STUDY PROTOCOL AND STATISTICAL ANALYSIS PLAN

Official title: Study to evaluate Celliant diabetic medical socks to INCREASE tissue oxygenation and INCIDENCE OF COMPLETE wound CLOSURE in diabetic foot wounds

NCT number: NCT04709419

IRB approved date: 01-03-23

STUDY TO EVALUATE CELLIANT DIABETIC MEDICAL SOCKS TO INCREASE TISSUE OXYGENATION AND INCIDENCE OF COMPLETE WOUND CLOSURE IN DIABETIC FOOT WOUNDS

PROTOCOL NUMBER:
HGX2020-01

SPONSOR:
HOLOGENIX, LLC
17383 SUNSET BOULEVARD, SUITE A420
PACIFIC PALISADES, CA 90272

ISSUE DATE:
NOVEMBER 8, 2022

SPONSOR SIGNATURE PAGE

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INVESTIGATOR SIGNATURE PAGE

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Site Name: _____

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Executive Summary

Title of Study	Study to Evaluate Celliant Diabetic Medical Socks to Increase Tissue Oxygenation and Incidence of Complete Wound Closure in Diabetic Foot Wounds
Protocol Number	HGX2020-01
Study Purpose	The purpose of this study is to demonstrate that the use of Celliant Socks increases tissue oxygenation (via oxygen saturation, StO_2) and incidence of wound closure in subjects with diabetic foot ulcers. This study will use hyperspectral imaging and wound assessment to measure these outcomes.
Study Design	This study is a prospective, multicenter, double-blind, 1:1 randomized clinical trial.
Enrollment	The study will enroll 254 evaluable subjects total, 127 per arm to meet the Primary Endpoint. Enrollment may continue up to twenty-five hundred (2500) evaluable subjects total to meet the Key Secondary Endpoint of complete wound closure.
Investigational Sites	UT Southwestern, Department of Plastic Surgery Parkland Hospital, Department of Plastic Surgery
Study Duration	Up to 28 weeks (including run-in and followup phase)
Efficacy Endpoints	Primary Endpoint: To compare the percent change from baseline in tissue oxygenation in Celliant and Control treated groups 12 weeks post initial application, as measured by hyperspectral imaging. Key Secondary Endpoint: To compare the incidence of complete wound closure of Celliant and Control treated groups 12 weeks post initial application. Other Secondary Endpoints: 1. Change in pain assessment as reported using the Visual Analog Scale (VAS) 2. 3-month follow-up to assess wound closure and reoccurrence 3. Accelerated wound closure as measured by changes in wound area, depth and volume. 4. Assessment of Compliance 5. Outcomes of Diabetic Foot Ulcer Scale – Short Form 6. Study related AE/SAEs
Safety Endpoints	The incidence of events reported
Study Groups	Subjects will be randomized 1:1 to either Active or Control medical sock for therapy. All participants will receive standard of care (SOC) for their wound therapy. This includes physician choice of selected dressings and offloading techniques. 1. Active Therapy: Celliant Sock 2. Control Therapy: Control Sock

Background and Rationale

There is a worldwide epidemic of diabetes. Foot ulcers are one of the most common complications in diabetic patients leading to amputation and hospitalization [1, 2]. Approximately one quarter of all hospital days for persons with diabetes are related to foot complications [1-3]. The majority of amputations (50%-83%) are in people with diabetes [4-7]. Foot ulcers are the most common underlying cause of amputation. It has been estimated that 15% of diabetics will have a lower extremity ulcer in their lifetime. In the United States there are approximately 120,000 non-traumatic lower extremity amputations performed each year [8, 9]. The direct cost of foot ulcers and amputations has been conservatively estimated to be approximately 1.6 billion dollars a year without consideration for physician fees, prosthetics, or rehabilitation costs [10].

Continuous supply of oxygen to the tissue through microcirculation is vital for the healing process and for resistance to infection [11]. Hypoxia of the wound primarily caused by vascular limitations is intensified by coincident conditions (e.g. infection, pain, anxiety and hyperthermia) and leads to poor healing outcomes [12, 13]. Sufficient oxygenation is especially important for cell proliferation, bacterial defense, angiogenesis, collagen synthesis and epithelialization [14]. From a diagnostic standpoint, measurements of wound oxygenation (transcutaneous O2 measurements or TCOM) are commonly used to guide treatment planning such as amputation decision [15-20]. In preventive applications, optimizing wound perfusion and providing supplemental O2 in the peri-operative period reduces the incidence of post-operative infections [21-23]. Hopf et al. [24] conducted a prospective study of 130 surgical patients to determine whether subcutaneous wound oxygen tension could predict the development of wound infection. Correction of wound pO2 (partial pressure of oxygen in the wound tissue) may, by itself, trigger some healing responses [25-33]. More importantly, approaches to correct wound pO2 favorably influence outcomes of other therapies such as responsiveness to growth factors and acceptance of grafts [25, 34, 35].

Infrared (IR) radiation is an electromagnetic spectrum with a wavelength of .70 to 100 μm. It has been observed that IR can stimulate cell proliferation, increase tissue regeneration in vitro, and increase the topical skin temperature [36-38]. Recent studies have been demonstrated that IR has been used for soothing effect and reducing pain of the wound site after standard medical wound treatments, even in clinical observations where wounds under exposure to IR decreased the wound healing times [39-42].

Summary of Findings from Previous Studies

The biological effects of infrared (IR) radiation (.70-100um) that have been observed in both in vitro and in vivo studies have launched IR as a promising modality for certain medical conditions (reviewed in [37]). Technological advances have provided new techniques for delivering IR radiation to the human body. Specialty lamps and saunas delivering pure mid-radiation (5-25 um; eliminating completely the near- and far-infrared bands) have become safe, attractive, and widely used devices to generate therapeutic effects. Additionally, studies have shown that IR-emitting apparel technology can also have a physiological effect on athletes during exercise [43].

For IR used as a therapeutic modality, the alternative terms “biogenetic radiation” and “biogenetic rays” have been widely used in the popular literature. IR wavelengths are too long to be perceived by the eyes, however, the body experiences its energy as a gentle radiant heat

which can penetrate over 1.5 inches (4 cm) beneath the skin [42]. In the IR radiation bands, only mid-IR transfers energy purely in the form of heat, which can be perceived by the thermoreceptors in human skin as radiant heat [44]. Not only is mid-IR absorbed by the human body, but it is also emitted by the body in the form of blackbody radiation (3 to 15 microns, with an output peak at 9.4 micron). IR energy is sufficient to increase the energy of rotational and vibrational modes of motion in bonds in many biological molecules (including the water molecules which predominate in tissue). Resulting epidermal temperature is higher when the skin is irradiated with IR than if similar energy densities delivered using shorter wavelengths are used. The prolonged erythemal response from IR exposure has been proposed to be due to increased epidermal temperatures associated with it, but levels of IR that do not produce any detectable skin heating can also have biological effects.

A possible explanation for these biological effects that occur without any detectable temperature change in the tissue is that the IR radiation is selectively absorbed by water molecules that are associated with ion channels in the membranes within the cell (both plasma membrane and organelle membranes)[42]. Water is by far the most important chromophore in the IR spectral range, both because of its high absorption coefficient, and its high abundance (70%) in tissue. These water molecules are absorbed on the surface of the so-called “heat-sensitive TRP (transient receptor potential) ion channels.” When these “structured” water molecules absorb IR, they change their pH by a small but significant amount, which affects the conformation of critical amino acids in the protein components of the ion channel. These TRP channels are calcium ion channels and an increase in cellular calcium can activate several important signaling pathways and even activate transcription factors. One specific TRP channel (TRPV3) has been linked to epithelial wound healing [45] through inducing release of nitric oxide (NO) from keratinocytes [46, 47]. Another, TRPC6, has been linked to regulate fibroblast-myofibroblast transdifferentiation and deletion of this protein results in delayed dermal wound healing in rodents [48].

As such, fibers embedded with materials and woven into fabrics are being used as garments, bedding and wraps to generate IR radiation, which is converted from the energy supplied by body heat, and provide health benefits from its effects. In a similar manner, discs of FIR emitting ceramic material have been applied to the human body to produce a beneficial effect. For instance, a blanket containing discs has been reported to improve quality of sleep [3] and single discs were applied to the breasts of women who encountered difficulty in producing sufficient breast milk during lactation [49]. Gloves have been made out of FIR-emitting fabrics and there have been reports that these gloves can be used to treat arthritis of the hands and Raynaud’s syndrome [50].

Celliant® Diabetic Medical Socks are intended to increase tissue oxygenation thereby increasing blood flow and local circulation leading to improved wound healing outcomes and possible pain reduction. Celliant® fibers, yarns and fabrics, like other products described above, are designed to re-emit infrared light derived from heat energy generated by the wearer back into the wearer’s skin. In 7 preliminary studies (including 3 published), application of the Celliant® Diabetic Medical Socks (or other Celliant®-containing systems) results in improved blood flow in healthy and diabetic subjects as measured by oxygen availability in skin and superficial tissue by transcutaneous partial pressure of oxygen (tcPO2) and reduced pain. A summary of these studies follows:

Published:

2019 tcPO2 increased by up to 10% and increase in grip strength of 12%, n=24, Hamblin and Gordon (shirts)[51]

2018 tcPO2 increased by 8.4% after 90 minutes, n=153, Hamblin and Gordon (shirts)[52]

2009 Reduction in foot pain in diabetic/neuropathic subjects, n=55 Gordan (socks)[53]

Papers:

2012 tcPO2 increase of 17% in healthy subjects, n=100, Shaojing (socks)[54]

2012 tcPO2 increase of 7% in healthy subjects, n=51, Gordon (shirts)[55]

2005 tcPO2 increase of 12%-24% in healthy subjects, n=13, McClue (gloves and socks)[56]

2003 tcPO2 increase of 12% in hands and 8% in feet of diabetic subjects, n=20 (gloves and socks) [57]

In all of these studies (n=7), measurements obtained while wearing Celliant® -either shirts, socks and/or gloves- were compared to measurements obtained while wearing control products identical in construction except lacking the center-loaded, thermo-reactive ceramic particles in the PET yarn. Furthermore, no significant differences in blood pressure, heart rate, or skin temperature were observed in measurements obtained while subjects wore control or Celliant® products.

Device Description

Celliant® Diabetic Medical Sock

Celliant® Diabetic Medical Socks (“Celliant Socks”) are intended to provide infrared radiation (IR) to increase tissue oxygen thereby increasing blood flow and circulation in the affected area leading to an effect on wound closure outcomes and possible pain reduction. Hologenix intends to produce Celliant fibers and yarns that are woven or knitted in combination with other materials, such as polyester, cotton and wool into hosiery (socks) and wraps or fabrics that are then used in garments such as shirts and pants; bed linens such as sheets, pillows and mattress pads; and clothing accessories such as sweatbands and shoe liners as well as seat covers, home and office furnishings.

Celliant® fibers are comprised of a proprietary blend of infrared (IR) emitting ceramic materials mixed with polyethylene terephthalate (PET). The proprietary blend consists of the following components: alumina oxide; silicon dioxide; and titanium dioxide. The Celliant fibers are woven or knitted into yarn and used to manufacture the Celliant Sock. The Celliant Sock content will be 82% Celliant polyester, 13% Nylon and 5% Spandex. Celliant fibers absorb energy (heat) generated by the wearer’s body by radiation, convection and conduction, and also from the environment, and re-emit the energy as infrared radiation back into the wearer’s body. Testing has shown that fabrics containing Celliant fibers have a higher emissivity (.25 mW/cm^2 at 35 Celsius) in the 2.5–20 μm wavelength range compared to similar PET-only fabrics [58, 59]. Moreover, several preliminary studies have shown that the application of Celliant Socks (or other Celliant-containing systems) results in improved blood flow in healthy and diabetic

subjects as discussed above. Therefore, it is believed that the increased infrared radiation emission from the Celliant garments is associated with this positive physiological impact of enhanced blood flow and circulation and increased tissue oxygenation (TcPO2), which may have a significant impact in wound healing.

Control Sock

The Control Sock will be visually identical to the Active Celliant® Sock. The sock will be made of identical materials without the Celliant® components. The Control sock will be 82% standard polyester, 13% Nylon, and 5% Spandex.

Regulatory Status

Overview

The Celliant® Diabetic Medical Sock is an investigational device and is not approved in the United States to provide infrared radiation (IR) to increase tissue oxygenation or to promote complete wound closure by increasing blood flow and circulation or to reduce pain in diabetic patients with foot ulcers.

However, the US FDA has determined the Celliant Products to be medical devices as defined in section 201(h) of the Act and are also general wellness products. The Celliant Products are medical devices because they are intended to affect the structure or function of the body of man by temporarily promoting increased local blood flow at the site of application in healthy individuals.

Proposed Indications for Use

The Celliant® Diabetic Medical Sock is intended to provide infrared radiation (IR) to increase tissue oxygenation (via oxygen saturation, S_tO_2) as measured by hyperspectral imaging (HSI) resulting in increased blood flow and local circulation in the affected area which may influence wound closure. In a clinical study, adult subjects (22 years and older) with diabetic foot ulcers showed an increase in tissue oxygenation and blood flow and local circulation with positive wound closure outcomes compared to subjects wearing non-Celliant® Diabetic Medical Socks. The Celliant® Diabetic Medical Sock may also reduce/improve pain

Benefits and Risks

Potential Benefits to the Subjects

There is no promise or guarantee that this product will help heal the subject's wound, but the potential benefit of wearing Celliant® Diabetic Medical Socks is that adults with diabetic foot ulcers may experience improved tissue oxygenation and improved wound closure outcomes and possible pain reduction compared to adults not wearing Celliant Socks. The clinical results may prove this hypothesis. Moreover, the potential benefits may extend beyond the immediate closure of chronic foot ulcers. The effects of IR on the tissue of the feet will tend to prevent any further lesions or ulcers developing. By increasing the tissue oxygenation, reducing inflammation, and stimulating collagen formation in the dermis, the skin may be "strengthened" and small traumas which would previously have progressed into a chronic wound may be healed up without even being noticed.

Additionally, the beneficial effect of Celliant ® Diabetic Medical Socks in wound closure can be derived from what is known about photobiomodulation (PBM). PBM involves the use of red light (600-700nm) or near-infrared light (760-1070nm) applied periodically to the tissue. By definition PBM is delivered using a powered light source such as a laser or a light-emitting diode (LED) array. There have been several successful clinical studies using PBM for healing of chronic wounds including diabetic foot ulcers. The difference between PBM and Celliant is the wavelength and the fact that the IR emitted from a Celliant ® Diabetic Medical Sock is being consistently delivered into the feet as long as the socks are worn.

Potential Risk

Participation in this clinical investigation presents low risks to subjects. These products have passed biocompatibility testing and there are no known risks of wearing Celliant® Diabetic Medical Socks. There have never been any reports of allergies developing to the fabric (PET) or to the minerals (Al_2O_3, SiO_2, TiO_2). Some general risks associated with any new wound dressing or skin-contact devices are listed in the table below.

Risks	Disorders/Conditions
Skin and Subcutaneous Tissue Reaction/Allergy	• Skin rash, irritation, blistering • Pruritus/itching • Skin excoriation/breakdown • Skin scarring if significant skin irritation were to occur • Skin hyper/hypo-pigmentation at and/or around dressing application area • Erythema/redness, edema, inflammation, or swelling at and/or around dressing application area
Mild Pain or Discomfort	• Tenderness/minor ache at and/or around dressing application area • Perspiration associated with wearing sock • Decreased sleep or sleep quality • Paresthesia (numbness, tingling, prickling, creeping sensation)
Other	• Risks to privacy • Loss of data confidentiality

Protection Against Risks

Protected health information (PHI) of subjects in clinical investigations are kept as confidential as possible in accordance with the Health Insurance Portability and Accountability Act of 1996 (HIPAA). However, confidentiality cannot be assured. The table below lists the entities granted access to protected health information (PHI).

Entity	Reason for Access
Sponsor's Clinical Representative (Monitor)	Assess data for accuracy and completeness
Institutional Review Board (IRB)	Ensure protection of research subjects
Regulatory Authorities (e.g. Food and Drug Administration	Audit clinical trial for Subject protection and data integrity
Investigator/Site Staff	Collection and assessment of data for accuracy and completeness

Alternatives to Participation

Subjects are not required to participate in this research study. As an alternative to study participation, subjects will receive wound care treatment per physician discretion.

1. Trial Objectives

The objective of this trial is to evaluate percent change from baseline in tissue oxygenation (via oxygen saturation, S_tO_2) in Celliant and Control treated groups 12 weeks post initial application, as measured by hyperspectral imaging in 254 evaluable patients total when a Celliant® Diabetic Medical Sock (n=127) is worn over the patients dressing as compared to wearing a Control Medical Sock (n=127). Enrollment may include up to 2500 total subjects to meet the Key Secondary Endpoint. The Key Secondary Endpoint is to compare the incidence of complete wound closure of Celliant and Control treated groups 12 weeks post initial application*. Other secondary outcomes include wound evaluation at 3 months post complete wound closure, accelerated wound closure as measured by changes in area, depth and volume as well as treatment compliance and assessment of pain reduction.

*if the study wound is deemed closed on the week 12 visit, the patient will continue using the sock and the wound will be evaluated on week 14 to confirm healed state.

2. Selection and Withdrawal of Subjects

Inclusion Criteria:

- Diagnosis of diabetes mellitus
- Subject is willing and able to wear a sports-style tube sock at least 22 hours a day.
- Ankle Brachial Index (ABI) ≥0.5 (bedside ABI is acceptable for screening purposes as the formal imaging ABI may not be resulted prior to surgery) or toe pressure of ≥30mmHg
- One or more diabetic foot ulcers (only one will be treated) that are located in the ankle area or below that has persisted a minimum of 30 days prior to the Screening visit
- Diabetic Foot Ulcers ≥0.5cm^2 and ≤16cm^2
- Ulcer grade I or II, Stage A, I or II Stage B, according to University of Texas Wound Classification System
- 22 years of age or older

Exclusion Criteria:

- Subject has untreated osteomyelitis
- Ulcers within 5cm of target ulcer or connected by fistulas
- Ulcer has decreased by 30% or more at the end of the run-in period
- Subject has active untreated cellulitis
- Subject has active untreated charcot
- Major immunodeficiency including HIV.
- Is pregnant or plans to become pregnant
- Is nursing or actively lactating

- Developmental disability/significant psychological disorder that in the opinion of the investigator could impair the subject's ability to provide informed consent, participate in the study protocol or record study measures, including untreated schizophrenia, bipolar disorder and psychiatric hospitalization within the last 2 years.
- Active alcohol or substance abuse in the opinion of the investigator that could impair the subject's ability to provide informed consent, participate in the study protocol or record study materials

Withdrawal of Subjects

Subjects may withdraw from the study at any time at their own request, or they may be withdrawn at any time at the discretion of the PI for safety, behavioral, or administrative reasons. If a subject does not return for a scheduled visit, every effort should be made to contact the subject, per institutional protocol. In any circumstance, every effort should be made to document subject outcome. The Investigator should inquire about the reason for withdrawal, request that the subject return for a final visit, if applicable, and follow up with the subject regarding any unresolved AE's.

Any related SAE occurring within 14 days following subject discontinuation must be reported to Hologenix and be followed up until stabilization or resolution. If the Subject withdraws from the study, and also withdraws consent for disclosure of future information, no further study evaluations will be performed, and no additional data will be collected. The PI may retain and continue to use any data collected before such withdrawal of consent.

The Investigator may choose to withdraw their own participation or discontinue a Subject from study with or without their consent for any of the following:

- Adverse Events
- Non-compliance
- Safety
- Complications
- Unforeseen events

A patient who experiences the noted changes, relative to previous measures, in the following parameters will be terminated early from study participation

- An increase in ulcer size by 50% or more prior to debridement from previous visit.
- An increase in pain rating by 50% or more from previous visit.
- Presence of infection as determined by 2 more bacterial infection indicators that requires treatment outside the study scope
- Presence of osteomyelitis that requires treatment outside the study scope.
- Need for wound management outside the study scope

- Or for any reason that may, in the opinion of the Investigator, affect negatively the wellbeing of the subject.

If for any reason the subject is withdrawn from the clinical investigation, the Investigator will inform the subject accordingly.

3. Clinical Investigation Plan

Description of Trial Design

This study is designed as a prospective, multi-center, double-blind, 1:1 randomized clinical trial to assess the clinical safety and efficacy of Celliant® Diabetic Medical Socks to increase tissue oxygenation and complete wound closure.

Investigational Sites

University of Texas Southwestern, Department of Plastic Surgery

PI: Lawrence A. Lavery, DPM, MPH
Location: Dallas, TX

Parkland Hospital, Department of Plastic Surgery

PI: Peter Crisologo, DPM
Location: Dallas, TX

Duration of Subject Participation

- Screening: Within 7 days Run-In initiation
- Run in Evaluation: 2 weeks of evaluation
- Treatment period: Up to 12 weeks
- Follow-up period: Up to 3 months post wound closure event
- Total duration of subject participation: Up to 28 weeks

Screening and Enrollment

Patients who present to the investigator's institution (through clinic admission, direct transfer from another facility, or through the emergency room) may be recruited to participate in the study. No direct marketing for subject recruitment will be done.

Patients approached for study participation will be at least 22 years of age at the time of consent, will undergo wound assessment, and meet all eligibility requirements. Those meeting eligibility criteria for the study will have the study explained to them by the Investigator. An Informed Consent Form will be provided to sign according to Section 11 prior to undergoing any study procedures. Patients will be encouraged to ask questions of the investigators. It will be made clear to the patient that not participating in the study will in no way influence the treatment plan or the relationship with the physician.

Wound Selection

Only one wound per subject will be included in the study. Subjects with multiple wounds will have each wound measured for volume. The wound with the largest volume meeting all eligibility requirements will be chosen for inclusion in the study. The etiology of the chosen wound, and if it is a new or recurring, will be documented.

Randomization and Blinding Procedure

Prior to study initiation, Active and Control samples will be prepared in sealed and labeled as A or B, also randomization envelopes will be provided to the research staff and used to obtain randomization assignment. Within each randomization envelope, there is an assignment of A or B. After run-in evaluation, the subjects wound will be debrided and dressed with SOC. At the end of the procedure, subjects who continue to meet all inclusion and no exclusion criteria will be randomized in a 1:1 ratio to be treated with either Control or Celliant therapies, stratified by wound size (greater than size of 4 cm^2) and study center. Study staff will use the randomization number labels and sock assignment of socks A or B contained in the envelope. The number will become the subject ID. The research staff will note the randomization number and the treatment assignment letter (A or B) on the CRF. The key, indicating therapy assignments (Active or Control), will only be available to staff not directly involved in the study until after the subject has completed study activities.

Visit Schedule

Study procedures for each phase of study are outlined below. In a later section, details on each study procedure are described, including equipment designation.

Screening

1. Explain purpose and nature of the study and obtain signature on the informed consent document.
2. Screen the subject against protocol inclusion and exclusion criteria, including all pertinent tests.
    a. ABI and/toe pressure
    b. Ulcer History, measurement, and classification
    c. Urine Pregnancy test (if indicated)

Baseline (may be done as same day as screening procedures)

1. Obtain general medical history and demographic information and social history
2. Complete a physical examination, body weight, height, and vital signs, including measurement of resting heart rate, respiratory rate, and blood pressure while seated.
3. Select the target ulcer
4. Obtain complete history pertinent to DFU disease including duration of the target ulcer,

previous and current treatment.

5. Perform debridement if indicated. Debridement is to be performed using curette, scissors, scalpel, or forceps.
6. Collect 4 tissue specimens. Two in RNA later and two in DNA/RNA shield.
7. Perform standardized photography and measurement of the study wound (eKare or available camera with ruler). Assess the post-debridement ulcer area (cm^2), perimeter (cm), and greatest depth (cm) using the inSight device (eKare, Fairfax, VA). These values are the baseline measurements for assessing accelerated wound closure at the two-week run-in visit.
8. Perform baseline SPP (note – this procedure can be performed at any visit from baseline up to therapy initiation but before therapy is started).
9. Perform baseline tcpO2 (note – this procedure does not need to be performed at baseline visit and can be performed at any visit through Run-In week 2/Therapy initiation visit).
10. Perform baseline hyperspectral imaging assessment on the dorsal and plantar aspects of the foot.
11. Perform neuropathy and pain assessment.
12. Obtain results from SOC hematology, blood chemistry, etc. that were not covered in inclusion/exclusion studies.
13. Subjects who do not meet eligibility and fail to qualify to enter the Run-in will be deemed Screen failures and discharged, noting the reasons for disqualification.
14. Collect all relevant concomitant medication (antibiotics, antifungals and other anti-infective therapies)
15. Wounds will be dressed SOC and offloaded per physician discretion.
16. Subjects who qualify to enter the run-in will be instructed to refrain from using excluded medications and return to the office for Run-in Visit 2 within 7 (±4) days.

<u>Run-In Procedures/Therapy Initiation</u>

Week 1 Run-in:

1. Assess target ulcer.
2. Perform debridement if indicated. Debridement is to be performed using a curette, scissors, scalpel, or forceps.
3. Perform standardized photography of the study wound.
4. Assess the post-debridement ulcer area (cm^2), perimeter (cm), and greatest depth (cm) using the inSight device.
5. If wound size (ulcer area) increases/decreases by 30% or more, patient will be deemed a screen failure and removed from the study.
6. If wound size (ulcer area) decreases by less than 30%, patient wound will be treated with standard of care and return for run-in Week 2 Run-In within 7 (±4) days.

7. Perform hyperspectral imaging assessment on the dorsal and plantar aspects of the foot.
8. Perform pain assessment.
9. Collect all relevant concomitant medication (antibiotics, antifungals and other anti-infective therapies)
10. Wounds will be dressed SOC and offloaded per physician discretion.

Week 2 Run-in/Therapy Initiation:

1. Assess target ulcer
2. Perform debridement if indicated. Debridement is to be performed using curette, scissors, scalpel, or forceps.
3. Perform standardized photography of the study wound.
4. Assess the post-debridement ulcer area (cm^2), perimeter (cm), and greatest depth (cm) using the inSight device.
5. If wound size (ulcer area) increases/decreases by 30% or more, patient will be deemed a screen failure and removed from the study.
6. If wound size (ulcer area) increases/decreases by less than 30%, patient wound will enter the study and randomized to either the Celliant Active or Control treatment arm.
7. Perform hyperspectral imaging of the dorsal and plantar aspects of the foot
8. Perform pain assessment.
9. Collect all relevant concomitant medication
10. Wound will be dressed with SOC and either Celliant Active or Control Sock applied.
11. Wound will be offloaded.
12. Provide subject with any additional medical socks needed until the next visit. Instruct the subject to change the socks if they become wet, soiled, or bloody and under any conditions dictated by PI instructions.

<u>Therapy/Treatment Phase</u>

Study Visit 1-11:

1. Document study compliance information from the subject
    a. Amount of time the sock was worn throughout the week
    b. Was it worn during the day/night?
    c. When/why the sock was changed.
    d. Has the sock been on in the 3 hours leading up to the appointment?
2. Remove sock and dressing and immediately perform hyperspectral imaging of the dorsal and plantar aspects of the foot if wound is active or closed.
3. Assess target ulcer (if wound has closed, document as such, perform steps 7 and 8 and

skip to Study Visit – Wound Closed).

4. Debridement is to be performed, if indicated, using curette, scissors, scalpel, or forceps. If debridement is necessary at weekly visits, all therapy must be removed and replaced post-debridement.
5. Collect 4 tissue specimens at week 3 and at week 6. Two in RNA later and two in DNA/RNA shield.
6. Perform standardized photography of the study wound.
7. Assess the ulcer area (cm^2), perimeter (cm), and greatest depth (cm) using the inSight device - if the wound is deemed closed by the physician, skip to EOS visit.
8. Perform SPP and tcPO2 measurements at Visit 4 (maybe be taken instead at visit 5 or 6 if subject has not been wearing their treatment sock leading up to the visit or misses this visit).
9. Collect all relevant concomitant medication
10. Perform pain assessment.
11. Redress the wound with standard of care (if wound is still active) and apply Celliant Active or Control Sock and apply offloading (if indicated).
12. Wound will be offloaded.
13. Provide subject with any additional medical socks needed until the next visit. Instruct the subject to change the socks if they become wet, soiled, or bloody and under any conditions dictated by PI instructions. Instruct the subject that they must wear the sock at all times and that including the 3 hours leading up to the next study visit.

Study Visit - Wound Closed

1. The study wound is defined as closed when there is complete skin re-epithelialization without drainage or dressing requirements confirmed by the evaluating physician at 2 consecutive study visits 2 weeks apart.
2. At this visit the subject will perform the EOS evaluation and enter the follow-up phase of the study.

Study Visit 12/EOS

1. If the study wound closes prior to the 12-week study mark, subjects will perform EOS visit at the time of wound closure. See Study Visit - Wound Closed.
2. A subject whose wound does not close completely by week 12 will exited from the study after the week 12 wound evaluation.
3. Document study compliance information from the subject
    a. Amount of time the sock was worn throughout the week
    b. Was it worn during the day/night?
    c. When/why the sock was changed.

d. Has the sock been on in the 3 hours leading up to the appointment?

4. Remove sock and dressing and immediately perform hyperspectral imaging of the dorsal and plantar aspects of the foot if wound is active or closed.
5. Assess target ulcer (if wound has closed, document as such, perform steps 7-12 and skip to Study Visit – Wound Closed).
6. If the subject still has a large enough wound, collect 4 tissue samples. Two in RNA later and two in DNA/RNA shield.
7. Perform standardized photography of the study wound.
8. Assess the ulcer area (cm^2), perimeter (cm), and greatest depth (cm) using the inSight device.
9. Perform SPP if wound is active or closed.
10. Perform tcpO2 if wound is active or closed.
11. Perform pain assessment
12. Ask the subject if they thought they were in the active or control group
13. Administer DFS-SF
14. Collect all relevant concomitant medication
15. Redress the wound per physician-directed standard of care.

Study Visit Follow up

1. If the subject's wound closes within the 12-week treatment window, they will enter the follow up phase of the study.
2. At 3 months post closure, wound evaluation will be performed as outlined below. This follow-up visit will be in person. In the event that an in-person visit is not possible, a telehealth visit may be performed and documented as such.
    a. Assess the study wound to determine that it remained closed
    b. If the study wound has reoccurred
        i. record the date of reoccurrence
        ii. measure the size of dehiscence or re-ulceration
        iii. exit the subject from the follow up phase
    c. Assess the study foot for new sites of ulceration
        i. record the date of reoccurrence
        ii. measure the size of dehiscence or re-ulceration
        iii. exit the subject from the follow up phase

Detailed Study Operations

Medical Status/History

- *The New York Heart Association (NYHA) Functional Classification System:* This system will be used to classify stage of heart failure. The New York Heart Association (NYHA) functional classification system is used to classify the stage of heart failure from Class I to IV (as shown in the table below). This system relates symptoms to everyday activities and the patient's quality of life.

Class	Patient Symptoms
Class I (Mild)	No limitation of physical activity. Ordinary physical activity does not cause undue fatigue, palpitation, or dyspnea (shortness of breath).
Class II (Mild)	Slight limitation of physical activity. Comfortable at rest, but ordinary physical activity results in fatigue, palpitation, or dyspnea.
Class III (Moderate)	Marked limitation of physical activity. Comfortable at rest, but less than ordinary activity causes fatigue, palpitation, or dyspnea.
Class IV (Severe	Unable to carry out any physical activity without discomfort. Symptoms of cardiac insufficiency at rest. If any physical activity is undertaken, discomfort is increased.

- *Body Mass Index (BMI):* BMI is calculated as the ratio of body weight in kilograms (kg) to height in meters squared (m^2).

- *Medical History:* The following information will be collected from each subject

  - previous ulcer history
  - amputation history
  - lower extremity bypass
  - lower extremity angioplasty
  - coronary artery bypass surgery
  - cardiac angioplasty
  - arthritis
  - liver disease
  - osteoporosis
  - malignancy
  - bone tumors
  - skin pathology
  - vital signs (weight, height, HR, BP)
  - diabetes history
    - duration
    - type
    - medication
  - date and type of surgery
  - vascular status
  - neuropathy

Clinical Lab Values

- BUN (blood urea nitrogen)
- Creatinine
- Estimated GFR (glomular filtration rate)
- Hemoglobin A1c
- Blood Glucose
- Prealbumin (transthyretin)
- Albumin
- CRP (C-reactive protein)

- ESR (Erythrocyte Sedimentation Rate)
- White blood cell count
- RBC
- Platelet Count
- Hemoglobin
- Hematocrit
- RDW
- Pregnancy test if subject is child bearing potential

Concomitant Medications

All antibiotics, antifungals and other anti-infective therapies engaged in by the subject will be recorded at screening visits and follow up visits

Social Factors

The following social factors will be recorded:

- marital status
- years of education
- occupation
- tobacco use history: number of years of smoking, current or previous smoker, use of chewing tobacco, average daily number of cigarettes for current smokers
- alcohol and drug use history: current and/or previous history, types of alcohol/drugs consumed in the past and/or present; frequency and amount of alcohol/drugs consumed in the past and/or present

Demographic Variables

The following demographics will be recorded for each subject:

- Gender: male or female.
- Age
- Language Spoken: English, Spanish, English and Spanish, other(specify)
- Race: Caucasian, African American, American Indian, Asian/Pacific Islander.
- Ethnicity: Hispanic or not

Wound History and Assessment

- *Wound History and Evaluation:* We will use the University of Texas Ulcer Classification to document ulcer severity. Our group developed and validated this classification system. We will enroll subjects with UT ulcer classification IA, IIA, IB, and IIB ulcers. These are ulcers without penetration to tendon, capsule or bone and without PAD. This classification system is a 4 x 4 matrix that includes the depth of the ulcer or the ulcer Grade: (0) pre/post-ulcerative site, (1) full thickness, (2) extends to tendon or capsule, (3) extends to bone and the Stage of the ulcer that identifies if there is infection and/or PAD: (A.) No PAD or infection, (B.) infection, (C.) PAD (D.) infection and PAD.

- *Wound Measurements and Debridement:* Debridement will be performed per physician discretion using a scalpel or bone curette to remove all surrounding callus, wound debris, necrotic tissue, fibrin, eschar and non-viable tissue to create a bleeding wound bed. We will objectively measure ulcer debridement and changes in wound size using changes in wound volume and area from wound measurements with the inSight digital 3D measurement device (eKare, Fairfax, VA). Digital photos will be taken after debridement. We will take post-debridement measurements of wound volume and area. Gardner et al reported that volume measurements with a digital image evaluation system was reliable.

- *Safety Measures:* Each of the following measures will be rated on the following 5-point Likert scale, as well as a descriptive evaluation being recorded, as applicable, for the study wound: 1) absence or 2) presence of the following:

    - Erythema
    - Discharge/drainage
    - Malodor
    - Tissue necrosis

Vascular Assessment

We will assess perfusion/tissue oxygenation with three approaches; ABI (bedside may be done for screening if formal ABIs have not yet been performed/resulted), Skin Perfusion Pressure, and Hyperspectral Imaging. We will evaluate Skin Perfusion Pressure (SPP) and Transcutaneous Oxygen (tcpO2) measurements using the 510(k) cleared PeriFlux 6000 Combined system, K131253 (Perimed Inc, Las Vegas, NV) on the dorsal and plantar angiosomes of the affected foot and obtain toe pressures and waveforms. Hyperspectral imaging using the 510(k)-cleared Kent SnapshotNIR Camera, K113507 (Calgary, AB, Canada) will be performed to evaluate the periwound tissue surrounding the ulcer and the angiosomes on the dorsal and plantar aspects of the foot.

Neurological Assessment

Semmes-Weinstein monofilament evaluation will be performed. We will assess light touch and pressure sensation at five sites on the study foot using a 10-gram monofilaments. We will perform Vibration Perception Threshold (VPT); a device that uses vibration to test how well your feet can detect gentle pressure.

Degree of Pain Rating

The subject will record his or her current degree/level of pain for the study wound using the

following 10cm (100mm) long Visual Analog Scale (VAS) labeled from '0': no pain to '10': worst pain imaginable

No pain 0_____10 Worst Pain Imaginable

The VAS is the most commonly used scale for assessing pain [60]. The VAS is a simple scale that consists of a 10cm line anchored at one end by a label that indicates total absence of the measure being evaluated and at the other end by a label that indicates the worse imaginable presence of the measure being evaluated. The subject marks on the line the spot for the intensity of the measure, which is then measured using a ruler (in mm units).

Infection Evaluation

The following clinical indicators of significant bacterial infection will be recorded as present or absent for the study wound. 'Significant Bacterial Infection Clinical Indicators' are defined in this study as those presenting as excessive beyond which would typically be expected given the clinical condition and nature of the wound type.

- local heat
- spreading redness
- swelling
- increased purulent exudate (draining pus)
- odor
- cellulitis
- loss of function
- fevers
- chills

Sock Use Instructions

The subjects will be provided with 10 pairs of socks on the initial visit and instructed that the socks may be washed as normal laundry as needed (bleach or no bleach). Subjects will be supplied additional pairs as needed on a case by case basis. The sock change frequency will be dictated by the physician; however, the subject will be instructed to change the sock as with any other sock if it becomes wet, soiled, or bloody.

Patient Reported Outcomes

The Diabetic Foot Ulcer Scale – Short Form (DFS-SF) (Appendix B) will be used to collect patient reported outcomes in this trial. The DFS questionnaire is designed to assess the impact of foot ulcers and their treatment on quality of life in people with diabetes.

Excluded Medications/Treatments

During the study procedures, the study subjects will refrain from using hyperbaric oxygen therapy (HBOT) and any cellular or tissue-based products (RegranX etc).

Standard of Care

- All study wounds will be debrided per physician discretion and documented.
- Wound Dressing: Two wound care treatments will be available, depending on wound status and physician discretion
    - **IODOSORB Gel** is a sterile antimicrobial dressing formulation of Cadexomer Iodine. When applied to the wound, IODOSORB absorbs fluids, removing exudate, slough and debris and forming a gel over the wound surface. As the gel absorbs exudate, iodine is released, killing bacteria and changing color as the iodine is used up. https://www.smith-nephew.com/professional/products/advanced-wound-management/iodosorb--iodoflex/iodosorb-gel/
    - **Hydrofera Blue READY Foam Dressing** protects wounds against infection with broad-spectrum antibacterial properties and helps with healing and managing chronic, acute and traumatic wounds. This ready-to-use, non-cytotoxic dressing assists with managing moisture, creating an ideal healing environment that does not draw out the process and helps control exudate. https://hydrofera.com
    - **AQUACEL EXTRA Hydrofiber Dressing** is composed of two layers of Hydrofiber technology stitched together. Specifically suited to help manage moderate to highly exuding wounds. https://www.convatec.com/products/pc-wound-leg-ulcers/aquacel-extra-hydrofiber-dressing
- Offloading: All subjects who can be offloaded will wear the DH OffLoading Walker. This is a removable offloading boot that reduces shear force by stabilizing the foot and ankle at 90°. https://www.ossur.com/injury-solutions/products/foot-and-ankle/walkers/dh-offloading-walker. If patients cannot be offloaded as defined above, a method of offloading will be used and documented, both type and reason why the Walker was not used.

Delegation of Responsibilities

Task	Responsibility		
	PI	Nurse	CRC
Determine eligibility of subjects	X		
Obtain informed consent	X	X	X
Randomization	X	X	X
Medical history	X	X	X
Dressing removal	X	X	X
Wound assessment of infection and closure	X		
Tissue Collection	X	X	X
Wound measurements	X	X	X
Study withdrawal assessment	X		
VPT	X	X	X
10-gram monofilament test	X	X	X
Hyperspectral imaging	X	X	X
Skin perfusion pressure measurements	X	X	X
Transcutaneous Oxygen measurements	X	X	X
Pain assessment	X	X	X
Concomitant medications	X	X	X
Subject compliance assessment and review	X	X	X
Administer patient reported outcomes	X	X	X
Dressing Application	X		
Study Device Application	X	X	X
Offloading	X		
CRF completion/corrections	X	X	X
CRF/eCRF signature	X		
SAE/AE assessments	X		
Update and maintain study documents	X	X	X
SAE reporting and data clarifications	X	X	X

Schedule of Events Table

The table below shows the schedule of planned events for each subject in this study.

		Weekly Visits (±4 days)					
	SC	BL/-2	-1	BL/0	1-11	*EOS	FUP @3mo
Informed Consent	X						
Inclusion/Exclusion	X						
Randomization				X			
Medical History, Social, Demographics, PE, Vitals		X					
Lab Values		X					
Vascular - SPP		X			X**	X	
Vascular – tcpO2		X			X**	X	
Vascular - HSI		X	X	X	X	X	
Monofilament, VPT		X					
Wound Assessment, Debridement		X	X	X	X	X	X
Tissue Collection (4 pieces)#		X			X##	X	
Wound Photography and Measurements		X	X	X	X	X	X
Pain Assessment		X		X	X	X	
Sock Application				X	X		
Offloading				X	X		
Adverse Events			X	X	X	X	X
Concomitant Medication		X	X	X	X	X	
DFS-SF						X	
Subject Stipend			X	X	X	X	X

*if subject is deemed closed at week 12, subject will be observed at week 14 and will enter FUP phase
**Visit 4 only, 5 or 6 if visit 4 is missed
Two for RNA later and two for DNA/RNA shield
Weeks 3 and 6

Trial Monitoring

Monitoring visits will be conducted by representatives of Hologenix according to the U.S. CFR Title 21 Parts 50, 56, and 812 and ICH Guidelines for GCP (E6). By signing this protocol, the Investigator grants permission to the Hologenix contact and appropriate regulatory authorities to conduct on-site monitoring and/or auditing of all appropriate study documentation.

Clinical monitors, qualified by training and experience, will be responsible for monitoring and

overseeing the conduct of this study. The clinical monitor will evaluate compliance with the protocol, FDA regulations, any specific recommendations made by the investigational site's IRB and the signed Investigator Agreement. Telephone, e-mail and fax communications, as well as on-site visits, will be conducted to ensure that this protocol is being followed and that any protocol deviations are properly documented. Clinical monitoring will include verification that the Informed Consent forms were properly completed for all subjects enrolled in this study, a review of clinical records for accuracy and completeness, resolution of missing or inconsistent results, reporting of complications, adverse events and unanticipated adverse device effects, and a review of source documents. The clinical monitor will verify that the Case Report Forms (CRFs) are in agreement with the source documentation and other study-related records available at the site. For record verification purposes, the clinical monitor will be provided access to relevant hospital records, original laboratory data, and other records and data as they relate to this study and as agreed to with the investigator prior to the initiation of the study. The investigator will make available to the clinical monitor for review all signed Informed Consent forms, all completed CRFs, source documentation and other relevant records for all subjects enrolled at the site. It is important that the investigator and other relevant site personnel are available for consultation with the clinical monitor during the monitoring visits and that sufficient time is devoted at the site for the monitoring process. Additionally, telephone, e-mail and fax communications will be conducted on a regular basis with the investigator and the site staff to ensure that the protocol is being followed and to address any issues that may occur during the course of the study. If a deficiency is noted during an on-site visit (or at any other time during the course of the study), the clinical monitor is required to discuss the situation with the investigator and the Sponsor (if required) to secure compliance.

Audits and Inspections

Internal audits are done periodically to assure regulations, guidelines and protocols are adhered to appropriately. Patient binders are reviewed for proper informed consent process documentation, case report forms are compared to the source documents and reviewed for accuracy and an independent review of the inclusion and exclusion criteria using a checklist. Additionally, the use of an auditing and monitoring tool which includes sections on consent, HIPAA, on study/treatment, off treatment/off study and a general section. Weekly meetings occur to go over each study. If there are any issues that arise, they are brought up and discussed at the meeting.

4. Statistical Analyses

General Statistical Considerations

This study is a prospective, sham-controlled, subject and evaluator blinded, 1:1 randomized, multicenter clinical study. The duration of the study is up to four months for randomized subjects. Sample size may not be reduced less than the expected total enrollment of 254 evaluable subjects total but may increase up to a total of 2500 evaluable subjects. Sample size will increase based on actual attrition that is expected to be between 5 and 15%.

The purpose of this study is to evaluate safety and efficacy outcomes related to the Celliant Sock (CS). The p-value thresholds for determining statistical significance are 0.05 and 0.025 for 2- and 1-sided tests, respectively, unless otherwise specified.

Data will be presented using summary statistics. For example, continuous data may be displayed with the mean, median, minimum, maximum, and/or standard deviation, while categorical data may be displayed as proportions and counts.

Any changes to the analysis of the trial data will be documented either in a future protocol amendment or in the SAP.

Randomization

Randomization will be conducted in a 1:1 ratio of the Treatment Group (Celliant Sock, n=127) or to a sham Control Group (placebo sock, n=127). Randomization will be performed within randomly selected block sizes of 3 and 6 with equal probability of group assignment stratified by study center and wound size (greater than size of 4 cm^2).

A Sponsor-designated independent statistical consultant will provide the randomization list. Sponsor personnel directly involved in the management, conduct, and reporting of the study will not have access to randomization assignments until the study is completed.

Analysis Population

The primary population for the effectiveness analyses will be the intent-to-treat (ITT) population, which will include all randomized subjects regardless of whether the socks were ever worn. Subjects will be assessed to the group to which they were randomized, regardless of whether they were provided the wrong socks.

A secondary population for effectiveness analyses will be the (AT) population, including subjects that attempted to put on the sock for any amount of time. These subjects will be analyzed according to the type of socks (Celliant or Placebo) they received. This population is also the safety population, which is the population used for all safety analyses.

A third population is the modified full analysis set (mFAS), which consists of all subjects that report wearing the sock as directed for at least 10 days.

Sample Size Justification

Assuming a 2-sided alpha at 0.05, and a power of 0.85, the true mean difference at 12 weeks between groups in percent change in tissue oxygenation as measured by hyperspectral imaging is estimated to be 8.2 with a standard deviation of 21.7. At a 1:1 allocation of experimental to control subjects, a total of 254 evaluable subjects are required. The sample size will increase to account for attrition at Week 12.

Analyzing the sample size for the key secondary endpoint of incidence of complete wound closure suggests a much large trial is need. Specifically, at 2500 evaluable total subjects, there would be approximately a power of 0.85 to detect a difference in incidence of complete wound closure at 12 weeks if the expected incidence of complete wound closure was 25% in the experimental group, and 20% in the control group, an absolute difference of 5% between the experimental group and the control group.

Primary Efficacy Analyses

The primary efficacy endpoint is an inferential test of whether the Treatment Group (Celliant® Diabetic Medical Sock) percent change in tissue oxygenation (via oxygen saturation, S_tO_2) compared to the Control Group (placebo sock) at 12 weeks is superior.

Effectiveness variables will be analyzed relative to the mean of the Weekly visit 0 and -1 tissue oxygenation measurements. For purposes of statistical analysis, this mean will be referred to as the baseline.

The hypotheses are:

H_0: $\%ChgPerf_{Celliant} \leq \%ChgPerf_{Placebo}$
H_a: $\%ChgPerf_{Celliant} > \%ChgPerf_{Placebo}$

A mixed model repeated measures (MMRM) will be utilized to model percent changes in tissue oxygenation by visit through 12 weeks from baseline to provide the contrast statement at 12 weeks between groups. Included in the model will be treatment group, the stratification variables of wound size (as a continuous variable) and study site. Success on this endpoint requires that the 12-week treatment group contrast difference is $\alpha<0.025$. There is no interim analysis for the primary endpoint. The primary endpoint is based on a fixed study design. Regulatory activities for all but the key secondary endpoint will begin if this primary endpoint is met.

Interim Analysis for the Key Secondary Endpoint

The key secondary endpoint will only be tested for significance if the success criterion for the primary endpoint are met at the time of the primary endpoint analysis in 254 evaluable subjects, which is the interim analysis for the key secondary endpoint.

The key secondary endpoint is an inferential test of whether the Treatment Group (Celliant sock) incidence of complete wound closure rate (CWCR) compared to the Control Group (placebo sock) at 12 weeks is superior.

The hypotheses for this endpoint are:

H_0: $CWCR_{Celliant} \leq CWCR_{Placebo}$
H_a: $CWCR_{Celliant} > CWCR_{Placebo}$

The key secondary endpoint of incidence of complete wound closure at 12 weeks will be blinded except to the unblinded Interim Analysis Committee (IAC) until the final analysis for this endpoint is conducted. Since the estimated sample size for the key secondary endpoint is anticipated to be many times larger than for the primary endpoint and there is some uncertainty relative to the anticipated treatment effect, the hierarchical design allowing for the testing of the key secondary endpoint, and then the use of the adaptive group sequential design to allow for the potential of sample size re-estimation is required for efficiency.

The IAC is comprised of an unblinded statistician and a subject area medical expert whose sole responsibility is to analyze the key secondary endpoint at 254 evaluable subjects to determine, according to the Lan-Demets O-Brien Fleming alpha spending function, tested at half of the trial information:

1) The criterion of early stopping for efficacy for CWCR is met at half of the trial information,

2) The trial will continue to the preplanned 508 planned evaluable subjects for assessing CWCR, based on the conditional power at the interim to be Cp≥ 0.85,

3) If neither of the first two criteria are met, then sample size re-estimation according to the inverse normal method (Lehmacher and Wassmer, 1999) will be conducted to allow for a sample size of up to 2500 evaluable subjects total for the study.

    a. The trial will be stopped for futility of the key secondary endpoint if the Cp assuming a maximum sample size of 1250 subjects for the study provides a CP<0.7.

    b. The trial will have a sample size increase as calculated by the inverse normal method to maintain a Cp=0.85.

With the exception of meeting criterion 1 or 3a, i.e. stopping further enrollment, the results of the interim analysis will not be shared with the clinical operations team involved directly in the study data at the patient or data management. This communication firewall is required to maintain the validity of the secondary endpoint in that subjects before and after the interim analysis are handled no differently in a way that would alter the estimated treatment effect.

A logistic model will be used to control for the stratification variables and assess for the treatment effect. Once again, as in the primary endpoint, the baseline wound size will be assessed as a continuous endpoint, although the stratification variable is dichotomous.

Type 1 error is controlled because of the hierarchical testing strategy of the primary endpoint and then key secondary endpoints, both at $\alpha_{1\text{-sided}}=0.025$.

An Interim Analysis Charter (IAC) will be provided solely to provide a communication plan and numerical example with SAS code to detail the calculations involved for the key secondary endpoint.

Hierarchical Testing of the VAS Pain Endpoint

The VAS pain endpoint will be tested as the third hierarchical endpoint, i.e., following testing of the primary endpoint and then the total wound closure endpoint. In order for the VAS pain endpoint to be tested, the primary endpoint and the TWCR endpoints would both need to be statistically significant. The final total sample size as determined by the sample size re-estimation for TWCR will be applied to the VAS pain endpoint without further modification.

Evaluability of Data

Prior to the database lock for the final primary endpoint analysis at 254 evaluable subjects, and again at any subsequent lock as dictated by the adaptive design for subjects evaluated after the final primary lock, an evaluability meeting will be held. The goal of the evaluability meeting is to identify the disposition of study subjects with respect to the predefined study populations.

Subject Demographic and Baseline Characteristics

Demographic and baseline characteristics of enrolled subjects will be summarized. The factors will include (but are not limited to): Age, sex, race, BMI (kg/m^2), baseline Diabetic and Foot Ulcer Scale-Short Form (DFS-SF). Medical history, PE, and vitals will be provided in a listing.

Additional Efficacy Analyses including Subgroup Analyses

Secondary Endpoints: To compare the incidence of the below between Celliant and Control treated groups:

1. For subjects whose wound closes within the 12-week period, wound evaluation at 3 months following complete wound closes for the following parameters
    a. Incidence of wound dehiscence/reoccurrence
    b. Size of reoccurrence
2. Accelerated wound closure as measured by changes in wound area, depth and volume.
3. Assessment of Compliance
4. Change in pain assessment as reported using VAS pain scale
5. Outcomes of Patient Reported Outcomes
6. Study related AE/SAEs

Additional, ad hoc exploratory analyses may also be conducted for efficacy endpoints.

Safety Endpoints and Analyses

An overview of events, to include total numbers of events, and total number of subjects will be tabulated and presented for all AEs, SAEs, procedure-related AEs by treatment type. Device specific risks such as “skin and subcutaneous tissue reaction/allergy” and “mild pain or discomfort” will also be tabulated in a separate summary table.

Additional, ad hoc exploratory analyses may also be conducted for safety endpoints.

Poolability

Pooling of outcome data from different clinical study sites will be performed by evaluating the homogeneity of the primary efficacy endpoint across sites. If the treatment by site interaction is $p \geq 0.2$, then homogeneity will be considered to be demonstrated across sites. If $p<0.2$, then site level variable will remain in the analysis models.

Handling of Missing Data

A list of subjects who were withdrawn from the study and their associated reasons for withdrawal will be provided. A list of subjects lost to follow-up will also be provided. If the missingness of tissue oxygenation data or total wound closure data at 12 weeks is missing in more than 7.5% of subjects, multiple imputation will be performed, creating n=5 fully imputed complete datasets for the primary and key secondary endpoint, to be evaluated combined through a t-test, after the

predefined analyses are provided for each imputed data set. A non-imputed analysis will also be provided.

5. Data Management

Standardized CRFs will be created at all participating sites. Investigators are responsible for the accurate completion and timely submission of the data collected during the trial. All data from the trial will be entered from the CRFs into a central database. Incoming data will be frequently reviewed to identify inconsistent or missing data and any adverse events. Any data issues are to be promptly addressed with the investigator Quality assurance procedures will be established to ensure that complete, accurate and timely data are submitted, that protocol requirements are followed, and that complications, adverse events and adverse device effects are correctly reported and investigated, as appropriate. Investigators are to maintain all source documents as required by the protocol, including laboratory results, supporting medical records, and signed Informed Consent Forms. The source documents will be used during the regular monitoring visits to verify information from the database against data contained on the completed CRFs.

6. Deviations from the Investigational Plan

Any deviations from the study protocol will be documented as Protocol Deviations in the Source Documentation.

7. Device Accountability

The Sponsor will ship investigational devices only to the designated investigators participating in this study. The Sponsor will not ship investigational devices to any site until evidence of IRB approval has been provided to the Sponsor. All investigators are responsible for providing a secure storage location for the investigational devices, supervising device use, as well as the disposal and/or return of the devices as instructed by the Sponsor. In addition, all investigators shall maintain records to document the receipt, use and disposition of all investigational devices received by their site. The Sponsor will maintain records of all shipments and disposition of the investigational devices and will routinely inspect for device accountability at the clinical sites participating in this study. The subjects will not be responsible for returning the investigational devices.

8. Ethics

Trial Conduct

The study will be conducted according to the Declaration of Helsinki, Protection of Human Volunteers (21 CFR 50), Institutional Review Boards (21 CFR 56), and Obligations of Clinical Investigators (21 CFR 812).

To maintain confidentiality, all laboratory specimens, evaluation forms, reports and other records will be identified by a coded number only. All study records will be kept in a locked research office with limited access to study personnel and code sheets linking a patient's name to a patient identification number will be stored separately in a secured area within the research office. Clinical information will not be released without written permission of the subject, except as necessary for monitoring by the FDA. The Investigator must also comply with all applicable

privacy regulations (e.g., Health Insurance Portability and Accountability Act of 1996, EU Data Protection Directive 95/46/EC).

Good clinical practice (GCP) is an international quality standard that is provided by International Conference of Harmonization (ICH), an international body that defines a set of standards, which governments can then transpose into regulations for clinical trials of medications as well as medical devices involving human subjects.

GCP guidelines are important as they provide the necessary enforcement of ethics of a clinical study, protection of human rights for the subjects (such as voluntariness) and assurance of the safety and efficacy of investigational products. High standards are required in terms of comprehensive documentation for the clinical protocol, record keeping (paper and electronic which includes computers and software), training, and facilities. Quality assurance and inspections ensure that these standards are achieved and maintained.

GCP guidelines include standards on how clinical trials should be conducted, define the roles and responsibilities of clinical trial sponsors, clinical research investigators, and monitors. As a research site, we follow the ICH-GCP guidelines. Our GCP includes review of each case to determine whether or not the patient would be a good candidate for a clinical trial and if there is an appropriate clinical trial for the patient. Important aspects include adequate time for the person to review the consent form and have all questions answered, voluntarily give consent and know that they can withdraw at any time. Maintaining the subject's privacy and confidentiality is handled by assigning a subject number for each study participant. Research staff responsibilities include: adherence to the protocol, proper documentation, accuracy of data and appropriate handling of the study product.

Institutional Review Board Review

The protocol and consent form will be reviewed and approved by the IRB/IEC of each participating center prior to study initiation. Serious adverse experiences regardless of causality will be reported to the IRB/IEC in accordance with the standard operating procedures and policies of the IRB/IEC, and the Investigator will keep the IRB/IEC informed as to the progress of the study. The Investigator will obtain assurance of IRB/IEC compliance with regulations.

Any documents that the IRB/IEC may need to fulfill its responsibilities (such as protocol, protocol amendments, Investigator's Brochure, consent forms, information concerning patient recruitment, payment or compensation procedures, or other pertinent information) will be submitted to the IRB/IEC. The IRB/IECs written unconditional approval of the study protocol and the informed consent form will be in the possession of the Investigator before the study is initiated. The IRB/IECs unconditional approval statement will be transmitted by the Investigator to Hologenix contact prior to the shipment of study supplies to the site. This approval must refer to the study by exact protocol title and number and should identify the documents reviewed and the date of review.

Protocol and/or informed consent modifications or changes may not be initiated without prior written IRB/IEC approval except when necessary to eliminate immediate hazards to the patients or when the change(s) involves only logistical or administrative aspects of the study. Such modifications will be submitted to the IRB/IEC and written verification that the modification was submitted and subsequently approved should be obtained.

The IRB/IEC must be informed of revisions to other documents originally submitted for review; serious and/or unexpected adverse experiences occurring during the study in accordance with the standard operating procedures and policies of the IRB; new information that may affect adversely the safety of the patients of the conduct of the study; an annual update and/or request for re-approval; and when the study has been completed.

Any amendment to the protocol will be written by the investigator and Hologenix contact. Protocol amendments cannot be implemented without prior written IRB/IEC approval except as necessary to eliminate immediate safety hazards to patients. A protocol amendment intended to eliminate an apparent immediate hazard to patients may be implemented immediately, provided the IRBs are notified within five working days.

Informed Consent

Informed consent will be obtained in accordance with the Declaration of Helsinki, ICH GCP, US Code of Federal Regulations for Protection of Human Subjects (21 CFR 50.25 [a,b], CFR 50.27, and CFR Part 56, Subpart A), the Health Insurance Portability and Accountability Act (HIPAA, if applicable), and local regulations.

Subjects cannot be asked to sign the Informed Consent form until the study has been fully approved by the investigational site's IRB and the Sponsor has received and reviewed the specific IRB approved Informed Consent form to be used by the site. The potential subject shall be given adequate time to read the consent form, have the study procedures explained, including risks and benefits, as well as alternative procedures, prior to signing the Informed Consent form. An example of the Informed Consent form for this study is provided in Appendix 1: Informed Consent Form. The consent form must be read by the subject, the subject's questions answered, and the form signed by the subject before the treatment can be performed. All subjects are to receive copies of their signed consent form. The date the subject signs the consent form is to be recorded on CRF.

Coverage of Expenses

Subject Compensation

Subjects will be paid 35 dollars for each week of participation in the study for each completed study visit. Payment will be made after each visit or within 30 days of the ending of their participation in the study. All subjects participating in this study will be provided with the Study Device listed in this protocol at no cost to them.

Cost to Subjects

There will be no cost incurred by the subject for participating in this study. All noted procedures are considered standard of care (SOC) and will be billed to the patient's insurance or whatever program the patient uses.

Confidentiality

Protected health information (PHI) of clinical investigation Subjects are kept as confidential as possible in accordance with the Health Insurance Portability and Accountability Act of 1996

(HIPAA). Members of the study team who will have access to the PHI information include the following: Research staff at UT Southwestern Hospital, UT Southwestern Wound Clinic, Parkland Wound Clinic, Parkland Inpatient Hospital, Sponsor (Hologenix, LLC), and any outside labs/facilities that may handle specimens etc. should be named here as well. When appropriate, specimens and data will only have subject ID. However, confidentiality cannot be assured.

9. Data Handling and Record Keeping

Source Documents

The investigator must maintain detailed source documents on all subjects who are enrolled or who undergo screening in the study. Source documents include subject medical records, hospital charts, clinic charts, investigator subject trial files, as well as the results of diagnostic tests (e.g., laboratory tests, hemodynamic studies).

The following minimum information should be entered into the subject's medical record:

- The date the subject entered the trial
- The IRB number and the name of the Sponsor
- The date that Informed Consent was obtained
- Evidence that the subject meets the trial eligibility requirements (e.g., medical history, study procedures and/or evaluations)
- The dates of all trial related subject visits
- Procedures performed that may affect clinical care
- Use of any concurrent medications (antibiotics, antifungals and other anti-infective therapies)
- Documentation of specific device used
- Occurrence and status of any adverse events (AEs)
- The date the subject exited the trial and a notation as to whether the subject completed the trial or was discontinued.

Data Collection

The investigator must maintain detailed records on all subjects who sign the Informed Consent Form and begin the pre-procedure evaluation. Data for enrolled subjects will be transcribed on to CRFs provided by the Sponsor. All data should be transcribed completely, promptly and legibly. Corrections should be made in a manner that does not obscure or eliminate the original error, by striking through the original data with one line, and initialing and dating the change,

along with the reason for the change (if not obvious). Original CRF pages will be collected by the Sponsor or Sponsor's designee after they are reviewed by the study monitor. The investigator should maintain a copy of all completed CRFs from this trial.

Trial exit forms will be completed for all enrolled subjects, regardless if they did or did not complete the trial (e.g., subject discontinuation, trial termination).

Record Retention

All records relating to the conduct of this trial are to be retained by the investigator until notified by the Sponsor or Sponsor's designee that the records may be destroyed.

10. Quality Control and Quality Assurance

Site Training

Research staff training involves being credentialed at our institution and other affiliates, CITI training, IATA training, training provided by Sponsors, in-house training and protocol training. Staff attend the monthly IRB presentations which keeps researchers up to date on regulatory news. Required self-training using a binder which contains information on background & regulations for research, IRB processes, review of historical cases and glossary of terms and a check off sheet for staff. Staff are also encouraged to seek out seminars, webinars & conferences that will further their education in research and area of indication (podiatry). The binder will be updated on an annual basis and when new information or changes occur to any regulations or guidelines.

Investigator Training

Investigator Responsibilities

The investigators are responsible for ensuring that this study is conducted according to this protocol and applicable regulations and that signed Informed Consent is obtained from each subject prior to his inclusion in this study. It is the investigator's responsibility to ensure that all staff assisting with this study have the appropriate qualifications and are fully instructed on the study procedures and respect subject confidentiality, as specified in the Investigator Agreement with the Sponsor

Investigator Records

Standardized Case Report Forms (CRFs) will be used to collect complete and accurate records of the clinical data generated from this study according to Good Clinical Practices (GCP) requirements. The investigators are responsible for collecting and accurately recording the clinical data generated for this study. Investigators are also responsible for maintaining records as required by FDA per Title 21 Code of Federal Regulations (CFR) §812.140.

Investigator Reports

The investigator will be responsible for providing the following reports, in accordance with CFR§812.150, to the Sponsor for this study:

- Serious Adverse Events (SAEs) and Unanticipated Adverse Device Effects (UADEs): The investigators will report by telephone, email or fax any SAEs or UADEs as soon as possible, within 24 hours of the investigator becoming aware of the event, to the Sponsor and the IRB. The Serious Adverse Event form is to be completed to document the SAE and it is to be faxed or express mailed to the Sponsor and the IRB within ten working days of the event.

- Withdrawal of Approval: If an IRB withdraws the approval to conduct this study for any reason, the investigator will notify the Sponsor as soon as possible, but in no event later than five working days after the withdrawal of the approval

- Progress Reports: The investigator will submit progress reports on the investigation to the Sponsor and the reviewing IRB at regular intervals, but in no event less often than yearly.

- Deviations from the Investigational Plan: The investigator must notify the Sponsor and the reviewing IRB of any deviation from the Investigational Plan to protect the life or physical well-being of a subject in an emergency. This notice must occur as soon as possible, but in no case longer than five working days following the occurrence of the deviation.

- Informed Consent: If the investigator uses a device without obtaining informed consent, the investigator shall report the use to the Sponsor and the IRB within 5 working days after the use occurs.

- Final Report: Within three months after the termination or completion of the study or the investigator's part in the study, the investigator shall submit a final report to the Sponsor and the IRB.

- Other Reports: Upon request from the IRB or the FDA, the investigator shall provide accurate, complete and current information about any aspect of the study.

Each investigator is responsible for ensuring that the study is conducted according to this protocol and that signed Informed Consent is obtained from each subject prior to their inclusion in this study.

11. Adverse Events and Serious Adverse Events

General

All observed or volunteered adverse events regardless of treatment group or suspected causal relationship to the investigational product(s) will be reported as described in the following sections.
For all adverse events, the investigator must pursue and obtain information adequate both to determine the outcome of the adverse event and to assess whether it meets the criteria for classification as a serious adverse event requiring immediate notification to the Hologenix contact. For all adverse events, sufficient information should be obtained by the investigator to determine the causality of the adverse event. The investigator is required to assess causality. For adverse events with a causal relationship to the device, follow-up by the investigator is

required until the event or its sequelae resolve or stabilize at a level acceptable to the investigator, and the Hologenix contact concurs with that assessment.

Adverse Event Reporting

For serious adverse events, the reporting period to Hologenix or its designated representative begins from the time that the subject provides informed consent, which is obtained prior to the subject's participation in the study, i.e., prior to undergoing any study-related procedure and/or receiving investigational product, through and including 28 calendar days after the last administration of the investigational product. Any serious adverse event occurring any time after the reporting period must be promptly reported if a causal relationship to investigational product is suspected. Adverse events (serious and non-serious) should be recorded on the CRF from the time the subject has undergone one treatment through last subject visit.

Definition of an Adverse Event (AE)

An adverse event is any untoward medical occurrence in a clinical investigation subject administered a product or medical device; the event need not necessarily have a causal relationship with the treatment or usage. Examples of adverse events include but are not limited to:

- Abnormal test findings;
- Clinically significant symptoms and signs or diagnosis;
- Changes in physical examination findings;
- Hypersensitivity;
- Progression/worsening of wound.

Protocol Specific AE Definition/Collection

For the purposes of this study, only AEs that are directly related to the study product, diabetes (including labs & kidney function) or wound healing (including infections related to the wound) will be collected and reported as these events may affect outcomes and analysis of data. AEs not related to the aforementioned product or diagnoses are not relevant to the subject outcomes of this research or data analysis (such as nausea, emesis, pain, constipation, diarrhea, mood disorders, respiratory diagnoses, cardiac related events, etc). AEs that do not fall under this protocol specific definition will be assessed and treated as per standard of care. SAEs will be collected as per the usual SAE guidelines (see section on reporting SAEs). In addition, planned procedures related to the study wound for a subject during the same hospitalization (as these subjects will be recruited while in the hospital because they have an infection in their foot wound/ulcer) for incision/debridement/amputation will be considered part of their continuation of SOC and not adverse events unless a complication, new infection or new wound occurs.

Definition of Serious Adverse Event (SAE)

A Serious Adverse Event (SAE) is any AE that has any serious unfavorable and unintended sign, symptom, or disease temporally associated with the use of the devices, whether or not considered related, including those that:

- results in death
- is life-threatening
- requires inpatient hospitalization or causes prolongation of existing hospitalization
- results in persistent or significant disability/incapacity
- is a congenital anomaly/birth defect
- requires intervention to prevent permanent impairment or damage

Medical and scientific judgment should be exercised in determining whether an event is an important medical event. An important medical event may not be immediately life-threatening and/or result in death or hospitalization. However, the important medical event should be reported as serious, if it is determined that the event may jeopardize the subject and/or may require intervention to prevent one of the other adverse event outcomes.

Causality Assessment of Adverse Events

The investigator's assessment of causality must be provided for all adverse events (serious and non-serious); the investigator must record the causal relationship in the CRF, as appropriate, and report such an assessment in accordance with the serious adverse reporting requirements if applicable. An investigator's causality assessment is the determination of whether there exists a reasonable possibility that the cleared medical device caused or contributed to an adverse event. If the investigator does not know whether or not medical device caused the event, then the event will be handled as "related to medical device" for reporting purposes. (see Section on Reporting Requirements). If the investigator's causality assessment is "unknown but not related to investigational product", this should be clearly documented on study records.

In addition, if the investigator determines a serious adverse event is associated with study procedures, the investigator must record this causal relationship in the source documents and CRF, as appropriate, and report such an assessment in accordance with the serious adverse event reporting requirements, if applicable.

Adverse Event Severity Assessment

The Investigator will provide an assessment of the severity of each adverse reaction by recording a severity rating on the appropriate SAE reporting page of the subject's file. Severity, which is a description of the intensity of manifestation of the SAE, is distinct from seriousness, which implies a patient outcome or SAE-required treatment measure associated with a threat to life or functionality. Severity will be assessed according to the following scale.

If required on the adverse event case report forms, the investigator will use the adjectives MILD, MODERATE, or SEVERE to describe the maximum intensity of the adverse event. For purposes of consistency, these intensity grades are defined as follows:	
MILD	Does not interfere with subject's usual function.
MODERATE	Interferes to some extent with subject's usual function.
SEVERE	Interferes significantly with subject's usual function.

Note the distinction between the severity and the seriousness of an adverse event. A severe event is not necessarily a serious event. For example, a headache may be severe (interferes significantly with subject's usual function) but would not be classified as serious unless it met one of the criteria for serious adverse events, listed above.

Withdrawal Due to Adverse Events (See Also Section on Subject Withdrawal)

Withdrawal due to adverse event should be distinguished from withdrawal due to insufficient response, according to the definition of adverse event noted earlier, and recorded on the appropriate adverse event CRF page.

When a subject withdraws due to a serious adverse event, the serious adverse event must be reported in accordance with the reporting requirements defined below.

Eliciting Adverse Event Information

The investigator is to report all directly observed adverse events and all adverse events spontaneously reported by the study subject. In addition, each study subject will be questioned about adverse events by a member of the research staff.

Reporting Requirements

Each adverse event is to be assessed to determine if it meets the criteria for serious adverse events. If a serious adverse event occurs, expedited reporting will follow local and international regulations, as appropriate.

Serious Adverse Event Reporting Requirements

If a serious adverse event occurs, Hologenix is to be notified within 24 hours of awareness of the event by the investigator. In particular, if the serious adverse event is fatal or life-threatening, notification to Hologenix must be made immediately, irrespective of the extent of available adverse event information. This timeframe also applies to additional new information (follow-up) on previously forwarded serious adverse event reports as well as to the initial and follow-up reporting of Exposure during pregnancy cases.

In the rare event that the investigator or member of the research team does not become aware of the occurrence of a serious adverse event immediately (e.g., if an outpatient study subject

initially seeks treatment elsewhere), the investigator or member of the research team is to report the event within 24 hours after learning of it and document the time of his/her first awareness of the adverse event.

For all serious adverse events, the investigator is obligated to pursue and provide information to Hologenix in accordance with the timeframes for reporting specified above. In addition, an investigator may be requested by Hologenix to obtain specific additional follow-up information in an expedited fashion. This information may be more detailed than that captured on the adverse event case report form. In general, this will include a description of the adverse event in sufficient detail to allow for a complete medical assessment of the case and independent determination of possible causality. Information on other possible causes of the event, such as concomitant medications and illnesses must be provided. In the case of a subject death, a summary of available autopsy findings and/or a copy of the death certificate must be submitted as soon as possible to Hologenix or its designated representative.

All AEs/SAEs should be followed until resolution.

Non-Serious Adverse Event Reporting Requirements

All adverse events will be reported on the adverse event page(s) of the CRF. It should be noted that the form for collection of serious adverse event information is not the same as the adverse event CRF. Where the same data are collected, the forms must be completed in a consistent manner. For example, the same adverse event term should be used on both forms. Adverse events should be reported using concise medical terminology on the CRFs as well as on the form for collection of serious adverse event information.

Reporting Requirements to Regulatory Authorities

Adverse events reporting, including suspected serious unexpected adverse reactions, will be carried out in accordance with applicable local regulations.

12. Committees

Data and Safety Monitoring Board

The study will be under the oversight of an independent DSMB that will assess the accumulated safety data on an ongoing basis. The role and responsibilities of the DSMB will be predefined in the DSMB charter.

13. Publication Policy

The preparation and submittal for publication of manuscripts containing the study results shall be in accordance with a process determined by mutual written agreement among the investigator and the Hologenix contact. 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 of 1996.

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15. Appendix 2 – Diabetic Foot Ulcer Scale – Short Form

Diabetic Foot Ulcer Scale - Short Form

INSTRUCTIONS:

These questions ask about the effect foot ulcers may have on your daily life and well-being. Please read each question carefully and think about the effect of your foot ulcers.

Answer every question by circling one number on each line. If you are unsure about how to answer a question, please give the best answer you can.

Diabetic Foot Ulcer - Short Form

1. How much have your foot ulcers:					
	Not at all	A little bit	Moder-ately	Quite a bit	A great deal
a) stopped you from doing the hobbies and recreational activities that you enjoy	1	2	3	4	5
b) changed the kinds of hobbies and recreational activities that you enjoy doing	1	2	3	4	5
c) stopped you from going away for a weekend, vacation, or holiday	1	2	3	4	5
d) made you choose a different kind of weekend, vacation, or holiday than you would have preferred	1	2	3	4	5
e) meant you had to spend more time planning and organizing leisure activities	1	2	3	4	5

2. Because of your foot ulcers, how often have you felt:					
	None of the time	A little of the time	Some of the time	Most of the time	All of the time
a) fatigued or tired	1	2	3	4	5
b) drained	1	2	3	4	5
c) that you had difficulty sleeping	1	2	3	4	5
d) pain while walking or standing	1	2	3	4	5
e) pain during the night	1	2	3	4	5

3. Because of your foot ulcers, how often have you:					
	None of the time	A little of the time	Some of the time	Most of the time	All of the time
a) had to depend on others to help you look after yourself (like washing and dressing)	1	2	3	4	5
b) had to depend on others to do household chores (like cooking, cleaning or laundry)	1	2	3	4	5
c) had to depend on others for getting out of the house	1	2	3	4	5
d) had to spend more time planning or organizing your daily life	1	2	3	4	5
e) felt that doing anything took longer than you would have liked	1	2	3	4	5

4. Because of your foot ulcers, have you felt:					
	Not at all	Slightly	Moder-ately	Quite a bit	Extre-mely
a) angry that you were not able to do what you wanted	1	2	3	4	5
b) frustrated by others doing things for you when you would rather do them yourself	1	2	3	4	5
c) frustrated because you were not able to do what you wanted	1	2	3	4	5
d) worried your ulcer(s) will never heal	1	2	3	4	5
e) worried you may have to have an amputation	1	2	3	4	5
f) worried about injury to your feet	1	2	3	4	5
g) depressed that you were not able to do what you wanted	1	2	3	4	5
h) worried about getting ulcers in the future	1	2	3	4	5
i) angry this has happened to you	1	2	3	4	5

j) frustrated because you have difficulty getting around	1	2	3	4	5
5. Because of your foot ulcers, how often were you bothered by:					
	None of the time	A little of the time	Some of the time	Most of the time	All of the time
a) having to keep weight off your foot ulcer	1	2	3	4	5
b) the amount of time involved in caring for your foot ulcer (including dressing changes, waiting for the home health care nurse, and keeping the ulcer clean)	1	2	3	4	5
c) the appearance, odor or weeping of your ulcer	1	2	3	4	5
d) having to depend on others to help care for your foot ulcer	1	2	3	4	5

THANK YOU FOR COMPLETING THIS QUESTIONNAIRE

DFS-SF - United States/English - Version of 22 Feb 08 - Mapi Research Institute. ID4557 / DFS-SFeng-USq_author