assessed by blinded evaluators regarding adverse events. Subjects will return 30 days after their last treatment and on Day 180 and 360 to fill out questionnaires, take pictures, and to be assessed by blinded evaluators regarding adverse events and improvement of static rhytids.

Keywords: Static horizontal neck rhytids, transverse neck lines, tech neck, Restylane Refyne, hyaluronic acid filler, necklace lines

Objectives: Primary Objective: To assess the efficacy of Restylane Refyne in the treatment of static horizontal neck rhytids.

Important Secondary Objectives:

1. To determine the safety and incidence of adverse events when using Restylane Refyne to treat static horizontal neck rhytids
2. To assess patient satisfaction when using Restylane Refyne to treat static horizontal neck rhytids
3. To assess the difference in adverse events, side effects, efficacy, and patient satisfaction when injecting Restylane Refyne to treat horizontal neck rhytids with a cannula versus sharp needle.
4. To assess the longevity of this treatment when treating horizontal neck rhytids

Endpoints: Primary Endpoint: The mean absolute change as graded by blinded evaluators doing live assessments on a 5-point transverse neck line validated scale¹ from baseline to 30 days after the last treatment

Important Secondary Endpoints:

1. The total number of adverse events during the trial
2. The mean absolute change as graded by subjects on a 5-point global aesthetic improvement scale (GAIS) from baseline to 30 days after the last treatment
3. The difference between the two groups on the mean number of adverse events and side effects 24-48 hours after the first treatment, the difference between the two groups in the mean absolute change on both a 5 point transverse neck line¹ scale as rated by blinded evaluators, and the difference between the two groups in the mean absolute change on the global aesthetic improvement scale (GAIS) as rated by subjects.
4. The mean absolute change as graded by subjects on a 5-point global aesthetic improvement scale (GAIS) and the live assessment using the transverse neck line validated scale from baseline to Day 180 and 360

Population: Healthy adults over the age of 21 living in Palm Beach County, Florida.

Sample Size & Sites: Sample size of 26. One Study Site (Boynton Beach, FL)

Duration of Study: 60 Days for the primary study and 360 days for long-term follow up

Study Agent: Restylane Refyne

SCHEMATIC OF STUDY DESIGN

- Total n=26
- Send out to those interested in the study a subject eligibility questionnaire with a request for photographs to be sent to an encrypted email to determine eligibility. This questionnaire will contain a screening consent for both the questionnaire and photographs.

Day 0 Randomization

- Obtain informed consent
- Treatment Group 1 Restylane Refyne: Right Side Cannula, Left Side Needle (n=10)
- Treatment Group 2 Restylane Refyne: Left Side Cannula, Right Side Needle (n=10)
- Treatment Group 3 Saline: Right Side Cannula, Left Side Needle (n=3)
- Treatment Group 4 Saline: Left Side Cannula, Right Side Needle (n=3)

Day 0 Baseline Assessments/ Study Intervention

- Baseline Assessments:
 - Review eligibility questionnaire with medical and cosmetic history, medications, and allergies
 - Pre-procedure pictures taken with Canfield's Intellistudio imaging system (Front, Right and Left Views)
 - Baseline assessment by blinded evaluators using a validated 5-point transverse neck line grading scale¹
 - Baseline safety assessment by examination
- Study Intervention:
 - Up to 1cc of Restylane Refyne or saline will be injected superficially into static horizontal rhytids on each side of the neck utilizing a needle or cannula based on random assignment to Group 1, 2, 3, or 4
 - Post-procedure pictures and questionnaires to be completed by subjects
 - Post-treatment safety analysis performed by injector and compliance analysis performed by study coordinator

Day 1-2 Follow-up Assessment

- Follow-up assessments of study endpoints and safety
 - 24-48 hours post-procedure pictures taken with Canfield's Intellistudio imaging system (Front, Right and Left View)
 - Questionnaires to be completed by subjects
 - Examination of subjects by evaluators assessing adverse effects
 - Safety analysis performed by injector and compliance analysis performed by study coordinator

Day 30 Assessment/Possible Second Intervention

- Follow-up assessments of study endpoints and safety
 - Day 30 post-procedure pictures taken with the Canfield's Intellistudio imaging system (Front, Right and Left Views)
 - Day 30 questionnaires to be completed by subjects
 - Review of subject diary
 - Examination of subjects by blinded evaluators for side effects and assessment using the 5-point transverse neck line grading scale¹
 - Assessment by subjects using GAIS questionnaire
 - Safety assessment by exam
 - Determination by treating investigator whether to continue to final visit assessment or whether additional treatment is needed
- Possible Second Study Intervention:
 - If patients have not achieved optimal correction according the treating investigator, then up to another 1cc of Restylane Refyne or saline can be used on either side according to the same group assignment.
 - Post-procedure Intellistudio pictures and post-procedure questionnaires will be done for these subjects
 - Safety analysis performed by injector and compliance analysis performed by study coordinator for these patients

Day 60 Final Short Term Follow Up for Those Receiving Second Study Intervention

- Follow-up assessments of study endpoints and safety
 - Post-procedure pictures taken with the Canfield's Intellistudio imaging system (Front, Right and Left Views)
 - Day 60 questionnaires to be completed by subjects
 - Review of subject diary

- Examination of subjects by blinded evaluators for side effects and assessment using the 5-point transverse neck line grading scale¹
- Assessment by subjects using GAIS questionnaire
- Safety Assessment by exam

Day 180 Assessment

- Long-Term Follow-up assessments of study endpoints and safety
 - Post-procedure pictures taken with the Canfield's Intellistudio imaging system (Front, Right and Left Views)
 - Day 180 questionnaires to be completed by subjects
 - Examination of subjects by blinded evaluators for side effects and assessment using the 5-point transverse neck line grading scale¹
 - Assessment by subjects using GAIS questionnaire
 - Safety Examination

Day 360 Assessment

- Final Long Term Follow-up assessments of study endpoints and safety
 - Post-procedure pictures taken with the Canfield's Intellistudio imaging system (Front, Right and Left Views)
 - Day 360 questionnaires to be completed by subjects
 - Examination of subjects by blinded evaluators for side effects and assessment using the 5-point transverse neck line grading scale¹
 - Assessment by subjects using GAIS questionnaire
 - Safety Examination

Principal Investigator

Robyn Siperstein, M.D.

Sub-Investigator

Lisa Grunebaum, M.D.

9897 Hagen Ranch Road

Boynton Beach, Florida, 33472

Site Monitor

Olivera Stojadinovic, M.D.

2001 Biscayne boulevard #2514

Miami, FL 33137

Medical Monitor

Julian Wiggan, M.D.

9897 Hagen Ranch Road

Boynton Beach, Florida, 33472

Study Coordinators

Veronica German, CCRC

Brurva Pierre, CCRC

Stacy Stankiewicz

9897 Hagen Ranch Road

Boynton Beach, Florida, 33472

Blinded Evaluators

Elizabeth Nestor M.D.

Suzanne Micciantuono Meran D.O.

Jennifer Richter P.A

Danyelle Dawes, MD

Heidi Gold, M.D.

Lainie Buck, P.A.

2 INTRODUCTION: BACKGROUND INFORMATION AND SCIENTIFIC RATIONALE

2.1 BACKGROUND INFORMATION

The demand for cosmetic treatments off the face continues to rise and patients often request the same cosmetic improvements they can achieve on their face to be matched on adjacent areas such as the neck. Multiple common treatment options are available for neck rhytides starting with minimally invasive procedures such as botulinum toxin and advancing to more invasive procedures such as radiofrequency and ultrasound devices or lasers, and the most invasive options which are surgical.

While most rhytids caused by muscle movements can be treated with minimally invasive treatments such as botulinum toxin injections, some patients have static etched-in lines from repetitive movements which cannot be entirely removed with this treatment. These types of lines can be treated with the correct hyaluronic acid (HA) filler; however, caution needs to be taken with most HA fillers in the neck due to the thin dermis and likelihood of the appearance of bumps if the wrong filler is selected.

There are only a few published reports of hyaluronic acid (HA) fillers being used alone in the neck, one report utilizing a cannula and thereby treating sub-dermally on only 2 patients, and a few others which were retrospective chart reviews limited to patients with Fitzpatrick skin types 4 and 5 and utilizing only photographs without live assessment²⁻⁵.

A validated scale has recently been created to assess transverse neck lines with standardized grading for the purpose of using in clinical trials.¹ However, to date there are no published randomized controlled clinical trials using HA fillers for horizontal neck rhytids.

While HA fillers seem to be a good choice to treat transverse horizontal neck rhytids, the potential for the appearance of bumps, even if transient, needs to be addressed through the proper selection of both filler and technique.

2.2 RATIONALE

There are many HA fillers with different properties allowing for specific use of each for different indications. XpresHAN technology, Restylane Refyne in particular, is an ideal fit for filling in etched lines in an area with a thin dermis due to its small particle size and also its ability to spread and integrate into surrounding tissue, therefore creating a smooth appearance and a natural look in this dynamic area.

While the use of a cannula is known to cause less side effects such as bruising, cannulas only treat sub-dermally, therefore treating only those rhytids caused by deeper volume loss. Utilizing a sharp needle more superficially in the dermis may be necessary to treat the most severe etched lines once thought to be non-effaceable (Level 3 patients on the Transverse Line Scale). In this study each technique will be used on one side and analyzed to see if one is superior in efficacy, safety, and cosmetic downtime.

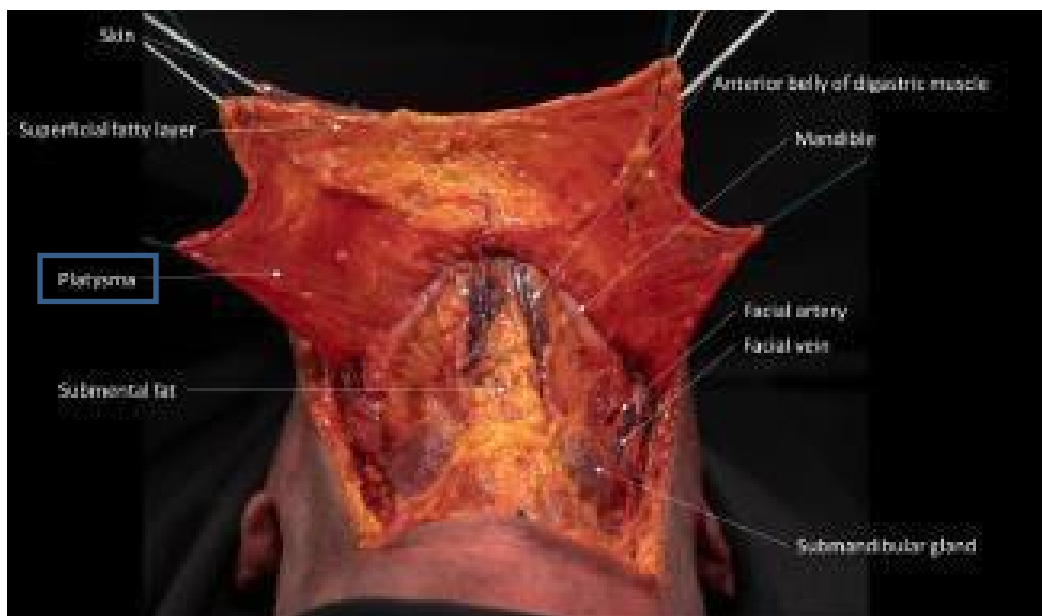
2.3 POTENTIAL RISKS AND BENEFITS

2.3.1 KNOWN POTENTIAL RISKS & RISK ANALYSIS

Common potential risks include swelling, urticaria, itching, pain, bumps, bruising, damage to underlying neck structures, atrophy/scarring, capillary disorders such as telangiectasia, dermatitis, discoloration, erythema, granuloma, hypersensitivity, infection, inflammation, neurological symptoms such as hypoesthesia, pain/tenderness, papules/nodules, pruritus, and reactivation of herpes infection. ⁶

Rare but potential risks with any hyaluronic acid gel include vascular occlusion from intravascular injection or vascular compression. This can cause necrosis at the site or in the area supplied by the affected blood vessels or rarely as changes in vision including blindness due to embolization, or cerebral ischemia or hemorrhage, leading to stroke. These rare adverse events have been reported immediately to a few days following injection and may be reversed with an injection of hyaluronidase. ⁶

In order to effectively treat neck rhytids, the injection of Restylane Refyne needs to be superficial in the dermis or superficial to the superficial fatty layer which is located above the platysma. This injection plane is a very low risk area. All concerning neurovascular structures course well below this area. The dermal plane of injection is the same as the FDA approved indication for this product, the only difference is the injection will be done on the neck, but the neck is a regular subcutaneous body region which follows the regular layered arrangement of subcutaneous architecture: skin, superficial fatty layer, platysma, deep fatty layer, deep cervical fascia. All significant arteries and veins are in the deep fatty layer. The superficial fatty layer above the platysma pictured below, is free of major neurovascular structures. The platysma is known to cover all cervical neurovascular structures like a blanket including the external jugular vein. See cadaveric dissection below which show the important structures underneath and protected by the platysma.



Risks will be minimized by using a superficial injection technique by holding a 4mm 32 gauge needle parallel to the skin and once inserted, pulling upward to ensure visualization in the proper plane during the injection (see photo below).

Photo of injection technique showing visualization of the needle in the skin and a small .01 cc bleb of HA filler



Photo of a ½ inch length needle used for injections on the face at the top of the picture and 4mm needle used in this protocol against the ruler in order to mitigate risk



To further mitigate any risks, the study investigator chosen is qualified by education and experience, and is highly skilled in the use of dermal fillers with an exceptional knowledge of the vascular anatomy and understanding of the depth and plane of injection, as well as knowledge of the signs and symptoms and management of potential complications to assure proper device implantation and management of study risk.

As the superficial fatty layer of the neck is free of neurovascular cervical structures, the risk for adverse events is reduced to other areas of the head and neck. No anatomical variation is known where major neurovascular cervical structures course superficial to the platysma. Any risk is minimized by using the injection technique shown in the picture above that ensures visualization in the proper plane well above the platysma during the entire procedure. Patient safety is assured with the proposed technique as no deep application of the product is performed.

The region for injection covers the total anterior cervical region depending on the extent of the cervical wrinkles. Of greater importance is the depth of the injection which is strictly subdermal. Deeper injection planes will not be effective for the desired aesthetic outcome. Cervical wrinkles are intradermal changes in dermal thickness and stability resulting from continuous neck movement and platysmal contractions. The observed wrinkles are classified as static wrinkles which can be best treated by intradermal or strictly subdermal soft tissue filler injections.

Photo of the treatment area outlined in the photo below and includes the entire anterior neck. Any horizontal rhytids in this area will be treated.



The organs of the neck (thyroid/parathyroid) are located deep to the deep cervical fascia whereas the cervical branches of the facial nerve are located deep to the platysma. The external jugular vein is located below the platysma through its entire course through the neck. The performed injection technique is superficial to all mentioned cervical structures. The primary investigator is knowledgeable of all these structures and will examine each subject's neck thoroughly including both visual assessment and palpation for underlying structures. These structures will be marked before the procedure begins in order for the primary investigator to be cognizant and mindful of these structures while injecting in order to further mitigate risks

There will be no deep application of the product and there will be close monitoring of all subjects before, during, and 30 minutes after the treatment with immediate halting of the study if any serious side effects occur (section 7.7). This safety assessment will include Snellen visual acuity, the ability to tighten the platysma when saying "E" and frowning, confrontational visual fields, ocular motility, oxygen saturation, heart rate, blood pressure, a visual exam looking for discoloration of the skin, and a pain scale. An Intravascular Treatment Protocol is available for reference (section 6.7) if an intravascular event occurs; and the subject diary and case report form includes instructions in case of severe intravascular events.

The following trials have already shown safety and efficacy treating this condition (neck rhytids) with hyaluronic acid filler.

- Tseng F, Hsiaoen, Y. Treatment of Horizontal Neck Wrinkles with Hyaluronic Acid Filler: A Retrospective Case Series. *PRS Global Open*. 2019 Aug.
- Han TY, Lee JW, Lee JH, Son SJ, Kim BJ, Mun SK, Kim MN, Lee CK. Subdermal minimalsurgery with hyaluronic acid as an effective treatment for neck wrinkles. *Dermatol Surg*. 2011 Sep;37(9):1291-6
- Lee SK, Kim HS. Correction of horizontal neck lines: Our preliminary experience with hyaluronic acid fillers. *J Cosmet Dermatol*. 2017; 00: 1–6.

In addition, the following trials have shown safety and efficacy with non-reversible fillers which are not as safe as the hyaluronic acid filler that we plan to use which can be reversed if needed with hyaluronidase.

- Yutskovskaya Y, Kogan E. Improved Neocollagenesis and Skin Mechanical Properties After Injection of Diluted Calcium Hydroxylapatite in the Neck and Decolletage: A Pilot Study. *Journals of Drugs in Dermatology*. 2017 Jan
- Chao YY, Chiu HH, Howell DJ. A Novel injection technique for horizontal neck lines correction using calcium hydroxylapatite. *Dermatol Surg*. 2011 Oct;37(10):1542-5.

2.3.2 KNOWN POTENTIAL BENEFITS

Improvement of the subject's neck rhytids leading to an aesthetic improvement. This aesthetic improvement may positively impact the subjects' psychological well-being.

3 STUDY DESIGN, OBJECTIVES, AND ENDPOINTS

3.1 DESCRIPTION OF THE STUDY DESIGN

- A Prospective, Evaluator-Blinded, Split-Neck, Randomized Placebo Controlled Clinical Study
- Single Center
- Study agent: Restylane Refyne
- Study Intervention on Day 0 and Possible Second Intervention on Day 30
- Evaluations by Blinded Investigators on Day 0, Day 1-2, Day 30, 60, 180 and 360 for those with a second intervention.

This will be a prospective, evaluator-blinded, split-neck, placebo controlled clinical study in which patients will be randomized to 4 groups.

The first group will receive up to 1cc of Restylane Refyne into the rhytids on the right side of the neck utilizing a 27-gauge cannula, while the left side will be injected with a sharp 32 gauge needle.

The second group will be reversed with the left side treated with Restylane Refyne utilizing a cannula and the right side treated with a sharp needle.

The third group will receive up to 1cc of saline into the rhytids on the right side of the neck utilizing a 27-gauge cannula, while the left side will be injected with a sharp 4mm 32 gauge needle.

The fourth group will be reversed with the left side treated with saline utilizing a cannula and the right side treated with a sharp needle.

All groups will be utilizing the same safety and comfort protocols established by the practice which include the use of topical numbing cream (B.L.T.), cleansing with alcohol, and a cold ice pack at the site of injection after the procedure.

Before and after the procedure a blinded evaluator will be grading the subjects' static horizontal neck lines using a validated 5-point grading scale. This assessment will be done by evaluating live subjects similar to the approach used in the validation study.¹ The blinded evaluators will also be grading the subjects side effects such as bruising and swelling 24-48 hours after the first treatment. A second treatment is allowed on Day 30 for those who have not achieved optimal correction. During and after every treatment, subjects will be evaluated for immediate side effects and will fill out post procedure questionnaires. Either on Day 30 or Day 60, a final short-term assessment and two long-term follow up assessments on Day 180 and 360 will be done. During these follow ups, subjects will have photos taken using Canfield's Intellistudio imaging system, along with live assessment by blinded evaluators regarding long-term side effects and improvement on a 5point neck line grading scale¹. Subject will also fill out long-term questionnaires on satisfaction, improvements, and side effects.

3.2 STUDY OBJECTIVES & ENDPOINTS

3.2.1 STUDY OBJECTIVES

Primary:

To assess the efficacy of Restylane Refyne in the treatment of static horizontal neck rhytids based on the improvement according to a validated 5-point grading scale.¹

Secondary:

• To assess the safety and number of adverse events when using Restylane Refyne to treat static horizontal neck rhytids

• To assess patient satisfaction when using Restylane Refyne to treat static horizontal neck rhytids

•• To assess the difference in efficacy, patient satisfaction, adverse events and side effects utilizing two different injection methods.

-- To assess the longevity of this treatment when treating horizontal neck rhytids

3.2.1 PRIMARY ENDPOINT

- Difference in the mean absolute change from baseline to 30 days after the last treatment on a 5-point validated transverse lines scale¹ as scored by blinded evaluators doing live assessments.

This endpoint will show if injections of Restylane Refyne in the horizontal static neck rhytids provides significant improvement in the aesthetic appearance of the neck area.

3.2.2 SECONDARY ENDPOINTS

- The number of reported adverse events
- Mean absolute change from baseline on the GAIS as rated by subjects
- The difference between the two groups in the mean number of adverse events and side effects 24-48 hours after the first treatment, the difference in the mean absolute change on both a 5-point transverse neck line scale¹ as rated by blinded evaluators doing live assessments, and the difference in the mean absolute change on the global aesthetic improvement scale (GAIS) as rated by subjects.
- Difference in the mean absolute change from baseline to 180 days and 360 days after the last treatment on a 5-point validated transverse lines scale¹ as scored by blinded evaluators doing live assessments.

These endpoints will show if injections of Restylane Refyne are safe in the neck, if patients believe the treatment leads to significant improvement of their neck rhytids, if there is a difference in the safety, side effects, and efficacy between the two different injection techniques, and the longevity of any significant improvements of neck rhytids with this device.

4 STUDY ENROLLMENT AND WITHDRAWAL

4.1 PARTICIPANT INCLUSION CRITERIA

In order to be eligible to participate in this study, an individual must meet all the following criteria:

- Provision of signed and dated informed consent form
- Stated willingness to comply with all study procedures and availability for the duration of the study
- Male or female, aged 22 or older
- In good general health as evidenced by medical history and brief exam
- For females of reproductive potential: use of highly effective contraception
- Score of 1-3 on a validated transverse neck line scale¹
- Negative urine pregnancy test for females of childbearing potential at screening and all injection visits.

4.2 PARTICIPANT EXCLUSION CRITERIA

An individual who meets any of the following criteria will be excluded from participation in this study:

- Subjects with allergies to hyaluronic acid filler or lidocaine.
- Subjects with autoimmune conditions
- Subjects with diabetes
- Subjects taking anti-coagulants, chemotherapy, immunosuppressive agents, immunomodulatory agents, diuretics, anti-histamines, or anti-inflammatory medications in the 2 weeks prior to the study or who will need to take these medications at any time during the first 60 days of the study.
- Subjects with any scheduled laser, light, or surgical procedures during the study, including dental surgery.
- Subjects who had neuromodulators in the past 6 months or fillers in the previous 2 years in the neck area
- Subjects who at any time had surgery or permanent fillers in the neck area
- Subjects with scars, tattoos, or many skin growths in the neck area
- Subjects unwilling or unable to sit still while an injector places Restylane Refyne in the neck
- Subjects unwilling or unable to keep their head still during the photos
- Subjects who are pregnant or nursing
- Female subjects unable to take or use some form of birth control
- Subjects with any neck bruising or swelling
- Subjects with current skin infections, tumors, herpes outbreak or dermatitis on the neck
- Subjects using skin-irritating topical preparations, pigmenting agents (self-tanning or bleaching creams), in the past two weeks or for the first 60 days of the study on the neck.
- Subjects using topical or oral retinoids or steroids for the last 6 months or for the duration of

the study on the neck

- Any medical condition that in the opinion of the Investigator would make the subject unsuitable for inclusion
- Women who are pregnant or breast feeding, or women of childbearing potential who are not practicing adequate contraception or planning to become pregnant during the study period.
- Study site personnel, close relatives of the study site personnel (e.g. parents, children, siblings, or spouse), employees, or close relatives of employee.

4.3 STRATEGIES FOR RECRUITMENT AND RETENTION

- Participants will be compensated for their participation in the study by having the opportunity to improve their neck rhytides by receiving up to 2cc of Restylane Refyne without any cost to the subject on each side of their neck.
- Subjects will receive both text, email, and phone call reminders for all their scheduled visits.
- 20 treatment subjects will be needed in the study to accommodate for a 10% expected dropout rate in order to have 18 subjects complete the entire study.
- The source of the subjects will be those who visited Siperstein Dermatology Group and allowed their information to be used in order to be contacted for promotions and trials. This list includes over 3000 patients.
- The subjects on this list will first be contacted via email to see if they are interested in the trial. If they respond, they will then be sent a subject questionnaire in order to screen for those who satisfy the inclusion/exclusion criteria. The first 26 subjects that satisfy these criteria will be scheduled for the trial.

4.4 PARTICIPANT WITHDRAWAL OR TERMINATION

4.4.1 REASONS FOR WITHDRAWAL OR TERMINATION

Participants are free to withdraw from participation in the study at any time upon request. An investigator may terminate participation in the study if:

- The participant's health declines requiring hospitalization
- The participant needs any surgery or other serious medical interventions
- The participant meets an exclusion criterion (either newly developed or not previously recognized) that precludes further study participation.
- The participant fails to show up for follow-up and final visits

4.4.2 HANDLING OF PARTICIPANT WITHDRAWALS OR TERMINATION

The primary investigator will attempt to visit all participants that have withdrawn or terminated from the trial if they are unable to make their follow-up visits to ensure as much data as possible has been collected and that all safety parameters have been met.

4.5 PREMATURE TERMINATION OR SUSPENSION OF STUDY

This study may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause. If the study is prematurely terminated or suspended, the PI will promptly inform the IRB and Galderma as well as provide the reason(s) for the termination or suspension.

Circumstances that may warrant termination or suspension include, but are not limited to:

- Determination of unexpected, significant, or unacceptable risk to participants
- Insufficient compliance to protocol requirements

Study may resume once concerns about safety, protocol compliance, or data quality is addressed and satisfy the IRB.

5 STUDY AGENT

5.1 STUDY AGENT(S), ACQUISITION & STORAGE

5.1.1 ACQUISITION

Restylane Refyne will be supplied by Galderma

5.1.2 FORMULATION

Restylane Refyne is a sterile, biodegradable, viscoelastic, non-pyrogenic, clear, colorless, and homogeneous soft hyaluronic acid gel with a moderate lifting capacity. The product has a sodium hyaluronate concentration of 20 mg/mL in phosphate buffered saline at pH 7 and contains 3 mg/mL lidocaine hydrochloride.⁶

5.1.3 PRODUCT STORAGE AND STABILITY

The product will be stored in its original package in an air-conditioned building with temperatures ranging from 72-78 degrees Fahrenheit. The product will remain in a locked cabinet until it is ready to be used on subjects.

5.1.4 PREPARATION

The product will be removed from original packaging and placed on a sterile field no longer than 60 minutes before it is injected into the subject. The subject's neck will be cleansed with Cetaphil wipes in order to remove all topicals such as make-up, sunscreen and moisturizers. A topical numbing cream will be applied for 30 minutes and then removed before the subject's neck will be cleansed with alcohol immediately prior to injection.

5.1.5 DOSING AND ADMINISTRATION

Each subject will receive up to 1cc of Restylane Refyne or saline for each side of the neck between 8am and 6pm on Day 0. Each subject will have the option of receiving up to an additional 1cc of Restylane Refyne or saline on Day 30+/-2 Days if the injector deems it necessary to achieve optimal correction.

5.1.6 ROUTE OF ADMINISTRATION

Restylane Refyne will be injected with either a 27 gauge cannula or a 32 gauge 4mm needle. The injections will be placed only in the dermis or superficial fatty layer, and the needle or the cannula will be injected perpendicular to the skin and directed upward so that the location of the tip of needle or cannula can be visualized during the entire treatment as shown in the picture below.



5.1.7 DURATION OF THERAPY

The subjects will be in the primary study which involves treatment interventions for 30-60 days.

5.1.10 TRACKING OF DOSE

The study coordinator will check each vial of Restylane Refyne after each procedure to ensure the correct documentation was done on each subject's chart.

5.1.11 DEVICE SPECIFIC CONSIDERATIONS

- TSK 32 Gauge x ½ inch needle
- TSK Blunt-Tipped MicroCannula 27 Gauge 1.5 Inch
- Canfield Photo-analysis System, Intellistudio

5.2 STUDY AGENT ACCOUNTABILITY PROCEDURES

The product will be shipped from Galderma through Mckesson Distributor. Once received the Study Coordinator will place the product in a locked cabinet until it is ready to be used on patients. The Study Coordinator will oversee assigning the product to each subject and ensuring before the subject leaves that they have received the correct amount.

6 STUDY PROCEDURES AND SCHEDULE

6.1 STUDY PROCEDURE/EVALUATIONS

6.1.1 STUDY SPECIFIC PROCEDURES

- Medical history obtained from baseline subject questionnaire and verbally verified
- Examination by blinded evaluators
- Canfield photo imaging
- Counseling the subjects on the product (Restylane Refyne) or placebo(saline) that they will be receiving
- Counseling the subjects that they will be injected with two different methods on each side of their necks until the end of the study
- The study coordinator will check before the patient leaves that all appropriate photos and paperwork have been completed.

6.1.2 STANDARD OF CARE STUDY PROCEDURES

The patients will be cleaned with Cetaphil wipes and then alcohol prior to injection. A cold ice pack will be applied after the procedure for several minutes.

6.2 STUDY SCHEDULE

6.2.1 SCREENING

Screening Visit (Day -30 to 0)

- Send out and review subject eligibility questionnaires containing a screening consent and request for photographs to be sent to an encrypted email to determine eligibility.
- Schedule study visits for participants who are eligible and available for the duration of the study.
- Provide participants with specific instructions needed to prepare for first study visit and all subsequent visits.

6.2.2 ENROLLMENT/BASELINE

Enrollment/Baseline Visit (Visit 1, Day 0)

- Review in depth the study design with subjects and obtain informed consent of potential participants verified by signature
- Perform examination including a baseline safety examination and verbally review subject's medications and medical history to determine eligibility based on inclusion/exclusion criteria.

- Verify all inclusion/exclusion criteria.
- Obtain demographic information
- Baseline transverse neck line scores recorded from blinded evaluators doing live assessments
- Pre-procedure photos
- Pre-procedure safety examination for baseline
- Administer up to 1cc of Restylane Refyne or saline on each side of the neck with either a needle or cannula according to randomized groups
- Record adverse events as reported by participant or observed by investigator.
- Perform protocol adherence check by study coordinator
- Collect post-procedure subject questionnaires
- Take post-procedure photos

6.2.3 FOLLOW-UP

Follow-up Visit 1 (Visit 2, Day 1-2) 24-48 hours after 1st injection

- Record adverse events as reported by participant or observed by investigator.
- Collect Day 1 subject questionnaires
- Take Photos
- Review the subject diary instructions
- Record immediate side effect scores from blinded evaluators.
- Safety Examination checking for AEs

Follow-up Visit 2 (Visit 3, Day 30 +/-2)

- Record adverse events as reported by participant or observed by investigator.
 - Take Photos
 - Collect questionnaires
 - Record participant's adherence to the study
 - Record Day 30 transverse neck line scores from blinded evaluators
 - Record Prolonged Side effect scores from blinded evaluators.
 - Review the subject's diary
 - Safety Examination checking for AEs
-
- Any subjects achieving optimal cosmetic improvement as determined by the injector (end of short term study), but in those subjects whose left and right sides differ in score on the transverse neck line scale, allow patients to have the side with the lower score to be injected with up to 2cc of Restylane Refyne with the technique used on the higher scoring side once the short term study has been completed.
 - Any subjects not achieving optimal cosmetic improvement as determined by the injector will be administered up to 1cc of additional Restylane Refyne or saline on either side of the neck with either a cannula or needle according to their original randomized grouping.
 - Perform protocol adherence check by study coordinator
 - Collect post-procedure subject questionnaires
 - Perform post-procedure safety examinations
 - Take Post-procedure photos
 - Safety Examination checking for AEs

Follow Up Visit 3 (Visit 4, Day 60 +/-2) for those receiving a second treatment on Day 30

- Record adverse events as reported by participant or observed by investigator.
- Collect questionnaires
- Take Photos
- Safety Examination checking for AEs
- Record participant's adherence to the study
- Record Side effect scores from blinded evaluators
- Record transverse neck line scores from blinded evaluators
- Record GAIS from patients and evaluators
- If subjects left and right sides differ in score on the transverse neck line scale, allow patients to have the side with the lower score to be injected with up to 2cc of Restylane Refyne with the technique used on the higher scoring side once the short term study has been completed.

Follow Up Visit 3 or 4 (Visit 4 or 5, Day 180 +/-5)

- Record adverse events as reported by participant or observed by investigator.
- Collect questionnaires
- Take Photos
- Safety Examination checking for AEs
- Record participant's adherence to the study
- Record Side effect scores from blinded evaluators
- Record transverse neck line scores from blinded evaluators
- Record GAIS from patients and evaluators

6.2.4 FINAL STUDY VISIT**Final Visit (Visit 5 or 6, Day 360 +/-7)**

- Record adverse events as reported by participant or observed by investigator.
- Collect questionnaires
- Take Photos
- Safety Examination checking for AEs
- Record participant's adherence to the study
- Record Side effect scores from blinded evaluators
- Record transverse neck line scores from blinded evaluators
- Record GAIS from patients and evaluators
- Provide final instructions

6.2.5 EARLY TERMINATION VISIT

- Record adverse events as reported by participant or observed by investigator.
- Collect questionnaires
- Take Photos

- Record participant's adherence to treatment program
- Provide final instructions

6.2.6 UNSCHEDULED VISIT

All unscheduled visits will be discouraged unless emergent. All unscheduled visits will be recorded and kept as brief as possible with no active intervention unless a vascular occlusion is suspected, or other emergent health condition is suspected.

6.2.7 SCHEDULE OF EVENTS TABLE

	Screening	Enrollment/Baseline Visit 1, Day 0	Visit 2, Day 1 ^a 2	Visit 3, Day 30	Visit 4, Day 60	Visit 5, Day 180	Visit 6, Day 360
Procedures							
Informed consent	x	X					
Demographics		X					
Medical history		X					
Subject Questionnaire	X	X	X	X	X	X	X
Safety Exam		X	X	X	X	X	X
Randomization		X					
Exam by blinded Evaluator		X	X	X	x	X	x
Intervention		X		+/- ^a X			
Evaluation for Adverse Effects		X	X	X	X	X	X
Photos		X	X	X	X	X	X

6.3 Photography

Photographs will be taken prior to treatments with study product and at every follow-up visit in order to document treatment effect. Photographs may also be taken to document AEs at the treating investigator's discretion. Baseline photographs may be used as a reference in the GAIS assessment by the treating investigator and subject. Site personnel will be thoroughly trained in the photographic equipment and techniques before study start.

A Photography Standard Operating Procedure will be provided in a separate user guide.

At the end of the study, the blinded evaluator will assess improvement in the neck by comparison of random, blinded pairings of the baseline and post-baseline photographs taken at 1 month after the last treatment and at months 6 and 12. The reviewer will answer the question "Which picture represents post treatment?" by either "Left", "Right", or "Neither". Analysis of the results will be translated to improvement "Yes" or "No".

6.4 Subject Questionnaires, GAIS, & Subject Diary

Subjects will be asked about their satisfaction with treatment using a subject Satisfaction questionnaire and GAIS at every follow up visit. The 30 days subject diary (starting on the day of each treatment) will include a question on when the subject feels comfortable returning to social engagement (public/social appearances) exemplified as returning to business office or other public workplace, having dinner in a public restaurant, attending a social event/gathering such as dinner party, etc.

6.5 PROHIBITED MEDICATIONS, TREATMENTS, AND PROCEDURES

Treatment with any oral anti-coagulants, anti-histamines, anti-inflammatory medications, corticosteroids, retinoids, diuretics, and topical retinoids or steroids are prohibited during the first 60 days of the study as well as any neck procedures or surgeries. In the event a subject takes these medications or has treatments or procedures, the primary investigator will make the final decision regarding early termination from the clinical trial.

6.6 PROPHYLACTIC MEDICATIONS, TREATMENTS, AND PROCEDURES

None will be used

6.7 RESCUE MEDICATIONS, TREATMENTS, AND PROCEDURES

If patients develop an infection during the trial, they will be prescribed the appropriate anti-viral or anti-bacterial medication but will continue in the study. If patients have symptoms suspicious for vascular occlusion at any time, they will be treated with hyaluronidase and excluded from the study, see vascular occlusion protocol below

EMERGENCY ASSISTANCE PROTOCOL

Vascular Occlusion Protocol

1. Flood the area with 300 units (2 vials) of Hylenex (hyaluronidase).
2. Observe the patient for capillary refill/clinical improvement. If no improvement is seen after one hour, inject another 1cc of Hylenex. Continue to observe patient in office and inject more Hylenex as needed.
3. Apply nitropaste to area after considering any contraindications/cautions (concurrent use of

- antihypertensive medication, mucosal application site, etc)
4. Aspirin 325mg PO x1 in office; Repeat daily for 5-7 days
 5. Frequent warm compresses and vigorous massage (patient should continue this treatment at home)
 6. Immediate transportation to a hospital that can manage pulmonary, embolic, cardiac, and cerebral embolic events if medical exam or vital signs are concerning (9655 W Boynton Beach Blvd, Boynton Beach, FL 33472)
 7. Follow up with patient daily in office or via photos if necessary
 8. Additional options if needed:
 - Prednisone 20-30mg PO daily x 2-3 days if significant swelling
 - Hyperbaric Oxygen: Boynton Beach: Bethesda Health City, 10301 Hagen Ranch Rd Ste B1 Boynton Beach, FL 33437, (561) 374-5441

Retinal artery Occlusion Protocol

(Vision loss within seconds of filler injection with or without pain)

1. Timolol 0.5%, one drop
2. Administer Acetazolamide 500mg PO x 1 (Pt CANNOT take if sulfa allergy)
3. Initiate ocular massage:
 - a. Ensure that the patient's eyes are closed.
 - b. Apply firm pressure on the eyeball through the closed eyelids. Pressure should be firm enough that the eyeball is indented about 2–3 mm.
 - c. Apply firm pressure for 5–15 seconds and quickly release.
 - d. Repeat this cycle for at least five minutes.
 - e. At the end of five minutes, check if the patient's vision in the affected eye is brighter.
4. Refer immediately to Dr. Steven Fagien for retrobulbar hyaluronidase
Injection and arrange transportation: (561) 393-9898, 660 Glades Rd # 210, Boca Raton, FL 33431

If any symptoms of a pulmonary, cardiac or cerebral ischemic event, immediately transfer to the closest hospital

7 ASSESSMENT OF SAFETY

7.1 ASSESSMENT OF AEs

Safety evaluations for this study include an interview of the subjects at each visit to obtain information about any medical occurrence that meets the definition of an AE. Each subject should be questioned about AEs at each study visit following the screening visit. The question should be asked: “Since your last clinical visit have you had any health problems?” Information on AEs can also be obtained from signs and symptoms detected during each examination by the Investigator or designee, which should include visual inspection of the treatment area.

AEs must be documented in the source document without regard for cause or relation to investigational product. If in the process of the interview, additional information regarding medical history or pre-planned medical or surgical procedures is revealed, it must be documented in the source documents.

It is the responsibility of the Investigator to determine severity of the AE and relatedness of the event to the study product.

7.2 ADVERSE EVENTS OF SPECIAL INTEREST

All incidences of visual disturbances, regardless of relationship to study product or seriousness, are considered Adverse Events of Special Interest (AESIs) and include but are not limited to the following: any change in vision, any loss of vision, blurry vision, double vision, pain in or around the eye, blind spot or shadow in the visual field, trouble moving eyes, any change in Snellen visual acuity including change of one line or greater.

AESIs should be reported within 24 hours to the IRB and FDA and appropriate follow up should be conducted in order to determine the cause, severity, seriousness, outcome, and relationship to the study product or procedures

7.2.1 Visual Function Assessments

Vision function assessments will be performed both prior to and 30 minutes post injection of the study product at baseline and optional touch-up treatment. These assessments include Snellen visual acuity, extraocular muscle function, and confrontation visual field testing.

Snellen visual acuity test

A Snellen Eye Chart will be used to assess visual acuity for distance vision. Visual Acuity will be conducted using the subject’s best distance correction (e.g. contacts or eye glasses) at a distance of approximately 6 feet from the chart. Each eye will be measured separately by either using an occluder or having the subject cover each eye. The subject will be asked to start reading the letters at the top of the chart working their way to the bottom. The smallest row of letters that the subject can read will indicate their visual acuity listed on the chart. Snellen Visual Acuity will be recorded in their source documentation. If there is one or more visual acuity line changes during the course of study, these will be reported within 24 hours to the IRB and FDA. These events should be reviewed and assessed by the Investigator as an Adverse Event.

Extraocular muscle function test

Extraocular muscle function testing examines the function of the eye muscles. This test observes the movement of the eyes in six specific directions to evaluate weakness or other problem in the extraocular muscles. The subject will be asked to sit or stand with their head up and looking straight ahead. The assessor will hold a pen or other object approximately 16 inches in front of the subject's face. The assessor will then move the object in several directions and ask the subject to follow it with their eyes, without moving their head. Normal or Abnormal will be recorded in the source documentation. Changes in movement of the eyes during the course of the study should be reviewed and assessed by the Investigator for a potential Adverse Event.

Confrontation visual field

The Confrontation Visual Field test will be used to assess the subject's peripheral vision. The examiner will sit facing the subject, about 3 to 4 feet away from the subject and ask the subject to fix their gaze on the examiner's eye. The examiner will hold their arms straight out to the side and bring their hands into the subject's visual field from the sides in each quadrant. The subject will signal as soon as the hand is seen. Each quadrant will be recorded as normal or abnormal in the source documentation. Changes in the visual field of the eyes during the course of the study should be reviewed and assessed by the Investigator for a potential Adverse Event

7.2.2 Subject Diary

The subject shall evaluate local tolerability in a 30 days diary, starting on the day of each treatment. The presence and maximum intensity of pre-defined, expected post-treatment events, i.e. bruising, redness, pain, tenderness, lumps/bumps, itching and swelling shall be assessed for the treated area. The Subject Diary will also have an area for the subject to report any other symptoms outside the pre-identified. Especially symptoms like change in vision change in skin color, dizziness, confusion, weakness or numbness in arms or legs, changes to consciousness or alertness, difficulty speaking/speech impairment, face droop, headache and fever should be recorded. The diary will also include instructions for the subject to:

- Contact the clinic and an eye doctor (retinal specialist or ophthalmologist) if experiencing change in vision (i.e. vision loss, blurriness, double vision, pain in or around eye, blindness, blind spots, problems moving your eyes).
- Call 911 and seek immediate medical attention if signs of stroke or subject experience dizziness, confusion, weakness or numbness in face, arms or legs, changes to consciousness or alertness, difficulty speaking/speech impairment, face droop

The Investigator or study staff should review the diary for completion and clarify any events as needed. For events that are ongoing at the time of diary completion, the Investigator should follow them and record resolution in the subject's study file.

The subject diary will also include a question on when the subject felt comfortable returning to social engagement after an injection.

7.2.3 Pregnancy Test

For all women of childbearing potential, including those currently using contraception, a urine pregnancy test is required prior to receiving any study treatment. The test result must be negative for the subject to receive any treatment with study product.

7.3 ADVERSE EVENTS

7.3.1 DEFINITION OF SERIOUS ADVERSE EVENTS (SAE)

Adverse event means any untoward medical occurrence associated with the use of an intervention in humans, whether considered intervention-related (21 CFR 312.32)

Serious adverse event or serious suspected adverse reaction.

An adverse event or suspected adverse reaction is considered "serious" if, in the view of either the investigator or sponsor, it results in any of the following outcomes: death, a life-threatening adverse event, inpatient hospitalization or prolongation of existing hospitalization, a persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions, or a congenital anomaly/birth defect. Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

7.3.2 DEFINITION OF UNANTICIPATED PROBLEMS (UP)

Unanticipated problems involving risks to participants or others to include, in general, any incident, experience, or outcome that meets all the following criteria:

- Unexpected in terms of nature, severity, or frequency given (a) the research procedures that are described in the protocol-related documents, such as the IRB-approved research protocol and informed consent document; and (b) the characteristics of the participant population being studied;
- Related or possibly related to participation in the research ("possibly related" means there is a reasonable possibility that the incident, experience, or outcome may have been caused by the procedures involved in the research); and
- Suggests that the research places participants or others at a greater risk of harm (including physical, psychological, economic, or social harm) than was previously known or recognized.

With these events the following will occur:

- *Modification of inclusion or exclusion criteria to mitigate the newly identified risks*
- *Implementation of additional safety monitoring procedures*
- *Suspension of enrollment of new participants or halting of study procedures for enrolled participants*
- *Modification of informed consent documents to include a description of newly recognized risks*
- *Provision of additional information about newly recognized risks to previously enrolled participants*

7.4 CLASSIFICATION OF AN ADVERSE EVENT

7.4.1 SEVERITY OF EVENT

- **Mild** – Events require minimal or no treatment and do not interfere with the participant's daily activities.
- **Moderate** – Events result in a low level of inconvenience or concern with the therapeutic measures. Moderate events may cause some interference with functioning.
- **Severe** – Events interrupt a participant's usual daily activity and may require systemic drug therapy or other treatment. Severe events are usually potentially life-threatening or incapacitating.

7.4.2 RELATIONSHIP TO STUDY AGENT

All AEs will have their relationship to study agent or study participation assessed. Evaluation of relatedness will consider etiologies such as natural history of the underlying disease, concurrent illness, concomitant therapy, study-related procedures, accidents, and other external factors.

The clinician's assessment of an AE's relationship to study agent is part of the documentation process, but it is not a factor in determining what is or is not reported in the study. If there is any doubt as to whether a clinical observation is an AE, the event should be reported. All AEs must have their relationship to study agent assessed. In a clinical trial, the study product must always be suspect. To help assess, the following guidelines are used.

- **Related** – The AE is known to occur with the study agent, there is a reasonable possibility that the study agent caused the AE, or there is a temporal relationship between the study agent and event. Reasonable possibility means that there is evidence to suggest a causal relationship between the study agent and the AE.
- **Not Related** – There is not a reasonable possibility that the administration of the study agent caused the event, there is no temporal relationship between the study agent and event onset, or an alternate etiology has been established.

7.4.3 EXPECTEDNESS

The primary investigator will be responsible for determining whether an AE is expected or unexpected. An AE will be considered unexpected if the nature, severity, or frequency of the event is not consistent with the risk information previously described for the study agent.

7.5 RECORDING INSTRUCTIONS

Investigators, or other study site personnel, shall record all AEs including:

- a) Event term (recorded in standard medical terminology and avoiding abbreviations)
- b) Affected area
- c) Start date (first day with symptoms)
- d) Stop date (last day with symptoms)
- e) Intensity (mild, moderate, or severe)
- f) Seriousness (serious or not serious)
- g) Causal relationship to study product or study product injection procedure (yes or no)
- h) Action taken (none, medication treatment, non-pharmacological treatment, or other procedures/tests, subject withdrawn)
- i) Outcome of the AE (ongoing, recovered, recovered with sequelae, death, chronic/stable, not recovered at the end of the study)

All AEs, non-serious as well as serious, are to be reported as an AE. The PI will record all reportable events with start dates occurring any time after informed consent is obtained until 7 (for non-serious AEs) or 14 days (for SAEs) after the last study intervention. At each study visit, the investigator will inquire about the occurrence of AE/SAEs since the last visit. Events will be followed for outcome information until resolution or stabilization.

7.6 REPORTING PROCEDURES

7.6.1 ADVERSE EVENT REPORTING

The FDA and IRB will receive notification of any UP or SAE within 24 hours of discovery by the primary investigator.

All SAEs will be followed until satisfactory resolution or until the site investigator deems the event to be chronic or the adherence to be stable. The study sponsor will be responsible for notifying FDA of any unexpected fatal or life-threatening suspected adverse reaction as soon as possible but in no case later than 7 calendar days after the sponsor's initial receipt of the information.

The study investigator shall complete an Unanticipated Adverse Device Effect Form and submit to the study sponsor and to the reviewing IRB as soon as possible, but in no event later than 7 working days after the investigator first learns of the effect. The study sponsor is responsible for investigating and reporting the results of such evaluation to FDA and to all reviewing IRBs and participating investigators within 10 working days after the sponsor first

receives notice of the effect.

A sponsor who conducts an evaluation of an unanticipated adverse device effect under 812.46(b) shall report the results of such evaluation to FDA and to all reviewing IRB's and participating investigators within 10 working days after the sponsor first receives notice of the effect. Thereafter the sponsor shall submit such additional reports concerning the effect as FDA requests (21 CFR 812.150(b)(1)).

The following information should be provided when reporting an AE:

- Subject identification (subject number and initials)
- Event description including observed symptoms
- Medical history related to event
- Event onset date and time
- Depth of injections
- Interventions implemented to treat event
- Event outcome (with resolution date and time if applicable)
- Relatedness to study product or procedure
- Seriousness of event
- Study treatment information such as number of injections, date of injections, name of product injected, volume injected, injection tool (needle or cannulae) etc.

7.6.2 REPORTING OF PREGNANCY

If it is discovered that a subject is pregnant, the subject will discontinue any interventions and continue in the study while continuing safety follow-up until the conclusion of her pregnancy.

7.7 STUDY HALTING RULES

Administration of the study agent will be halted when two SAEs determined to be related are reported. The primary investigator will notify the IRB and FDA and investigators immediately when the second serious adverse event is reported, and enrollment screens will stop accepting new study participants. The study sponsor will inform the FDA of the temporary halt and the disposition of the study.

Additionally, enrollment and treatment into the study will be temporarily halted if the Sponsor receives a SAE for a vascular embolic event that lead to skin necrosis, vision loss, stroke, pulmonary or cardiac complications or death and is determined by the Investigator to be directly or possibly related to the study device or injection procedure. The SAE will be investigated by the Sponsor. If the Sponsor's investigation concludes:

- The SAE was unanticipated, directly related to the study product or device injection procedure, and presents an unreasonable risk to study subjects, the study will be

terminated and the Investigators notified. The IRB and FDA will also be notified if the study is prematurely terminated due to safety concerns.

- If the SAE does not meet the above criteria, then enrollment in the study will continue.

7.8 SAFETY OVERSIGHT & CLINICAL MONITORING

An independent medical expert (medical monitor) will advise the study investigator and monitor participant safety. The role of the Medical Monitor is to 1) Review all AEs on a regular basis throughout the trial; 2) be available to advise the investigators on trial-related medical questions or problems, and 3) evaluate cumulative participant safety data and make recommendations regarding the safe continuation of the study. The Medical Monitor will remain blinded throughout the conduct of the clinical trial unless unblinding is warranted to optimize management of an adverse event or for other safety reasons. Clinical site monitoring is conducted to ensure that the rights and well-being of human subjects are protected, that the reported trial data are accurate, complete, and verifiable, and that the conduct of the trial follows the currently approved protocol/amendment(s), with GCP, and with applicable regulatory requirement(s).

The subjects will fill out a safety diary where they can record an adverse event (AE) when they are not in the office for 60 days after the initial treatment. Specific instructions will be included to seek immediate medical attention if any of the below symptoms or signs are observed. Any incidence of vascular occlusion, any symptomatic embolic event including but not limited to incidence of visual disturbances (including but not limited to vision loss, blurry vision, double vision, pain in or around the eye, blind spot or shadow in the visual field, trouble moving eyes, change in the Snellen Visual acuity of one line or greater), pulmonary embolism, cerebrovascular event, or death will be immediately reported to the agency along with depth of injection, the volume and tools used to inject, symptoms, time to onset and resolution, and any interventions and there will be immediate halting of the study if any serious side effects occur.

Any vascular embolic event leading to skin necrosis, change in vision, stroke, pulmonary or cardiac complications, or death will lead to halting the study. Further, a root cause investigation would then be conducted to determine the cause and whether it was anticipated or unanticipated and if the latter, the study would be suspended until an avoidance strategy for the unanticipated event can be devised.

An assessment of pain as a safety endpoint will be done using a quantitative tool to assess pain at baseline, immediately after treatment, 30 minutes after treatment, daily in the patient diary, and at each follow up visit.

An independent medical expert (medical monitor) will advise the study investigator and monitor participant safety. The role of the Medical Monitor is to 1) Review all AEs on a regular basis throughout the trial; 2) be available to advise the investigators on trial-related medical questions or problems, and 3) evaluate cumulative participant safety data and make recommendations regarding the safe continuation of the study. The Medical Monitor will remain blinded throughout the conduct of the clinical trial unless unblinding is warranted to optimize management of an adverse event or for other safety reasons. Clinical site monitoring

is conducted to ensure that the rights and well-being of human subjects are protected, that the reported trial data are accurate, complete, and verifiable, and that the conduct of the trial follows the currently approved protocol/amendment(s), with GCP, and with applicable regulatory requirement(s).

The emergency assistance protocol (Section 6.7) includes instructions for investigators to follow in the event of an intravascular injection. These instructions will include immediate transportation to a hospital that can manage pulmonary, embolic, cardiac, and cerebral embolic events. In the event of blindness or any ophthalmic signs or symptoms, subjects will undergo immediate evaluation by an ocular specialist and this specialist will be informed of the study prior to starting.

8.1 DESCRIPTION OF STATISTICAL METHODS

This is a randomized clinical study. Continuous data will be expressed as means with standard deviations. The p-value for statistical significance will be 0.05. For the primary objective, the test is one-tailed. Normality checks will be performed using the Shapiro-Wilk Test.

Confidence intervals (CIs) will be two-tailed and constructed at a confidence level of 95%. Statistical tests will be performed at a significance level of 5%, and p-values will be two-sided, unless otherwise specified.

The following populations will be defined:

- Intention-to-treat (ITT) Includes all subjects who are randomized and will be analyzed according to the randomization scheme
- Per protocol (PP) Includes all subjects in ITT who complete 3 months after baseline visit without any deviations considered to have substantial impact on the primary effectiveness
- Safety Includes all subjects who were treated with *Restylane Refyne* or randomized to the control group, and will be analyzed according to as-treated principle

Intention-to-treat is the primary population for all efficacy analyses. If the PP population contains less than 90% of the subjects in the ITT, a sensitivity analysis of the primary efficacy endpoint will be performed based on the PP population. Safety analysis is performed based on the safety population set.

The disposition of subjects will be presented in tables and/or figures as appropriate. The number of screened, treated, completed, and withdrawn subjects will be presented, as well as number of subjects in each analysis population set.

Demographic endpoints, baseline assessments, and subject characteristics will be presented based on the ITT analysis set using descriptive statistics by treatment, as appropriate.

8.2 STATISTICAL HYPOTHESES

Primary Endpoint

- The alternative hypothesis is the difference in the absolute mean from baseline to 30 days after the last treatment on a 5-point validated transverse neck line scale is greater than zero
- The null hypothesis is the difference in the absolute mean from baseline to 30 days after the last treatment on a 5-point validated transverse neck line scale is zero.

For the primary endpoint, a validated 5-point transverse neck scale developed by Allergan will be used.¹ The scale is shown below:

0 None No transverse neck lines



1 Minimal Superficial transverse neck lines



2 Moderate Moderate, effaceable transverse neck lines



3 Severe Deep, non-effaceable transverse neck lines



4 Extreme Non-effaceable transverse neck furrows with redundant skin



Secondary Endpoints

1.

- The alternative hypothesis is the difference between Group 1 and Group 2 mean change from baseline to 30 days after the last treatment on a 5-point validated transverse neck line scale is greater than zero
- The null hypothesis is the difference in the two group's mean change from baseline to 30 days after the last treatment on a 5-point validated transverse neck line scale is zero

2.

- The alternative hypothesis is the difference in the absolute mean change from baseline to 180 and 360 days after the last treatment on a 5-point validated transverse neck line scale is greater than zero
- The null hypothesis is the difference in the absolute mean change from baseline to 180 and 360 days after the last treatment on a 5-point validated transverse neck line scale is zero

All other secondary effectiveness analyses will be done descriptively as appropriate.

8.3 SAFETY ANALYSIS

Safety analysis will be descriptive only.

The number and percentage of subjects reporting each pre-defined, expected, post-treatment symptoms, as collected in the 30-day diary, will be presented in total and by maximum severity. Number of days with the event will be presented by treatment group and category: 1, 2-7, 8-13, and 14-28.

A summary of all AEs will be provided, which will include:

- number of subjects with at least one AE and number of events (in total as well as serious AEs)
- number of subjects with at least one related AE and number of events (in total as well as serious AEs)
- number of subjects with at least one mild, moderate and severe AE and number of events (in total as well as serious AEs)
- number of subjects with at least one unrelated AE and number of events (in total as well as serious AEs)
- number of subjects who did not have an AE

The number of subjects with AEs related to study product or injection procedure as well as the number of events will be summarized, and action taken for related AEs will also be summarized. Serious AEs will be listed. AEs with onset >21 days after most recent treatment will be listed.

In order to evaluate the safety of treatment using needle and cannula respectively, AE data will be summarized by needle and cannula separately.

8.4 DROP OUTS & MISSING DATA

Number of missing values will be summarized and reported as appropriate.

For ITT analysis of the primary endpoint, missing values will be assumed to be missing due to lack of effect. Therefore, the primary method of imputation will use the baseline observation carried forward (BOCF) method. Impact of missing data on the primary endpoint will be evaluated by performing sensitivity analysis based on the observed cases, as well as worst-case and best- case imputation, and tipping point analyses in the ITT set.

All other effectiveness endpoints will be evaluated based on the observed cases in ITT.

Descriptive statistics of all safety data will be performed on observed cases in the Safety population.

8.5 WITHDRAWALS AND DEVIATIONS

All withdrawn subjects will be listed individually, including at least subject number, date and reason for withdrawal, and last visit performed.

Subjects with deviations will be listed individually, including subject number and observed deviation. Depending on the seriousness of the deviation, subject might be excluded from the PP population.

8.6 SAMPLE SIZE

Sample size calculation where $\alpha = 0.05$, $\beta = 0.2$, $\sigma = .76$, $\mu_1 - \mu_2 = 0.5$

$$n = \frac{(z_{1-\alpha} + z_{\beta})^2 \sigma^2}{(\mu_1 - \mu_2)^2}$$

Plugging in the parameters into the above calculation $n=18$.

If we account for a 10% dropout rate, then the sample size should be 20 patients.

According to a pilot study with 5 patients at Siperstein Dermatology Group who were receiving hyaluronic acid injections for their transverse neck lines, the standard deviation on the transverse neck line scale was 0.76 .

According to a second pilot study done at Siperstein Dermatology group, 44 patients at Siperstein Dermatology Group would need to be screened to obtain 1 clinical trial subject. Therefore 880 subjects would need to be screened to obtain the 20 clinical trial subjects needed so that with a 10 % drop put rate there would be 18 clinical trial subjects remaining to complete the study.

Outcome measure used for calculations: The mean absolute change in a validated 5-point scale between baseline and 30 days after the last injection of Restylane Refyne.

- Test statistic: t-test
- Type I error rate (alpha) 0.05
- Power level (e.g., 80% power) 80%

A Breslow-Day test will be used to test the homogeneity of the odds ratios for the needle and cannula groups (versus the pooled control subjects). If the odds ratios are found not to be homogenous, the treatment effect will be tested separately for the needle and cannula group, at the 2.5% level of significance (to account for multiplicity).

The primary endpoint will be the mean change between Day 0 and 30 days after the last treatment on a 5-point neck line scale above. A Paired t-test will be used to analyze the primary endpoint. The results of the statistical analysis will be presented as an odds ratio with 95% Confidence intervals. A Shapiro-Wilk Test will be used to assess the normality of data. There will be no outliers since we are using a 5-point scale.

A Paired t-test will also be used to analyze the two secondary endpoints also utilizing the 5-point neck line scale. The results of the statistical analysis will be presented as an odds ratio with 95% Confidence intervals. A Shapiro-Wilk Test will be used to assess the normality of data. All other secondary effectiveness analyses will be done descriptively as appropriate.

8.5 MEASURES TO MINIMIZE BIAS

8.6.1 ENROLLMENT/ RANDOMIZATION/ MASKING PROCEDURES

In order to avoid bias, the subjects will be randomized to receive the treatment on one side with one technique and the other side with the other technique as well as randomized to receive treatment or placebo. All evaluators will be blinded as to which technique was used on which side and who received the treatment and who received the placebo. The blinded evaluators will observe the patients, but both the evaluator and subjects will be told that no discussions are to take place between the two and that the subjects are to remain silent during the evaluator examination.

To ensure only the clinical investigator performing the procedures and the study coordinator know the assignment of techniques, each subject will have an envelope assigned to them to be held by the study coordinator with the subject's name on the outside and the assignment on the inside. Only during injections does the study coordinator allow the clinical investigator to see the inside of the envelope with the randomized group assignment.

9 ETHICS/PROTECTION OF HUMAN SUBJECTS

9.1 ETHICAL STANDARD

The investigator will ensure that this study is conducted in full conformity with Regulations for the Protection of Human Subjects of Research codified in 45 CFR Part 46, 21 CFR Part 50, 21 CFR Part 56, and the ICH E6.

9.2 INSTITUTIONAL REVIEW BOARD

The protocol, informed consent forms, recruitment materials, and all participant materials will be submitted to the IRB for review and approval. Approval of both the protocol and the consent form must be obtained before any participant is enrolled. Any amendment to the protocol will require review and approval by the IRB before the changes are implemented to the study. All changes to the consent form will be IRB approved; a determination will be made regarding whether previously consented participants need to be re-consented.

9.3 INFORMED CONSENT PROCESS

Informed consent is required for all participants. In obtaining and documenting informed consent, the investigator will comply with applicable regulatory requirements and should adhere to 45 CFR Part 46, 21 CFR Part 50, 21 CFR Part 56, and ICH GCP. Prior to the written informed consent, the consent forms will be provided to the participants. Consent forms describing in detail the study agent, study procedures, and risks will be given to the participant and written documentation of informed consent is required prior to starting intervention/administering study product.

The investigator will explain the research study to the participant and answer any questions that may arise. All participants will receive a verbal explanation in terms suited to their comprehension of the purposes, procedures, and potential risks of the study and of their rights as research participants. Participants will have the opportunity to carefully review the written consent form and ask questions prior to signing. The participants will have the opportunity to think about being a participant prior to agreeing to sign. The participant will sign the informed consent document prior to any procedures being done specifically for the study. The participants may withdraw consent at any time throughout the course of the trial. A copy of the informed consent document will be given to the participants for their records. The rights and welfare of the participants will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this study.

9.4 PARTICIPANT AND DATA CONFIDENTIALITY

Participant confidentiality is strictly held in trust by the participating investigators, their staff, and the sponsor(s) and their agents. Therefore, the study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the sponsor.

The study monitor, other authorized representatives of the sponsor, representatives of the IRB or pharmaceutical company supplying study product may inspect all documents and records required to be maintained by the investigator, including but not limited to, medical records for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at each clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by local IRB and Institutional regulations.

Study participant research data, which is for purposes of statistical analysis and scientific reporting, will be transmitted to and stored at the Siperstein Dermatology Group Boynton research office. This will not include the participant's contact or identifying information. Rather, individual participants and their research data will be identified by a unique study identification number. The study data entry and study management systems used by clinical sites and by Siperstein Dermatology Group research staff will be secured and password protected. At the end of the study, all study databases will be de-identified and archived at the Siperstein Dermatology's Boynton Beach research office.

9.4.1 RESEARCH USE OF STORED HUMAN DATA

Pictures and data collected under this protocol may be used in future publications. Access to stored pictures and data will be stored using codes assigned by the investigators. Data will be kept in password protected computers. Only investigators will have access to the samples and data.

10 DATA HANDLING AND RECORD KEEPING

10.1 DATA COLLECTION AND MANAGEMENT RESPONSIBILITIES

Data collection is the responsibility of the clinical trial staff at the site under the supervision of the site PI. The investigator is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported.

All source documents should be completed in a neat, legible manner to ensure accurate interpretation of data. Black ink is required to ensure clarity of reproduced copies. When making changes or corrections, cross out the original entry with a single line, and initial and date the change.

Clinical data (including AEs, concomitant medications, and expected adverse reactions data) will be entered into a 21 CFR Part 11-compliant data capture system. The data system includes password protection and internal quality checks, such as automatic range checks, to identify data that appear inconsistent, incomplete, or inaccurate. Clinical data will be entered directly from the source documents.

10.2 STUDY RECORDS RETENTION

Study documents will be retained for a minimum of 2 years after the formal discontinuation of the clinical study. These documents will be retained for a longer period, however, if required by local regulations. No records will be destroyed without the written consent of the sponsor, if applicable. It is the responsibility of the sponsor to inform the investigator when these documents no longer need to be retained.

10.3 PROTOCOL DEVIATIONS

A protocol deviation is any noncompliance with the clinical trial protocol. The noncompliance may be either on the part of the participant, the investigator, or the study site staff. As a result of deviations, corrective actions are to be developed by the site and implemented promptly. It is the responsibility of the site to use continuous vigilance to identify and report deviations within 7 working days of identification of the protocol deviation, or within 7 working days of the scheduled protocol-required activity. All deviations must be addressed in study source documents, reported to the IRB and sponsor. The site PI/study staff is responsible for knowing and adhering to their IRB requirements.

10.4 PUBLICATION

The PI will be responsible for submitting the data to a peer-reviewed journal within 3 months of the end of the trial.

11 CONFLICT OF INTEREST POLICY

The independence of this study from any actual or perceived influence, such as by the pharmaceutical industry, is critical. Therefore, any actual conflict of interest of persons who have a role in the design, conduct, analysis, publication, or any aspect of this trial will be disclosed and managed. Furthermore, persons who have a perceived conflict of interest will be required to have such conflicts managed in a way that is appropriate to their participation in the trial.

1. Jones D, Carruthers A, Hardas B, Murphy DK, et al. Development and validation of a photonic scale for evaluation of transverse neck lines. *Dermatol Surg*. 2016;;42(Suppl 1):S235–S42.
2. Minokadeh A, Black J, Jones D. Effacement of Transverse Neck Lines With VYC-15L and Cohesive Polydensified Matrix Hyaluronic Acid. *Dermatologic Surgery*. 2018;44:S53–S56.
3. Lee, SK., Kim, HS. Correction of horizontal neck lines: Our preliminary experience with hyaluronic acid fillers. *J Cosmet Dermatol*. 2017;00:1–6.
4. Han TY, Lee JW, Lee JH, Son SJ, Kim BJ, Mun SK, Kim MN, Lee CK. Subdermal minimal surgery with hyaluronic acid as an effective treatment for neck wrinkles. *Dermatol Surg*. 2011Sep;37(9):1291-6
5. Tseng F, Hsiaoen, Y. Treatment of Horizontal Neck Wrinkles with Hyaluronic Acid Filler: A Retrospective Case Series. *PRS Global Open*. 2019 Aug
6. Restylane Refyne [package insert]. Galderma, Inc., San Diego, CA; October 2007