UCI Office of Research	**Institutional Review Board** **Human Research Protections** **Protocol Narrative – Expedited/Full Committee Biomedical/Clinical Research** *November 2021*
Upload this completed narrative and any supplemental documentation to the IRB Application.	**IRB USE ONLY –** **HS#: 2021-6555**
Lead Researcher Name: Raj Vyas, MD, FACS	
Study Title: Micro/nanobubbles (MNBs) and Wound Therapy: A Pilot Study Involving a Novel Oxygen Delivery System for Treatment of Acute and Chronic Wounds	

CLINICAL TRIAL MASTER PROTOCOL AND INVESTIGATIONAL BROCHURE INFORMATION *

	Master Protocol	**Investigator Brochure:** <Specify Drug/Device>	**Investigator Brochure:** <Specify Drug/Device>	**Sponsor Consent Form Template(s)**
Version #:				
Version Date:				

[ ] This study is investigator-authored (investigator developed the study and is conducting the study at UCI and/or with other non-UCI sites).

* Add columns as applicable

NON-TECHNICAL SUMMARY

Provide a brief non-technical summary or synopsis of the study that can be understood by IRB members with varied research backgrounds, including non-scientists and non-affiliated members.

Oxygen delivery by means of perfusion is one of the primary factors in wound healing. The usual irrigation used in managing wounds is limited by its oxygen carrying capacity. Micro/nanobubbles (MNBs) can be used to increase the oxygen dissolved in solution to assist the body's inherent means of perfusion in delivering oxygen to a wound by directly delivering an oxygen rich solution to the wound bed. In our recent animal study, irrigation with MNBs showed a favorable response in wound healing. Our aim is to take these results forward and apply them to human wounds. MNBs irrigation will be used to improve tissue oxygenation and potentially lead to improved healing as well. This will be assessed by indirect tissue oxygen measurements and photographs evaluating overall appearance of the wound. This pilot study has the potential to add to the wound healing armamentarium for treatment of many different types of wounds.

SECTION 1: PURPOSE AND BACKGROUND OF THE RESEARCH

1. Provide the scientific or scholarly rationale for the research. Describe the relevant background information and the specific gaps in current knowledge that this study intends to address.

Acute wounds complicated by a prolonged state of wound healing and poor tissue oxygenation often transition to a chronic state. Acute tissue ischemia can be encountered in situations such as, but not limited to, microvascular free tissue transfer, replantation of digits after traumatic injury, and local tissue rearrangement. Generally, acute wounds heal without sequela. However, where oxygen is a limiting reagent, salvage modalities to improve local oxygenation are used to prevent progression of wound ischemia. These are often limited and include reoperation, hyperbaric oxygen therapy (HBOT), and nitroglycerin paste.

Common risk factors leading to wound ischemia include, but are not limited to, smoking, diabetes mellitus, and increased tissue pressure. Management of these wounds requires weeks to months of local wound care, negative pressure wound therapy and secondary surgeries for wound debridement and coverage. Not surprisingly, clinicians have searched for techniques to improve wound oxygenation such as (HBOT) and Topical Oxygen Therapy (TOT) to improve chronic wound healing. However, such therapies are costly, not portable, and are only offered at specialized centers.

Micro/nanobubbles (MNBs) are miniature oxygen-filled bubbles, which offer a new agent for improving both wound oxygenation and healing. Our team has extensively studied micro/nanobubbles (MNBs) and their application in wound healing in the animal model.[1] MNBs are particularly favorable for improving wound oxygenation due to the following properties: (1) stability in solution (>12 hours), (2) efficient oxygen distribution in wound beds, (3) ability to be internalized by cells, (4) potential to debride tissue and destroy bacteria.[1-3] Given our success with MNBs in animal wounds we expect this to be beneficial for the treatment of chronic wounds in humans.

Negative pressure wound therapy (NPWT) is currently used widely in the hospital for wounds and facilitates wound healing by improving angiogenesis, clearing pro-inflammatory cytokines, and generating healthy granulation tissue.[4] A recent advent in NPWT is the addition of intermittent irrigation of wounds through the device with a variety of fluids and is called NPWT with instillation (NPWTi).[5] NPWTi has been shown to reduce bioburden and biofilm present in wounds improving the wound mileau.[6] Our team, in collaboration with UCI Bioengineering, has designed a NPWTi that is capable of delivering MNBs solution as an irrigation solution, thereby increasing delivering oxygen to wounds through the NPWTi device.

In this pilot study, we aim to assess the efficacy of MNBs in delivering oxygen to human wounds and thereby improving their healing. If outcomes are favorable, we will have heralded a new, inexpensive, and portable solution for the oxygenation of wounds.

2. Provide relevant preliminary data (animal and/or human).

At the Center for Tissue Engineering at UC Irvine, we have demonstrated that MNBs can be generated in solutions, such as saline, achieving oxygen saturations up to 800 mmHg (Figure 1). Additionally, when full-thickness rat burn wounds are irrigated with MNBs, there is improved tissue oxygenation, wound healing, and collagen organization when compared to standard, non-oxygenated saline solutions (Figure 2).

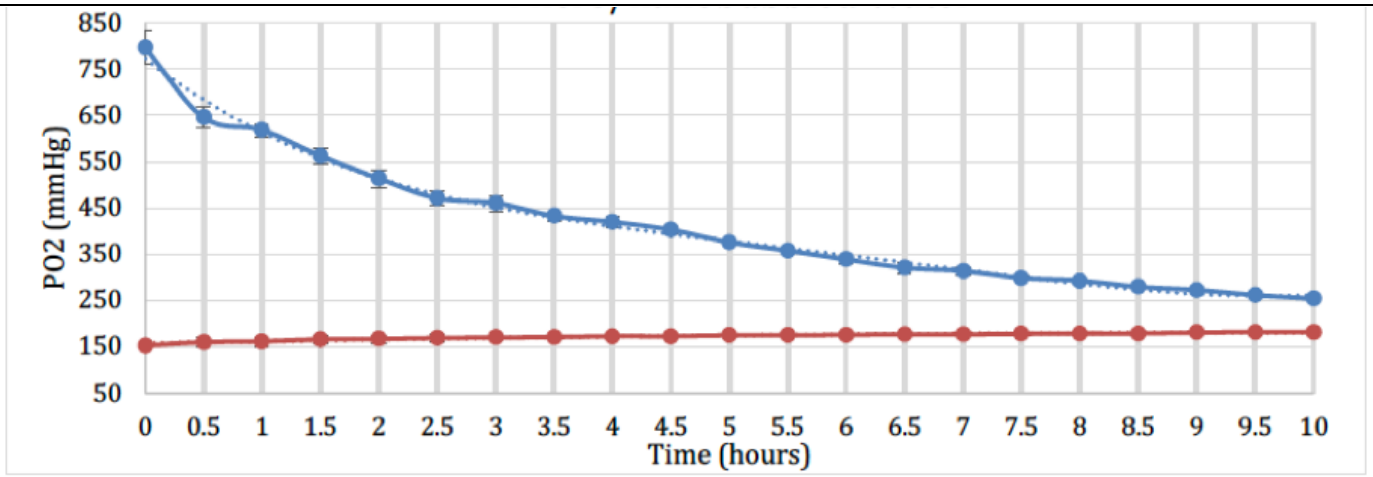

Figure 1: Micro/nanobubbled saline oxygen carrying capacity (blue/top graph) vs. standard saline (red/bottom graph) over 10-hour period (n=4).

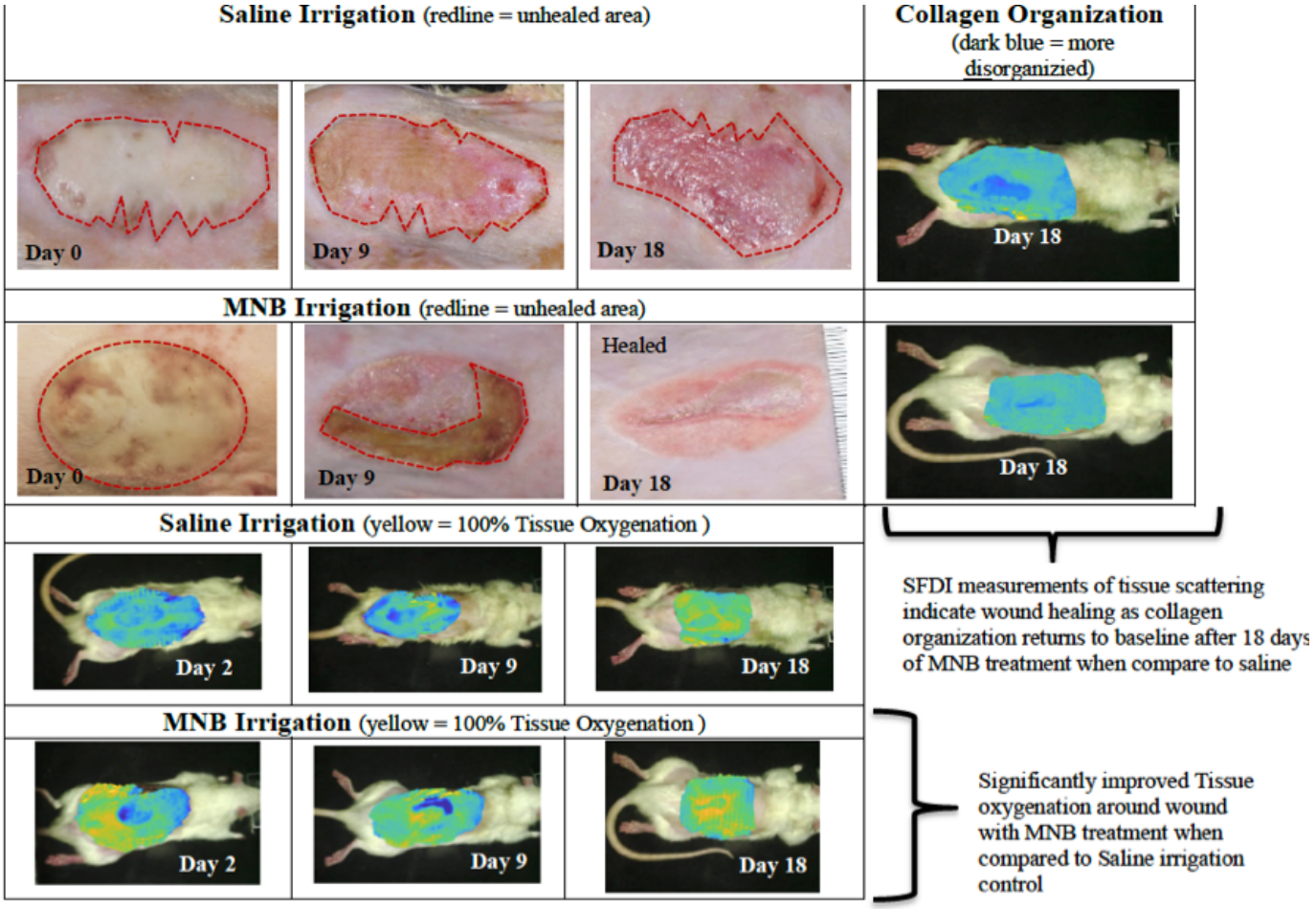

Figure 2: 4 rats with 3-cm full-thickness burn wounds were treated with either continuous saline (control) or MNB irrigation (bubbles made in saline) for a period of 18 days. Every third day, wounds were imaged with Spatial Frequency Domain Imaging (SFDI) to determine the degree of tissue oxygenation, collagen organization, and overall wound healing. Over the course of 18 days, the MNB-treated rats had significantly improved wound healing, tissue oxygenation (yellow=100% tissue oxygenation), and collagen organization when compared to the saline-treated control.

3. Describe the purpose, specific aims or objectives. Specify the hypotheses or research questions to be studied.

Specific Aim 1: Utilize an MNB solution as an irrigation solution to improve wound oxygenation in ischemic tissue (e.g00. ischemic surgical tissue, traumatic wounds, and failing replants).

Hypothesis: Tissue immersed in MNBs will have improved oxygenation when compared to tissue immersed in normal saline alone.

Study Design:

1. Acutely ischemic tissue will be identified by the practitioners listed in this IRB based on the selection criteria.
2. Baseline oxygen assessment: Near Infrared Spectroscopy Imaging (NIRS) (KENT SnapShot https://www.kentimaging.com/product/) will be used to assess wound oxygenation saturation, oxy, and deoxyhemoglobin levels prior to the MNB or normal saline application. This will provide us with a baseline oxygen tension measurement. The NIRS KENT SnapShot is an FDA approved non-contact-based imaging modality used to assess wound/tissue oxygenation in the clinical setting and is currently available in our research laboratory.
3. The MNB and normal saline solutions will be used in wet-to-dry wound care dressings with daily scheduled dressing changes. Wound oxygen measurements will be recorded using Near Infrared Spectroscopy Imaging (NIRS) twice daily for both the experimental and control groups. In addition, photographs will be taken daily to assess wound healing progress.
4. With each dressing change, a non-traumatic vidal curette will be used to collect wound exudate in both groups, and a pH strip will be used to measure the pH. The proteins will then be extracted by standard methods, and ELISA kits will be used to assess cytokines, chemokines, and MMPs.

Specific Aim 2: Utilize MNBs as an installation solution in NPWTi (VERAFLO) to improve wound oxygenation and healing in chronic wounds.

Hypothesis: Chronic wounds treated with NPWTi and MNB irrigation showed improved tissue oxygenation and healing when compared to wounds treated with NPWTi and irrigated with normal saline.

Study Design:

1. Chronic wounds that require NPWTi will be identified by the practitioners listed on this IRB based on the selection criteria.
2. Baseline oxygen assessment: Near Infrared Spectroscopy Imaging (NIRS) (KENT SnapShot https://www.kentimaging.com/product/) will be used to assess wound oxygenation saturation, oxy, and deoxyhemoglobin levels prior to the NPTWi/MNB or NPTWi/saline application. This will provide a baseline wound oxygen measurement. The NIRS KENT SnapShot is an FDA approved non-contact-based imaging modality used to assess wound/tissue oxygenation in the clinical setting and is currently available in our research laboratory.
3. NPWTi with MNB or NPWTi with normal saline will be applied to the wound with 20-minute instillation periods every 3 hours (standard instillation settings).
4. Near Infrared Spectroscopy Imaging (NIRS) will be used to assess oxygen at each wound vacuum dressing change every 3 days in both treatment branches.
5. Photographs will be taken daily to assess wound healing progress.
6. With each dressing change, a non-traumatic vidal curette will be used to collect wound exudate in both groups, and a pH strip will be used to measure the pH. The proteins will then be extracted by standard methods, and ELISA kits will be used to assess cytokines, chemokines, and MMPs.

4. Describe the primary outcome variable(s), secondary outcome variables, and predictors and/or comparison groups as appropriate for the stated study objectives/specific aims.

Primary outcomes:

1. Wound oxygenation measurements using NIRS (KENT SnapShot Imaging) to make comparisons to baseline (pre-treatment) levels and prior readings.
2. Assess ischemia reversal and circulatory recovery following MNB treatment vs normal saline over a period of time.
3. Wound size/area analysis using daily photographs taken before initiation of treatment and during treatment.
4. Analysis of wound chemical properties, including cytokines (e.g. VEGF, TGF-B, etc.), proteases (e.g. MMP9), and pH in both groups before and after treatment. These will be measured at each dressing change using the procedure described in the Study Design Section.

Secondary outcomes:

1. Readmission to the hospital for the same wound
2. Length of hospital stay
3. Return to the operating room
4. Need for surgical debridement

5. List up to ten relevant references/articles to support the rationale for the research. Do not append an extensive NIH-grant-style bibliography.

1. Sayadi LR, Banyard DA, Ziegler ME, et al. Topical oxygen therapy & micro/nanobubbles: a new modality for tissue oxygen delivery. *Int Wound J.* 2018;15(3):363-374.
2. Tsuge H. *Micro- and Nanobubbles: Fundamentals and Applications.* Boca Raton: CRC Press; 2014.
3. Khan MS, Hwang J, Lee K, et al. Oxygen-Carrying Micro/Nanobubbles: Composition, Synthesis Techniques and Potential Prospects in Photo-Triggered Theranostics. *Molecules.* 2018;23(9).
4. Lalezari S, Lee CJ, Borovikova AA, et al. Deconstructing negative pressure wound therapy. *Int Wound J.* 2017;14(4):649-657.
5. Back DA, Scheuermann-Poley C, Willy C. Recommendations on negative pressure wound therapy with instillation and antimicrobial solutions - when, where and how to use: what does the evidence show? *Int Wound J.* 2013;10 Suppl 1:32-42.
6. Matiasek J, Djedovic G, Kiehlmann M, Verstappen R, Rieger UM. Negative pressure wound therapy with instillation: effects on healing of category 4 pressure ulcers. *Plastic and Aesthetic Research.* 2018;5.
7. Jarbrink K, Ni G, Sonnergren H, et al. The humanistic and economic burden of chronic wounds: a protocol for a systematic review. *Syst Rev.* 2017;6(1):15.

SECTION 2: ROLES AND EXPERTISE OF THE STUDY TEAM

1. **List the Lead Researcher and Co-Researchers who will engage in human subject research.** *Co-Researchers are faculty, staff, students and other academic appointees who the Lead Researcher (LR) considers to be key personnel for conducting the research study. These individuals work closely with the LR to design, conduct, and/or report on the research.*
2. ***UPDATED!*** **List Research Personnel as required per the Research Personnel Heat Map.**
3. **In lieu of listing Research Personnel (as required per the Research Personnel Heat Map), the LR must maintain the Study Team Tracking Log (or something similar) listing all Research Personnel who are engaged in the research.**
4. For each research team member, indicate all applicable research activities the individual will perform. *Finalizing informed consent is reviewing, answering/asking questions, confirming competency, as necessary, and signing/confirming the informed consent.*
5. If applicable, list the Faculty Sponsor as a Co-Researcher who will have research oversight responsibilities.

Lead Researcher:

Name and Degree: Raj Vyas, MD, FACS

Position/Title and Department: Attending, Dept of Plastic Surgery, University of California, Irvine

Team Member will: **[x]** Screen/Recruit **[x]** Finalize Informed Consent

[x] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Oversee entire project, coordinate CTE manufacture of MNBs, oversee presentation and publications. Involved in selecting patients based on inclusion and exclusion criteria outlined in this study. Oversee clinical efficacy and safety of the study while recruiting and consenting appropriate patients for the study.

Co-Researcher:

Name and Degree: Alan Widgerow, MBBCh, MMed, FCS, FACS

Position/Title and Department: Adjunct Professor, Director Center for Tissue Engineering UCI Dept of Plastic Surgery, School of Medicine

Team Member will: **[ ]** serve as Faculty Sponsor with research oversight responsibilities

[x] Screen/Recruit **[x]** Finalize Informed Consent

[x] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Involved in selecting patients based on inclusion and exclusion criteria outlined in this study. Oversee clinical efficacy and safety of the study while recruiting and consenting appropriate patients for the study.

Co-Researcher:

Name and Degree: Garrett A Wirth, MD

Position/Title and Department: Prof. of Plastic Surgery, Dept of Plastic Surgery, University of California, Irvine

Team Member will: **[x]** Screen/Recruit **[x]** Finalize Informed Consent

[x] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Involved in selecting patients based on inclusion and exclusion criteria outlined in this study. Oversee clinical efficacy and safety of the study while recruiting and consenting appropriate patients for the study. Oversee wound dressing changes as outlined in the protocol study procedure.

Co-Researcher:

Name and Degree: Mark Robert Kobayashi, MD FACS

Position/Title and Department: Attending, Dept of Plastic Surgery, University of California, Irvine

Team Member will: **[x]** Screen/Recruit **[x]** Finalize Informed Consent

[x] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Involved in selecting patients based on inclusion and exclusion criteria outlined in this study. Oversee clinical efficacy and safety of the study while recruiting and consenting appropriate patients for the study.

Co-Researcher:

Name and Degree: Cathy Joyce Tang, MD

Position/Title and Department: Assistant Prof., Dept of Plastic Surgery, University of California, Irvine

Team Member will: **[x]** Screen/Recruit **[x]** Finalize Informed Consent

[x] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Involved in selecting patients based on inclusion and exclusion criteria outlined in this study. Oversee clinical efficacy and safety of the study while recruiting and consenting appropriate patients for the study. Oversee wound dressing changes as outlined in the protocol study procedure.

Co-Researcher:

Name and Degree: Amber Rachel Coffey-Leis, MD FACS

Position/Title and Department: Attending, Dept of Plastic Surgery, University of California, Irvine

Team Member will: **[x]** Screen/Recruit **[x]** Finalize Informed Consent

[x] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Involved in selecting patients based on inclusion and exclusion criteria outlined in this study. Oversee clinical efficacy and safety of the study while recruiting and consenting

appropriate patients for the study.

Co-Researcher:

Name and Degree: Eric D. Wang, MD

Position/Title and Department: Assistant Professor, Dept of Plastic Surgery, University of California, Irvine

Team Member will: **[x]** Screen/Recruit **[x]** Finalize Informed Consent

[x] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Involved in selecting patients based on inclusion and exclusion criteria outlined in this study. Oversee clinical efficacy and safety of the study while recruiting and consenting appropriate patients for the study. Oversee wound dressing changes as outlined in the protocol study procedure.

Co-Researcher:

Name and Degree: Jagmeet S. Aurora, BS

Position/Title and Department: Medical Student, University of California, Irvine – School of Medicine

Team Member will: **[x]** Screen/Recruit **[ ]** Finalize Informed Consent

[x] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Assist with subject recruitment, research procedures, statistical analysis of data, and thus will have access to subjects' identifiable information.

Co-Researcher:

Name and Degree: Jason Pham, BS

Position/Title and Department: Research Fellow, Dept of Plastic Surgery, University of California, Irvine

Team Member will: **[x]** Screen/Recruit **[ ]** Finalize Informed Consent

[x] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Assist with subject recruitment, research procedures, statistical analysis of data, and thus will have access to subjects' identifiable information.

Co-Researcher:

Name and Degree: Nima Khoshab, MD

Position/Title and Department: PGY-1, Dept of Plastic Surgery, University of California, Irvine

Team Member will: **[x]** Screen/Recruit **[x]** Finalize Informed Consent

[x] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Involved in selecting patients based on inclusion and exclusion criteria outlined in this study. Oversee clinical efficacy and safety of the study while recruiting and consenting appropriate patients for the study. Oversee wound dressing changes as outlined in the protocol study procedure.

Co-Researcher:

Name and Degree: Ekaterina Tiourin, MD

Position/Title and Department: Attending, Dept of Plastic Surgery, University of California, Irvine

Team Member will: **[x]** Screen/Recruit **[x]** Finalize Informed Consent

[x] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Involved in selecting patients based on inclusion and exclusion criteria outlined in this study. Oversee clinical efficacy and safety of the study while recruiting and consenting appropriate patients for the study. Oversee wound dressing changes as outlined in the protocol study procedure.

Co-Researcher:

Name and Degree: Mikhail Kunal Pakvasa, MD

Position/Title and Department: PGY-1, Dept of Plastic Surgery, University of California, Irvine

Team Member will: **[x]** Screen/Recruit **[x]** Finalize Informed Consent

[x] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Involved in selecting patients based on inclusion and exclusion criteria outlined in this study. Oversee clinical efficacy and safety of the study while recruiting and consenting appropriate patients for the study. Oversee wound dressing changes as outlined in the protocol study procedure.

Co-Researcher

Name and Degree: Brock Lanier, MD

Position/Title and Department: Attending, Dept of Plastic Surgery, University of California, Irvine

Team Member will: **[ ]** serve as Faculty Sponsor with research oversight responsibilities

[X] Screen/Recruit **[X]** Finalize Informed Consent

[X] Perform Research Activities *(describe below)* **[X]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Involved in selecting patients based on inclusion and exclusion criteria outlined in this study. Oversee clinical efficacy and safety of the study while recruiting and consenting appropriate patients for the study.

Co-Researcher

Name and Degree: Lohrasb Ross Sayadi, MD

Position/Title and Department: Resident, Plastic Surgery, University of California, Irvine

Team Member will: **[ ]** Screen/Recruit **[X]** Finalize Informed Consent

[X] Perform Research Activities *(describe below)* **[X]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Lead scientist involved in conducting animal studies and building MNB generation device. Over 4 years of dedicated research experience. Will be responsible for MNB generation, treatment, and analysis of wounds meeting inclusion criteria for the study.

Co-Researcher

Name and Degree: Ruben A Castro MD

Position/Title and Department: Resident, Plastic Surgery, University of California, Irvine

Team Member will: **[ ]** Screen/Recruit **[X]** Finalize Informed Consent

[X] Perform Research Activities *(describe below)* **[X]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Veteran and physician with extensive experience in the treatment of chronic wounds in the battlefield. Will be responsible for MNB generation, treatment, and analysis of wounds meeting inclusion criteria for the study.

Co-Researcher

Name and Degree: Omotayo Arowojolu MD, PhD

Position/Title and Department: Resident, Plastic Surgery, University of California, Irvine

Team Member will: **[ ]** Screen/Recruit **[X]** Finalize Informed Consent

[X] Perform Research Activities *(describe below)* **[X]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Physician scientist with expertise in molecular and cellular biology, signal transduction, pharmacology and their applications to skin and soft tissue survival and stress responses. Will be responsible for identification, treatment, and analysis of wounds meeting inclusion criteria.

Co-Researcher

Name and Degree: Leonardo Alaniz, BBA

Position/Title and Department: Medical Student (Year 2), University of California, Irvine

Team Member will: **[x]** Screen/Recruit **[ ]** Finalize Informed Consent

[X] Perform Research Activities *(describe below)* **[x]** Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications

(training, experience): Serve as the research administrative point of contact. This entails filing the IRB, translating the informed consent form, obtaining FDA IND, and ensuring that research guidelines are met. Additionally, he will assist with subject recruitment, research procedures, statistical analysis of data, and thus will need to have access to subjects' identifiable information.

Co-Researcher:

Name and Degree: Gabrielle Labove, MD

Position/Title and Department: Resident (PGY2), Plastic Surgery, University of California, Irvine

Team Member will: [ ] Screen/Recruit [X] Finalize Informed Consent

[X] Perform Research Activities (describe below) [ ] Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Assist in consenting patients (where applicable) and application of irrigation or dressings when necessary.

Co-Researcher:

Name and Degree: Jennifer Fligor, MD

Position/Title and Department: Resident (PGY2), Plastic Surgery, University of California, Irvine

Team Member will: [ ] Screen/Recruit [X] Finalize Informed Consent

[X] Perform Research Activities (describe below) [ ] Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Assist in consenting patients (where applicable) and application of irrigation or dressings when necessary.

Co-Researcher:

Name and Degree: Seung Ah Lee, MD

Position/Title and Department: Resident (PGY3), Plastic Surgery, University of California, Irvine

Team Member will: [ ] Screen/Recruit [X] Finalize Informed Consent

[X] Perform Research Activities (describe below) [ ] Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Assist in consenting patients (where applicable) and application of irrigation or dressings when necessary.

Co-Researcher:

Name and Degree: Daniel Gardner, MD

Position/Title and Department: Resident (PGY3), Plastic Surgery, University of California, Irvine

Team Member will: [ ] Screen/Recruit [X] Finalize Informed Consent

[X] Perform Research Activities (describe below) [ ] Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Assist in consenting patients (where applicable) and application of irrigation or dressings when necessary.

Co-Researcher:

Name and Degree: Pauline Santos, MD

Position/Title and Department: Resident (PGY3), Plastic Surgery, University of California, Irvine

Team Member will: [ ] Screen/Recruit [X] Finalize Informed Consent

[X] Perform Research Activities (describe below) [ ] Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Assist in consenting patients (where applicable) and application of irrigation or dressings when necessary.

Co-Researcher:

Name and Degree: Lauren Patty, MD

Position/Title and Department: Resident (PGY4), Plastic Surgery, University of California, Irvine

Team Member will: [ ] Screen/Recruit [X] Finalize Informed Consent

[X] Perform Research Activities (describe below) [ ] Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Assist in consenting patients (where applicable) and application of irrigation or dressings when necessary.

Co-Researcher:

Name and Degree: Shadi Lalezari, MD

Position/Title and Department: Resident (PGY4), Plastic Surgery, University of California, Irvine

Team Member will: [ ] Screen/Recruit [X] Finalize Informed Consent

[X] Perform Research Activities (describe below) [ ] Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Assist in consenting patients (where applicable) and application of irrigation or dressings when necessary.

Co-Researcher:

Name and Degree: Emily Borsting, MD

Position/Title and Department: Resident (PGY4), Plastic Surgery, University of California, Irvine

Team Member will: [ ] Screen/Recruit [X] Finalize Informed Consent

[X] Perform Research Activities (describe below) [ ] Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Assist in consenting patients (where applicable) and application of irrigation or dressings when necessary.

Research Personnel:

Name and Degree: Mary Ziegler, PhD

Position/Title and Department: Chief Project Scientist, Center for Tissue Engineering, Department of Plastic Surgery, University of California, Irvine

Team Member will: [ ] Screen/Recruit [ ] Finalize Informed Consent

[X] Perform Research Activities (describe below) [ ] Access subject identifiable data

List the research activities/procedures to be performed and the individual's relevant qualifications (training, experience): Chief scientist at the center for tissue engineering will oversee data analysis.

SECTION 3: SUBJECT POPULATION(S) (INDIVIDUALS/RECORDS/BIOSPECIMENS)

A. Subjects To Be Enrolled on this UCI protocol (Persons/Records/Biospecimens)

1. Complete the table of subject enrollments below. *Include additional rows for subject category/group, as needed.*
2. If the study involves the use of existing records or biospecimens, specify the maximum number to be reviewed/collected, and the number needed to address the research question.

Category/Group (e.g., adults, controls, parents, children)	**Age Range** (e.g., 7-12, 13–17, adults)	**Maximum Number to be Consented or Reviewed/Collected** (include withdrawals and screen failures)	**Number Expected to Complete the Study *or Needed to Address the Research Question***
Adults	>18 year of age	40	26

Total: 40	

B. Overall Study Sample Size

If this is a multi-site study, provide the total number of subjects to be enrolled from all sites.

[x] Not applicable: This study will only take place at UCI, and does not involve other sites.

Total number of subjects across all sites: <Type here>

C. Eligibility Criteria

1. Identify the criteria for inclusion and exclusion.

Criteria for inclusion:

Specific Aim 1: Acutely Ischemic Tissue

- Traumatic wounds with skin defects deemed suitable for healing by secondary or tertiary intention.
- Surgical wounds or reconstructions with threatened ischemic skin changes including split and full thickness skin grafts, local flaps, free flaps, and tissue displaying circulatory insufficiency. Surgical wounds complicated by dehiscence, epidermolysis, and necrosis.
- Upper and lower extremity wounds with injury to nerve, tendon, and vascular structures requiring coaptation or microsurgical replantation of any of the mentioned structures, specifically wounds deemed vascularly compromised after revascularization.
- Surgical or traumatic with compromised tissue graft; where xenograft, allograft, or autograft sourced tissue were used for the purpose of tissue regeneration and/or revascularization.
- Patient must be a current inpatient at UCI Health.

Specific Aim 2: Chronic Ischemic Tissue

- Decubitus pressure ulcers (stage 2 and above), diabetic foot ulcers (stage 2 and above), venous insufficiency ulcers, arterial ulcers, and neuropathic skin ulcers.
- Cutaneous thermal, chemical, and electrical burn injuries requiring hospital admission for acute management of the burn injury.
- Chronic wounds complicated by or directly related to radiotherapy related tissue injury.
- Patient must be a current inpatient at UCI Health.

Criteria for exclusion:

- Wounds associated with malignancy in the wound bed.
- Grossly infected wounds.
- Wounds with exposed vital structures such as nerves, arteries, and veins.

Wounds which can be managed in the outpatient setting.

2. If eligibility is based on age, gender, pregnancy/childbearing potential, social/ethnic group, or language spoken (e.g., English Speakers only), provide a scientific rationale.

[x] Not applicable: Subject eligibility is not based on these factors.

<Type here>

3. If American Indian or Alaska Native Tribes will be included in the research:
   a. Specify the name of the Tribe and
   b. Specify whether there is Tribal Law that may be applicable to this research and that provides additional protections for subjects (i.e., additional information to be disclosed in the consent process).

[x] Not applicable: American Indian or Alaska Native Tribes are not included in the research.

<Type here>

D. PRE-SCREENING AND DETERMINING ELIGIBILITY WITHOUT INFORMED CONSENT

1. **IMPORTANT NOTES:**
   a. This section is **Not applicable** to research that is funded/supported by the Department of Justice (**DOJ**)
   b. This section addresses pre-screening activities that are performed **without the written informed consent of the prospective subject or legally authorized representative (LAR).** This may be allowed without requesting a waiver of informed consent **IF the following guidelines are utilized**:

[ ] Not applicable: Information and/or biospecimens will not be obtained for the purpose of screening, recruiting, or determining eligibility of prospective subjects. *Skip to Section 4.*

[ ] Study team will obtain information through **oral or written communication** with the prospective subject or LAR (i.e. self-report of medical information; medical records will not be screened).

*Submit recruitment script/s for IRB approval. Be sure to address minimum recruitment requirements and address **the following guidelines**:*

i. Privacy: The script must address the case where someone other than the potential subject receives the communication. Please be mindful of privacy considerations (i.e., do not

disclose any private information – such as a patient diagnosis). Limit phone contact / messages to no more than 5 attempts.

ii. *Expertise: Study team member/s contacting potential subject must be knowledgeable and able to answer questions related to the screening and the main study.*

iii. *Specific Information: Include a description of the information and/or biospecimens that will be obtained for the purpose of screening, recruiting, or determining eligibility and the reasons for performing the screening tests.*

iv. *Confidentiality: Include a statement that informs the potential subject that if they are not eligible to participate in the study that the identifiable information and / or biospecimens will not be used for research purposes and will be destroyed at the earliest opportunity consistent with conduct of the research.*

[x] Study team will **screen medical records** to determine subject eligibility.

Complete Appendix T to request a partial waiver of HIPAA Authorization.

[ ] Study team will **screen medical records** to determine subject eligibility **under IRB approved screening protocol.** Specify HS#: <Type here>

[ ] Study team will **screen non-medical records** (i.e., student records) to which they have access to determine subject eligibility. Specify: <Type here>

[ ] For research accessing student records, check here to confirm that evidence of FERPA[1] compliance has been / will be obtained (and on file) from the local school/district site or the UCI Registrar prior to the initiation of research.

[ ] Study team will **access stored identifiable biospecimens**.

2. For studies that will **screen medical records**, explain how the study team will access the clinical data. *Access to UCI Medical Center medical records for research purposes outside the capacity of the Honest Broker Services, such as access to physician notes, must be obtained from the Health Information Management Services.*

[1] *34 CRF 99: Family Educational Rights and Privacy Act (FERPA) applies to this research.*

[ ] Not applicable: This study does not involve the screening of medical records.

How Obtained: Indicate <u>all</u> that apply:

[ ] The study team will request specific patient information/data from UCIMC Health Information Management Services.

[x] The study team will review their UCI patients' records and abstract data directly from those records.

[ ] The study team will request specific patient information/data from UCI Health Honest Broker Services. Describe the following:

Cohort selection criteria (e.g., use the available Clinical Terms from the Cohort Discovery Tool such as Demographics: Gender, Diagnoses: Asthma, Procedures: Operations on digestive system): <Type here>

Expected cohort size/patient count: <Type here>

Cohort attributes or data elements (e.g., lab test values, medication, etc.): <Type here>

[ ] The study team will review non-UCI Health records and abstract data directly from those records. Describe the following:

Specify the non-UCI Health records that will be screened: <Type here>

Explain how the study team has access to this clinical data: <Type here>

[ ] Other; explain: <Type here>

3. For studies that will **screen existing biospecimens**:
    a. Indicate the source of the biospecimens and explain how the existing biospecimens will be obtained.
    b. Indicate whether the biospecimens were originally collected for research purposes.

[x] Not applicable: This study does not screen existing biospecimens.

How Obtained: Indicate <u>all</u> that apply:
[ ] UCI Health Pathology Biorepository

[ ] Other UCI-Health Entity; specify: <Type here>

[ ] Non-UCI Entity; specify: <Type here>

[ ] Other; explain: <Type here>

Originally collected for research purposes:
[ ] NO – Please explain: <Type here>

[ ] YES – UCI IRB approval granted under IRB protocol number (i.e. HS#): <Type here>

[ ] YES – Non-UCI IRB approval granted. Confirm **<u>one</u>** of the following:

[ ] A copy of the IRB Approval Notice and Consent Form for the original research collection will be submitted with the IRB application (APP). The IRB Approved Consent Form does not preclude the proposed activity.

[ ] A copy of the commercial Vendor Policy or a Letter from the Vendor attesting that the information was collected and will be shared in an appropriate and ethical manner will be submitted with the APP. The vendor's policy does not preclude the proposed activity.

<u>SECTION 4</u>: RECRUITMENT METHODS

Check any of the following methods that will be used to recruit subjects for this study:

[ ] Not applicable: This study involves no direct contact with subjects (i.e., use of existing records, charts, biospecimens).

Specify database or IRB-approved protocol number (HS#), if applicable: <Type here>

[ ] Advertisements, flyers, brochures, email, Facebook, and/or other media.

Specify where recruitment materials will be posted: <Type here>

If subjects will be recruited by mail, e-mail, or phone, specify how their contact information will be obtained: <Type here>

Submit recruitment materials for IRB approval.

[x] The study will be listed on Clinicaltrials.gov. *All Applicable Clinical Trials must be registered.*
[ ] The study will be listed on the Center for Clinical Research (CCR) Find a Trial web page. *This webpage is for UCI School of Medicine departments as well as the clinical research conducted at the Chao Family Comprehensive Cancer Center and the Alpha Stem Cell Center.*
[ ] The study will be listed on the UC Irvine Health Clinical Trials web page.  *Submit the UCIMC Standard Research Recruitment Advertisement for IRB approval.*
[ ] The UCI Social Sciences Human Subjects Lab/Sona Systems will be used.  *Ensure that all applicable consent documents include reference to use SONA.*
[ ] Referral from colleagues • Study team will provide colleagues with UCI IRB-approved recruitment materials for distribution to potential subjects (e.g., recruitment flyer, introductory letter); • An IRB-approved recruitment letter will be sent by the treating physician. The letter will be signed by the treating physician and sent to his/her patients to inform them about how to contact study team members; and/or • Colleagues obtain permission from interested patient to release contact information to researchers. • Study team does not have access to patient names and addresses for mailing. • If colleagues will screen their patients' medical records to determine subject eligibility and approach patients directly about study participation: *Complete Appendix T to request a partial waiver of HIPAA Authorization.*  *Submit recruitment materials for IRB approval.*
[ ] Study team will contact potential subjects who *have given prior permission to be contacted* for research studies. Specify when and how these individuals granted permission for future contact: <Type here> Specify database or IRB-approved protocol number (HS#): <Type here>
[x] Study team members will approach their own patients, students, employees for participation in the study.
[ ] Other Recruitment Methods: <Indicate the recruitment method(s) here>

SECTION 5: INFORMED CONSENT PROCESS

A. Methods of Informed Consent

1. Indicate all applicable informed consent methods for this study. *Submit the consent/assent document(s) with your e-IRB Application (e.g., Study Information Sheet, Recruitment script, Consent Form, etc.). Only IRB approved consent forms (containing the IRB approval footer) may be used to consent human subjects at UCI.*

[x] Signed informed consent will be obtained from subjects. Signed informed consent and/or parental permission will be obtained from subjects, as applicable.

[ ] Requesting a waiver of signed informed consent. Signed consent will not be obtained; consent will be **obtained verbally or via the web**. Informed consent, parental permission and/or child assent will be obtained from subjects, as applicable.

Complete Appendix P.

[ ] Requesting to seek surrogate consent from the subjects' LAR. Surrogate consent may be considered only in research studies relating to the cognitive impairment, lack of capacity or serious or life-threatening disease and conditions of the research subjects.

Complete Appendix E.

[ ] Requesting a waiver of the consent process. Consent will not be obtained. *Skip to Section 5.B.*

Complete Appendix O.

2. Indicate where the consent process will take place.

[x] In a private room
[ ] In a waiting room
[ ] In an open unit
[ ] In a group setting
[ ] The internet
[ ] In public setting
[ ] Over the phone
[ ] Other (specify): <Type here>

3. Specify how the research team will assure that subjects or their LAR have sufficient time to consider whether to participate in the research.

[ ] Subjects or their LAR will be allowed to take home the unsigned consent form for review prior to signing it.

[x] Subjects or their LAR will be allowed 24 hours to consider whether to consent.

[ ] Other (specify): <Type here>

4. If children are enrolled in this study, describe the parental permission process and the child assent process.

[x] Not applicable: Children are not enrolled in this study.

<Type here>

5. Some subjects may be vulnerable to coercion or undue influence, such as those who are economically or educationally disadvantaged, have impaired decision-making capacity, or students (undergraduate, graduate, and medical students) and employees of UCI (administrative, clerical, nursing, lab technicians, post-doctoral fellows and house staff, etc.), describe the procedures to ensure the voluntary participation of these individuals.

[ ] Not applicable: Subjects are not vulnerable to coercion or undue influence.

[x] Other (specify): The potential subjects will be informed that no matter what they decide regarding participation, their care received from their physician will not be affected.

B. Health Insurance Portability and Accountability Act (HIPAA) Authorization

Indicate all applicable HIPAA authorization methods for this study.

[ ] Not applicable: Study does not involve the creation, use, or disclosure of Protected or Personal Health Information (PHI).

[ ] Requesting a Total waiver of HIPAA Authorization. HIPAA authorization will not be obtained at all for the study.

Complete Appendix T.

[x] Requesting a Partial waiver of HIPAA Authorization. HIPAA authorization will not be obtained for screening/recruitment purposes. However, written (signed) HIPAA research authorization is obtained for further access to personal health information.

Complete Appendix T.

[x] Written (signed) HIPAA Research Authorization will be obtained from subjects. Signed authorization, parental authorization, and/or child assent will be obtained from subjects or their LAR, as applicable.

Complete the HIPAA Research Authorization form.

C. Methods of Informed Consent for non-English Speakers

1. Indicate the applicable informed consent method for non-English speakers.

[ ] **Not applicable**: Only individuals who can read and speak English are eligible for this study. *Scientific justification must be provided in Section 3.C.2.*

[x] The English version of the consent form will be translated into appropriate languages for non-English speaking subjects or their LAR once IRB approval is granted. *The translated consent form must be submitted to the IRB for review prior to use with human subjects. Only IRB approved consent forms (containing the IRB approval stamp) may be used to consent human subjects at UCI.*

[ ] Requesting a short form consent process.


Complete Appendix Q.

The short form process will be used for the following occasional and unexpected languages:

[ ] All non-English languages

[ ] All non-English languages except Spanish

[ ] Other languages (specify): <Type here>

2. Explain how non-English speaking subjects or their LAR will be consented in their language and who will be responsible for interpreting and facilitating the informed consent discussion for the non-English speaking subjects.

[ ] At least one member of the study team is fluent in the language that will be used for communication, and that study team member(s) will be available during emergencies.


For all members of the study team responsible for obtaining informed consent from non-English speaking subjects, provide their qualifications to serve in this capacity (i.e. language fluency) in Section 2.

[x] The study team has 24-hour access to a translation service with sufficient medical expertise to discuss the research in this study.

[ ] Other (explain): <Type here>

SECTION 6: RESEARCH METHODOLOGY/STUDY PROCEDURES

A. Study Location

Specify where the research procedures will take place (e.g. UCI Douglas Hospital – Cardiac Care Unit, UCI Main Campus Hewitt Hall, UCI Health – Pavilion II, UCI Family Health Center, Anaheim, Irvine High School).


If research activities will also be conducted at non-UCI locations (e.g., educational institutions, businesses, organizations, etc.), Complete Appendix A. Letters of Permission or other documentation may be required (e.g. Off-site Research Agreements or IRB Authorization Agreements).

UCI Douglas Hospital and Tower Building

B. Study Design

1. Include an explanation of the study design (e.g., randomized placebo-controlled, cross-over, cross-sectional, longitudinal, etc.) and, if appropriate, describe stratification/ randomization/blinding scheme.

This is a double-blind, controlled study. Participants will be selected and matched based on similar wound profiles, pathology mechanisms, comorbidity profiles, and age. They will randomly be assigned to the experimental (MNB) or control (normal saline) group. The randomization ratio between both groups will be 1:1. Measurements of objective datapoints (e.g. StO2), cytokines, proteases, and pH will be taken by blinded research personnel

2. Provide precise definitions of the study endpoints and criteria for evaluation; if the primary outcomes are derived from several measurements (i.e., composite variables) or if endpoints are based composite variables, then describe precisely how the composite variables are derived.

Endpoints include:
-Wound healing measured by wound area less than 10% of original
-Additional wound measurements: cytokines, proteases, and pH
-Re-operation and debridement
-Infection requiring discontinuation of NPWTi
-Sepsis
-Death from any other cause

C. Research Procedures

1. Provide a detailed chronological description of all research procedures.

1. Medical records will be pre-screened by research personnel to review each potential subject's medical history.
2. Wounds will be evaluated by research personnel to assess for inclusion and exclusion criteria. Only wounds meeting inclusion criteria will be included in the study.
3. Prior to MNB or normal saline treatment initiation Near Infrared Spectroscopy (NIRS), a non-contact based and non-invasive imaging modality, will be used to assess each wound's pretreatment oxygen saturation (StO2), relative oxyhemoglobin level (HbO2), and relative deoxyhemoglobin (Hb) levels.
4. Pretreatment photographs will be taken of each wound and any personal identifying information will be covered and excluded.
5. MNB solutions will be generated at the Center for Tissue Engineering lab at UC Irvine medical center building 55. Oxygen-filled MNBs will be produced in saline solution using a shear generation system based upon a gear pump design. This design first pre-mixes saline and compressed oxygen in a metered fashion, followed by shear generation, and subsequent controlled decompression to produce MNBs. To achieve adequate decompression, a multi-stage decompression nozzle with 0.5 mm and 0.2 μm nozzle elements are used. A 1 L volume of recirculating saline solution is continuously treated in this manner with oxygen for 10 minutes. Post-production, the MNB solutions are sterile-filtered with a 0.2 μm mesh filter followed by 10 minutes of UV radiation. This solution will then be transported by research personnel to UC Irvine medical center. Normal saline will be acquired from UC Irvine medical center.

Specific Aim 1:

a. Absorbent wound dressings (Kelix) will be saturated with MNB or normal saline solutions and wounds will be dressed per standard of care. Dressing will be changed daily by research personnel.
b. NIRS recording is captured every 12 hours to obtain oxygen saturation (StO2), relative oxyhemoglobin level (HbO2), and relative deoxyhemoglobin (Hb) levels.
c. With each dressing change, a non-traumatic vidal curette will be used to collect wound exudate in both groups, and a pH strip will be used to measure the pH. The proteins will then be extracted by standard methods, and ELISA kits will be used to assess cytokine, chemokines, and MMP's.
d. Photographs will be taken every 24 hours, and any personal identity information will be covered.
e. Treatment will be terminated at maximum of 30 days or when the patient is deemed ready to discharge.
f. Patients will be seen for follow-up as necessary, and wounds will be photographed at each visit.

Specific Aim 2:

a. NPWTi will be connected to 1L of MNB saline or plain normal saline solution. Wounds will be dressed with wound vacuum per standard of care with negative pressure at -125 mmHg.
b. MNB or normal saline instillation will take place 20 minutes at every 3 hours. Volume of MNB or normal saline instillation will be assessed individually depending on wound size.
c. NIRS recording will be performed at each wound vacuum change (every 3 days) to obtain oxygen saturation (StO2), relative oxyhemoglobin level (HbO2), and relative deoxyhemoglobin (Hb) levels.
d. With each dressing change, a non-traumatic vidal curette will be used to collect wound exudate in both groups, and a pH strip will be used to measure the pH. The proteins will then be extracted by standard methods, and ELISA kits will be used to assess cytokine, chemokines, and MMP's.
e. Photographs will be taken at every wound vacuum dressing change, and any personal identifying information will be covered and excluded.
f. Treatment will be terminated at maximum of 30 days or when the patient is deemed ready to discharge.
g. Patients will be seen for follow-up as necessary, and wounds will be photographed at each visit.

2. Describe the duration of a subject's participation in the study. If there are sub-studies, include duration of participation in each sub-study.

The duration of participation is 2-4 weeks. Wound therapy will be performed for a maximum of 4 weeks or while the patient is admitted as an inpatient at UCI Medical Center. Patients deemed fit for discharge by attending physicians will not remain hospitalized for the study, but will instead be discharged home and treatment will be terminated.

3. List data collection instruments (e.g., measures, questionnaires, interview questions, observational tool, etc.).

Investigator-authored, non-standardized, or un-validated measures must be submitted for review.

-Near Infrared Spectroscopy Imaging (NIRS) (KENT SnapShot https://www.kentimaging.com/product/) will be used to measure tissue oxygenation.
-Non-traumatic, vidal curettes will be used for exudate collection.
-pH strips will be used to measure pH.
-ELISA kits will be used for assessment of cytokines, chemokines, and MMP's.
-Wound photographs will be collected for wound measurement.

D. UCIMC Supplementary Clinical Services

If a UCIMC clinical unit/department (e.g., phlebotomy for blood draws, pharmacy for dispensing study drug(s), radiation services for X-rays, MRIs, CT scans, and Neurology for lumbar punctures) will perform research-related procedures:

1. List the research procedure (e.g. lumbar puncture, MRI, CT Scan), and
2. Identify the unit/department that will perform the procedure.

[x] Not applicable: This study does not involve the services of a UCIMC clinical unit/department.

<Type here>

E. Privacy

Privacy is about the subject's ability to control how much others see, touch, or collect information about the subject. Indicate all of the following methods that will be used to assure subject privacy. *Violations of privacy include accessing a subject's private information without consent, asking personal sensitive information in a public setting, being audio recorded or photographed without consent.*

[x] Research procedures (including recruitment) are conducted in a private room.

[ ] Use of drapes or other barriers for subjects who are required to disrobe.

[ ] Only sensitive information directly related to the research is collected about subjects.

[ ] When information is collected from internet sources, the internet site's privacy statement will be reviewed and followed.

Provide a copy of the Data Use Policy to the IRB.

[ ] Other (specify): <Type here>

F. Use of Identifiable Private Information and/or Identifiable Biospecimens as Part of the Main Study

1. For studies that will use **existing identifiable biospecimens** as part of the **main study** (not for determining eligibility):
   a. Indicate the source of the biospecimens and explain how the existing biospecimens will be obtained.
   b. Indicate whether the biospecimens were originally collected for research purposes.

[x] Not applicable: This study does not use existing biological specimens as part of the main study.

How Obtained: Indicate <u>all</u> that apply:

[ ] UCI Health Pathology Biorepository

[ ] Other UCI-Health Entity; specify: <Type here>

[ ] Non-UCI Entity; specify: <Type here>

[ ] Other; explain: <Type here>

Originally collected for research purposes:

[ ] NO – Please explain: <Type here>

[ ] YES – UCI IRB approval granted under IRB protocol number (i.e. HS#): <Type here>

[ ] YES – Non-UCI IRB approval granted. Confirm **<u>one</u>** of the following:

[ ] A copy of the IRB Approval Notice and Consent Form for the original research collection will be submitted with the IRB application (APP). The IRB Approved Consent Form does not preclude the proposed activity.

[ ] A copy of the commercial Vendor Policy or a Letter from the Vendor attesting that the information was collected and will be shared in an appropriate and ethical manner will be submitted with the APP. The vendor's policy does not preclude the proposed activity.

2. For studies that will **use identifiable clinical data** as part of the **main study** (not for determining eligibility), indicate the source and how the study team will access the medical records. *Access to UCI Medical Center medical records for research purposes outside the capacity of the Honest Broker Services, such as access to physician notes, must be obtained from the Health Information Management Services.*

For investigator initiated/authored studies <u>only</u>, submit a data abstraction sheet that includes a complete list of data elements/information that will be collected from (existing) records or submit the case report form (CRF; eCRF).

[ ] Not applicable: This study does not involve the use of identifiable clinical data as part of the main study. *Skip to Section 6.G.*

How Obtained: Indicate <u>all</u> that apply:

[ ] The study team will request specific patient information/data from UCIMC Health Information Management Services.

[x] The study team will access their UCI patients' records and abstract data directly from those records.

[ ] The study team will request specific patient information/data from UCI Health Honest Broker Services. Describe the following:

Cohort selection criteria (e.g., use the available Clinical Terms from the Cohort Discovery Tool such as Demographics: Gender, Diagnoses: Asthma, Procedures: Operations on digestive system): <Type here>

Expected cohort size/patient count: <Type here>

Cohort attributes or data elements (e.g., lab test values, medication, etc.): <Type here>

[ ] The study team will request non-UCI Health records and abstract data directly from those records. Describe the following:

Specify the non-UCI Health records that will be screened: <Type here>

Explain how the study team has access to this clinical data: <Type here>

[ ] Other; explain: <Type here>

3. For studies that involve use of existing (i.e. on the shelf; currently available) clinical data, specify the time frame of the clinical data to be accessed (e.g. records from January 2002 to initial IRB approval).

N/A

G. Collection of Photographs, or Audio/Video Recording

1. Describe all procedures involving the use and/or collection of photographs, or audio/video recording.

[ ] Not applicable: This study does not involve photographs or audio/video recording. *Skip to Section 6.H.*

<Type here>

2. Specify if photographs or audio/video recording will include subject identifiable information (e.g., name, facial image). If so, indicate which identifiers will be collected.

Daily assessment of wounds will be captured by photography – names and facial images will not be included or be part of the data set.

3. Explain whether the photographs or audio/video recording will be included in subsequent presentations and/or publications and, if so, whether subject identifiers will be included.

Photographs will be included in presentations and publications, but all identifying information will be excluded.

H. Sharing Results with Subjects

1. Describe whether individual results (results of investigational diagnostic tests, genetic tests, or incidental findings) will be shared with subject or others (e.g., the subject's primary care physician). *Only tests ordered by a physician and conducted in a CLIA certified lab may be shared.*
2. Explain what information will be shared and how the results will be shared.

[x] Not applicable: Individual results will not be shared with subjects.

<Type here>

3. Describe whether overall study results will be shared with subjects.
4. Explain how results will be shared.

[x] Not applicable: Final study results will not be shared with subjects.

[ ] The overall study results will be listed on Clinicaltrials.gov. *All Applicable Clinical Trials must be registered.*

[ ] Other: <Type here>

I. Statistical Considerations *(This section is required for Investigator-Authored Research)*

1. Statistical Analysis Plan: Describe the statistical method(s) for the stated specific aims and hypotheses.

   The Biostatistics, Epidemiology and Research Design (BERD) Unit under the Institute for Clinical and Translational Science (ICTS) can assist in developing power and sample size calculation. Visit: http://www.icts.uci.edu/services/berd%20request.php *for a consultation.*

   Your analysis plans should match the stated study specific aims and hypotheses in Section 1.

   For cancer related research: The Chao Family Comprehensive Cancer Center's Biostatistics Shared Resource (BSR) assists investigators with the design of new studies, power and sample size calculations, and data analyses. To request BSR support, visit: https://www.cancer.uci.edu/biostatistics/consultation.asp"

[ ] Not applicable: A statistical analysis plan is not appropriate for this qualitative study design. Plan for assessing study results:
Skip to Section 7.

<Type here>

2. Describe the primary statistical method(s) that will be used to analyze the primary outcome(s) or endpoints.

The primary outcome is wound oxygenation and healing. The following variables will be used for assessment of the primary outcome: oxygen saturation (stO2), relative oxy/de-oxyhemoglobin levels, cytokine/protease concentrations, and re-epithelization. Two-tailed t-tests will be used for evaluation of each variable in the experimental vs. control groups. A significance of <0.05 has been selected for this trial.

3. Describe the secondary statistical method(s) that will be used to analyze the secondary outcome(s) or endpoints.

Chi-square analysis will be used to compare qualitative variables (coded as present or absent) between both groups: readmission for the same wound, return to the OR, and need for surgical debridement. Mann-Whitney U-Tests will be used to compare length of hospital stay (a non-normal quantitative variable) between groups.

4. If appropriate describe secondary or post hoc analyses of primary outcome(s) or other exploratory analysis.

<Type here>

5. Sample Size Determination**:** Explain how the overall target sample size was determined (e.g., power analysis; precision estimation), providing justification of the effect size for the primary outcome based on preliminary data, current knowledge/literature and/or cost consideration; if appropriate, provide sample size justification for secondary outcomes. Power analysis should (at least) match the primary outcome/endpoint.

Sample size was determined by a power analysis to match the primary outcome: wound oxygenation and healing. Existing literature on animal models has demonstrated up to a 47.11% difference in reduced scattering (i.e. improved collagen organization) – (control mean: 1.032 mm^{-1}, control standard deviation: 0.1715 mm^{-1}, test mean: 1.668 mm^{-1}, test standard deviation: 0.1596 mm^{-1}). When assessing wound healing, animal studies showed that wounds treated with MNB's healed as early as 18 days vs. wounds treated with normal saline, which still had not completely healed. Using this existing data, power analysis results state that n=13 in each group is sufficient sample size given a 0.9 power.

SECTION 7: RISK ASSESSMENT AND POSSIBLE BENEFITS

A. Risk Assessment

1. Indicate the appropriate level of review of this study, based upon your risk assessment.

[x] This study involves greater than minimal risk to subjects and requires Full Committee review. *Skip to Section 7.B.*

[ ] This study involves no more than minimal risk and qualifies as **Expedited research**.

2. If this study involves no more than minimal risk, provide justification for the level of review and for all applicable Expedited Categories you have chosen.

-

B. Risks and Discomforts

1. Describe and assess any reasonably foreseeable risks and discomforts — physical, psychological, social, legal or other. Include an assessment of their expected frequency (e.g., common – 65%, less common – 40%, unlikely – 5%, rare - <1%) and the seriousness (mild, moderate, severe). *A bullet point list is recommended. If this study will involve the collection of identifiable private information, even temporarily, for which the disclosure of the data outside of the research could reasonably place the subjects at risk, include the risk of a potential breach of confidentiality.*

- Infection <1% - equal to the risk of irrigating wounds with saline alone. In fact, irrigation reduces the risk of infection.
- Poor wound healing, < 1% - Same risk as irrigation of wounds with normal saline.
- Scarring < 1% - Same risk as irrigation of wounds with normal saline.

2. Discuss what steps have been taken and/or will be taken to prevent and minimize any risks/ potential discomforts to subjects. *Examples include: designing the study to make use of procedures involving less risk when appropriate; minimizing study procedures by taking advantage of clinical procedures conducted on the subjects; mitigating risks by planning special monitoring or conducting supportive inventions for the study; implement security provisions to protect confidential information.*

Irrigation of the wounds will occur per standard of care as described in the research procedures.

C. Potential Benefits

1. Describe the potential benefits subjects may expect to receive from participation in this study. *Compensation is not a benefit; do not include it in this section.*

[ ] There is no direct benefit anticipated for the subjects.

Patients treated with MNB have the potential to have improved healing and a shorter healing period.

2. Specify the expected potential societal/scientific benefit(s) of this study.

Reversal of acute ischemia, prevention of tissue necrosis, promote healing in chronic wounds (all in a cost-effective way).

SECTION 8: ALTERNATIVES TO PARTICIPATION

Describe the alternatives to participation in the study available to prospective subjects. Include routine (standard of care) options as well as other experimental options, as applicable.

[ ] No alternatives exist. The only alternative to study participation is not to participate in the study.

[x] There are routine standard of care alternatives available; specify: NPWT without instillation, normal saline irrigation of wounds

[ ] There are other alternatives to study participation; specify: <Type here>

SECTION 9: SUBJECT COSTS

1. Indicate below if subjects or their insurers will be charged for study procedures. Identify and describe those costs.

[ ] Not applicable: This study involves no interaction/intervention with research subjects. *Skip to Section 10.*

[x] This study involves interaction/intervention with research subjects; however there are no costs to subjects/insurers.

[ ] This study involves interaction/intervention with research subjects, and there are costs to subjects/insurers: <Type here>

2. If subjects or their insurers will be responsible for study-related costs, explain why it is appropriate to charge those costs to the subjects or their insurers. Provide supporting documentation as applicable (e.g., study procedures include routine (standard of care) procedures; FDA IDE/HDE/IND letter that supports billing to subjects).

[x] Not applicable: The study involves no costs to subjects for study participation.

[ ] Study related costs will be billed to subjects or their insurers for the following reasons: <Type here>

SECTION 10: SUBJECT COMPENSATION AND REIMBURSEMENT

1. If subjects will be compensated for their participation, explain the method/terms of payment (e.g., money; check; extra credit; gift certificate).

[ ] **Not applicable**: This study involves no interaction/intervention with research subjects. *Skip to Section 11.*

[x] No compensation will be provided to subjects.

[ ] Compensation will be provided to subjects in the form of cash/gift certificate.

[ ] Compensation will be provided to subjects in the form of a check issued to the subjects through the UCI Accounting Office. The subject's name, address, and social security number, will be released to the UCI Accounting Office for the purpose of payment and for tax reporting to the Internal Revenue Service (IRS).

[ ] Other: <Type here>

2. Specify the schedule and amounts of compensation (e.g., at end of study; after each session/visit) including the total amount subjects can receive for completing the study. *Compensation should be offered on a prorated basis when the research involves multiple visits.*

   For compensation ≥ $600, subject names and social security numbers must be collected. This information must be reported to UCI Accounting for tax-reporting purposes.

[x] **Not applicable**: This study involves no compensation to subjects.

Subjects will be compensated with the following schedule and amounts: <Type here>

3. Specify whether subjects will be reimbursed for out-of pocket expenses. If so, describe any requirements for reimbursement (e.g., receipt).

[x] **Not applicable**: This study involves no reimbursement to subjects.

Subjects will be reimbursed; specify: <Type here>

SECTION 11: CONFIDENTIALITY OF RESEARCH BIOSPECIMENS/DATA

A. Information and/or Biospecimens Storage

1. Indicate how information and/or biospecimens will be stored and secured. Check all that apply:

[x] Information will be maintained electronically. Information will be password protected and maintained in an encrypted format. *Researchers may access UCI-contracted data sharing and storage tools through UCI OIT.*

[ ] Information will be maintained in hard copy. Information will be stored in a locked area that is not accessible to non-study team members.

[ ] Biospecimens will be stored in a locked lab/refrigerator/freezer that is not accessible to non-study team members.

2. List the location(s) where the data and/or biospecimens will be stored.

UC Irvine Center for Tissue Engineering Building 55 computers - data will be stored on these UCI computers

3. Indicate all subject identifiers that may be retained with the information and/or biospecimens collected for the research study. *If any study-related data will be derived from a medical record, added to a medical record, created or collected as part of health care, or used to make health care decisions the HIPAA policy applies. The subject's HIPAA Research Authorization is required or a waiver of HIPAA Research Authorization must be requested by completing Appendix T.*

[ ] This study does not involve the collection of subject identifiers.

Check all the following identifiers will be used, created, collected, disclosed as part of the research:

[x] Names	**[ ]** Social Security Numbers	**[ ]** Device identifiers/Serial numbers
[x] Dates*	**[x]** Medical record numbers	**[ ]** Web URLs
[x] Postal address	**[ ]** Health plan numbers	**[ ]** IP address numbers
[x] Phone numbers	**[ ]** Account numbers	**[ ]** Biometric identifiers
[ ] Fax numbers	**[ ]** License/Certificate numbers	**[ ]** Facial Photos/Images
[ ] Email address	**[ ]** Vehicle id numbers	**[ ]** Any other unique identifier

[ ] Other (Specify all): <Type here>

* *birth date, treatment/hospitalization dates*

4. Indicate if a code will be used to link subject identifiers with the information and/or biospecimens.

[ ] Not applicable: No subject identifiers will be collected.

[x] A code will be used (i.e. information and/or biospecimens will be coded). Subject **identifiers** will be **kept separately** from the information and/or biospecimens. The code key will be destroyed at the earliest opportunity, consistent with the conduct of this research.

[ ] A code will not be used. Subject **identifiers** will be **kept directly** with the information/biospecimens.

5. If **subject identifiable data** will be transported or maintained on **portable devices**, explain why it is necessary use these devices. *Only the "minimum data necessary" should be stored on portable devices as these devices are particularly susceptible to loss or theft. If there is a necessity to use a portable device for the initial collection of identifiable private information, the research files must be encrypted, and subject identifiers transferred to a secure system as soon as possible.*

[x] Not applicable: Research data will not be transported or maintained on portable devices.

[ ] Research data will need to be maintained on the following portable device(s) for the following reason(s): <Type here>

B. Information and/or Biospecimens Access

1. Specify who will have **access to subject identifiable information and/or identifiable biospecimens** as part of this study. Check all that apply.

[ ] Not applicable: No subject identifiers will be collected.

[x] Authorized UCI personnel such as the research team and appropriate institutional officials, the study sponsor or the sponsor's agents (if applicable), and regulatory entities such as the Food and Drug Administration (FDA), the Office of Human Research Protections (OHRP), and the National Institutes of Health (NIH).

[ ] Other: <Type here>

2. Specify whether subject identifiers be disclosed in presentations and/or publications.

[x] Not applicable: No subject identifiers will be collected.

[ ] Subject identifiers will **not** be disclosed.

[ ] Subject identifiers will be disclosed. Text regarding the disclosure will be included in the consent document and specific permission to disclose will be discussed with subjects.

3. Specify whether **information and/or biospecimens be shared** with other researchers **outside of the study team** (i.e., UCI / non-UCI researchers) for secondary research purposes. *When accessing/transferring data from/to a non-profit, please contact Grace J. Park at parkgj@uci.edu. When accessing/transferring data from/to a for-profit, please contact the Industry Contract Officer at UCI Beall Applied Innovation assigned to your department.*

[ ] Not applicable: information and/or biospecimens will not be shared.

[ ] Identifiable information and/or identifiable biospecimens may be shared. Text regarding the information/specimens sharing will be included in the consent document and specific permission to share information will be discussed with subjects.

Check one of the following:

[ ] A biorepository will be established and manage by the UCI study team. ***Submit Appendix M.***

[ ] Subject identifiers will be retained in an established non-UCI biorepository (i.e. not managed by the UCI study team). The non-UCI biorepository has a current IRB approval on file. Specify the non-UCI biorepository: <Type here>

[x] De-identified information and/or de-identified biospecimens may be shared (i.e. research participants cannot be identified by other researchers). Text regarding the information/biospecimens sharing will be included in the consent document, as applicable.

Check one of the following:

[x] No subject identifiers will be retained by the study team beyond initial collection (i.e. information/biospecimens cannot be linked to an individual and a key code does not exist). Requests for de-identified information and/or de-identified biospecimens will be managed by the UCI study team.

[ ] Subject identifiers will be retained by the study team beyond initial collection (i.e. information/biospecimens can be linked to an individual and/or a key code exists). A biorepository will be established and managed by the UCI study team. ***Submit Appendix M.***

[ ] Subject identifiers will be retained by the study team beyond initial collection (i.e. information/biospecimens can be linked to an individual and/or a key code exists). De-identified information/biospecimens will be retained and managed in an established non-UCI biorepository (i.e. not managed by the UCI study team). The study team will remove any information that could potentially allow for the re-identification of participants prior to sending the information/biospecimens to the non-UCI biorepository. Specify the non-UCI biorepository: <Type here>

[ ] Other: <Type here>

C. Research Information and/or Biospecimens Retention

1. Indicate how long research information and/or biospecimens will be retained.
2. If more than one option applies, indicate accordingly.
3. If research involves Protected Health Information (PHI): Investigators must destroy PHI at the earliest opportunity, consistent with the conduct of this study, unless there is an appropriate justification for retaining the identifiers or as required by law. Otherwise, identifiable data is to be retained as noted below.

UPDATED! In accordance with UCOP policy, information/biospecimens will be retained for 10 years after the end of the calendar year in which the research is completed, unless otherwise specified in the award agreement. Choose the longest retention period applicable to the study:

[ ] There is no contract or award associated with this research. Information/biospecimens will be retained for 10 years after the end of the calendar year in which the research is completed.

[ ] The contract or award associated with this research requires that information/biospecimens be retained for the following period; specify time frame: <Type here>.

[x] The study is conducted under an IND or an IDE investigation, information/biospecimens will be retained for two years after an approved marketing application. If approval is not received, the information/biospecimens will be kept for 2 years after the investigation is discontinued and the FDA is notified per FDA sponsor requirements.

[ ] This research includes the potential for future **secondary research using information/biospecimens** which will be stored and maintained indefinitely.

D. Audio/Video Recordings & Photographs

1. If subject identifiable audio/video recordings will be collected, specify the timeframe for the transcription and/or de-identification.
2. If subject identifiable photographs will be collected, specify the timeframe for de-identification.

UPDATED!

[x] Not applicable: Identifiable audio/video recordings and/or photographs will not be collected.

Transcription:

[ ] Audio/video recordings transcribed; specify time frame: <Type here>

[ ] Audio/video recordings will NOT be transcribed; specify why: <Type here>

De-Identification:

[ ] Subject identifiable audio/video recordings & photographs will be de-identified;

- specify time frame: <Type here>
- specify how (ex. real name replace with pseudonym during transcription; blurred facial features): <Type here>

[ ] Subject identifiable audio/video recordings & photographs will NOT be de-identified; specify why: <Type here>

E. Certificate of Confidentiality

1. Indicate whether a Certificate of Confidentiality (CoC) has been or will be requested.
[x] Not applicable: No CoC has been requested for this study. **[ ] This is a non-NIH funded/supported study. Choose one of the following:** **[ ]** A CoC will be requested for this study. *The CoC application must be submitted to the IRB staff for review after IRB approval.* **[ ]** A CoC has been obtained for this study. *Provide a copy of the CoC Approval Letter.* The expiration date of this CoC is: <Type here> **[ ] This is an NIH funded/supported study** and a CoC will be automatically issued for studies that involve identifiable, private, and sensitive information.
2. Explain in what situations the UCI study team will disclose identifiable private information protected by a CoC.
None