CLINICAL STUDY PROTOCOL

An Open-Label Phase 2 Study to Investigate the Efficacy, Tolerability, and Safety of the HTS-519 Insert in the Treatment of Subjects with Distal Lateral Subungual Onychomycosis of the Great Toenail

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Authors	Hendrik A Kroon, Jay E Birnbaum
Sponsor	Hallux, Inc. 23052 Alcalde Drive, Suite A Laguna Hills, CA 92653 United States

This study will be conducted in compliance with Good Clinical Practice and the Declaration of Helsinki (with amendments), in accordance with local legal and regulatory requirements and in compliance with the applicable parts of the United States Code of Federal Regulations.

1 PROTOCOL SYNOPSIS

PROTOCOL TITLE	An Open-Label Phase 2 Study to Investigate the Efficacy, Tolerability, and Safety of the HTS-519 Insert in the Treatment of Subjects with Distal Lateral Subungual Onychomycosis of the Great Toenail
PROTOCOL No.	HTS-002B
SPONSOR	Hallux, Inc.
INDICATION (PHASE)	Distal lateral subungual onychomycosis (Phase 2)
STUDY CENTERS	One investigational site in the US
INDICATION AND RATIONALE	The purpose of this study is to evaluate the efficacy, clinical benefit, and safety of the HTS-519 Insert for the treatment of subjects with laboratory-confirmed distal lateral subungual onychomycosis (DLSO) of the great toenail.
STUDY DESIGN	This is a single-center open-label study to determine the efficacy of the maximal feasible dose (MFD) of the HTS-519 Insert using an optimal insertion technique. All subjects will be screened for DLSO and will meet inclusion and exclusion criteria. Subjects will receive up to 4 HTS-519 Inserts based on a protocol-specific treatment algorithm. Treatment will be administered to all subjects on Day 1, Day 29, and Day 57 under local digital anesthesia. Efficacy variables are derived from KOH tests / mycological culture and digital planimetric quantification of investigator evaluations of nail involvement and extent of unaffected clear nail growth. Safety variables include adverse events, assessments of local tolerability, and results from physical examination and vital signs measurements.
INVESTIGATIONAL PRODUCT	HTS-519 (Hallux Terbinafine Subungual) Insert, a solid 6-mm cylindrical pellet containing 519 µg (70%) terbinafine hydrochloride, 21% polyethylene glycol 3350, 5% triethyl citrate, and 4% stearic acid, assembled in a 25-gauge needle single-use applicator.
ROUTE AND FREQUENCY OF ADMINISTRATION	Up to 4 HTS-519 Inserts will be administered subungually 3 times at intervals of 4 weeks (at treatment visits Days 1, 29, and 57).
STUDY OBJECTIVES	The primary efficacy objective is to provide a preliminary estimate of the rate of complete cure of DLSO at Week 48 after treatment with the HTS-519 Insert. Secondary objectives will be to provide estimates of mycological and clinical cure rates, and other responder rates. Reports of physician and subject experiences with the procedure will also be elicited as an exploratory objective. Subjects will report their pain scores during and following HTS-519 Insert placement. Physicians will assess ease of insertion. Local tolerability and safety of administration of subungual inserts will be assessed. With an effective fungicidal treatment, the timing of attaining a clear nail is determined by the subject's normal nail growth rate (emergence of clear nail) and baseline linear involvement of the nail. In order to maximize the opportunity for an early clinical "readout" on product efficacy, pre-identified subjects with longitudinal nail involvement and nail growth rates capable of achieving a clear nail by Week 48 will be treated. Therefore, Therapeutic Efficacy will be determined as the ratio of the rate of unaffected nail growth to the rate of total nail growth. This ratio will be frequently assessed to monitor efficacy.
PLANNED SAMPLE SIZE AND STATISTICAL CONSIDERATIONS	It is planned that at least 30 subjects will complete the 3 treatments specified. No sample size calculation was conducted.

PATIENT POPULATION Inclusion Criteria	1. Male or female between 18 and 74 years of age inclusive. 2. Clinical diagnosis of DLSO in at least one great toenail. If both great toenails meet inclusion criteria, the toenail with the greater involvement will be designated the target toenail. 3. Linear nail involvement of at least 4 mm, but without involvement of the proximal 3 mm of the nail, based on the investigator's visual assessment. Overall nail involvement should be at least 25% but not more than 75% as per the investigator's visual assessment (Grade 3). Nail thickness ≤ 2 mm or total thickness of the nail plus subungual debris measuring ≤ 3 mm. 4. Subject's nails are required to have a normal rate of growth (approximately 1 mm / month or greater) in order to be physiologically capable of achieving clinical cure/near clinical cure within the 48-week study period. 5. Laboratory confirmation of DLSO with direct microscopy of subungual debris positive for hyphal elements (KOH test) and fungal culture positive for dermatophytes. 6. Generally of good health. 7. Willing to comply with protocol requirements and sign a statement of informed consent. 8. Willing to refrain from using any lotions, creams, liquids, or polish on the large toenail or on the skin immediately adjacent to the large toenail during the treatment period unless directed to do so by the investigator. 9. Willing to refrain from using on the toenails topical products to which the subject has a high risk of developing an allergic reaction and/or dermatitis; 10. Willing to refrain from receiving pedicures for the duration of the study. 11. Willing to refrain from using topical steroids or topical antifungals on the great toenail or the skin immediately adjacent to the great toenail, or systemic antifungals for the duration of the study.
PATIENT POPULATION Exclusion Criteria	1. History of severe or chronic immunosuppression, an immunocompromised condition, known or suspected HIV, extensive dermatomycoses, extensive recurrent herpes zoster or severe dermatitis affecting the feet that would interfere with safety and tolerability assessments. 2. Any systemic or dermatologic disorder, such as uncontrolled psoriasis, severe eczema, or severe atopic dermatitis which, in the opinion of the investigator, will interfere with the study results or increase the risk of adverse events (subjects with mild, controlled psoriasis, eczema, or dermatitis may be included if the condition would not interfere with safety and tolerability assessments). 3. Any severe and/or chronic disease that would affect a subject's nail growth or interfere with the subject's ability to complete the trial, such as severe renal failure, peripheral vascular disease, severe chronic obstructive pulmonary disease, severe heart disease, uncontrolled diabetes mellitus or other endocrine disease, or uncontrolled malignancy. 4. History of toenail surgery or any significant injury to the target toenail matrix. 5. Treatment with any investigational drug within 1 month prior to Screening. 6. Topical antifungal treatment applied to the feet within 1 month prior to the start of study treatment. 7. Use of oral terbinafine within 6 months, or use of any other oral antifungal drug within 3 months, prior to the start of study treatment. 8. Hypersensitivity to terbinafine or to any other ingredients of the formulation. 9. Active onychomycosis of the fingernail. 10. Symptomatic tinea pedis requiring treatment at Day 1 of the study. 11. Superficial white, proximal subungual onychomycosis, lichen planus,

	psoriasis, or any condition that interfere with the treatment procedure or assessment of clear nail. 12. Structural deformities of the target toenail or foot (eg, genetic or pigment disorders, chemical damage, tumors) that would interfere with treatment procedures or with assessments of efficacy, safety, or tolerability. 13. Suspected subungual dermatophytoma. 14. Any other condition that in the opinion of the investigator renders the subject unsuitable for participation in this study.
EFFICACY ASSESSMENTS	Efficacy is determined in the target great toenail by • evaluation of clinical cure (complete clearance of signs and symptoms) using the Investigator Global Assessment Scale, further documented by digital photography analysis, and • laboratory confirmation of mycological cure (negative culture for dermatophytes and negative KOH microscopy). From these assessments the efficacy outcomes of complete cure (mycological cure and clinical cure), mycological cure, clinical cure and effective treatment (no more than 10% affected nail) will be determined. Therapeutic Efficacy (ratio of the rate of unaffected nail growth to the rate of total nail growth) will be assessed using digital image analysis.
SAFETY ASSESSMENTS	Safety assessments include monitoring AEs and local tolerability at the administration site using a 4-point scale. Pain assessments at time of treatment and 7-days post procedure. Results of physical examination and vital signs measurements will be assessed.
STATISTICAL METHODS	Cure rates will be estimated along with 95% confidence intervals. Interval parameters such as length and ratios will be characterized using descriptive statistics. The primary analysis population will be the intent-to-treat population. The analysis of safety is conducted in the population of all treated subjects. Rates of AEs and local tolerance assessments will be presented descriptively. Events of clinical significance will be identified individually and described without inferential statistics.

Table 1-1 Schedule of Study Procedures

	Screening	Allocation and Treatment			Post-Treatment					
Procedure	Visit 1	Visit 2	Visit 3	Visit 4	Visit 5	Visit 6	Visit 7	Visit 8	Visit 9	Visit 10
	Screening	Baseline Day 1	Week 4 Day 29 ±3	Week 8 Day 57 ±3	Week 12 Day 85 ±7	Week 16 Day 113 ±7	Week 20 Day 141 ±7	Week 24 Day 169 ±7	Week 36 Day 253 ±7	Week 48 Day 337 ±7
ENROLLMENT AND SCREENING										
Informed consent	X									
Demographic, medical/medication history	X	X[a]								
Inclusion / exclusion criteria	X	X[a]								
Target toenail measurements	X	X								
INTERVENTIONS										
Notch target great toenail[b]		X								
Administer study treatment		X	X	X						
Administration site assessment		X	X	X						
Pain assessment		X	X	X						
ASSESSMENTS										
Physical examination and vital signs	X									X
Subungual sample for KOH microscopy and fungal culture	X[c]							X	X	X
Investigator Global Assessment of clear nail[d]	X				X	X	X	X	X	X
Digital photography[d]	X	X	X	X	X	X	X	X	X	X
Dispense subject diary		X[e]	X[e]	X[e]						
Review and record adverse events		X	X[f]	X[f]	X[f]	X	X	X	X	X
Monitor concomitant medications		X	X	X	X	X	X	X	X	X

[a] Record any change from baseline
[b] When the notch is approaching the area of the distal groove due to outgrowth of the nail, re-notch the superficial nail plate and continue measurements
[c] Subungual sampling may be repeated to confirm diagnosis of DLSO by fungal culture
[d] Nails should be clipped prior to assessments
[e] Next day follow-up call to check for adverse events, changes in medication
[f] Includes review of subject diary

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	Post-Treatment-Extension	
Procedure	**Visit 11**	**Visit 12**
	Week 60 Day 420 ±7	**Week 72 Day 504 ±7**
ENROLLMENT AND SCREENING		
Informed consent		
Demographic, medical/medication history		
Inclusion / exclusion criteria		
Target toenail measurements		
INTERVENTIONS		
Notch target great toenail[b]		
Administer study treatment		
Administration site assessment		
Pain assessment		
ASSESSMENTS		
Physical examination and vital signs		
Subungual sample for KOH microscopy and fungal culture	X	X
Investigator Global Assessment of clear nail[d]	X	X
Digital photography[d]	X	X
Dispense subject diary		
Review and record adverse events		
Monitor concomitant medications		

[a] Record any change from baseline
[b] When the notch is approaching the area of the distal groove due to outgrowth of the nail, re-notch the superficial nail plate and continue measurements
[c] Subungual sampling may be repeated to confirm diagnosis of DLSO by fungal culture
[d] Nails should be clipped prior to assessments
[e] Next day follow-up call to check for adverse events, changes in medication
[f] Includes review of subject diary

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LIST OF ABBREVIATIONS

ADL	Activities of daily living
AE	Adverse event
AR	Adverse reaction
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
BUMC	Banner University Medical Center
CFR	Code of Federal Regulations
CMM	Center of Medical Mycology
CRF	Case report form
DLSO	Distal lateral subungual onychomycosis
EDC	Electronic data capture
EOS	End of Study
FDA	U.S. Food and Drug Administration
GCP	Good Clinical Practice
HIPAA	Health Insurance Portability and Accountability Act
HTS	Hallux Terbinafine Subungual
ICH	International Conference on Harmonization
IGA	investigator Global Assessment
IRB	Institutional Review Board
KOH	Potassium hydroxide
MedDRA	Medical Dictionary for Regulatory Activities
MFD	Maximum feasible dose
NRS	Numerical rating scale
OTC	Over-the-Counter (non-prescription)
PEG	Polyethylene Glycol
PK	Pharmacokinetic
PRG	Physician Research Group
SAE	Serious adverse event
SAP	Statistical Analysis Plan
SAR	Suspected adverse reaction
SOP	Standard operating procedure
SQL	Sonora Quest Laboratories
TBF	Terbinafine
WMA	World Medical Association

2 KEY ROLES AND CONTACT INFORMATION

Name / Role	Address
Sponsor	Hallux, Inc. (Hallux) 23052 Alcalde Drive, Suite A Laguna Hills, CA 92653 Telephone: (800) 380 9130 Fax: (949) 315 7373
Sponsor Medical Monitor	Henk-André Kroon, MD, MBA Executive VP, Chief Medical Officer 23052 Alcalde Drive, Suite A Laguna Hills, CA 92653 Telephone: (949) 353 9538 Fax: (949) 315 7373
Contract Research Organization	Physicians Research Group, Inc. (PRG) 2155 East Conference Drive, Suite 110 Tempe, AZ 85284 Telephone: (623) 565 8624
Clinical Laboratory	Center for Medical Mycology (CMM) 11100 Euclid Avenue Cleveland, OH 44106 Telephone: (888) 469 2522 Fax: (216) 844 1076 Laboratory Sciences of Arizona / Sonora Quest Laboratories (SQL) 1255 W Washington Street Tempe, AZ 85281 Telephone: (602) 685 5211 Fax: (602) 685 5028 Laboratory Sciences of Arizona/Banner University Medical Center (BUMC) 1111 E. McDowell Road Phoenix, AZ 85006 Telephone: (602) 839 2872 Fax: (602) 839 2674
Digital Imaging	Canfield Scientific, Inc. (Canfield) 4 Wood Hollow Road Parsippany, NJ 07054 Telephone: (973) 434 1200 Fax: (973) 887 0549

3 INTRODUCTION: BACKGROUND INFORMATION AND SCIENTIFIC RATIONALE

3.1 Background Information

Onychomycosis or *tinea unguium* is a chronic fungal infection of the nail unit affecting 2-18% of the population worldwide. The observed incidence increases with diminished levels of immune competence (eg, diabetes, malignancy), age (prevalence rate in children is 0.2%-2.6% vs 48% in age over 70) or trauma to the nail unit.[1]

The most common form of fungal nail disease is Distal Lateral Subungual Onychomycosis (DLSO), which as the name implies is caused by the subungual infiltration of the distal and lateral nail unit by dermatophytes (*Trichophyton rubrum* and *mentagrophytes*, and *Epidermophyton floccosum*).[2]

Initial fungal penetration of the distal nail bed or lateral nail groove is followed by advancement proximally along the longitudinal streaks of the nail bed and ventral nail plate. Mild inflammation ensues in the nail bed, resulting in focal parakeratosis and subungual hyperkeratosis, with the consequence of onycholysis (painless detachment of the nail plate from the nail bed) and thickening of the subungual region. The inflammatory response creates a subungual space that serves as a reservoir for fungi and superinfecting bacteria and molds.[3]

The clinical diagnosis of DLSO relies on recognition of its signs (eg, discoloration of the nail plate, onycholysis, and subungual hyperkeratosis). Onycholysis can be distinguished by its white color, because of the presence of air under the detached nail plate.[2] Subungual hyperkeratosis can be easily identified as thickening of the nail bed or hyponychium beneath the preformed nail plate, and measured with a caliper.[4] The opaque white, yellow to brown discoloration of the nail plate provides a visual indicator of diseased and unaffected nail area.[5]

Laboratory confirmation of fungal nail infection by dermatophytes is performed through potassium hydroxide (KOH) direct microscopic evaluation and fungal culture of subungual material. The subungual specimen must be obtained from the nail bed and, if insufficient, from the ventral nail plate as well, using a small curette or a #15 scalpel blade. The KOH test merely confirms the presence of septate fungal hyphae (dead or alive) and arthroconidia in the collected subungual debris and therefore necessitates subsequent fungal identification through culture.[3]

Two systemic antifungal treatments have been approved by FDA for the treatment of DLSO and are considered the most effective in its treatment. Oral terbinafine (Lamisil®, Novartis Pharmaceuticals) is considered the gold standard, yielding complete cure rates of 38% after daily dosing for 12 weeks with a 250 mg dose.[6] In clinical studies, 14% of subjects receiving oral itraconazole (Sporanox®) achieved complete cure after 12 weeks of daily 200 mg oral therapy.[7]

Topical FDA-approved treatments for toenail onychomycosis have demonstrated disappointingly low efficacy rates and have been associated with poor compliance.

These topical treatments include recently introduced 10% efinaconazole topical solution (Jublia®) and 5% tavaborole topical solution, (Kerydyn®), as well as 8% ciclopirox nail lacquer (Penlac®), marketed since 1999. Complete cure rates in controlled clinical studies for these 3 compounds were 15.2-17.8% for Jublia®,[8] 6.5-9.1% for Kerydyn®,[9] and 5.5-8.5% for Penlac®[10] after daily application to the entire toenail area for 48 weeks.

Patient's use of the most effective systemic agents may be limited due to drug-drug interactions, toxicity concerns such as rare cases of liver failure, isolated reports of serious skin reactions, FDA-recommended liver enzyme monitoring for terbinafine[6,11] and liver and cardiac toxicity for itraconazole.[7] Application site reactions are the most common side effects of the three topical antifungal treatments.[8-10]

While these approved antifungal agents, whether applied systemically or topically, cover a broad spectrum of fungi and display high *in vitro* activity against dermatophytes, DLSO remains extremely refractory to antifungal treatment because of the difficulty of achieving and maintaining sufficient concentrations of antifungal agents at the sites of infection.

3.2 Rationale

Hallux is developing an investigational treatment for DLSO that utilizes areas of reduced adherence between nail bed and nail plate to deliver a biodegradable insert containing the potent fungicidal terbinafine to the subungual space, targeting areas with high fungal viability, located at or beyond the visible leading edge of disease.

The HTS-519 Insert contains the maximum dose (70%) of terbinafine that can be reliably and technically formulated in a polymeric biodegradable matrix. Terbinafine HCl was chosen as the active pharmaceutical ingredient because its properties are well characterized, and because it is the most clinically effective pharmaceutical against *Trichophyton rubrum* and *Trichophyton mentagrophytes*, the dermatophytes accounting for over 90% of fungal nail infections.[12-15] Terbinafine is a highly lipophilic and keratinophilic synthetic allylamine that interferes with fungal ergosterol biosynthesis through inhibition of squalene epoxidase, thereby blocking the conversion of squalene to squalene epoxide.[16] The depletion of ergosterol in the fungal cell membrane has a fungistatic effect, and the intracellular accumulation of the precursor squalene appears to have a fungicidal effect *in vitro*.[17]

In a healthy volunteer pharmacokinetic study (TMI-7005) using TMI-358, a prototype of the HTS-519 Insert, terbinafine drug release from the HTS-519 Insert was characterized by an initial rapid burst followed by a prolonged and slow dissolution phase that resulted in nail tissue terbinafine levels in excess of those seen with oral terbinafine for at least 23 days. These data support a treatment regimen of 3 repeated doses at intervals of 4 weeks in order to maintain adequate nail tissue terbinafine levels for at least 3 consecutive months.

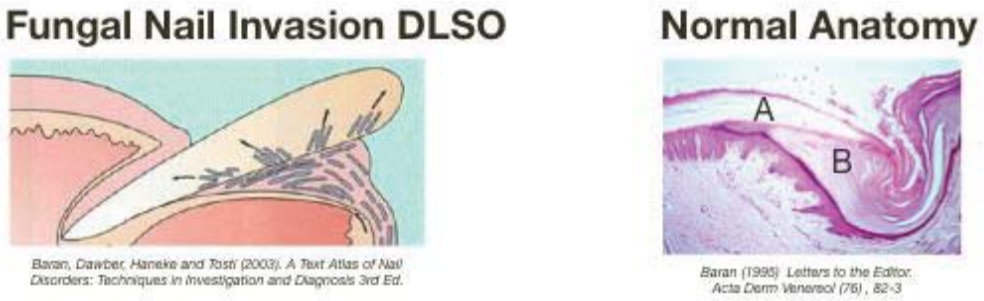
The administration procedure is unique in that the HTS-519 Insert is delivered with a needle applicator in a channel created by microcannulation in between the nail bed and nail plate. This subungual administration constitutes topical drug delivery to the nail

bed, intentionally avoiding live tissue. In addition, while the clinical manifestations of the disease are clearly visible, the insertion procedure relies on tactile feedback to gauge the extent of the subungual space into which the HTS-519 Insert is deposited.

Access points to the subungual space are found in areas of reduced adherence of the nail plate to the nail bed caused by:

- Response to disease in the nail bed or nail plate (onycholysis, subungual hyperkeratosis / debris – Fig. 3-1 left).
- Normal anatomical features of the nail unit: nail plate with greatest radius of curvature (A- Fig. 3-1 right) and lateral groove (B – Fig. 3-1 right), which are natural entry points for an invasive fungal infection.[18,19]

Figure 3-1 Sites of Reduced Nail Adherence

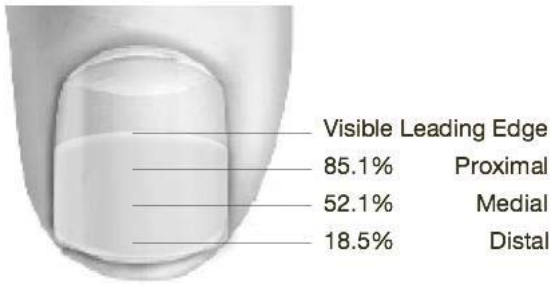


Efficacy of the HTS-519 Insert is dependent upon the ability of terbinafine to diffuse from the insert's biodegradable matrix. In order to achieve a uniform distribution of terbinafine in the target nail area and maximize antifungal tissue concentrations of terbinafine in target tissue, equidistant placement of HTS-519 Inserts from each other was chosen to be most effective. The anatomical width of the nail (typically ranging from 10-20 mm) constrained the maximum feasible dose (MFD) or number of inserts to 4. This would require terbinafine to diffuse and reach fungicidal tissue levels up to 2.5 mm from the HTS-519 Insert and HTS-519 Inserts to be separated ≤ 5 mm from each other, as described in Section 7.2. An important advantage of this approach is that dose can be defined based on diffusion distance and data be interpreted independent of the number of inserts administered. Allowable adaptations to the dosing regimen based on heterogeneity of disease presentation are described in Section 7.3.

The 2-step insertion technique was informed by standard techniques used in (cosmetic) dermatology procedures to reduce local complications, and optimized for subungual access by the Principal Investigator, Lewis Freed, DPM. The cannulation step uses a blunt-tip DermaSculpt® microcannula, a device commonly used in cosmetic facial procedures with dermal fillers in order to avoid tissue trauma and bruising. Proper placement technique (eg, speed of insertion / withdrawal, insertion depth), sterile barrier precautions, as well as proper physician education and training have also been incorporated into the procedure.[20] In the subungual procedure, cannulation also prevents potential clogging of the device needle orifice with subungual debris and tissue trauma that can be caused by the beveled needle applicator.

The intended target of the HTS-519 Insert is the area close to the visible leading edge of disease (Figure 3-2) that contains the largest percentage of viable hyphae.[3,21]

Figure 3-2 Sample Location and Percentage Viable Organisms by Fungal Culture



While the visible leading edge provides a visual guide for the HTS-519 Insert placement, the actual insertion depth into the subungual space is more precisely determined by tactile feedback from initial probing with an atraumatic small-gauge (27-30 G) microcannula.

After probing, at each insertion point, a cannulation step with a 22-23 G blunt-tipped microcannula (DermaSculpt®, CosmoFrance) creates a channel in the subungual space. The insertion depth is declared and recorded when a clear "stop" is felt (Appendix 6). This will be the predefined insertion depth for the applicator. The beveled tip 25 G needle applicator (containing the 6 mm cylindrical drug insert) should not extend beyond that depth in order to prevent potential trauma to the nail bed and reduce rapid elimination of delivered drug by permeation into vasculature, as well as to facilitate patient's acceptance.[22]

Pretreatment will include hydration of the toenail with a normal saline impregnated gauze to increase nail flexibility. In selected subjects, and in addition to the prescribed nail clipping to the distal groove, minimal distal debridement in order to remove impacted subungual material will be allowed as per instructions in Appendix 5.

3.3 Potential Risks and Benefits

Terbinafine has been approved for use as a topical agent for skin infections since 1993 and orally for the treatment of onychomycosis since 1996. Both the topical and oral formulations are generally regarded as safe and effective.

HTS-002B is the first clinical study that uses the HTS-519 Insert. However, relevant safety information is available from 6 clinical studies that have been performed with the TMI-358 Micro Implant, the prototype to HTS-519, that contained 358 µg terbinafine instead of 519 µg, was 2 mm shorter (4 mm vs 6 mm) and did not contain triethyl citrate or stearic acid, excipients added to improve manufacturability at scale.

3.3.1 Potential Risks

Nonclinical studies in guinea pigs and minipigs have demonstrated no evidence of toxicity with intradermal administration. Treatment of minipigs with intradermal administration of up to twelve TMI-358 Micro Implants (up to three TMI-358 Micro

Implants per site) on up to 3 occasions was well tolerated. A guinea pig maximization study demonstrated no evidence of sensitization potential.

The existing clinical database includes safety and tolerability data from 238 subjects, 186 subjects with toenail DLSO, 15 subjects with fingernail DLSO, and 37 healthy volunteers, who have received intradermal treatment with between one and nine TMI-358 Implants.

The expected adverse events (AEs) are application site reactions, including pain, swelling, irritation, and hematoma; although many patients show none of these reactions. The implantation procedure in all 6 clinical trials conducted to date has been preceded by local anesthesia, either topical ethyl chloride or a lidocaine digital block. Those patients who reported post-procedural pain usually experienced discomfort within 2 days following the procedure.

Plasma concentration levels of terbinafine and the major metabolite, N-desmethyl terbinafine, were obtained for 59 study subjects with DLSO, who had been treated with 1, 2, or 3 implants at Day 1, Week 4, and Week 8 administered intradermally. The maximum systemic terbinafine plasma levels in patients receiving TMI-358 (3.35 ng/mL) was about 300-fold lower than systemic levels in patients taking oral terbinafine (1000 ng/mL). Four weeks after dosing with three TMI-358 Implants, the median plasma values were below the limit of quantification (0.04 ng/mL) and the maximum observed value was 0.067 ng/mL. In addition, no noteworthy accumulation was noted at Weeks 8 and 12.

In the referenced nonclinical and prior human studies, an intradermal, rather than subungual, administration was investigated. The 2-step atraumatic (cannulation) placement technique used in this study is novel and designed to deliver drug to the subungual space and minimize trauma to the nail bed. This proposed procedure constitutes the topical administration of terbinafine to the nail bed and nail plate, not administration of a treatment into a living site or tissue. The treatment procedure is described in full in Appendix 6.

As before, this study will use a distal digital block in all subjects to render the subungual insertion procedure itself painless (Appendix 3). Risks associated with local anesthesia are pain at the injection site, local injury or infection, and rare allergic reactions.

3.3.2 Potential Benefits

Subungual administration of the HTS-519 Insert has the potential benefit of delivering the effective antifungal terbinafine in high concentrations directly to the site of infection, without systemic exposure. Earlier pharmacokinetic data in healthy subjects collected in study TMI-7005, suggests that after administration of one TMI-358 Micro Implant administered subungually, terbinafine tissue concentrations are as high as those seen with continuous oral Lamisil® for at least 23 days. The clinical effect of subungual administration of the HTS-519 Insert may deliver a complete cure rate that is at least comparable to the current gold standard.

4 STUDY OBJECTIVES

4.1 Study Objectives

The primary efficacy objective of this study is to provide a preliminary estimate of the rate of complete cure of DLSO at Week 48 after treatment with the HTS-519 Insert. Secondary objectives will be to provide estimates of mycological and clinical cure rates, and other responder rates.

Reports of physician and subject experiences with the procedure will also be elicited as an exploratory objective. Subjects will report their pain scores during and following HTS-519 Insert placement. Physicians will assess ease of insertion. Local tolerability and safety of administration of subungual inserts will be assessed.

With an effective fungicidal treatment, the timing of attaining a clear nail is determined by the subject's normal nail growth rate (emergence of clear nail) and baseline linear involvement of the nail. In order to maximize the opportunity for an early clinical "readout" on product efficacy, pre-identified subjects with longitudinal nail involvement and nail growth rates capable of achieving a clear nail by Week 48 will be treated. Therefore, Therapeutic Efficacy will be determined as the ratio of the rate of unaffected nail growth to the rate of total nail growth. This ratio will be frequently assessed to monitor efficacy.

4.2 Study Outcome Measures

4.2.1 Primary

The primary outcome measure, complete cure at Visit 10 (Week 48), is based on clinical evaluation of "clear nail" using the Investigator's Global Assessment (IGA) Scale (Section 9.1.1) and laboratory confirmation of mycological cure of the target toenail (Section 9.1.2).

The investigator will evaluate the extent of clear nail or absence of signs of residual onychomycosis using the IGA Scale (Table 4-1), with a Grade 0 representative of a clinical cure. The investigator's assessment will be documented by digital photography planimetric analysis by Canfield Scientific, Inc.

Table 4-1 Investigator Global Assessment Scale

Grade 0	Completely clear nail, with nail area clear of signs of residual disease (ie, nail dystrophy, onycholysis, or subungual hyperkeratosis) attributable to onychomycosis
Grade 1	Almost clear nail with ≤ 10% affected nail
Grade 2	11%-24% affected nail
Grade 3	25%-75% affected nail
Grade 4	>75% affected nail

Subungual samples will be assessed for KOH visualization by microscopy and fungal culture for dermatophytes by a laboratory. Since subjects enroll with both a positive KOH (fungal elements present at microscopy) and positive culture for dermatophytes, a mycological cure is declared when both laboratory assessments are negative.

4.2.2 Secondary

The four secondary outcome measures are combinations of the same parameters used for evaluating the primary efficacy outcome of complete cure (Table 4-2).

Table 4-2 Secondary Efficacy Outcomes

Efficacy Outcome	Efficacy Criteria	
	investigator Assessment / Planimetry	**Laboratory Outcome**
Complete Cure other than at Week 48 (EOS)	Grade 0 - Completely clear nail, with nail area clear of signs of residual disease	Negative KOH microscopy and Negative culture for dermatophytes
Effective Treatment	Grade 1 - Almost clear nail with ≤ 10% affected nail	
Mycological Cure	Not a determinant in outcome	
Clinical Cure	Grade 0 - Completely clear nail, with nail area clear of signs of residual disease	Not a determinant in the outcome

4.2.3 Exploratory

Exploratory efficacy outcomes are summarized in Table 4-3.

Table 4-3 Exploratory Efficacy Outcomes

Efficacy Objective	Efficacy Variables
Therapeutic Efficacy	Ratio of the rate of unaffected nail growth to the rate of total nail growth
Therapeutic Response	Achievement of ≥ 5mm unaffected nail length
Mycological Relapse	Positive mycology determined at any time point subsequent to a mycological cure
Clinical Relapse	Clinical signs of infection determined at any time point subsequent to a clinical cure
Ease of Insertion	An investigator rating measured on 5-point Likert scale

4.2.3.1 Therapeutic Efficacy

While the target of the fungicidal agent in the treatment of DLSO is a pathogen, efficacy is not only established by negative fungal culture for dermatophytes, but also by the physiological or clinical manifestation of a cure – the appearance of unaffected clear nail and ultimately clear nail.[23]

In DLSO, the fungal infection starts at the distal edge of the nail and extends proximally in the subungual environment. With an effective treatment, fungal growth ceases and the emergence of clear or unaffected nail occurs at a rate no faster than the individual's baseline linear nail growth rate.[24]

Nail growth rates can be established by monitoring the outgrowth of a notch cut into the surface of the nail plate and placed in the midline 3 mm from the proximal fold,[25,26] as described in Appendix 2.

Monitoring the emergence of clear nail and outgrowth of the notch with digital photography allows for the determination of "Therapeutic Efficacy," defined as the ratio of rate of unaffected nail growth to the rate of total nail growth. This outcome is defined by the rate of unaffected nail growth with the contemporaneous assessment of rate of total nail growth. As a physiological biomarker or diagnostic, the ratio can be used as an

early and repeated clinical measure of efficacy of the investigational product, and to predict the likelihood of attaining complete cure prior to end of study.[24] In addition, the ratio can be used to compare therapeutic progress between subjects, irrespective of their baseline extent of diseased nail.

4.2.3.2 Therapeutic Response Rate

Therapeutic response rate is defined as the percentage of subjects achieving ≥ 5mm unaffected nail length regardless of mycological status.

4.2.3.3 Mycological and Clinical Relapse Rate

Persistence of treatment effect will be assessed starting after the last treatment, using the following definition of relapse. A subject is said to have had a mycological relapse who shows positive mycology at any time point after having achieved mycological cure. A subject is said to have had a clinical relapse who shows any clinical signs of infection at any time point after having achieved clinical cure.

4.2.3.4 Ease of Insertion

For each insertion point the investigator will assess and record ease of insertion of the treatment procedure as described in Section 9.2.3.

5 SELECTION AND WITHDRAWAL OF PATIENTS

Subjects with a clinical diagnosis of DLSO of the large toenails caused by dermatophytes, laboratory confirmed by KOH visualization and positive culture for dermatophytes, will be enrolled until 30 subjects have completed the 3 treatments specified by the protocol.

Approximately 60 subjects will be screened for entrance to the study. Based on previous studies in this indication, it is expected that the majority of subjects will be male. Subjects will be recruited from a single podiatric practice, the OrthoArizona East Valley Foot & Ankle Specialists, based in Mesa, Arizona. The practice operates from additional locations serving Gilbert, Scottsdale and San Tan Valley that may serve as referral (not treatment) centers.

Subjects must meet all of the following inclusion criteria (Section 5.1) to be eligible for enrollment into the study and must not meet any of the exclusion criteria listed in Section 5.2 in order to be eligible.

5.1 Subject Inclusion Criteria

1. Male or female between 18 and 74 years of age inclusive.
2. Clinical diagnosis of DLSO in at least one great toenail. If both great toenails meet inclusion criteria, the toenail with the greater involvement will be designated the target toenail.
3. Linear nail involvement of at least 4 mm, but without involvement of the proximal 3 mm of the nail, based on the investigator's visual assessment. Overall nail involvement should be at least 25% but not more than 75% as per the investigator's visual assessment (Grade 3). Nail thickness ≤ 2 mm or total thickness of the nail plus subungual debris measuring ≤ 3 mm.
4. Subject's nails are required to have a normal rate of growth (approximately 1 mm / month or greater) in order to be physiologically capable of achieving clinical cure/near clinical cure within the 48-week study period.
5. Laboratory confirmation of DLSO with direct microscopy of subungual debris positive for hyphal elements (KOH test) and fungal culture positive for dermatophytes.
6. Generally of good health.
7. Willing to comply with protocol requirements and sign a statement of informed consent.
8. Willing to refrain from using any lotions, creams, liquids, or polish on the large toenail or on the skin immediately adjacent to the large toenail during the treatment period unless directed to do so by the investigator.
9. Willing to refrain from using on the toenails topical products to which the subject has a high risk of developing an allergic reaction and/or dermatitis;
10. Willing to refrain from receiving pedicures for the duration of the study.
11. Willing to refrain from using topical steroids or topical antifungals on the great toenail or the skin immediately adjacent to the great toenail, or systemic antifungals for the duration of the study.

5.2 Subject Exclusion Criteria

1. History of severe or chronic immunosuppression, an immunocompromised condition, known or suspected HIV, extensive dermatomycoses, extensive recurrent herpes zoster or severe dermatitis affecting the feet that would interfere with safety and tolerability assessments.
2. Any systemic or dermatologic disorder, such as uncontrolled psoriasis, severe eczema, or severe atopic dermatitis which, in the opinion of the investigator, will interfere with the study results or increase the risk of adverse events (subjects with mild, controlled psoriasis, eczema, or dermatitis may be included if the condition would not interfere with safety and tolerability assessments).
3. Any severe and/or chronic disease that would affect a subject's nail growth or interfere with the subject's ability to complete the trial, such as severe renal failure, peripheral vascular disease, severe chronic obstructive pulmonary disease, severe heart disease, uncontrolled diabetes mellitus or other endocrine disease, or uncontrolled malignancy.
4. History of toenail surgery or any significant injury to the target toenail matrix.
5. Treatment with any investigational drug within 1 month prior to Screening.
6. Topical antifungal treatment applied to the feet within 1 month prior to the start of study treatment.
7. Use of oral terbinafine within 6 months, or use of any other oral antifungal drug within 3 months, prior to the start of study treatment.
8. Hypersensitivity to terbinafine or to any other ingredients of the formulation.
9. Active onychomycosis of the fingernail.
10. Symptomatic tinea pedis requiring treatment at Day 1 of the study.
11. Superficial white, proximal subungual onychomycosis, lichen planus, psoriasis, or any condition that interfere with the treatment procedure or assessment of clear nail.
12. Structural deformities of the target toenail or foot (eg, genetic or pigment disorders, chemical damage, tumors) that would interfere with treatment procedures or with assessments of efficacy, safety, or tolerability.
13. Suspected subungual dermatophytoma.
14. Any other condition that in the opinion of the investigator renders the subject unsuitable for participation in this study.

5.3 Subject Recruitment and Retention

Subjects will be primarily recruited from a single podiatric practice and 3 (non-treatment) locations that are part of the practice. This site will conduct a prescreening study (HTS-001), which will precede and run concurrently with the conduct of study HTS-002B, until enrollment is complete.

This pre-screening study will establish the rate of normal nail growth in up to 400 subjects with clinically confirmed DLSO of the great toenail, after appropriate consent has been obtained. Based on nail growth rates observed in HTS-001, subjects will be invited to the Screening visit for study HTS-002B. Inclusion of a population with a nail growth rate potentially capable of clearing diseased nail within 48 weeks, will provide an opportunity to establish a treatment effect in a small sample size earlier than in a study with an unselected population and improve compliance by providing subjects an approximate target date for a clinical cure.

5.4 Subject Withdrawal and Stopping Criteria

In accordance with legal requirements and ICH GCP, subjects may withdraw from the study at any time without stating a reason and without prejudice to further treatment. A subject's participation is to be terminated immediately upon his/her request. If, at the time of refusal, HTS-519 has already been administered, the subject will be advised on follow-up safety procedures.

The investigator may withdraw a subject from the study and discontinue study treatment and assessments at any time.

The subject and the investigator are advised to be alert to any adverse effects that may be attributable to the study medication or procedures, and not to continue study treatments when there is any perceived risk of harm to the subject.

A subject's discontinuation from the study will be considered if a subject experiences a serious adverse event (SAE) or develops conditions which would have prevented his/her entry into the study according to the safety-related medical exclusion criteria.

The investigator must discontinue a subject from the study if the following criteria are met:

- Any subject who develops intolerable adverse effects attributable to the study procedures - an "intolerable adverse effect" is one that is regarded by either the subject or the investigator or both as necessitating the discontinuation of study treatment.
- Biochemical or clinical evidence of liver injury with total bilirubin levels greater than 3 times the upper limit of normal is detected.

If a subject is discontinued because of an AE, the event will be followed until it is resolved or it is considered to have stabilized and does not require further follow-up.

If a subject is discontinued at any time after entering the study, the subject will stop receiving further treatment. The investigator will make every effort to see the subject

and complete a final evaluation, following the end of study assessments (Section 8.3.3). This is not required if a subject is withdrawn prior to dosing.

Early discontinuation of each subject who has given informed consent to participate will be recorded in the case report form (CRF) including the primary reason for discontinuation, other than study completion, as described in Table 5-1:

Table 5-1 Reason and Description of Subject Withdrawal or Discontinuation

Reason	Description
Failure to meet eligibility criteria	Failure to meet inclusion and / or exclusion criteria
Adverse Events (AE)	Clinical events occurred or laboratory results are reported that in the medical judgment of the investigator are grounds for discontinuation in the best interests of the subject.
Lost to Follow-Up	The subject stopped coming for visits and study personnel were unable to contact the subject.
Withdrawal of Consent	The subject desired to withdraw from further participation in the study. The subject is not obliged to provide any reason for withdrawal of consent, but where a reason is given this will be recorded in the CRF.
Study Terminated by Sponsor	An indication that a clinical study was stopped by its sponsor.
Physician Decision	A position, opinion or judgment to withdraw or discontinue the subject from the study reached after consideration by a physician.
Protocol Deviation	The subject failed to adhere to the protocol requirements, and, in the investigator's opinion, the best interest of the subject or study assessment of safety of the subject would be affected by continued involvement.
Lost to Follow-Up	The subject stopped coming for visits and study personnel were unable to contact the subject.
Death	Outcome was death
Other	The subject was terminated for a reason, other than one previously listed, that could potentially impact data or subject safety.

5.5 Handling of Subject Withdrawals or Discontinuation of Study Intervention

All enrolled subjects will be included in the study analysis whether or not they withdraw from the study or discontinue the study procedures after enrollment. However, enrollment of subjects will continue until 30 subjects have received all 3 of the prescribed treatments.

5.6 Study Discontinuation

This study may be suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided by the suspending or terminating party to the investigator and regulatory authorities. If the study is prematurely terminated or suspended, the principal investigator will promptly inform the IRB and will provide the reasons for the termination or suspension.

6 STUDY DESIGN

6.1 Study Design

This is a single-center, open-label study intended to establish preliminary point estimates of efficacy and assess the clinical benefits and safety of HTS-519 Insert, administered in the subungual space, in subjects with clinically and laboratory-confirmed DLSO of the great toenail.

All subjects will be screened for DLSO and will meet specific inclusion and exclusion criteria.

If both great toenails have clinical and laboratory evidence of onychomycosis and fulfill the inclusion / exclusion criteria, the one with the greater involvement will be designated as the target nail for efficacy evaluation, but both of the great toenails may be treated and evaluated for safety if the subject and the investigator consent.

Treatment will be administered to all subjects on Day 1, 29, and 57. Each subject will receive a dose of from 1 to 4 HTS-519 Inserts based on the width of the nail involved by disease (Section 7.2). Each treatment visit will be followed up with a phone contact the next day to enquire about adverse events.

After the last treatment on Day 57, subjects will return for follow-up observations 4 weeks apart, until Visit 8 (Week 24). Thereafter, subjects are followed-up every 12 weeks until End of Study Visit 10 (Week 48). Study design is summarized in Table 6-1.

Table 6-1 Study Design

Screening	Treatment HTS-519 Administration			Post Treatment			
Screening	Baseline First Treatment	Second Treatment	Third Treatment	Intermediate Visits			End of Study
	Day 1	Day 29 (±3)	Day 57 (±3)	Days 85, 113 and 141 (±7)	Day 169 (±7)	Day 253 (±7)	Day 337 (±7)
Visit 1	Visit 2	Visit 3	Visit 4	Visit 5, 6, 7	Visit 8	Visit 9	Visit 10

At Week 48, subjects that in the opinion of the investigator have a reasonable chance to reach a complete cure in the 24 weeks following the End of Study Visit or have reached a complete cure at Visit 10, will be invited to complete additional visits. Subjects that meet the extension criterion at Visit 10 will be evaluated again at Visit 11 (Week 60) to determine their eligibility to remain in the study until Visit 12 (Week 72).

Treatment efficacy will be evaluated on the basis of mycology (KOH test results and culture for dermatophytes) and clinical signs (investigator assessment of clear nail and length of unaffected nail as determined by planimetric photographic measurements). Persistence of treatment effect will be assessed during the post-treatment phase (Section 9).

Safety measurements will include local clinical signs and symptoms, including erythema, edema, hematoma, bruising, itching, and pain; and reports of adverse events (Section 10).

6.2 Procedures and Assessments Flow

The major procedure and assessments performed by the investigator or delegated staff on the (target) great toenail during the study are summarized and sequenced in Table 6-2 for all visits categorized as either screening, treatment and post-treatment visits.

Table 6-2 Procedure and Assessment Flow

Procedure				Visits and Order of Procedure		
	Aim	Procedure / Tool	Subjects	Screen	Treatment	Post-Treatment
Target toenail assessment, nail clipping and notching (Appendix 2)						
	Standardized assessment of clear nail (growth) & treatment	Clipping and measurements of the nail with caliper / ruler; notching of the nail	All	1	1	1
Digital photography (Canfield Manual)						
	Visual evidence of disease and emergence of clear nail	Digital image capture of the target great toenail per Canfield Manual	All	2	2	2
Distal Digital Block (Appendix 3)						
	Local anesthesia of nail unit	Infiltration anesthesia (liocaine 2%)	All		3	
Subungual Sampling for Mycology (Appendix 4)						
	Laboratory confirmation DLSO	Subungual material sampling by curettage	All	3		3
Precannulation Procedure (Appendix 5)						
	Increase nail flexibility	Soaking with normal saline	All		4a	
	Access to subungual space	Debridement	Selected		Optional	
	Determine optimal insertion points	Optimal insertion point algorithm based on involved nail width using photographic ruler and skin marking	All		4b	
	Subungual depth assessment	Small gauge probe/cannula to determine most proximal edge of subungual space (leading edge)	All		4c	
		27G or 30G .406mm x 50mm				
		27G or 30G .305mm x 25mm				
HTS-519 Insert Administration Procedure (Appendix 6)						
	Usability of needle applicator / HTS-519 Insert	Inspect HTS-519 Insert assembled in applicator	All		5a	
	Microcannulation	Atraumatic creation of subungual channel	All		5b	
		22G or 23G .712mm x 50mm				
		22G or 23G .635mm x 29mm				
	HTS-519 Insert administration	Advance needle applicator through subungual channel already created and deposit HTS-519 Insert proximally into subungual channel / confirmation of placement	All		5c	
		25G .508mm x 44.5mm				
End of Assessment of Procedure(s) – Site Administration Reaction (Appendix 7)						
	Local tolerability assessment of procedural site	Site Administration Assessment Scale	All		6	
End of Assessment of Procedure(s) – Digital Photography (Canfield Manual)						
	Documentation of areas of diseased nail, emergence of clear nail and status pre- and post procedure	Outline diseased nail and obtain digital image of the dorsal surface of the great toenail per Canfield Manual	All	4	7	4

7 STUDY INTERVENTION

7.1 Investigational Product Description

The HTS-519 (Hallux Terbinafine Subungual) Insert is a white to off-white cylindrical pellet, 0.394 mm in diameter and 6 mm in length, containing 519 µg terbinafine hydrochloride constituting 70% of total weight (740 µg). Terbinafine hydrochloride is formulated in the finished product for immediate release. The insert further consists of 21% polyethylene glycol 3350 (PEG), 5% triethyl citrate, and 4% stearic acid. The HTS-519 Insert is preassembled in a 25-gauge needle applicator, designed to deposit the insert into the subungual space between the nail plate and nail bed.

7.1.1 Acquisition

The HTS-519 Insert will be supplied by the sponsor, Hallux, Inc. Hallux will also provide flexible blunt-tipped microcannulae of increasing sizes (22G-30G), enabling the safe administration of HTS-519 Insert in a subungual channel. Clinical supplies required for the distal digital block will be sourced from the site's pharmacy stock.

7.1.2 Formulation, Packaging and Labelling

The HTS-519 Insert is supplied assembled in a 25G needle single-use applicator. Each applicator is packaged in a sealed pouch for single use. A label with the following information will be affixed to each unit package:

Hallux, Inc.
Laguna Hills, CA 92653
HTS
P/N 02-100-01
Protocol HTS-002B
Contents: 1 applicator, each containing 519µg terbinafine
Sterile
Caution: New Drug – for Investigational Use Only
Store at room temperature (59-86^{0}F / 15-30^{0}C)
Lot No. 16B001
Mfg. Date: 16/02/11
Label P/N: 20-100-02

7.1.3 Product Storage and Stability

The HTS-519 Insert should be stored at room temperature 59-86^{0}F / 15-30^{0}C.

7.2 Dosage, Preparation and Administration of Study Product

Placement of HTS-519 Inserts equidistant from each other is the most objective and effective means to achieve uniform distribution of terbinafine to the target nail area after dissolution and diffusion of the active ingredient from the biodegradable polymer insert.

Administration of four HTS-519 Inserts is considered the maximum number of inserts or maximum feasible dose (MFD) that a large toenail with a typical anatomical width of between 10-20 mm can practically accommodate. For complete and uniform antifungal coverage of a 20-mm-wide nail with a maximum of 4 inserts, each insert would be required to provide 5 mm of coverage, or 2.5 mm bi-directionally. However, the extent of diseased or involved nail width (INW) is typically less than the anatomical width of the toenail. Assuming that only the INW requires coverage (ie, not the entire nail width) and that each insert provides 5 mm of coverage, smaller nails/nails with reduced INW will require proportionally fewer Inserts for complete coverage of the INW. For this protocol, a standardized dose of 1 insert/5 mm INW will be utilized.

Dose determination therefore starts with measuring the width of diseased or involved nail (INW) using a (transparent) ruler. The measured width of diseased nail divided by 5, rounded up to the nearest integer, yields the number of inserts or MFD to be administered to any diseased nail area and limits the maximum required coverage for each HTS-519 Insert to 5 mm width (0-5 / 1, 6-10 / 2, 11-15 / 3 and 16-20 / 4 as in Table 7-1).

Table 7-1 Determination of Maximal Feasible Dose (MFD) by Involved Nail Width (INW)

	Involved Nail Width (INW) of Affected Great Toenail (mm) and MFD																			
INW (mm)	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
MFD (number of Inserts)	0	1					2					3					4				

After topical administration of HTS-519, terbinafine dissolves from the biodegradable polymer Insert. The ability to reach fungicidal tissue concentrations depends on terbinafine diffusion bi-directionally from the insert and adequate tissue penetration. An optimal insertion point algorithm was developed to maximize terbinafine delivery to target tissue by efficiently distributing the terbinafine diffusion distance from each HTS-519 Insert over the width of diseased nail. The algorithm calculates a diffusion distance (displayed in **bold** in Table 7-2) for any involved nail width for any number (*d)* of inserts as follows: involved nail width / (2 x *d*). This number is used to determine the actual insertion point from the edge of the diseased nail (eg, 2 mm, for a 12-mm involved nail width requiring 3 inserts [Table 7-1]). Spacing between HTS-519 Inserts is twice the calculated distance (eg, 4 mm for nail referenced above, resulting in optimal insertion points at 2, 6 and 10 mm from the medial edge of disease [Table 7-2]).

Table 7-2 Optimal Insertion Point Algorithm Based on Involved Nail Width (mm)

INSERTS	1	2		3			4			
INW (mm)	I	I	II	I	II	III	I	II	III	IIII
20							2.5	7.5	12.5	17.5
19							2.4	7.1	11.9	16.6
18							2.3	6.8	11.3	15.8
17							2.1	6.4	10.6	14.9
16							2.0	6.0	10.0	14.0
15				2.5	7.5	12.5				
14				2.3	7.0	11.7				
13				2.2	6.5	10.8				
12				2.0	6.0	10.0				
11				1.8	5.5	9.2				
10		2.5	7.5							
9		2.3	6.8							
8		2.0	6.0							
7		1.8	5.3							
6		1.5	4.5							
5	2.5									
4	2.0									
3	1.5									
2	1.0									
1	0.5									
0	0.0									

The actual insertion point for each insert (I-IIII in Table 7-2) will be recorded from the medial side of the toenail's lateral fold and subsequently marked on the adhesive-backed photo scale tape (rounded to the nearest 0.5mm) folded in order to determine its anatomical position and administration in a consistent and standardized manner (see Appendix 5).

The diffusion distance is not only used to determine optimal insertion points. It also serves as a measure of a fixed dose (dose = mm diseased nail width covered), allowing a comparison of safety and efficacy between treatments independent of the number of inserts administered and involved nail width treated.

With a maximum feasible number of four HTS-519 Inserts per toe, a single treatment would deliver 4 (inserts) x 519 µg = 2,076 µg or 2.076 mg and 4,052 µg or 4.152 mg terbinafine if both large toenails are treated. This compares to 13.2-45.0 mg/day for daily dosing with marketed topical terbinafine creams, gels or spray, and 250mg/day for systemically administered terbinafine for 3 months. The treatment with HTS-519 Inserts is repeated 3 times, 4 weeks apart, for a total exposure over approximately 8 weeks of 6.228 mg, or 12.456 mg when 2 great toenails are being treated.

7.3 Modification of Study Product Administration

Small deviations (± 1 mm) from the insertion point determined by the optimal insertion point algorithm to facilitate atraumatic placement of the maximum feasible dose (MFD) or number of inserts is anticipated and referred to as opportunistic placement.

Anatomical nail and disease characteristics necessitating a different insertion point (opportunistic placement) than determined from Table 7-2 could include the points of greatest radius of curvature of the nail plate in the region defined by the longitudinal midline of the nail plate and a lateral nail fold and areas of onycholysis or hyperkeratosis.

The investigator may decide to administer fewer HTS-519 Inserts than determined by involved nail width based on clinical considerations. For example, the investigator may decide to omit administration of a HTS-519 Insert, if it appears that the subungual space is not accessible after pretreatment of the nail or reasonable effort by the investigator and even an opportunistic placement is not feasible.

With clearing of the nail the width of diseased nail may diminish and the number of HTS-519 Inserts should be adjusted accordingly.

The longitudinal depth of the accessible subungual space at the predetermined insertion point may be less than 6 mm, the length of the HTS-519 Insert. In that instance, after administration, the HTS-519 Insert may be projecting out beyond the level of the distal groove and distal nail plate. The part of the HTS-519 Insert not covered by the nail plate will be removed manually.

Any modification to the optimal insertion point algorithm or administration as described above must be documented in the CRF.

7.4 Treatment Accountability and Compliance Checks

It is the investigator/Institution's responsibility to establish a system for handling study treatments and products. This includes:

- Maintaining accurate records of receipt of all test articles by a responsible person, including dates of receipt.
- Providing treatment to subjects only as directed in the protocol. Reasons for departure from the expected dispensing regimen must be recorded.
- Storing all study treatments and supplies at site in a secure, restricted access area (eg, a locked room) under room temperature conditions and protected from light, moisture, and freezing as stated on the label.
- Reconciling delivery records with records of usage and destroyed/returned stock. Records of usage should include the identification of the person to whom the study treatment was dispensed and the quantity and date of dispensing. This record is in addition to any drug accountability information recorded in the CRF. Any discrepancies must be accounted for on the appropriate forms. Certificates of delivery and return must be signed, preferably by the investigator or a pharmacist, and copies retained in the investigator Site File.

7.5 Procedures for Training of Clinicians on Procedural Intervention

Clinical experience has demonstrated that tactile rather than visual feedback guides the advancement of devices through subungual debris, with a clear tactile "stop" signal felt when reaching the proximal boundary of the accessible subungual space. In this initial study of HTS-519 Insert, treatment will be administered by a single investigator, Lewis Freed, DPM. Dr. Freed has been involved in the optimization of the subungual procedure and is well qualified to conduct this study.

7.6 Assessment of Clinician and/or Subject Compliance with Study Procedural Intervention

Study treatments are administered by the treating investigator at Day 1, Day 29, and Day 57. Immediately after each treatment, the investigator's will record in the CRF the number of inserts inserted, the insertion point and depth of each insert, and the aspects of the disease presentation which informed the investigator's choice of number and placement as per Section 7.2 and Section 7.3.

The presence of HTS-519 Inserts after placement will be confirmed visually if not obscured by discolored nail by the investigator and documented in the CRF. Transillumination (or diaphanoscopy) of the nail unit with a standard pocket penlight or similar light source may be used to visualize an opaque HTS-519 Insert obscured by nail disease and aid with the subungual localization of the administered inserts. Post-procedure digital image capture, which also includes a transillumination sequence, is used to document the presence of a HTS-519 Insert in the subungual space.

7.7 Prior and Concomitant Medications/Treatments

Any concomitant medication deemed necessary for the welfare of the subject during the study may be given at the discretion of the investigator. It is the responsibility of the investigator to ensure that details regarding the medication are recorded in full in the CRF.

Prohibited medications include:

- Concomitant use of systemic antimycotics during the study. Topical antifungal medicines will be permitted for subjects who develop symptomatic tinea pedis during the course of the study but contact with the large target toe must be avoided.
- Use of lotions, creams, liquids, or polish on the target great toenail or the skin immediately adjacent to the target great toenail during the study is prohibited.

7.8 Allocation to Treatment

Each subject who satisfies the criteria to participate in the study will be assigned a unique subject number. The subject numbers will be assigned sequentially in the order in which subjects are enrolled. The investigator will administer the required number of HTS-519 Inserts based on diseased nail width (Section 7.2).

7.9 Treatment Blinding Code

This is an open label study in which investigator and subject are not blinded to treatment.

8 STUDY VISIT SCHEDULE AND CONTACTS

Study visits are based on the number of days from the date of the study visit at Baseline (Day 1) and summarized in Table 1-1. During the treatment phase a window of ±3 days is allowed for the study visit based on the date of Baseline. For all other study visits, a window of ±7 days is allowed for each study visit based on the date of Baseline.

8.1 Screening (Visit 1)

The screening visit will occur within approximately 6 weeks of the Baseline visit. At Screening, the following procedures will be completed for each subject:

- Explain the particulars of the study to potential subjects.
- Obtain and document informed consent from potential subjects before performing any study-related procedures.
- Obtain demographic, medical history and concomitant medication information.
- Review inclusion/exclusion criteria for eligibility.
- Perform a physical examination and obtain vital signs needed to determine eligibility.
- Evaluate the involved great toenail(s) for anatomical (nail width) and disease characteristics as per Appendix 2 and complete the Investigator Global Assessment Scale (Section 9.1.1) to confirm eligibility.
- Collect subungual samples by curettage from each great toenail for which there is clinical suspicion of the presence of onychomycosis. Examine subungual samples using KOH microscopy. Confirm the presence of hyphae and ship unused subungual material to the central mycology laboratory for confirmatory fungal culture for dermatophytes (Appendix 4) and enroll the subject into the study. The tissue sample for screening mycology may be collected up to 8 weeks before the start of study therapy.
- Outline affected area of the great toenail(s) and obtain a digital photograph of the dorsal surface of the toenail(s) for analysis by Canfield Scientific as described in the Canfield manual.

8.2 Allocation and Treatment Visits

8.2.1 Enrolment / Baseline, Day 1, (Visit 2)

Results of fungal cultures, clinical laboratory assessments, and digital photography analysis will be reviewed by the Principal investigator in order to notify subjects of their eligibility to continue in the study. Subjects meeting eligibility criteria will return for their Baseline visit within approximately 6 weeks of Screening. At Baseline, the following procedures are to be completed:

- Review inclusion/exclusion criteria for eligibility.
- Document any changes to medical history, physical condition, concomitant medications according to the subject's response since the Screening visit.
- Evaluate the involved great toenail(s) for anatomical (nail width) and disease characteristics as per Appendix 2 and complete the Investigator Global Assessment Scale (Section 9.1.1) to confirm eligibility.
- Determine the location of the great toenail designated as the target toenail for the duration the study. If both great toenails have clinical and laboratory evidence of

onychomycosis, the one with the greater involvement will be designated as the target nail for the primary efficacy evaluation. Both great toenails may be treated.
- Notch the target toenail 3 mm from the lunula as a baseline for nail growth measurement (Appendix 2).
- Obtain subject evaluation of pain before, during and after the infiltration anesthetic and cannulation procedure (Section 10.2).
- Administer study treatment as described in Appendix 3, 5 and 6 and record site administration reactions (Appendix 7) if any.
- Obtain a digital photograph of the dorsal surface of the toenail(s) for analysis by Canfield Scientific as described in the Canfield manual.
- Dispense subject 7-day diary.
- Schedule the next day follow-up phone contact and Day 29 visit.

8.2.2 Day 29 ±3 days, (Visit 3), and Day 57 ±3 days, (Visit 4)

At study visit 3, and visit 4, the following procedures will be performed:
- Document any changes to concomitant medications and occurrence of adverse events according to the subject's response since the last visit.
- Retrieve the subject 7-day diary and review for completeness.
- Administer study treatment as described in Appendix 3, 5 and 6 and record site administration reactions (Appendix 7) if any.
- Obtain subject evaluation of pain before, during and after the infiltration anesthetic and cannulation procedure (Section 10.2).
- Obtain a digital photograph of the dorsal surface of the toenail(s) for analysis by Canfield Scientific as described in the Canfield manual.
- Dispense subject 7-day diary.
- Schedule the next day follow-up phone contact and the Day 57 and Day 85 Visit as appropriate for the visit.

8.3 Post-Treatment Visits

8.3.1 Intermediate Visits: Day 85 ±7 days, (Visit 5 – Week 12), 113 ±7 days, (Visit 6 – Week 16), and 141 ±7 days, (Visit 7 – Week 20)

At study Visits 5, 6, and 7, the following procedures will be completed:
- Document any changes to concomitant medications and occurrence of adverse events according to the subject's response since the last visit.
- Retrieve the subject 7-day diary and review for completeness (Visit 5 only).
- Evaluate the (target) great toenail for clear nail and extent of clear nail using the Investigator Global Assessment Scale as described in Section 9.1.1.
- Obtain a digital photograph of the dorsal surface of the toenail(s) for analysis by Canfield Scientific as described in the Canfield manual.
- Schedule the Day 113 and 141 Visits respectively.

8.3.2 Intermediate Visits: Day 169 ±7 days (Visit 8 – Week 24) and Day 253 ±7 days (Visit 9 – Week 36)

At the study visit 8 and 9, the following procedures will be conducted:

- Document any changes to concomitant medications and occurrence of adverse events according to the subject's response since the last visit.
- Evaluate the (target) great toenail for clear nail and extent of clear nail using the investigator Global Assessment Scale as described in Section 9.1.1.
- Collect subungual samples by curettage from each treated great toenail and ship to the central mycology laboratory for confirmatory fungal culture for dermatophytes and KOH microscopy as described in Appendix 4.
- Obtain a digital photograph of the surface area of the target great toenail for analysis by Canfield Scientific as per the Canfield Manual.
- Schedule the Day 253 and 337 Visits respectively.

8.3.3 End of Study, Day 337 ±7 days (Visit 10 – Week 48)

When the subject returns for the End of Follow-up Visit, the following procedures will be completed:

- Document any changes to concomitant medications and occurrence of adverse events according to the subject's response since the last visit.
- Perform a physical examination, including vital signs. Collect subungual samples by curettage from each treated great toenail and ship to the central mycology laboratory for confirmatory fungal culture for dermatophytes and KOH microscopy as described in Appendix 4.
- Evaluate the (target) great toenail for clear nail and extent of clear nail using the investigator Global Assessment Scale as described in Section 9.1.1.
- Obtain a digital photograph of the surface area of the target great toenail for analysis by Canfield Scientific as per the Canfield Manual.
- Schedule the Day 420 Visit for those subjects that in the opinion of the investigator have the ability to reach a complete cure over the next 24 weeks based on clinical improvement observed during the entire study duration and mycological status, or have reached a complete cure at this Visit.

8.4 Post-Treatment Extension Visits

8.4.1 Study Extension Visit, Day 420 ±7 days (Visit 11 – Week 60)

For those subjects returning for the Study Extension Visit, the following procedures will be completed:

- Evaluate the (target) great toenail for clear nail and extent of clear nail using the investigator Global Assessment Scale as described in Section 9.1.1.
- Collect subungual samples by curettage from each treated great toenail and ship to the central mycology laboratory for fungal culture for dermatophytes and KOH microscopy as described in Appendix 4.
- Obtain a digital photograph of the surface area of the (target) great toenail for analysis by Canfield Scientific as per the Canfield Manual.
- Schedule the Day 504 Visit for subjects with a complete cure at this visit and for those subjects that in the opinion of the investigator have the ability to achieve a complete cure by Visit 12.

8.4.2 Study Extension Visit, Day 504 ±7 days (Visit 12 – Week 72)

For those subjects returning for the Study Extension Visit, the following procedures will be completed:

- Evaluate the (target) great toenail for clear nail and extent of clear nail using the investigator Global Assessment Scale as described in Section 9.1.1.
- Collect subungual samples by curettage from each treated great toenail and ship to the central mycology laboratory for confirmatory fungal culture for dermatophytes and KOH microscopy as described in Appendix 4.
- Obtain a digital photograph of the surface area of the (target) great toenail for analysis by Canfield Scientific as per the Canfield Manual.

8.5 Withdrawal Visit

For any subject who withdraws from the study prior to Week 48, the End of Study (Visit 10) assessments should be performed and recorded to the extent possible and appropriate.

8.6 Unscheduled Visit

Although every effort will be made to see subjects at the per protocol visits unplanned visits may occur. These unscheduled visits may occur to perform additional assessments or procedures outside of the protocol specific visit calendar. These assessments could be related to the emergence of new clinical signs and symptoms or adverse events, lab results or questions the subject may have about the study or for any other clinical reason.

Unscheduled visits will be assigned the designation Visit “0”. The date of the visit and observations relevant to the study will be recorded in the CRF (eg, adverse events, change in medication) under “unscheduled visit”. If there are no specific observations to be captured the reason of the visit is recorded in the medical records only.

9 EFFICACY ASSESSMENTS

9.1 Evaluation of the Target Toenail

The investigator will clinically establish the presence of DLSO of the great toenail as well as any clinical involvement of other toenails. Thickness of subungual hyperkeratosis will be measured in millimeters (one decimal point) from the top of the nail plate to the nail bed, using a caliper. Nail width will be recorded in millimeters using a ruler (Appendix 2).

The affected area of the nail or visible leading edge of disease may be outlined by the investigator prior to target toenail evaluation and digital image collection to facilitate central image review.

The assessments listed in Sections 9.1.1, 9.1.2 and 9.1.3 determine subject eligibility and treatment efficacy and are conducted after clipping the nail to the distal groove (the transverse demarcation extending across the distal digit that divides the hyponychium from the volar epidermis). Subjects are therefore allowed to cut their toenails, but not beyond the target nail's distal groove so as not to interfere with the assessment of clear nail.

9.1.1 Investigator Global Assessment of Clear Nail

Evaluation of clear nail will be based upon that portion of surface area of the nail that is clear of signs of disease (ie, nail dystrophy, onycholysis or subungual hyperkeratosis) attributable to onychomycosis and active infection. The investigator will assess the involved great toenail for the extent of clear nail using the investigator Global Assessment (IGA) Scale. Missing nail is considered diseased nail.

Table 9-1 Investigator Global Assessment Scale

Grade 0	Completely clear nail, with nail area clear of signs of residual disease (ie, nail dystrophy, onycholysis or subungual hyperkeratosis) attributable to onychomycosis
Grade 1	Almost clear nail with ≤ 10% affected nail
Grade 2	11%-24% affected nail
Grade 3	25%-75% affected nail
Grade 4	>75% affected nail

Note that a completely clear nail is not necessarily a "perfect nail" as the appearance of the nail may be affected by comorbid and pre-existing conditions unrelated to DLSO, such as longitudinal ridging, lamellar nail splitting, trachyonychia, brittle nails, and trauma, that will not respond to antifungal treatment.

9.1.2 Digital Photography

A high resolution digital photograph of the target nail is taken with specialized photographic equipment and computer hardware standardized for distance and lighting provided by Canfield Scientific (central imaging lab) to document visual evidence of nail disease, nail clearance or status post subungual sampling or HTS-519 Insert administration.

Each subject will place the target foot in the toe stop that also functions as an internal scale included in all images to allow objective measurements of clear nail. The digital image will be captured before and after treatment administration as per the Canfield manual provided as a separate document. The image will be electronically uploaded instantaneously to Canfield Scientific for immediate assessment of quality.

9.1.3 Subungual Sampling for Mycology

As described in Appendix 4 the investigator collects subungual samples by curettage from each great toenail for which there is clinical suspicion of the presence of onychomycosis.

At Screening samples will be examined by the investigator using KOH microscopy to confirm the presence of septate hyphae, and remaining subungual material will be sent to the mycology laboratory for KOH visualization and confirmatory fungal culture for dermatophytes. After the Screening visits, the samples may be sent to the laboratory without the on-site KOH microscopy.

If at Screening the KOH microscopy exhibits septate hyphae, but the culture fails to grow dermatophytes the culture may be repeated. If 2 sequential cultures are negative, it is recommended that the nail not be included in the study and no further cultures are attempted.

Cultures grown from samples taken during the study will be stored at the central mycology laboratory for future susceptibility testing at the end of the study. Results will be reported separately from the final study report.

9.2 Evaluations Associated with the Insertion Procedure

9.2.1 Insertion Point

The insertion point for each HTS-519 Insert administration is determined by an optimal insertion point algorithm (Section 7.2). Its exact location relative to the medial side of involved nail (in mm) is marked on a self-adhesive (transparent) photo scale ruler referred to in Appendix 5 to guide placement. In case of an opportunistic placement (Section 7.3), the position of the actual insertion point is marked on the ruler as well and recorded in the CRF.

9.2.2 Insertion Depth

The insertion depth will be determined by marking the microcannula with a black marker pen at the level of the distal groove when the micro-cannula is advanced to the “stop” position and measuring the distance between the mark and the tip or mark and hub. The insertion depth that was thus established is used to mark the maximum insertion depth on the shaft of the needle applicator with a marker pen.

9.2.3 Investigator Insertion Rating

A 5-point Likert scale will be used for the investigator’s assessment of ease of insertion. The scoring ranges from 1-5 with (1) = very easy, (2) = easy, (3) = difficult, (4) = very difficult and (5) = impossible to administer the HTS-519 Insert.

10 SAFETY ASSESSMENTS

The safety of HTS-519, administered subungually, will be assessed by monitoring local tolerability at the administration site, adverse events, and evaluating data from physical examinations and vital signs measurements.

10.1 Local Tolerability

Before and after placement of HTS-519 Insert(s) and at each visit the investigator will examine the nail unit and surrounding tissue for adverse signs and symptoms. Immediately following the administration of an insert the event of bleeding will be evaluated specifically by the investigator using the following scale: 0 = none, 1 = punctate bleeding not extending beyond the distal groove, 2 = active bleeding extending beyond the distal groove and resolving within 5 minutes, and 3 = active bleeding extending beyond the distal groove and lasting longer than 5 minutes (Appendix 7).

Within 1 hour of treatment the subject will record the level of burning/stinging, itching, redness, swelling, oozing, and bruising. The signs and symptoms will be rated as 0 = none, 1 = mild, 2 = moderate, or 3 = severe as defined in Appendix 7.

After each treatment the subject will be provided a 7-day diary to record the same assessments every evening using the rating scale described in detail in Appendix 7. The day after treatment a phone contact will take place to ensure that the diary is completed and to collect any changes in medication and health status.

The presence or absence of a subungual hematoma will be documented by the investigator at every visit. In addition, over the course of the study, the investigator will observe the nail unit for signs of nail dystrophy, foreign body reactions, and infectious processes such as paronychia and osteomyelitis.

10.2 Patient Assessment of Pain

The numerical rating scale (NRS) is a one-dimensional pain intensity scale with a number 0 (no pain) located on the left and a number 10 (worst pain imaginable) displayed on the right of the horizontal scale.[27] The subject will be instructed to select a number that best represents their perception of the degree of pain. The magnitude of the pain associated with anesthesia, access to the subungual space and placement of a HTS-519 Insert is collected on paper by the subject and subsequently recorded in the CRF.

The subject will be provided a subject diary for seven days following each insertion procedure to record pain levels using the same numerical rating scale every evening.

10.3 Physical Examination and Vital Signs Measurements

An abbreviated physical examination will be performed at Screening and Week 48. The examination will focus on the essential aspects of the subjects' illness and general physical condition, including examination of the foot for the presence of tinea pedis and adequacy of circulation etc..

Vital signs assessed include heart rate and blood pressure using a blood pressure recording device with an appropriate cuff size. Measurements will be made after the

subject has been resting supine for a minimum of 5 minutes. Weight will be measured as well.

10.4 Adverse Events

10.4.1 Adverse Event Definition and Documentation

An adverse event (AE) is any untoward medical occurrence in a subject, administered an investigational product and which does not necessarily have to have a causal relationship with this treatment (21 CFR 312.32). An AE (also referred to as an adverse experience) can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of an investigational product, whether or not considered related to the investigational product.

Adverse events may be volunteered spontaneously by the subject or discovered as a result of general, non-leading questioning, through physical examination, laboratory test, or other means.

Adverse events resulting from concurrent illnesses, reactions to concurrent illnesses, reactions to concurrent medications, or progression of disease states must be recorded. Any medical condition that is present at the time that the subject is screened but does not deteriorate should not be reported as an AE. However, if the signs or symptoms associated with the medical condition deteriorate at any time during the study, it should be recorded as an AE.

Abnormal laboratory values or test results will be recorded as AEs only if (1) they induce clinical signs or symptoms, (2) require therapy, or (3) are considered by the investigator to be of clinical significance. Prior medical conditions/diseases are considered AEs only if they worsen after the start of protocol-specified study procedures.

All AEs reported or observed from the time a subject signs informed consent until exit from the study must be recorded in detail on the appropriate page of the CRF and followed until the event is resolved, the event reaches a clinically stable outcome, or the subject is lost to follow-up. For each AE, the AE record will include type of event, start and stop dates, severity (mild, moderate, or severe), seriousness, relationship to the study treatment (definite, probably, possibly, unlikely, or not related), actions taken (required treatment or observations), and outcome.

Adverse events that are prospectively collected as outcomes or observations include procedure related pain, post-treatment symptoms collected in a subject diary and an investigator assessment of the nail before and after treatment. For the purpose of this study this prospectively collected information will not be again reported as AEs unless:

- Considered clinically significant by the Investigator,
- Requiring treatment or study treatment modification, or
- Meeting the criteria for a Serious Adverse Event (SAE)

A temporary change in nail appearance reasonably associated with healing or clearing of disease is not considered an adverse event, unless any of the 3 criteria above apply.

10.4.2 Serious Adverse Events

An AE or SAR is considered “serious” (see 21 CFR 312.32) if, in the view of either the investigator or sponsor, the adverse event:

- Results in death.
- Is life threatening (i.e., the subject was at risk of death at the time of the event; it does not refer to an event that hypothetically might have caused death if it was more severe).
- Requires insubject hospitalization or prolongation of existing hospitalization
- Results in persistent or significant incapacity or substantial disruption of the ability to conduct activities of normal life.
- Is a congenital anomaly / birth defect.
- Is considered to be an important medical event.

Based upon medical and scientific judgment, important medical events that may not be immediately life threatening, or result in death or hospitalization, but may jeopardize the subject or may require intervention to prevent one of the other outcomes listed in the definition above may be considered a serious adverse event (SAE).

Hospitalizations are defined as official admission, not necessarily an overnight stay for observation or for an out-subject procedure. If, however, the investigator feels that the cause of the out-subject procedure or overnight stay is medically significant, then the event will be considered an SAE with the serious criteria of Important Medical Event. A hospitalization for social reasons in the absence of an AE is not an SAE. Additionally, if a subject is hospitalized for a procedure that was planned prior to the study, this will not be considered an SAE. If a hospitalization is prolonged, such as for a fever, then that fever will be considered an SAE.

10.5 Time Period and Frequency for Event Assessment and Follow-up

The investigator will record all reportable events with start dates occurring any time after informed consent is obtained until 7 (for non-serious AEs) or 30 days (for SAEs) after the last day of study participation in the CRF. 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.

10.6 Adverse Event Characteristics

Each AE has three attributes as described in Sections 10.6.1, 10.6.2 and 10.6.3. Severity and relationship to study treatment are recorded in the CRF by the investigator. The Medical Monitor and the Study PI will be responsible for determining whether an SAE is expected or unexpected. An adverse event will be considered unexpected if the nature, severity, or frequency of the event is not consistent with the risk information previously described for the intervention.

10.6.1 Adverse Event Severity

The investigator will classify the severity (intensity) of each AE according to the following definitions summarized in Table 10-1:

Table 10-1 Adverse Event Severity

Classification	Definition
Mild	Awareness of sign or symptom, but easily tolerated; no impact on activities of daily living
Moderate	Sign(s) or symptom(s) cause discomfort, but do not interfere with or prevent normal activities
Severe	Sign(s) or symptom(s) of sufficient intensity to interfere with or perform normal activities

It should be pointed out that the term “severe” is a measure of intensity and that a severe AE is not necessarily serious.

10.6.2 Adverse Event Relationship to Study Treatment

All events considered to be noxious and unintended responses to an investigational product related to any dose should be considered an Adverse Reaction (AR). The phrase “responses to a medicinal product” means that a causal relationship between a medicinal product and an AE is at least a reasonable possibility.

A suspected adverse reaction (SAR) therefore indicates any AE for which there is a reasonable possibility that the drug caused the adverse event, ie, there is evidence to suggest a causal relationship between the drug and the adverse event.

The determination of the causal relationship between the drug and the adverse event will be based in part on the investigator’s assessment of the relationship of the AE to the investigational treatment. An AE considered by the investigator to be probably or definitely related to the study treatment is to be considered an AR, and an AE considered by the investigator to be possibly related to the study treatment is a SAR, but not an AR. An AE considered by the investigator unlikely to be related, or to be unrelated, to the study treatment, is not a SAR, unless the sponsor determines otherwise based on other evidence.

The investigator will assess the likely relationship of each AE to the investigational product according to the definitions outlined in Table 10-2.

Table 10-2 Terms for Defining Relationship of Adverse Events to Study Product

Association	Definition	Interpretation
Not Related	(1) the existence of a clear alternative explanation (eg, mechanical bleeding at surgical site) or (2) non-plausibility, eg, the subject is struck by an automobile or develops cancer a few days after product administration, event occurred before dosing.	Adverse Event (AE) - Unrelated
Unlikely Related	A clinical event, including laboratory test abnormality (if applicable), with an improbable time sequence to product administration and in which other drugs, chemicals for underlying disease provide plausible explanations.	

Association	Definition	Interpretation
Possibly Related	A clinical event, including laboratory test abnormality (if applicable), with a reasonable time sequence to administration of the drug, but which could also be explained by concurrent disease or other drugs or chemicals.	Suspected Adverse Reaction (SAR) - Related
Probably Related	A clinical event, including laboratory test abnormality (if applicable), with a reasonable time sequence to administration of the drug, unlikely to be attributed to concurrent disease or other drugs or chemicals, and which follows a clinically reasonable response on withdrawal.	Adverse Reaction (AR) - Related
Definitely Related	A clinical event, including laboratory test abnormality (if applicable), that is clearly related to the investigational agent(s) or research intervention: the adverse event has a temporal relationship to the administration of the investigational agent(s) or research intervention, follows a known pattern of response, and no alternative cause is present.	

10.6.3 Expectedness of Adverse Events

An AE or SAR is considered “unexpected” if it is not listed in the Investigator’s Brochure or is not listed at the specificity or severity that has been observed; or, if an Investigator’s Brochure is not required or available, is not consistent with the risk information described in the general investigational plan or elsewhere in the current application, as amended.

“Unexpected” as used in this definition, also refers to AEs or SARs that are mentioned in the Investigator’s Brochure as occurring with a class of drugs or as anticipated from the pharmacological properties of the drug, but are not specifically mentioned as occurring with the particular drug under investigation.

10.7 Reporting Procedures

Clinical safety personnel will be available for SAE reporting on a 24-hour basis. Reports will be reviewed during normal business hours. Investigator instructions and sponsor requirements for safety reporting are outlined in Sections 10.7.1 and 10.7.2, respectively.

The Medical Monitor for this study is Dr. Henk-André Kroon, Chief Medical Officer at Hallux, Inc. who can be reached on hak@halluxinc.com, by phone (949) 353 9538 or fax (949) 315 7373.

10.7.1 Investigator Reporting Requirements - SAE

Site personnel must report any AE that meets SAE criteria or unusual frequency of AEs within 24 hours from the time staff is aware of the event to the medical monitor (or designee for this study), even if the event(s) appear to be unrelated to study treatment.

Follow-up information about a previously reported SAE must also be reported to the medical monitor (or designee) within 24 hours of receiving it. Follow-up reports regarding the status of the SAE and the subject’s subsequent course should be submitted until the SAE has subsided, the condition stabilized (in the case of persistent impairment), the subject receives alternative therapy, or the subject dies.

Electronic submission of initial and follow-up information and automatic e-mail notification of the Medical Monitor will occur when the investigator completes the SAE report form in the CRF. Once submitted, the medical monitor or designee will send a confirmation email to the investigator within 1 business day. In the event a confirmation email is not received, contact the Medical Monitor directly by phone, email or fax. A confirmation of receipt will be sent within 1 business day.

If the SAE has not been previously documented (ie, is a new occurrence) and it is thought to be related to the investigational product (or therapy), the medical monitor may contact the investigator to obtain further information. If warranted, an investigator alert may be issued, to inform all investigators involved in any study with the same product (or therapy) that this SAE has been reported.

The IRB should also be notified of SAEs and of any follow-up information in writing according to their reporting requirements.

10.7.2 Sponsor Reporting Requirements

Hallux must notify the FDA and all participating investigators of any potential serious risks associated with use of the study product as they are identified during this clinical trial. This notification must be made as soon as possible but in no case later than 15 calendar days (7 calendar days for fatal or life-threatening SARs) after determining that the information qualifies for reporting based on the following criteria:

10.7.2.1 Serious and unexpected suspected adverse reactions.

The sponsor must report any suspected adverse reaction that is both serious and unexpected to all concerned Investigators/Institutions, to the Institutional Review Boards (IRBs), where required, and to the regulatory authority(ies). A single or small number of events of a specific AE must be reported as a SAR only if there is a reasonable possibility that study product caused the event (ie, there is evidence to suggest a causal relationship between the study product and the AE). For example:

- A single occurrence of an event that is uncommon and known to be strongly associated with drug exposure (eg, angioedema, hepatic injury, Stevens-Johnson Syndrome and acute liver failure);
- One or more occurrences of an event that is not commonly associated with drug exposure, but is otherwise uncommon in the population exposed to the drug (eg, tendon rupture);
- An aggregate analysis of specific events observed in a clinical trial (such as known consequences of the underlying disease or condition under investigation or other events that commonly occur in the study population independent of drug therapy) that indicates those events occur more frequently in the drug treatment group than a concurrent or historical control group.

10.7.2.2 Increased rate of serious suspected adverse reactions

The sponsor must also report any clinically important increase in the frequency of a serious SAR over that which is listed in the protocol or Investigator’s Brochure.

10.7.2.3 Unexpected fatal or life-threatening suspected adverse reactions

The sponsor must notify the FDA of any unexpected fatal or life-threatening suspected adverse reactions as soon as possible but in no case later than 7 calendar days after the sponsor's initial receipt of the information.

Expedited reports should comply with the applicable regulatory requirement(s) and with the International Conference on Harmonisation (ICH) Guideline for Clinical Safety Data Management: Definitions and Standards for Expedited Reporting (E2A). The Sponsor should submit to the regulatory authority(ies) all safety updates and periodic reports, as required by applicable regulatory requirement(s).

11 STATISTICAL CONSIDERATIONS

A detailed and comprehensive Statistical Analysis Plan (SAP), describing all statistical analyses will be prepared and approved prior to database lock. The statistical analysis and report will conform to relevant ICH guidance. Any changes to the statistical methods need not be reported as a protocol amendment but must be documented in the clinical study report.

11.1 Statistical Hypotheses

Statistical hypothesis testing is not planned for this study.

11.2 Sample Size Determination

No formal sample size calculation was performed for this study. The sample size of 30 subjects completing 3 treatments is considered sufficient to provide initial point estimates of treatment-specific effects in this phase 2 study.

11.3 Final Analysis Plan

The standard summary statistics for analysis of the categorical baseline and outcome variables (mycological, clinical cure, and Therapeutic Efficacy) will be count and proportion (expressed as percentage). The following standard summary statistics will be used for analysis of continuous baseline and outcome variables: N, mean, standard deviation, median, quartiles, and maximum and minimum.

11.3.1 Analysis Populations

The *intent-to-treat analysis set* comprises all subjects who have been assigned to study treatment. This will be the primary analysis set for efficacy. The effect of missing data points will be analyzed on an exploratory basis.

The *per-protocol analysis set* comprises all subjects within the intent-to-treat analysis set who have completed the assessments for clear nail and mycology for dermatophytes at Week 48 and who have no major protocol violations. This will be the secondary analysis set for efficacy. Allocation of subjects to the per-protocol analysis set will be determined at a meeting convened by the Sponsor after the database is locked.

The *safety analysis set* comprises all subjects who have received at least 1 dose of study treatment.

11.3.2 Demographics and Baseline Characteristics

Subject demography and baseline characteristics will be presented in by-subject listings and summarized by descriptive statistics. A separate summary and listing will be provided for subjects discontinued before randomization (Screen Failures).

11.3.3 Prior and Concomitant Medications

Prior and concomitant medication information for all randomized subjects will be presented in a by-subject listing.

11.3.4 Statistical Methods for Efficacy Parameters

The appropriate standard summary statistics will be used for all efficacy variables. Cure rate endpoints will be estimated with 95% confidence intervals. Parametric endpoints (eg, ratios and lengths) will be summarized using descriptive statistics.

11.3.5 Safety Analysis

Extent of exposure to study treatments will be summarized according to the number of subjects exposed and the total dose administered.

Incidence, relatedness, and severity of AEs and SAEs will be tabulated by preferred term and system organ class using the most recent version of MedDRA. Adverse events will be summarized by presenting the number and percentage of subjects having any AE, having an AE in each body system and having each individual AE.

Of particular interest are the AEs involving administration site reactions (ie erythema, burning/stinging, itching, scaling, swelling/edema) and self-reported pain. These will be listed separately and summarized by frequency counts at each visit and at each time point of the subject diary. In addition, the incidence of SAEs, drug related AEs, serious and drug-related AEs, and any AEs resulting in discontinuation from the study will be listed.

Vital signs measurements and physical examination findings, together with changes from baseline assessments, will be described using standard summary statistics.

Baseline for all physical examination evaluations and vital signs measurements will be defined as the last evaluation just prior to the first administration of study treatment.

12 SOURCE DOCUMENTS AND ACCESS TO SOURCE DATA/DOCUMENTS

Source data are all information, original records of clinical findings, observations, or other activities in a clinical trial necessary for the reconstruction and evaluation of the trial. Source data may be written or typed by authorized and qualified study personnel directly from clinical observation, or they may be generated, on paper or electronically, by an automated measurement device or system of devices.

Source documents may include, for example, subject medical records, screening or enrollment logs, laboratory reports, electrocardiograms, subject diaries, drug dispensing and collection records, drug accounting logs, study notes, and study-related correspondence.

In order to ensure the integrity of the study data, procedures must be in place to allow every element of the study data to be traced to its source. It is the responsibility of the study monitor to verify that data recorded on the CRF are an accurate transcription of the source data. The source documents are typically the property of the study site itself, and may include data that are not relevant to the study database and are therefore not subject to study-related quality control procedures.

The study site must maintain a list of originators (persons, devices, and instruments) of all source data. The list for persons should include signatures, initials, and user identification (user ID) tags that will permit a reviewer to trace the source data records to their originator.

In order to facilitate source data verification of the CRF, all source documents must be maintained at the study site in individual subject folders. While not strictly a source document, the original signed Informed Consent document is to be kept in the same folder for ease of review. Source documents must be maintained in the same way for all enrolled study subjects and for screen failures (individuals who are screened for the study but who do not meet the entrance criteria).

The investigator must assure that the subject's anonymity will be maintained. On all study documentation, with the exception of the consent form and subject identification logs, subjects will only be identified by their unique identification code and initials and will not be referred to by name.

All records will be kept in a secure storage area with limited access. Clinical information will not be released without the written permission of the subject (or the subject's legal guardian), except as necessary for monitoring and auditing by the Sponsor, its designee, U.S. Food and Drug Administration (FDA) or other regulatory agencies, or the IRB.

13 QUALITY CONTROL AND QUALITY ASSURANCE

As specified in the investigator's agreement, the investigator agrees to allow the sponsor's (or designated CRO's) study monitor, quality assurance auditor, health authority inspector, and/or IRBs inspector direct access to all relevant source documents, and to allocate his/her time and the time of his/her staff to the auditor/inspector to discuss findings and any issues.

13.1 Monitoring Procedures

During the study, the study monitor will review the progress of the study on a regular basis to ensure adequate and accurate data collections. Specific data verification procedures will be described in a study-specific monitoring plan that includes aspects of ongoing remote and periodical on-site monitoring activities.

Monitoring site visits to review CRFs, subject case notes, administrative documentation including the investigator Site File, and telephone communications with site staff, will be performed throughout the study.

At each study monitoring visit, the investigator will make available all records pertaining to the study. To allow sufficient time to assemble documentation for the study monitor, monitoring visits will be confirmed in advance of planned visits.

Significant or relevant communications with the Sponsor or sponsor's designated study representative should be documented by the site and retained for the study file.

13.2 Quality Assurance

In addition to the routine monitoring procedures and to ensure compliance with GCP and all applicable regulatory requirements, the investigator(s)/Institution(s) will permit study-related audits, IRB review, and regulatory inspection(s), providing direct access to all source documents, drug records, and original CRFs at some or all of the study sites used in the study.

The sponsor or sponsor's representative, will provide prior notice of such a planned GCP audit, during or after completion of the study. The investigator should promptly notify the sponsor or CRO of any audits scheduled by any regulatory authorities or IRB and promptly forward copies of any audit reports received to the sponsor.

14 ETHICS COMMITTEE REVIEW/INFORMED CONSENT

14.1 Ethical Conduct of the Study

The study will be conducted in accordance with ethical principles expressed in ICH E6(R1): Good Clinical Practice (GCP) guidelines, the WMA Declaration of Helsinki,[28] IRBs, and in accordance with the United States Title 21 Code of Federal Regulations (CFR) Part 50 Protection of Human Subjects.

This study must be carried out in compliance with the protocol, applicable laws and US regulatory requirements.

14.2 Institutional Review Board

The final study protocol, subject informed consent form and other documentation or information provided subjects will be reviewed and approved by the IRB in compliance with local regulations. Approval will be received in writing before initiation of the study.

Any changes to the study design will be formally documented in protocol amendments and approved by the IRB prior to implementation, except in the case of changes made to protect subject safety, which will be implemented immediately.

14.3 Informed Consent

The investigator is responsible for obtaining informed consent from the subject after adequate explanation of the aims, methods, anticipated benefits, and potential hazards of the study and before any protocol-specific screening procedures or any study medications are administered. The investigator or designee will answer any questions that may arise. Subjects will be given the opportunity to discuss the study with their surrogates or think about it prior to agreeing to participate.

This informed consent should be given by means of a standard written statement, provided in non-technical language. The subject is required to read and review the document or have the document read to him or her before signing and dating it. If written consent is not possible, oral consent can be obtained, if witnessed by a signed statement from one or more persons not involved in the study, mentioning why the subject was unable to sign the form. No subject can enter the study before informed consent has been obtained from him/her, or his/her legally authorized representative.

Subjects will also be asked to consent to allow the sponsor, sponsor representative, or external regulatory auditor to review their medical records to confirm compliance with GCP.

The acquisition of informed consent should be documented in the subject's clinical or research record and the informed consent form should be signed and personally dated by the subject and by the person who conducted the informed consent discussion. The original signed informed consent form should be retained in the investigator Site File and a copy of the signed consent should be provided to the subject prior to participation in the trial.

The subjects will be informed that they may withdraw from the study at any time without prejudice to further treatment. They will receive all information that is required by local regulations and ICH guidelines.

14.4 Exclusion of Women, Minorities, and Children (Special Populations)

Individuals of any gender or racial/ethnic group may participate in this study as long as their age is between 18-74 inclusive.

14.5 Disclosure and Confidentiality

14.5.1 Confidentiality of Study Documentation

By signing the protocol, the investigator agrees to keep all information provided by the sponsor in strict confidence and to request similar confidentiality from his/her staff and the IRB. Study documents provided by the study sponsor (ie, protocols, investigators' brochures, CRFs and other material) will be stored appropriately to ensure their confidentiality. The information provided by the sponsor to the investigator may not be disclosed to others without direct written authorization from the sponsor, except to the extent necessary to obtain informed consent from subjects who wish to participate in the trial.

14.5.2 Privacy of Individual Health Information

Subject confidentiality is strictly held in trust by the investigators, study staff, and the sponsor(s) and their agents. This confidentiality is extended to cover testing of biological samples in addition to any study information relating to subjects.

While all data records will be identified by the corresponding subject number, the identity of the subject will be held in confidential source documents at the study site. No information concerning the study or the data will be released to any unauthorized third party except as specifically authorized by each individual subject in the written informed consent.

The study monitor or other authorized representatives of the sponsor may inspect all study documents and records required to be maintained by the investigator, including but not limited to, medical records (office, clinic, or hospital) for the study subjects. The clinical study site will permit access to such records. All study personnel with access to this information are legally bound not to disclose it.

14.6 Future Use of Stored Specimens and Other Identifiable Data

De-identified digital images of the subject’s toenail(s) will be stored during the duration of the study at Canfield Scientific, before being archived at the sponsor after study completion.

No biological samples will be stored for future use and no genetic testing will be performed.

15 DATA HANDLING AND RECORD KEEPING

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. The investigators will maintain adequate case histories of study subjects, including accurate case report forms (CRFs), and source documentation.

15.1 Data Management Responsibilities

Data collection and accurate documentation are the responsibility of the study staff under the supervision of the investigator. All source documents and laboratory reports must be reviewed by the study team and data entry staff, who will ensure that they are accurate and complete. Unanticipated problems and adverse events must be reviewed by the investigator or designee.

Once recorded, the study data must be protected from unauthorized modification or deletion, and all authorized modifications and deletions must be securely linked in the permanent record with their author, time of change, and reason for change (ie, the audit trail must be maintained). There must be a procedure whereby the PI certifies the data to be accurate and complete and releases the data for transmittal to the sponsor or CRO.

Procedures and specifications for management of the study data once released to sponsor or CRO will be described in detail in a separate Data Management Plan to be approved by the sponsor. This document will include definitions of the data sets and variables, references to prevailing SOPs, and descriptions of the following procedures:

- Procedures for data management review and query processing;
- Procedures for assignment and medical review of standard preferred-term coding of adverse events, concomitant medications, or other terminology collected in the study data;
- Procedures for electronic data transfer of study data elements from external non-CRF data sources, such as analytical, imaging, or other specialized laboratory services;
- Procedures for certification and closure of the database prior to unblinding.

Procedures for data collection and data management will be designed to ensure that each data element may be traced with a high level of confidence from its originator or recorder to its representation in the study database and then to its place in the analysis and integrated clinical study report according to GCP.

At each stage in the data collection and management process, the data must be accessible for review by authorized parties, such as the study monitor, a designated auditor, and an FDA inspector.

15.2 Data Capture Methods

The primary data collection tool for the study is an electronic data capture (EDC) or CRF system designed to record all the data required by the protocol in designated labeled fields. A computer generated password and specific roles to access and use the EDC will be provided to study staff prior to study start.

Each page of the CRF is headed by identifying information including the study number, subject number, and study visit number or time point. Entries to the CRF are made by authorized site staff according to written CRF Completion Guidelines for the study or as instructed by a qualified trainer at study initiation.

Case report form entries may be written directly from clinical observation or they may be transcribed from source data recorded at an earlier time. The originator of the data (ie, the individual making the observation or evaluation) date and time of the recorded event are captured electronically.

Each CRF must be completed within 5 days of a visit, reviewed, and subsequently signed, and dated by the investigator as outlined in the project timeline. The completed CRF will be reviewed by data management after monitoring is complete. A copy of the completed CRF will be provided to the investigator. The copy will remain at the site in the investigator's files.

Computer systems and devices which are used to produce electronic source documents must conform to the principles described in relevant regulatory guidances (eg, the FDA's Draft Guidance [December 2010] on Electronic Source Documentation in Clinical Investigations, and references therein), which are designed to ensure the security and integrity of those data sources.

15.3 Types of Data

During the conduct of this study, safety and efficacy data will be collected in the CRF. Safety data consist of standard adverse event data as well as specific site administration reaction data. Efficacy data consist of investigator assessments of clear nail and laboratory data from the Center of Medical Mycology and emergence of clear nail. Outcome data include assessments of pain and ease of insertion.

15.4 Schedule and Content of Reports

The following reports will be generated during the conduct of this open label study to monitor study progress and quality.

A screening log will be kept to monitor and analyze reasons for screen failure.

The quality of subungual sampling will be assessed for the ability of the investigator to visualize septate hyphae. In addition, results from the Center of Medical Mycology will be reviewed for evidence of bacterial contamination of the sample and the ability of a clinical diagnosis of DLSO to be confirmed by laboratory means. The review may lead to modifications to the subungual sampling technique at the site.

Canfield's digital image result reports issued for each subject at each visit, will be reviewed for photo quality, correct trimming of the nail, subungual sampling technique and congruence with sequential IGA results.

Early terminations will be tracked in order to ensure that subjects return for their final visit. Protocol deviations will be monitored and the determination of protocol deviations will be determined prior to unlocking the database.

15.5 Study Records Retention

The investigator is required to maintain all study documentation, including regulatory documents, copies of CRFs, signed informed consent forms, and records for the receipt and disposition of study medications, for a period of at least 2 years following the last approval date of a marketing application in an ICH region or until at least 2 years after the formal discontinuation of clinical development of the investigational product, whichever comes later.

15.6 Publication and Data Sharing Policy

Following completion of the study, the data may be considered for a formal presentation at a scientific meeting or for publication in a scientific journal. In these cases, the Sponsor will be responsible for these activities and will work with the investigator(s) to determine how the manuscript is written and edited, the number and order of authors, the publication to which it will be submitted, and other related issues. Data are the property of the Sponsor and cannot be published without prior authorization from the Sponsor, but data and publication thereof will not be unduly withheld.

The publication or presentation of any study results shall comply with all applicable privacy laws, including, but not limited to, the Health Insurance Portability and Accountability Act (HIPAA) of 1996.

15.7 Protocol Deviations

The investigator or designee must document and explain in the subject's source documentation any deviation, intentional or unintentional, from the approved protocol that may affect the subject's rights, safety, or well being and/or the completeness, accuracy, and reliability of the study data.

Protocol deviations will be documented by the clinical monitor throughout the course of monitoring visits. Investigators will be notified of deviations in writing by the monitor and corrective action and preventative action plan will be instituted in accordance with sponsor's SOPs. The IRB will be notified of all protocol violations in a timely manner as per its guidelines.

16 LITERATURE REFERENCES

1. Foster, K. W., Ghannoum, M. A., & Elewski, B. E. (2004). Epidemiologic surveillance if cutaneous fungal infection in the United States from 1999 to 2002. J Am Acad Dermatol(50), 748-752.
2. Elewski, B. E., & Hay, R. J. (1996). Update on the management of onychomycosis: highlights of the third international summit on cutaneous antifungal therapy. Clin Infect Dis, 23, 305-13.
3. Elewski, B. E. (1998, July). Onychomycosis: pathogenesis, diagnosis and management. Clin Microbiol Rev, 415-429.
4. Tosti, A., Baran, R., Dawber, R. P., & Haneke, E. (2003). Onychomycosis and its treatment. In R. Baran, R. P. Dawber, E. Haneke, A. Tosti, & I. Bristow, Text Atlas of Nail Disorders (pp. 197-220). London, England: Martin Dunitz.
5. Scher, R. K. (1996). Onychomycosis: a significant medical disorder. J Am Acad Dermatol, 35(3 Pt 2), S2-5.
6. Novartis Pharmaceuticals Corporation. (2015, February). Lamisil® Tablets 250 mg [Package Insert]. East Hanover, NJ, USA.
7. Ortho-McNeil-Janssen Pharmaceuticals. (2014). Sporanox® Capsules, 100mg [Package Insert]. Titusville, NJ, USA.
8. Valeant Pharmaceuticals North America LLC. (2014, June). Jublia® topical solution 10% [Package Insert]. Bridgewater, NJ, USA.
9. Anacor Pharmaceuticals, Inc. (2014, July). Kerydyn® topical solution 5% [Package Insert]. Palo Alto, CA, USA.
10. Dermik Laboratories. (2004, December). Penlac® Nail Lacquer [Package Insert]. Berwyn, PA, USA.
11. Haneke, E., Tausch, I., Brautigam, M., Weidinger, G., & Welzel, D. (1995). Short-duration treatment of fingernail dermatophytosis: a randomized, double-blind study with terbinafine and griseofulvin. LAGOS III Study Group. J Am Acad Dermatol(32), 72-77.
12. Roberts, D. T. (1994). Oral therapeutic agents in fungal nail disease. J Am Acad Dermatol(31), S78-S81.
13. Baran, R., & Hay, R. J. (2001, Oct). New evidence for the efficacy of combination therapy in onychomycosis. Br J Dermatol, 145(Suppl), 60:1.
14. Villars, V., & Jones, T. C. (1989). Clinical Efficacy and tolerability of terbinafine (Lamisil) a new topical and systemic fungicidal drug for treatment of dermatomycoses. Clin Exp Dermatol(14), 124-27.
15. Sigurgeirsson, B., Billstein, S., Rantanen, T., & et al. (1999). Study: efficacy and tolerability of continuous terbinafine (Lamisil®) compared to intermittent itraconazole in the treatment of toenail onychomycosis. Br J Dermatol, 141(Suppl 56), 5-14.
16. Ryder, N. S. (1992). Terbinafine: mode of action and properties of teh squalene epoxidase inhibition. Br J Dermatol, 126(Suppl 39), 2-7.
17. Birnbaum, J. E. (1990). Pharmacology of the allylamines. J Am Acad Dermatol(23), 782-85.
18. Baran, R., Dawber, R. P., Hanecke, E., & Tosti, A. (2003). Nail plate and soft tissue abnormalities. In R. Baran, R. P. Dawber, E. Hanecke, A. Tosti, & I. Bristow, A Text Atlas of Nail Disorders: Techniques in investigation and diagnosis (p. 87). London: Martin Dunitz.

19. Baran, R., & De Doncker, P. (1995). Lateral edge nail involvement indicates poor prognosis for treating onychomycosis with the new systemic antifungals. Acta Derm Venereol, 76, 82-83.
20. Levy, L. L., & Emer, J. J. (2012, Apr-Jun). Complications of Minimally Invasive Cosmetic Procedures: Prevention and Management. J Cutan Aesthet Surg, 5(2), 121-132.
21. Shemer, A., Trau, H., Davidovici, B., Grunwald, M. H., & Amichai, B. (2008). Collection of fungi samples from nails: comparative study of currettage and drilling techniques. JEADV(22), 182-185.
22. Wolinsky, J. B., Colson, Y. L., & Grinstaff, M. W. (2012, April 10). Local Drug Delivery Strategies for Cancer Treatment: Gels, Nanoparticles, Polymeric Films, Rods, and Wafers . J Control Release, 159(1), 14-26.
23. Sigurgeirrson, B. (2010). Prognostic factors for cure following treatment of onychomycosis. J Eur Acad Dermatol Venereol, 24, 679-84.
24. Shuster, S. (1998). Onychomycosis: making sense of the assessment of anti-fungal drugs. Acta Derm Venereol, 78, 1-4.
25. Zaias, N., & Drachman, D. (1983). A method for the determination of drug effectiveness in onychomycosis. J Am Acad Dermatol, 9, 912-9.
26. Zaias, N., & Rebell, G. (2004). The successful treatment of trichophytum rubrum nail bed (distal subungual) onychomycosis with intermittent pulse dosed terbinafine. Arch Dermatol, 140, 691-5.
27. Flaherty, S. A. (1996). Pain measurement tools for clinical practice and research. J Am Assoc Nurse An, 133-40.
28. World Medical Association General Assembly. (2013, October). World Medical Association Declaration of Helsinki - Ethical principles for Medical Research Involving Human Subjects. Retrieved March 21, 2016, from World Medical Association: http://www.wma.net/en/30publications/10policies/b3/index.html.pdf?print-media-type&footer-right=[page]/[toPage]
29. Flarity-Reed, K. (2002, Aug). Methods of digital block. J Emerg Nurs, 28(4), 351-4.
30. Sigurgeirsson, B. (2011, January 23). Onychomycosis - How to taka a good sample - v2.1. Retrieved March 21, 2016, from Vimeo: https://vimeo.com/19090068

SIGNATURES AND AGREEMENT WITH THE PROTOCOL

Sponsor Approval

The signature of any of the representatives below confirms the fact that I have reviewed and approved protocol 3.0 and confirm that the protocol follows GCP.

Signature: DocuSigned by: 30ACB8DEFF5947D... Date: 9/14/2017

Henk-André Kroon, MD, MBA
Chief Medical Officer, Hallux, Inc.

Signature: DocuSigned by: Jay E Birnbaum 1161026BAD7E4A6... Date: 9/14/2017

Jay Birnbaum, PhD
Chief Scientific Officer, Hallux, Inc.

Principal investigator Approval

I agree to conduct the study according to the terms and conditions of this protocol, current ICH Guidelines for GCP, the Code of Federal regulations, the Health Insurance Portability and Accountability Act, and with applicable regulatory requirements. All information pertaining to the study shall be treated in a confidential manner.

Signature: DocuSigned by: Lewis H Freed 3560CFCBD70E4A1... Date: 9/27/2017

Lewis H Freed, DPM
Principal investigator
OrthoArizona - East Valley Foot & Ankle Specialists

APPENDIX 1: GENERAL PROCEDURE AND ASSESSMENT CONDITIONS

The following equipment is needed in the procedure room(s) at the treatment site:

- A barrier free examination chair with an instrument tray at the foot section covered with a black cloth or material.
- Non-sterile clippers, transparent ruler and caliper (eg, Carrera Precision CP9806-TF Titanium 0-6-Inch Electronic Fractional & Decimal Digital Caliper)
- Sterile gloves, antiseptic solution, local anesthetic (lidocaine 2% solution), small gauge needles, and 2-5 ml syringe for administering local anesthetic.
- Sterile gloves, soap (eg,“Dove”), water, (disposable) emesis basin, chuck pads, paper towel, alcohol swabs, sterile serrated curette or No. 15 scalpel blade, DermaPak® and sterile specimen container for subungual sampling.
- Adhesive-backed photo scale tape, permanent marker to the nail plate (eg, Lumocolor® Permanent Marker Superfine Black), (non)-sterile surgical marker (eg, Classical Style Surgical or Surgeons Fine Tip Skin Marking Pen) to mark the optimal insertion points on the skin, penlight for transillumination.
- Laminated optimal insertion point algorithm table.
- Sterile gauze, (normal) saline wound wash/flush (0.9% sodium chloride solution, drug and preservative free).
- DermaSculpt® blunt-tipped microcannulas sized 22-30G and HTS-519 Insert assembled in a 25G needle applicator.
- Photo-equipment, camera unit tethered to a dedicated laptop which is connected to the internet, provided by Canfield Scientific.
- Other data collection and recording equipment as needed.

At any visit explain the procedure and assessments that will be performed or conducted at the visit to the subject and ensure informed consent was obtained. Check the records for data from earlier treatment visits or treatments and verify which toe is the target toenail.

APPENDIX 2: QUANTITATIVE ASSESSMENT OF NAIL GROWTH, SUBUNGUAL HYPERKERATOSIS AND NAIL WIDTH

Nails will be clipped to within 1 mm of the hyponychium, the thickened stratum corneum of the epidermis, which lies under the free edge of the nail - discard the nail clippings.

Subungual hyperkeratosis will be evaluated with a caliper and measured in millimeters from the hyponychium to the top of the nail plate at the site of maximum involvement. All measurements are to be recorded in one-decimal point (eg,1.4 mm). If no subungual hyperkeratosis is present, the measurement would be recorded as zero.

Nail width (diseased and non-affected nail) is measured at the distal nail groove using a transparent ruler or caliper and recorded in millimeters.

The method to measure linear nail growth used is adapted from Zaias et al. (Zaias & Drachman, 1983) (Zaias & Rebell, 2004) and involves following the distal outgrowth of a transverse notch in the superficial nail plate. This is achieved as follows:

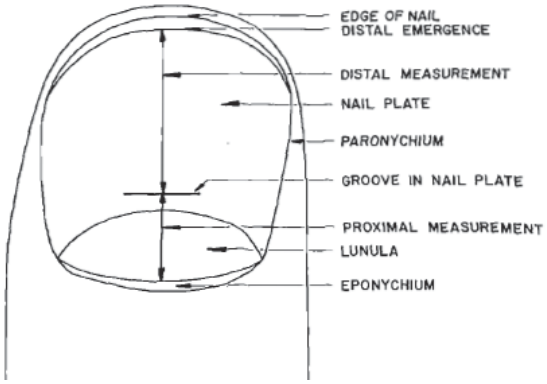
1. Wear gloves.

2. Wash the nail and surrounding skin with soap and water and dry them with a clean towel.

3. Identify an area of clear nail in the midline, approximately 3 mm distal from the nail lunula but proximal to the infected nail area to place the notch.

4. Use a sterile scalpel#15 to cut a notch (Fig. 16-1 – “groove in nail plate”) into the nail plate surface plate perpendicular to the longitudinal nail axis at the site identified in step 3.

Figure 16-1 Placement of a Notch or Groove in the Midline Relative to Lunula



As the initial notch moves distally over time, it may grow out of the nail or be clipped away when trimming the nail. At that time a new notch will have be created following the same procedure described ins steps 1-4 above in order to enable continued measurement of the rate of nail growth.

APPENDIX 3: DISTAL DIGITAL BLOCK

The three-sided digital block[29] creates an adequate block of the 4 digital dorsal and volar digital nerves that innervate the nail unit.

1. The digit is sterilized by swabbing all areas to be treated with a surgical soap, 0.5% chlorhexidine gluconate solution in 70% isopropanol or alcohol. A tourniquet is not used in this procedure.

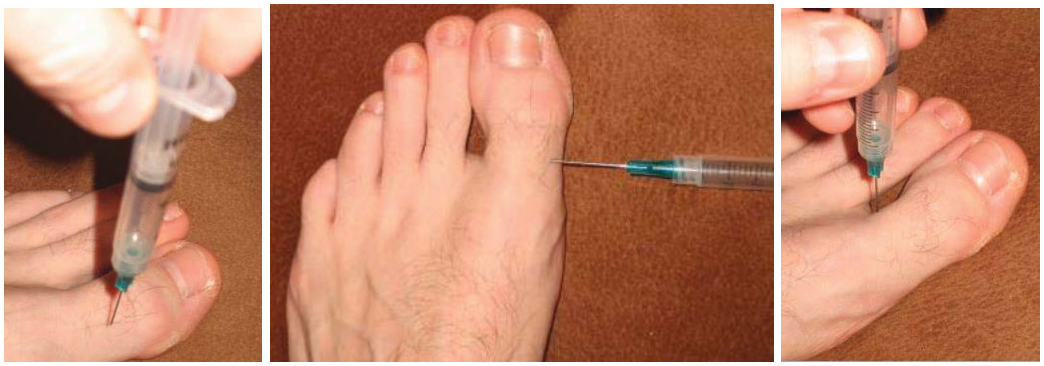
2. Prepare lidocaine 2% for injection without epinephrine in a 3 mL or dental syringe and apply a 30-gauge injection for injection.

3. Place the patient's extremity volar/plantar side down.

4. Administer a topical cryogen spray (50% ethyl chloride/50% dichlorotetrafluorethane spray) for 1 to 7 seconds just prior to needle insertion to create a light frost at the intended injection site (2-3 mm proximal to the junction of proximal and lateral nail fold).

5. Perform a Three-Sided Digital Block: a total of 1.0-2.0mL of Lidocaine HCl 2% is administered distal from the metatarsal-phalangeal joint (Fig. 16-2):
    i. Insert a 30G needle at a 90° angle at the medial aspect of the digit, just distal to the metatarsal-phalangeal joint (see image below).
    ii. Slowly inject the anesthetic as the needle is advanced toward the volar/plantar side, without piercing the volar skin.
    iii. Slowly withdraw the needle and redirect it medially.
    iv. Advance the needle slowly from medial to lateral side while the anesthetic is injected (see middle image below)
    v. Withdraw the needle.
    vi. Make another injection over the already anesthetized skin at the lateral aspect of the digit, with the needle at 90 degrees, advancing it from the dorsal to ventral aspect, as was done medially (see right image below).

Figure 16-2 Medial Injection (left), Medial to Lateral Injection (center), Lateral Injection (right)



APPENDIX 4: MYCOLOGY SPECIMEN SAMPLING AND HANDLING

The aim of the subungual sample procedure described below is to collect a most proximal subungual sample of sufficient quantity that in a high percentage of samples result in single cultures of dermatophytes.

An instructive on-line video of the subungual sampling procedure described in detail below is available by clicking on the link, courtesy of the Center or Medical Mycology in collaboration with Dr. Sigurgeirsson.[30]

1. Wear sterile gloves when collecting nail samples. Bacteria and fungi living on the skin as normal flora (eg,*Candida parapsilosis)* are a significant source of contamination.

2. Use sterile instruments to trim nails and collect subungual debris. DO NOT use the same instruments to collect a nail sample from two different subjects before sterilizing them. Using sterile instruments will reduce the contamination rate.

3. Before you collect subungual debris, wash the nail and surrounding skin with soap and water and dry them with a clean towel. Next, wipe the nail and the surrounding skin well with a freshly opened, pre-moistened alcohol prep and allow the nail and skin to air dry.

4. If not already done so, clip the nail to within 1 mm of the hyponychium, the thickened stratum corneum of the epidermis, which lies under the free edge of the nail - discard the nail clippings.

5. Wipe the trimmed nail with freshly opened, pre-moistened alcohol prep again and allow to air dry.

6. Using a clean sterilized 1 or 2 mm serrated curette, remove the outermost distal subungual material and DISCARD IT.

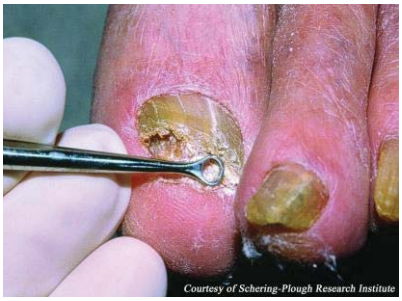
7. Thoroughly wipe the curette with freshly opened pre-moistened alcohol prep.

8. Cut back the nail plate, using sterile scissors and discard the nail portions.

9. Position the Dermapak® collection envelope under the nail.

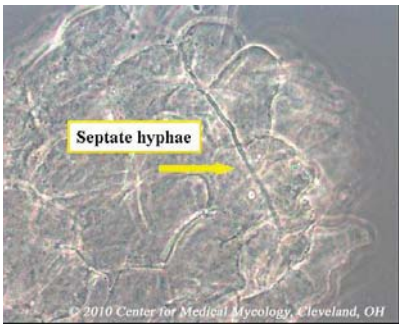
10. Use the clean sterilized curette to obtain an adequate and more proximal sample of subungual debris. (Fig.16-3). Label each sample to identify the great toenail (left or right) from which the subungual sample was collected.

Figure 16-3 Proximal Collection of Subungual Debris



11. Transfer a small portion of the debris to a drop of KOH on a glass slide - cover with a coverglass. (Make sure not to use all the subungual debris for KOH examination – there must be an adequate amount of sample to send to the laboratory for culture). Allow the slide to set for 10 minutes before microscopic examination. Examine the collected specimen under the microscope looking for the presence of septate hyphae indicative of dermatophytes. (Fig. 16-4)

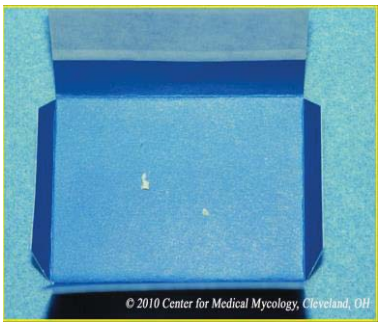
Figure 16-4 Septate Hyphae in Subungual Debris (10% KOH Microscopy Preparation)



12. If you must prepare a second KOH slide, be sure to clean the curette again with alcohol before sampling the debris again - KOH solution can be a source of bacterial contamination.

Common pitfalls in specimen collection such as excessive nail clippings and too little subungual material are depicted in Fig. 16-5.

Figure 16-5 Poor Specimen Collection: Inclusion of Large Nail Clipping and Insufficient Material



An example of an optimal and adequate amount of sample, provided it is obtained from the “right place”, collected in a DermaPak® is displayed in Fig. 16-6.

Figure 16-6 Optimal Subungual Specimen

A repeat subungual sample may be collected.

The collected subungual material is transferred from the DermaPak® to a sterile specimen container for shipment to the lab. The sample collected in the DermaPak® may be stored on-site under ambient conditions for up to 4 weeks.

APPENDIX 5: PRECANNULATION PROCEDURE

1. Soaking of the Nail Unit

Soak surgical gauze in sterile normal saline solution (0.9% sodium chloride solution, drug and preservative free) and wrap around the toenail. Apply after administration of the digital block and keep in situ for approx.10 minutes while adequate anesthesia is established.

2. Debridement

Debridement is not routinely carried out in this study. If necessary to facilitate access to the subungual space, it is recommended that debridement not be carried out more proximally than the most proximal margin of involved or dystrophic nail.

Nail debridement is permitted when the thickness or length of diseased or viable nail, in the opinion of the investigator, limits access to the subungual space with an atraumatic cannula or when it is difficult to locate an insertion point due to poor delineation of the nail bed from the nail plate.

If the nail becomes loose from the nail bed and secondary infection is likely to occur debridement of the loose nail plate may be instituted to prevent this complication.

3. Determine and Mark Optimal Insertion Point
    a. Measure involved nail width using a transparent ruler.
    b. Determine how many HTS-519 Inserts are required based on the topography of the infected nail area as described in Section 7.2.

Figure 16-7 Photo Ruler

    c. Indicate the equidistant insertion points on the self-adhesive photographic ruler guide as measured from the medial lateral fold. Apply the ruler to the tip of the toenail (Fig. 16-7) to mark the insertion points on the proximal skin fold with a surgical / surgeon's skin marker to facilitate consistent placement and create a reference point for digital photography. If a subungual channel is visible from a previous administration or treatment visit this channel may be used.

4. Subungual Depth Assessment

A 27G or 30G DermaSculpt® atraumatic microcannula is used to gauge the maximum depth of the subungual space. This is determined by advancing the microcannula slowly until a "stop" is felt by tactile feedback. Cannula size to be determined by the investigator.

    a. Place the blunt-tipped microcannula at each predetermined insertion point between nail plate and the nail bed/hyponychium.
    b. Advance slowly proximally and parallel to the nail bed until a clear "stop" is felt. Rotation of the cannula may be used to advance the cannula.

Repeat steps 4a and 4b for each insertion point.

APPENDIX 6: HTS-519 ADMINISTRATION PROCEDURE

The handling and placement procedure details the administration of one HTS-519 Insert in the subungual space at any insertion point. The administration procedure will be followed for each administration placement and consists of unpacking and a 2-step cannulation step.

1. Unpacking the needle-applicator (Fig. 16-8)

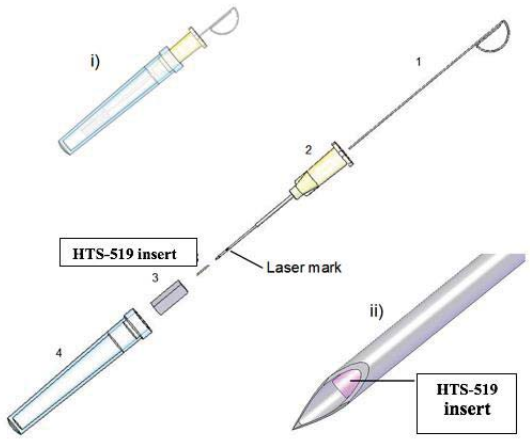
    a. Inspect each foil for package integrity. Do not use if there is evidence of damage to the foil pouch and / or sterility is in question.

    b. Hold the pouch in a horizontal position and open the pouch by separating the foil leaves located at the top of the pouch.

    c. Remove the sterile preloaded needle applicator from the pouch and place in the foot-end tray. Carefully, holding onto the plastic hub of the applicator (ii-2), remove the needle cover (ii-4), with the bevel up.

2. Inspection of the needle applicator and insert

Figure 16-8 HTS-519 Insert Assembled in Needle Applicator



i		Assembled Applicator
ii	1	Piston or Mandrel
	2	25G Needle inside 21G Housing with Hub
	3	Sponge Foam Needle Tip Protector
	4	Needle Cover

    a. The following needs to be successfully performed in order to verify that the insert did not dislodge from the assembled applicator needle and is present intact in the needle applicator. Conduct above the foot tray covered by black material, so that any HTS-519 Insert that falls out can be easily identified.

i. Inspect the needle cover for integrity and for inserts that may be visible in the needle cover (ii-4). Needle needs to be centered relative to the sponge foam needle protector (ii-3). Detach the cover in the horizontal plane. Once removed, invert the needle cover (ii-4) in your hand above the tray to ensure that the needle cover did not contain HTS-519 Insert material.
ii. Inspect and visualize the white colored tip of the HTS-519 Insert in the bevel of the needle. If not visible move the piston (ii-1) proximally until the insert is visible. Do not move any part of the HTS-519 Insert beyond the tip of the needle.

b. If the above is successfully completed place the needle applicator horizontally in the tray. Do not use if there is any doubt about the presence of an intact insert in the applicator. Do not push or touch the mandrel (ii-1) as it may release the HTS-519 Insert from the applicator.

3. Unpacking the Dermasculpt® microcannulas

Each Dermasculpt® cannula comes packaged together with a sharp needle as follows (30G atraumatic needle with 25G sharp needle; 27G and 25G microcannula with a 23G sharp needle; 22G cannula with a 20G sharp needle)

a. Inspect for package integrity. Do not use if there is evidence of damage to the foil pouch and / or sterility is in question.

b. Ensure that the correct gauge microcannula is unpacked and that the sharp beveled and larger needle is discarded unopened.

4. Cannulation and Insertion Technique

The cannulation and insertion technique is completed in a single sequence for each insertion point. For a right handed operator the insertion procedure is conducted from right to left, starting with the left toe, working from lateral to medial. This is followed by treating the right toe, administering treatment from medial to lateral.

A DermaSculpt® atraumatic 22G or 23G sized microcannula is used to create a subungual channel at each insertion point, large enough to accommodate a 25G needle applicator. Use of the smallest cannula size is preferred, but final determination of cannula gauge is made by the investigator.

a. Place the 22G or 23G blunt-tipped microcannula at each predetermined insertion point between nail plate and the nail bed.

b. Advance slowly proximally and parallel to the nail bed until a clear “stop” is felt. The stop may be just beyond the visible leading edge and close to the depth achieved with a 27G or 30G cannula.

c. Measure the insertion depth achieved by the cannula with a transparent ruler and record to use for marking the maximum insertion depth on the shaft of the needle applicator (ii-2).

d. Withdraw the cannula and place in the tray.

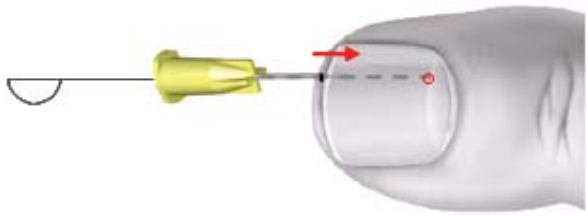
A 25 gauge Needle Applicator is used to deposit the 6 mm long biodegradable cylindrical pellet into the subungual channel immediately following microcannulation at each insertion point.

a. Mark the applicator needle (ii-2) with the desired insertion depth as determined in the preceding the 22G or 23G DermaSculpt® cannulation step. Note the position of the mark relative to the laser mark (Fig. 16-9), which is located approx. 6 mm from the tip of the needle applicator. If the mark has to be placed proximal to the laser mark anticipate that the HTS-519 Insert may project beyond the distal groove after administration.

b. With the insert visible in the needle invert the needle, so that the tip is pointing downwards and check that the HTS-519 Insert remains in situ. Conduct this action above the tray that is covered with a black material, so that any insert that falls out can be easily identified.

c. Visualize the subungual channel and place the needle at the channel entry point between the nail plate and the nail bed / hyponychium.

d. Holding the hub (ii-2) advance the applicator needle slowly into the already created subungual channel (gray dotted lines) underneath the nail plate preferably in the bevel-down direction. Do NOT angle upwards or downwards. The needle can NOT extend into the lunula and can NOT be advanced beyond the insertion depth mark (black in picture) on the needle shaft (ii-2) in order to avoid tissue trauma.



e. Withdraw the needle hub (ii-2) completely, while holding the piston steady to deposit the HTS-519 Insert into the subungual channel (gray dotted lines). Do NOT push the piston or mandrel (ii-1).

f. Remove the entire needle applicator slowly.

g. Check the position of the HTS-519 Insert visually and by transillumination. If the HTS-519 Insert, as anticipated from insert depth marking, projects beyond the distal groove, remove the part of the insert not covered by the nail plate manually.

h. Ensure that the HTS-519 Insert is no longer in the needle applicator by expressing the piston. Also note the collection of subungual debris in the tip of the bevel if any before disposing the applicator in a sharps container.

i. Check the insertion areas for local administration site reactions. Simple compression with a gauze or an ice pack may be applied to the insertion site for a short period if necessary.

j. Repeat the same procedure for each insertion

APPENDIX 7: ADMINISTRATION SITE ASSESSMENT SCALE

Subject Diary

Score	Assessment	Description
Burning/Stinging		
0	None	No burning/stinging
1	Mild	Slight warm; tingling sensation; not really bothersome
2	Moderate	Definite warm; tingling/stinging sensation that is somewhat bothersome
3	Severe	Hot; tingling/stinging sensation that has caused definite discomfort
Redness		
0	None	No redness present
1	Mild	Faintly detectable erythema; very lightly pink
2	Moderate	Dull red, clearly distinguishable
3	Severe	Deep/dark red
Oozing and Crusting		
0	None	Absent
1	Mild	Faint signs of oozing
2	Moderate	Definite oozing or crust with 5 or fewer sites per area
3	Severe	Marked and extensive oozing and crusting
Bruising		
0	None	Absent
1	Mild	Slight, barely perceptible
2	Moderate	1 or 2 bruises present, limited and discernible
3	Severe	Multiple bruises, diffuse and easily discernible
Itching		
0	None	No Pruritus
1	Mild	Occasional, slight itching, scratching
2	Moderate	Constant or intermittent itching/scratching that is not disturbing sleep
3	Severe	Bothersome itching/scratching that is disturbing sleep
Swelling		
0	None	No elevation
1	Mild	Barely perceptible elevation
2	Moderate	Clearly perceptible elevation but not extensive
3	Severe	Marked and extensive elevation

Investigator Assessment

Score	Assessment	Description
Bleeding		
0	None	None
1	Mild	Punctate bleeding not extending beyond distal groove - compression not needed
2	Moderate	Active bleeding extending beyond the distal groove and resolving within 5 minutes – may require compression
3	Severe	Active bleeding extending beyond the distal groove and lasting longer than 5 minutes – may require compression and bandage