# Clinical and Histological Outcomes of Allogenic Amnion Chorion Membrane in the Healing of Free Gingival Graft Donor Site

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## STUDY PROTOCOL

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Clinical and Histological Outcomes of Allogenic Amnion Chorion Membrane in the Healing of Free Gingival Graft Donor Site

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## STUDY PROTOCOL

# 1. BACKGROUND AND RATIONALE

## 1.1. General Introduction

Free gingival graft (FGG) procedure is commonly conducted to increase the width of keratinized gingiva around teeth and implants. Although FGG procedure has predictable clinical outcomes, morbidities and complications on the donor sites (palate) usually affect patients' willingness to receive the procedure (Griffin et al. 2006). Several alternative materials, such as allogenic dermal matrix or xenogeneic collagen matrix, have been used to prevent these complications, but these substitutes have inferior clinical results to FGG (McGuire and Scheyer 2014; Wei et al. 2000). Since FGG has an irreplaceable position in augmenting keratinized gingiva, clinicians utilize dressings to improve palatal wound healing (Shanmugam et al. 2010; Thoma et al. 2016). Allogenic amnion chorion membrane (ACM) has been used to facilitate wound healing, such as diabetic ulcer (Zelen et al. 2013), chronic wounds(Forbes and Fetterolf 2012) and perforated sinus membrane (Holtzclaw 2015) because this placental membrane retains extracelluar matrix proteins, growth factors and cytokines to promote cell proliferation and control inflammation at the wound (Koob et al. 2013). Additionally, allogenic amnion chorion membrane can be well attached to the wound because of its thin thickness and flexibility. Due to these characteristics, allogenic amnion chorion membrane might be an excellent dressing for palatal wound after harvesting a FGG.

## 1.2. Rationale and justification for the Study

### a. Rationale for the Study Purpose

So far, there is no strong evidence supporting the clinical impact of any dressings or membranes on the healing of the FGG donor site. It is very important to assess the effects of the allogenic amnion chorion membrane on the wound of the FGG donor site given this membrane has the potential to improve wound healing and reduce complications.

### b. Rationale for Materials Selected

The ACM (allogenic amnion chorion membrane, BioXclude®, SNOASIS MEDICAL) is a commercial product and has multiple sizes available. It has been used for tissue healing and tissue regeneration in dentistry. Based on the available evidence, it has the potential to improve wound healing of the FGG donor site. The collagen dressing, CLD (Collagen Patch®, Zimmer), is a common product used for tissue healing in dentistry(Shanmugam et al. 2010). It is selected as a dressing material to cover the wound in the control group.

### c. Rationale for Study Population

The study will recruit subjects who need free gingival graft surgery. The dressings, ACM or CLD, will be placed in the wound of the FGG donor site. These subjects

should be systemically healthy and are able to receive FGG surgery. .

### d. Rationale for Study Design

It is a randomized controlled and split-mouth design study. The randomized controlled trail (RCT) is usually considered the gold standard for a clinical trial and it is able to provide the highest level of evidence within all the study designs (Howick et al. 2011). The split-mouth design is a popular design in oral health research. Two treatments (test and control) are randomly assigned to either the right or left halves of the dentition/palate. This design removes a lot of inter-individual variability from the estimates of the treatment effect given two treatments are performed in the same subject (Lesaffre et al. 2009).

## 2. HYPOTHESIS AND OBJECTIVES

### 2.1. Hypothesis

Null hypothesis:
Healing at the palatal wound covered with an allogenic amnion chorion membrane and the collagen dressing has no significant difference. Alternative hypothesis:
The palatal wound covered with an allogenic amnion chorion membrane heals faster than the palatal wound covered with a collagen dressing. The patients with an allogenic amnion chorion membrane feel less pain and have fewer complications than the patients with a collagen dressing.

### 2.2. Primary Objectives

This study aims to compare an allogenic amnion chorion membrane (BioXclude®, SNOASIS MEDICAL) to a collagen dressing (Collagen Patch®, Zimmer) in the palatal wound healing from the clinical and histological perspectives.

### 2.3. Secondary Objectives

In addition to the outcomes of wound healing, the clinician's feedback with these two materials (ACM and CLD), such as handling and operation time, will also be evaluated.

### 2.4. Potential Risks and Benefits:

### a. End Points - Efficacy

All eligible patients need FGG procedure and will receive the appropriate treatments. The patients may have improved healing or may not have additional clinical benefits by participating in this clinical study.

### b. End Points - Safety

1) Study related risks:
Initial infection, pain, swelling, and bleeding related to FGG procedure and gingival

biopsy; adverse outcomes of the receipt sites; The patients will have the same risks if they receive the same treatment without participating in the study.
2) Protection against risks:
All efforts will be made to minimize risks to all and every participant: only patients with the need for FGG procedure will qualify for the study.

## 3. STUDY POPULATION

### 3.1. List the number of subjects to be enrolled.

The proposed project is a pilot study. We use a convenient sample size based on the budget. We will recruit 19 patients in total, and with a dropout rate of 15%, we will have a sample size of 15.
With 15 patients, we can detect an effect size as small as 0.7 between two treatments for the two main outcomes (the area of healing and VAS pain score at week 2), by a paired t test assuming a within-subject correlation of 0.6 at the significance level of 0.05 with 80% power.

### 3.2. Criteria for Recruitment

The patients who need FGG procedure will be recruited in the study. These patients should be systemically healthy to receive this procedure.

### 3.3. Inclusion Criteria

Nineteen subjects will be recruited among the patients attending the Clinic for Graduate Periodontics, UTSD, who are in need of a FGG procedure to augment keratinized gingiva, augment ridge or cover recession defects. The size of the FGG that the patient needs will not be bigger the size of FGGs that have to be harvested in this study (two 8mm (width) x 10mm (length) x ≈1.5mm (thickness) FGG). All subjects are ≥18 year-old and systemically healthy or with controlled common systemic conditions, such as hypertension, that will not affect wound healing.

### 3.4. Exclusion Criteria

Patients will be excluded if they are current heavy smokers(>10 cigarettes/day), have diabetes or other systemic diseases that may comprise healing, take antibiotics and/or analgesics within one week before the procedure and have loss of sensation on the palate. Patients who stop smoking more than one year are eligible.

### 3.5. Withdrawal Criteria

A subject may be discontinued from participation in the study for any of the following reasons:
1. Withdrawal of consent
2. Subject noncompliance with the protocol, as determined by the investigator
3. Any event or condition that would make continued participation in the study not in the best interest of the subject, as determined by the investigator
4. Pregnancy
5. Development of any medical condition that might affect the treatment and clinical outcomes, as determined by the investigator.
6. Initiation of any treatment or exposure that might affect the healing outcomes of the FGG donor site, as determined by the investigator.

7. Investigator discretion

### 3.6. Subject Replacement

Subjects who withdraw from the study can be replaced. However, to complete the study within the time allocated, the center will not enroll subjects after 24 months from enrollment initiation.

## 4. TRIAL SCHEDULE

There will be eight appointments including the baseline appointment and seven follow-up appointments (4 days, 10 days, 2, 3, 4, 6, 8 weeks) after the surgery. The details of each visit will be mentioned in 6.3. Study Visits and Procedures.

## 5. STUDY DESIGN

### 5.1. Summary of Study Design

This randomized controlled and split-mouth design study aims to compare an allogenic amnion chorion membrane (BioXclude®, SNOASIS MEDICAL) to a collagen dressing (Collagen Patch®, Zimmer) in the palatal wound healing from the clinical and histological perspectives. Nineteen patients will be recruited. The wound healing would be assessed using pictures and questionnaires at multiple time points. A biopsy will be done to evaluate histological healing. Patients will not have additional complications other than the complications from FGG procedure. However, the size of wounds will be larger than the size required to treat defects. The hypothesis is that the patients with an allogenic amnion chorion membrane feel less pain and have fewer complications than the patients with a collagen matrix.
The potential application of the allogenic amnion chorion membrane in palatal wound will benefit patients who receive FGG procedure. The results can be applied in managing various wounds in the oral cavity in the future.

## 6. METHODS AND ASSESSMENTS

### 6.1. Randomization and Blinding

Site allocation (right or left side of the palate) to the two groups (ACM or CLD) will be performed by the investigators before the surgeon harvests FGGs, based on computer-generated randomization (R Statistical Software). Patients will be blinded because they will not know which dressing is in the test group or the control group.

### 6.2. Contraception and Pregnancy Testing

Pregnancy status of subjects who are women with childbearing potential will be orally confirmed at the screening. The pregnant subjects will be excluded from the study.

## 6.3. Study Visits and Procedures

*a. Screening Visits and Procedures*

Study protocol and consent forms will be approved by the Institutional Review Board at the University of Texas Health Science Center at Houston. The trial will be registered with ClinicalTrials.gov. The clinicians in UTSD will be told the information of this clinical trial. The potential subjects will be identified in the clinic of Department of Periodontics, for initial screening. The principal investigator will confirm the eligibility of these patients.
All patients will sign the consent forms and are informed of the details of study procedures as well as potential complications. After informed consent is obtained, the surgery will be scheduled as the first visit. An impression will be taken to make the stent before the surgery.

*b. Study Visits and Procedures*

Patients will have FGGs harvested from two sites on the palate (right and left; the area between canine to first molar). In addition to the FGG wounds, two palatal gingiva biopsies will be harvested. The size of the FGG will be standardized (8mm (width) x 10mm (length) x ≈1.5mm thickness). FGGs will be harvested by the #15 blade with the assistance of a template stent. Two FGGs will be used based on the patient's need. If the patients need grafts larger than two standardized FGGs in one procedure, the patients will be excluded from the study. The biopsies of palatal gingiva will be harvested by a tissue punch and the size is standardized (4mm (diameter) x ≈1.5mm thickness). The area of biopsy will be at least 3mm away from the FGG wound. Biopsies and FGGs will be harvested in the area between maxillary canine and first molar where tissue grafts are commonly harvested from (Reiser et al. 1996). These wounds can be completely healed even without placing any dressing materials (Harris et al. 2007). Dressing materials are used to improve healing and reduce complications. The wounds of FGG and biopsy will be covered with an allogenic amnion chorion membrane (ACM, BioXclude®) or a collagen dressing (CLD, Collagen Patch®) based on the randomization table.

Clinical Measurements

The donor sites of FGG will be evaluated on 4 days, 10 days, 2 weeks, 3 weeks, 4 weeks, 6 weeks, 8 weeks after the surgery (7 follow-up visits in total). Healing of the donor site will be analyzed based on (1) the degree of epithelialization, (2) the size of wound area, (3) color match and (4) bleeding condition.
(1) Epithelization will be evaluated at each follow-up visit until the wound is completely epithelized. It is measured by means of bubble formation after dripping hydrogen peroxide (3%) to the wound surface and epithelization will be ranked as total, partial or none (Keceli et al. 2015; Marucha et al. 1998);
(2) The wound area will be assessed by means of a clinical picture taken at each follow-up visit (Del Pizzo et al. 2002; Thoma et al. 2012). The picture will be taken with an angulation perpendicular to the wound. A periodontal probe will be placed by the wound as a reference scale. Wound area is defined as the area without

epithelization. The wound area on the picture will be measured by ImageJ;
(3) Color match of the donor site will be identified at every visit by using a 4-point scale: 1. Obvious difference; 2. Noticeable difference; 3. Disguisable difference; 4 No noticeable difference in comparison with adjacent gingiva. Three measurements, (1), (2), and (3), will be performed by one independent examiner who does not know the dressing (ACM or CLD) for each wound;
(4) Haemostasis has to be achieved when no bleeding is actively seen. Delayed bleeding, prolonged haemorrhaging from the palate during the postsurgical period, reported by the patient, will be documented (Del Pizzo et al. 2002).

The wound healing will also be evaluated from the patient's perspective by means of (5) pain and (6) sensibility. Patients will be given a questionnaire to answer these questions every day or at the scheduled appointments (4 days, 10 days, 2 weeks, 3 weeks, 4 weeks, 6 weeks, 8 weeks) until the patient completes the study.
(5) Pain will be assessed by using VAS pain scores (0 to 10. 0: no pain, 1: minimal pain, 10: severe pain) (Keceli et al. 2015);
(6) Sensitivity will be assessed by means of a periodontal probe (XP 23/UNC 15, Hu-Friedy) using a 4-point discrimination scale (coronal, apical, mesial, distal) around the donor area, before the surgery and at the follow-up visits. Sensibility will be recorded using a rubbing movement and a pin-pressure nociception. Patients will be asked to give a rating of their loss of sensibility based on a three-point verbal descriptor scale (VDS): none, mild or moderate, severe) (Del Pizzo et al. 2002);
(7) The surgeons will be given a questionnaire to answer questions regarding handling of the two different materials after performing the surgery.

Volumetric Analysis

The impression of FGG donor site will be obtained using intraoral scanner at pre and post-operatively 4 days, 10 days, 2 weeks, 3 weeks, 4 weeks, 6 weeks, 8 weeks. The STL files of pre-op and post-op will be imported into the reverse engineering software. Then, three-dimensional images will be reconstructed and superimposed to allow for the measurement of volumetric change within the initial harvested area during wound healing phase. 10% of the sites will be randomly selected for repeated measurements.

Histological and Histomorphometric Analyses

Gingival specimens (4mm in diameter, 1.5mm in thickness) will be harvested at baseline visit and the 10-day follow-up visit for histological and histomorphometric analyses. The second biopsy will be performed at the 10-day follow-up visit because epithelization will not be completed before two weeks. The tenth day is a proper timing to evaluate early healing. Specimens will be cut in sections (5μm in thickness) and stained with either H&E to identify the cellular composition of inflammatory infiltrates or Masson's trichrome to detect new collagen depositions. For qualitative histological analysis, the histological sections will be evaluated using a Nikon light microscope, using Nikon NIS Element AR Imagine Software (Nikon Metrology, Inc. Brighton, MI) to calculate the percentages of the new collagen and matured collagen, as well as the thickness of the epithelium automatically. Three regions of interest will be analyzed on each specimen corresponding to the superficial, central, and apical third. Immunohistochemical staining will be performed to detect factor VIII (von Willebrandt factor) to detect endothelial cells and therefore angiogenesis. The area of angiogenesis will be quantified using the same Nikon NIS Element AR Imagine Software.

*c. Final Study Visit:*

The 8-week follow-up will be the subject's last visit for this clinical trial. The patients will have clinical measurements as previously mentioned and received the gift card.

*d. Post Study Follow up and Procedures*

The subjects will continue having routine maintenance appointments to follow up the outcomes related to the surgery and periodontal health in the periodontics clinic of UTSD. If the patients have symptoms or complications, the necessary treatments, will be performed.

*e. Discontinuation Visit and Procedures*

Subjects are free to withdraw from participation in the study at any time upon request. A subject may be discontinued from participation in the study for any of the following reasons:

1. Withdrawal of consent
2. Subject noncompliance with the protocol, as determined by the investigator
3. Any event or condition that would make continued participation in the study not in the best interest of the subject, as determined by the investigator
4. Pregnancy
5. Development of any medical condition that might affect the treatment and clinical outcomes, as determined by the investigator.
6. Initiation of any treatment or exposure that might affect the outcomes of implant therapy, as determined by the investigator.
7. Investigator discretion

Any subject with a serious adverse event, such as life-threating diseases, hospitalization, that is ongoing at the time of discontinuation will be followed until the event returns to baseline, resolves, or stabilizes. If the serious adverse event does not meet these outcomes within 30 days after discontinuation or after study completion, the subject will be referred to an appropriate practitioner for continued care. If the study is discontinued, subjects will be referred back to the qualified clinicians for necessary dental care.

## 7. TRIAL MATERIALS

### 7.1. Trial Product (s)

Allogenic amnion chorion membrane (ACM), BioXclude, is a minimally manipulated allograft amnion chorion tissue for use as a wound covering in dental surgery. It has been widely used in dentistry. Its unique physical and biological properties provide the benefits of a growth factor and an occlusive barrier. The tissue used in BioXclude is obtained from consenting mothers who donate their placentas after elective caesarian section delivery. The amnion chorion tissue is processed using a patented tissue processing technology (Purion®,) designed to cleanse and maintain the delicate structures of the tissue. Following processing and dehydration, the allografts are packaged and terminally sterilized. This processing methodology allows for

retention of the biological factors found in native amnion chorion tissue.

Collagen dressing (Collagen Patch®, Zimmer) is used to cover wound in dental surgery. It can adhere to the wound and retain its structural integrity even when wet. It is known to control bleeding and stabilize blood clots as well as protect the wound bed while accelerating the healing process.

### 7.2. Storage and Drug Accountability

BioXclude membrane and Collagen dressing will be stored at ambient temperature in its original packaging following the manual instruction. The expiration date for the product is recorded on the product container labeling as year (4 digits) and month (2 digits) and the product expires on the last day of the month indicated. Expiration date printed on the labeling is valid as long as product is stored at ambient temperature and in an unopened foil pouch/packaging. Once the product is expired, the material will be discarded.

## 8. TREATMENT

### 8.1. Rationale for Selection of Treatments

All subjects need FGG procedure and FGGs are harvested for clinical and research purposes. The treatments are conducted following general clinical principles. The details of procedure are mentioned in section 6.3.

### 8.2. Specific Restrictions / Requirements

Patients will be prescribed an analgesic (Ibuprofen, 600 mg, q.i.d; if the patient cannot take ibuprofen, 500mg acetaminophen, q.i.d will be prescribed). A disinfectant solution (Chlorhexidine digluconate, 0.2% solution) will be prescribed but the patient will only apply the solution to the recipient site using a monojet syringe (q 8 hours, 10 days). An individualized protective stent will be given to the patient. The patient will be instructed to wear the stent overnight after the surgery and continue wearing it for 10 days. The patient will be instructed to take liquid diet for the first 2 days then soft diet till the 10th day. Sutures at the FGG site will be removed 4 days after surgery.

### 8.3. Blinding

The surgeons cannot be blinded because the surgeons will be told which dressing should be placed on the specific site. Patients will be blinded given they will not know how the surgical group will be assigned. The patients will be informed of benefits of all the procedures and realize all procedures are supported by scientific evidence.

## 9. SAFETY MEASUREMENTS

### 9.1. Definitions

All unanticipated problems will be reported in this study. The Committee For the Protection of Human Subjects (CPHS) considers unanticipated problems to be any

incident, experience, or outcome that meets all of the following criteria:

Is unexpected in terms of nature, severity, or frequency given a) the research procedures that are described in the IRB-approved research protocol and informed consent, and b) the characteristics of the subject population being studied;

Is related or possibly related to participation in the research (possibly related means there is a reasonable possibility that the incident, experience, or outcome may have been caused by the procedures involved in the research); and

Places subjects or others at a greater risk for physical, psychological, economic, or social harm than was previously known or recognized.

An incident, experience, or outcome that meets the 3 criteria above will generally warrant consideration of substantive changes in order to protect the safety, welfare, or rights of subjects or others. Examples of corrective actions or substantive changes that might need to be considered in response to an unanticipated problem include the following:

Changes to the research protocol initiated by the investigator prior to obtaining IRB approval to eliminate apparent immediate hazards to subjects

Modification of inclusion or exclusion criteria to mitigate newly identified

risks Implementation of additional procedures for monitoring subjects

Suspension of enrollment of new subjects

Suspension of research procedures in currently enrolled subjects

For this study, an severe adverse event (SAE) is defined as an unanticipated problem occurring during the study that fulfils 1 or more of the following criteria:

1. Results in death
2. Is immediately life-threatening†
3. Requires inpatient hospitalization or prolongation of existing hospitalization
4. Results in persistent or significant disability or incapacity
5. Is a congenital abnormality or birth defect
6. Is an important medical event that may jeopardize the subject or may require medical intervention to prevent one of the outcomes listed above

Hospitalization for elective procedures or surgeries will not be considered SAEs, nor will inpatient hospitalizations for convenience.

Pregnancy in women with childbearing potential should not be reported as an SAE, but if pregnancy occurs, it must be reported in accordance with the procedures described in Section 6.2. Pregnancy will not be regarded as an SAE unless there is suspicion that a study intervention may have interfered with the effectiveness of a contraceptive medication and the event meets the criteria for an unanticipated problem. If the pregnancy results in an outcome other than a normal birth or elective abortion of a healthy fetus, it will be reported as an SAE.

### 9.2. Collecting, Recording and Reporting of Adverse Events

Examination and close follow-up of parameters capturing subjects' oral health will be collected on case report forms (CRFs). These will be completed at every study visit, and data will be compiled into a pre-specified format and reviewed monthly by the PI for safety oversight.

Serious adverse events (as defined in Section 9.1) will be collected from the time of enrollment until the last clinic visit and will be recorded in the electronic health records (EHR) system. At each study visit, the clinician or investigator will inquire about the occurrence of SAEs since the last assessment. The investigator will review all source documentation related to study procedures for evidence of SAEs. Events will be followed for outcome information until they return to baseline or stabilize, or

until 30 days after study completion or subject discontinuation. Subjects who have an SAE that is ongoing 30 days after study completion or discontinuation will be referred to an appropriate practitioner for continued care.
Upon learning that a subject has experienced an SAE, the investigator must report the event to CPHS within 24 hours after becoming aware of the event.

On a monthly basis, the following events will be reported to every PI:

Number of subjects experience severe complications and number of subjects enrolled. Severe complications include severe pain (VAS>6), continuous bleeding and severe swelling that needs prescription to control.

Duration of observation of subjects experiencing severe complications and duration of observation of subjects enrolled.

Any tooth loss, abscess, or other adverse oral health development requiring therapy or other intervention and the etiology (as captured in the dental history)

Every PI will review the monthly reports for any safety signals.

### 9.3. Safety Monitoring Plan

The purposes of the clinical monitoring activities are to ensure that the rights of human subjects are protected, the study is implemented in accordance with the protocol, and the integrity of study data is maintained.
All subjects will be monitored for postoperative healing and tissue response at a regular interval while the entire oral health will be maintained throughout the study period.

## 10. DATA ANALYSIS

### 10.1. Data Quality Assurance

Data and measurements will be checked by two separate investigators (Chun-Teh Lee, Seonghong Min) as well as analyzed statistically to ensure that the data obtained is accurate, complete and reliable.

### 10.2. Data Entry and Storage

Case report forms (CRFs) will be completed and stored in a locked file cabinet in the PI's office located at UTSD. Data will be entered electronically in excel spreadsheets, and images will be stored electronically; both will be stored on the PIs work computer in a locked office and password protected.

## 11. SAMPLE SIZE AND STATISTICAL METHODS

### 11.1. Determination of Sample Size

The pilot project will use a convenient sample size based on the budget. We will recruit 19 patients in total, and with a dropout rate of 15%, we will have a sample size of 15.
With 15 patients, we can detect an effect size as small as 0.7 between two treatments for the two main outcomes (the area of healing and VAS pain score at

week 2), by a paired t test assuming a within-subject correlation of 0.6 at the significance level of 0.05 with 80% power. Considering this is a pilot study, Bonferroni correction is not necessary.

### 11.2. Statistical and Analytical Plans

*a. General Considerations*

We will present means and standard deviations for completeness of the report. The statistical significance level to test the primary endpoint was set at $p<0.05$, a priori.

*b. Safety Analyses*

Safety will be evaluated by tabulations of adverse events and will be presented with descriptive statistics at baseline and follow-up visits each month.

Adverse events will be classified as severe complications and summarized for baseline and follow-up visits.

All information pertaining to adverse events noted during the study will be listed by subject, detailing verbatim given by the investigator, preferred term, date of onset, date of resolution, severity, and relationship to procedure. The onset of adverse events will also be shown relative (in number of days) to the day of performing the surgery.

A tabulation of Serious Adverse Events (SAEs) will be provided by subject within treatment groups. The proportion of subjects in each treatment group reporting adverse events that occur in ~ 3% in either treatment group will be compared using Bayesian methods. The specific preferred terms analysed will be those that are reported by at least five percent of the subjects in either treatment group.

*c. Statistical Analysis Plan:*

We will explore the difference between the two treatments for outcomes including the degree of epithelialization, the size of wound area, color match, bleeding condition, pain, sensibility, and outcomes from Gingival specimens by paired t test or McNemar's test depending on the variables.

## 12. ETHICAL CONSIDERATIONS

### 12.1. Informed Consent

Informed consent is a process that is initiated prior to the individual's agreeing to participate in the study and continues throughout the individual's study participation. Extensive discussion of risks and possible benefits of study participation will be provided to the subjects and their families. A consent form describing in detail the study interventions, procedures, and risks will be given to the subject. Consent forms will be IRB-approved, and the subject will be asked to read and review the document. The investigator or designee will explain the research study to the subject and answer any questions that may arise. The subject will sign the informed consent document prior to any study-related assessments or procedures. Subjects will be given the opportunity to discuss the study with their surrogates or think about it prior

to agreeing to participate. They may withdraw consent at any time throughout the course of the trial. A copy of the signed informed consent document will be given to subjects for their records. The rights and welfare of the subjects will be protected by emphasizing to them that the quality of their clinical care will not be adversely affected if they decline to participate in this study.
The consent process will be documented in the clinical or research record.
This documentation will include the following:
A notation of the date that the consent was obtained

A statement that the consent was obtained prior to the initiation of study procedures

A statement that the subject had adequate time to review the consent and that all questions were answered prior to initiation of study procedures
A notation confirming that a copy of the signed consent was given to the subject

### 12.2. IRB review

The protocol, informed consent form(s), and all advertising and subject materials will be submitted to the IRB for review and approval. Approval of both the protocol and informed consent form must be obtained before the enrollment of any subject. Any amendment to the protocol will require review and approval by the IRB before the changes are implemented in the clinic.

### 12.3. Confidentiality of Data and Patient Records

The subject's name will appear only on the consent form and clinical record, both of which will be kept separate from collected study data. All subject files will be kept confidential and placed in a double-locked office. A unique coded study number will be assigned to each subject for data collection. The number will not contain any personal information (e.g., dates, age) to further ensure protection.
The study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the PI. No subject names will be used in publications or presentations.

## 13. PUBLICATIONS

Following completion of the study, the investigator is expected to publish the results of this research in a scientific journal. The International Committee of Medical Journal Editors (ICMJE) member journals have adopted a trials-registration policy as a condition for publication. This policy requires that all clinical trials be registered in a public trials registry such as ClinicalTrials.gov (De Angelis et al. 2004), which is sponsored by the National Library of Medicine. Other biomedical journals are considering adopting similar policies. For grants and cooperative agreements, it is the institution's responsibility to register the trial in an acceptable registry.
The ICMJE defines a clinical trial as any research project that prospectively assigns human subjects to intervention or comparison groups to study the cause-and-effect relationship between a medical intervention and a health outcome. Studies designed for other purposes, such as to study pharmacokinetics or major toxicity (e.g., phase I trials), would be exempt from registering trials in a public registry such as ClinicalTrials.gov.

## 14. RETENTION OF TRIAL DOCUMENTS

Patients will be assigned identifying codes that will be linked to all collected study data, stored in secured database by PI. All the electronic files will be encrypted and are stored in primary investigator's external drive, that will be locked in the PI's office cabinet. Stents will be stored in a locked cabinet in the PI's office. The following individuals/ institutions will have access to the records: the Principal Investigator and coinvestigators, and the University of Texas Health Science Center at Houston, including the Institutional Review Board. Absolute confidentiality cannot be guaranteed because of potential need to share this information with the above parties. The aggregate results of this study, with preservation of patient confidentiality, may be used for teaching, meeting presentation or publishing purpose. Records will be maintained for at least 6 years from the starting date of each subject.

### List of Possible Attachments

**Appendix 1** **Case report form**

**Appendix 2** **Patient report form**

**Appendix 3** **Flyer**

**Appendix 4** **Informed Consent Form**

**Appendix 5** **Schematic of Study Design**

**Appendix 6** **Study Schedule**

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