EXTRACTION SITE PRESERVATION USING FDBA WITH BIOXCLUDE VS. FDBA WITH BIO-GIDE: A RANDOMIZED CLINICAL TRIAL.

by

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CERTIFICATE OF APPROVAL

MASTER'S THESIS

This is to certify that the Master's thesis of

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has been approved by the Examining Committee for the thesis requirement for the Master of Science degree in Oral Biology at the June 2013 graduation.

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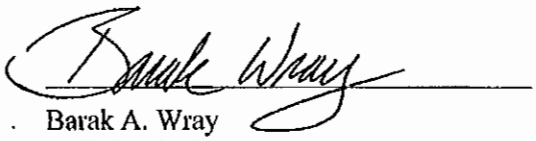

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ABSTRACT

EXTRACTION SITE PRESERVATION USING FDBA WITH BIOXCLUDE VS. FDBA WITH BIO-GIDE: A RANDOMIZED CLINICAL TRIAL.

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Introduction:

This randomized, controlled, blinded, clinical investigation will compare extraction site preservation performed with freeze dried bone allograft (FDBA) and either the Bio-Gide membrane or the BioXclude membrane. Alveolar ridge dimensional changes from the time of extraction until implant placement at 6 months will be assessed. Up to 70 patients treatment planned for an extraction with site preservation and implant placement will be enrolled. Participants will be randomized to receive Bio-Gide or BioXclude. Each participant will have a jaw impression made of the extraction site to provide a stone model for the fabrication of a customized acrylic stent. The stent will be used by blinded investigators to obtain standardized clinical measurements of the alveolar ridge at 6 points around the extraction site, immediately after the extraction, and at 6 months right before implant placement. In addition to the pre-extraction impression, impressions will also be taken at 4 and 12 weeks post-extraction, and at the time of

implant placement. The four sets of models from these impressions will be used to measure dimensional changes in the alveolar ridge. Soft tissue closure across the membranes will also be assessed. Study investigators, blinded to the membrane material, will make all clinical measurements. Standard clinical procedure evaluates postoperative healing at 1, 2, 4, 6, 8, 12 and 16 weeks and again at 6 months. For study participants, impressions and research measurements will be made only during these post-operative visits. Data analysis will compare changes in site dimension and soft tissue closure.

Methods:

Up to 70 patients treatment planned for an extraction with site preservation and implant placement will be included in the study. Up to 35 patients will receive FDBA and Bio-Gide and up to 35 patients receive FDBA and BioXclude . In both groups, no attempt will be made to obtain primary closure. Participants will have impressions made of the anticipated extraction site to provide stone models for the fabrication of a customized acrylic stent. Acrylic stents will be used by blinded investigators to obtain standardized clinical measurements of the alveolar ridge at 6 points around the extraction site following extraction and 6 months later at implant placement. In addition to the pre-extraction impression, impressions will also be taken at 4 and 12 weeks post-extraction and at the time of implant placement. The four sets of models from these impressions will be used to measure dimensional changes in the alveolar ridge. Soft tissue closure across the membranes will also be assessed. Standard clinical procedure evaluates postoperative healing at 1, 2, 4, 6, 8, 12 and 16 weeks and again at 6 months. For study participants, impressions and research measurements will be made only during these

post-operative visits. Data analysis will compare changes in ridge dimension and soft tissue closure.

Results:

Currently this research protocol has been prepared to be submitted to the IRB. Following results of correspondence with Snoasis, the makers of BioXclude, the protocol will be submitted.

Discussion:

Research will commence following IRB approval.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF ABBREVIATIONS

GBR Guided Bone Regeneration

FDBA Freeze-dried Bone Allograft

PD Probing Depth

CAL Clinical Attachment Level

CEJ Cementoenamel Junction

WRNMMC Walter Reed National Medical Military Center

PO Post-Operative

NSAIDS Nonsteroidal Anti-Inflammatory Drugs

q6h Every 6 Hours

q8h Every 8 Hours

mg Milligram

TBSP Tablespoon

VAS Visual Analog Scale

CHAPTER I: INTRODUCTION

Both the esthetic and functional outcomes of dental implant therapy are highly dependent on sufficient alveolar bone volume following tooth extraction. Although bone loss prior to extraction may have occurred due to trauma, periapical pathosis, or periodontal disease, it is important to understand that alveolar ridge bone loss occurs after extraction. In the normal post extraction remodeling process, without an intervention to preserve alveolar ridge contours, bone loss occurs in both horizontal and vertical dimensions.[1] Considering that the dimensions of the alveolar ridge are critical for implant placement, it is imperative to recognize that site (ridge) preservation following tooth extraction is needed to maintain as ideal an alveolar ridge anatomy as possible. If an implant is placed at a site in the jaw with less ideal jaw dimension, then the implant may not be completely integrated (embedded) in bone. When this occurs, and implant threads are not covered as seen in Figure 1, the threads become locations where bacterial plaque can grow. Such a situation clinically exposes the patient to recurrent infection around the implant and increased risk to implant loss.

Figure 1: Implant placed with insufficient alveolar bone.

Ridge Preservation Therapy

Bone loss following tooth extraction can be minimized by ridge preservation therapy utilizing an osseous graft material and membrane over the grafted bone. Without site (ridge) preservation interventions such as bone grafting with a membrane that try to minimize post extraction bone loss (Figures 2 and 3), a significant reduction in both horizontal and vertical bone volume can be witnessed. Ridge preservation therapy is a

viable procedure to augment bone following a dental extraction and follows principles similar to guided bone regeneration. As defined by the American Academy of Periodontology, guided bone regeneration refers to ridge augmentation or other bone regenerative procedures. Barrier techniques, such as placement of a membrane at the graft site, are employed in the attempt to exclude the epithelium from covering the existing bone surface.[4] Use of a barrier prevents the faster growing epithelium from invading the site where regeneration is desired and provides an increased opportunity for slower growing connective tissue and osteoid to fill in an extraction site, a deficient alveolar ridge or a bone loss defect around an existing tooth produced by periodontal disease.

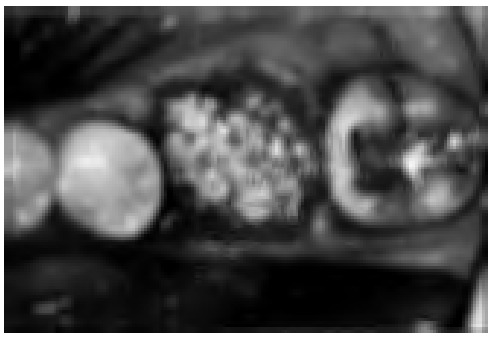


Figure 2: Bone graft in extraction site.

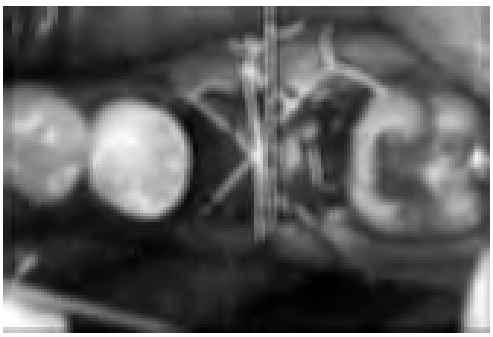


Figure 3: Bone graft covered with a membrane.

Tissue flap primary closure is difficult and sometimes impossible to achieve when performing site preservation after an extraction. Resorbable collagen membranes function optimally when primary closure is achieved, but when exposed to the oral cavity they are subject to increased resorption, which can allow loss of graft material and soft tissue invagination of the extraction socket. This loss of graft material and soft tissue invagination results in greater alveolar ridge loss at the extraction site; leading to a less

favorable outcome for future implant placement. These potential pitfalls with collagen membranes may make the amnion-chorion membrane an attractive alternative. The amnion-chorion membrane has been shown to enhance wound healing properties due to the varying growth factors contained within the membrane itself, which in turn, accelerate epithelialization over the treated site. By accelerating epithelialization across the membrane, improved wound closure time could aid in bone graft containment, and lead to greater gains alveolar ridge dimensions during post extraction remodeling.

Collagen Membranes vs. Amnion Chorion Membranes

Amnion-chorion membranes and collagen barriers are both utilized extensively with FDBA for preservation of extraction sockets in dentistry. There are no published studies that have directly compared these two membranes as they relate to both soft and hard tissue healing.

CHAPTER II: REVIEW OF THE LITERATURE

Healing Following Tooth Extraction

Both the esthetic and functional outcomes of dental implant therapy are highly dependent on sufficient alveolar bone volume following tooth extraction. Although bone loss prior to extraction may have occurred due to trauma, periapical pathosis, or periodontal disease, it is important to understand that alveolar ridge bone loss occurs after extraction. In the normal post extraction remodeling process, without an intervention to preserve alveolar ridge contours, bone loss occurs in both horizontal and vertical dimensions.[1] Considering that the dimensions of the alveolar ridge are critical for implant placement, it is imperative to recognize that site (ridge) preservation following tooth extraction is needed to maintain as ideal an alveolar ridge anatomy as possible. If an implant is placed at a site in the jaw with less ideal jaw dimension, then the implant may not be completely integrated (embedded) in bone. When this occurs, and implant threads are not covered by bone as seen in the photograph to the right, the threads become locations where bacterial plaque can grow. Such a situation clinically exposes the patient to recurrent infection around the iinplant and increased risk to implant loss.

Wound healing following extraction has been evaluated extensively in humans. Shortly following extraction, clot formation occurs that is replaced by granulation tissue within seven days. By the twentieth day, the granulation tissue is replaced by connective tissue. While this connective tissue replacement is occurring, osteoid formation begins at the base of the socket by seven days, and eventually, two thirds of the original bony socket that held the tooth will be filled with bony trabeculae at thirty-eight days. During healing, epithelium from the gingiva adjacent to the extracted tooth quickly migrates into

the extraction site. This process is called apical proliferation and it occurs much more quickly than bone deposition. It can be seen as early as four days after extraction.[2] Inhibition of epithelial apical proliferation into the extraction site affords connective tissue and bone greater opportunity to regenerate the extraction site.

Following extraction, dimensional changes in the bone surrounding the extraction site occur and this resorption of alveolar crest can adversely influence subsequent restorative treatment.[3] Without site (ridge) preservation interventions such as bone grafting (Figure 2) that try to minimize post extraction bone loss, a significant reduction in both horizontal and vertical bone volume can be witnessed. In non-bone grafted extraction sites, a reduction in ridge height of 1 mm and nearly a 50% of alveolar ridge width loss can be measured clinically over a twelve month period. However, most of the alveolar ridge resorption occurs within the first three months following extraction.[1]

Osseous Allograft Material

Osseous allograft material has become the gold standard when preserving the dimensions of an edentulous area that is intended to receive future implant placement. Bone allograft is defined by the American Academy of Periodontology as a graft between genetically dissimilar members of the same species.[4] Bone allograft can be cadaver in origin and can also be obtained from living donors undergoing such procedures as hip replacement surgery. Allograft material is available in both block and particulate form. A commonly utilized particulate osseous allograft is freeze-dried bone allograft (FDBA). FDBA is a versatile graft material that is used extensively in dentistry and medicine for many different procedures.

Allograft materials are safe. Mellonig's comprehensive review of bone graft donor selection, and the testing and inactivation of transmissible diseases in bone allograft material, concluded that the chance of obtaining a bone graft from an HIV infective donor is 1 in 1.67 million. Furthermore, the calculated risk of HIV transmission after receiving a bone graft was 1 in 2.8 billion. At the time of Mellonig's 1995 article, FDBA had been used for over 25 years and there had been no documented cases of any type of disease transmission.[5] This finding of FDBA safety was re-enforced by a 25 October 2013 posting by the Centers for Disease Control and Prevention.[6] It is recommended that only allografts from sources accredited by the American Association of Tissue Banks be used.[7]

FDBA has proven to be osteoconductive. Osteoconduction is the physical effect in which the matrix of the graft material forms a scaffold allowing cells from the recipient to penetrate the graft and form new bone. The mineralized nature of freeze-dried bone allows for excellent space maintenance properties at graft sites compared to demineralized FDBA. Moreover, freeze-dried bone allograft may stimulate earlier, more rapid and substantial new bone formation.[8] Due to its mineralized nature, freeze-dried bone allograft maintains a grafted area with a denser allograft material as seen in Beck & Mealey's study in 2010. Histologic samples of extraction sites grafted with mineralized freeze-dried bone allograft at three and six months post operatively depicted how residual grafting material was surrounded by new woven bone.[9]

Usage of Allograft and a Membrane

As shown by Hämmerle and Jung in 2003, using a bone graft material in

conjunction with a membrane has a higher success rate in the treatment of horizontal ridge defects when compared to bone grafting without the use of an epithelial excluding membrane.[10] The ideal barrier membrane for use with guided bone regeneration in extraction sites should have several characteristics. The membrane should be biologically inert, help stabilize the clot formation, and be able to contain the graft material at the surgical site. It should have sufficient rigidity to maintain space in the extraction site, but should be flexible enough to allow for ease in adaption to the treatment area. The membrane should produce predictable results and be cost effective. Further, it should be resorbable, eliminating the need for surgical re-entry to remove the membrane. Most importantly, the membrane should last long enough before it resorbs to permit epithelium to grow over the healing surface, yet still prevent the epithelium from growing into the grafted site. This epithelial exclusion property allows for selective cell repopulation in which cells associated with bone healing can predominate in the location where bone deposition is needed.

In extraction sites, grafting with an allograft and a membrane enhances horizontal ridge preservation by significantly reducing the loss of the bucco-lingual (cheek-tongue) dimension at extraction sites. Ridge preservation techniques minimize horizontal ridge dimension loss to 27 to 30% as opposed to nearly a 50% loss following extractions without ridge preservation.[11, 1] When a membrane is used in studies on guided tissue regeneration, fibroblasts and epithelial cells are precluded from entering the grafted site which allows the more slowly migrating osteogenic cells to produce bone in the defect area.[12]

Collagen Membranes

Many studies have utilized a collagen material, such as Bio-Gide, when performing site preservation procedures. Since collagen is a resorbable, naturally occurring biological substance, collagen membranes are highly versatile and used extensively in dentistry and medicine. Collagen is a protein composed of three polypeptide chains. Each polypeptide chain contains nearly one thousand amino acids. Collagen can be assembled into various larger molecules that have different properties. This functional diversity allows collagen to be prepared into lattice-like gels or cross-linked compacted solids that can be fabricated into membranes.[13]

Many collagen membranes in various shapes and sizes are available to the practitioner. These membranes are defined as xenograft materials because the collagen is derived from other species. Although membranes authorized for commercial use by the FDA are derived mostly from porcine and bovine collagen sources, membranes made with bovine collagen are predominantly used. Collagen membranes are resorbable and possess the ability to promote wound healing through stabilization of the clot. In addition, they enhance wound closure by attracting fibroblasts and providing a scaffold for cellular growth and movement.[14]

Different collagen membranes demonstrate varying degrees of longevity before they are fully resorbed or degraded during the healing process. Cross-linked, resorbable collagen membranes can last up to 24 weeks after their application to a surgical site while non-cross-linked collagen membranes persist from eight to twelve weeks. As epithelium covers the outer surface of the collagen membrane a layer of dense fibrous tissue infiltrate forms below the membrane. Therefore, during the optimal healing process as the membrane is resorbed the grafted bone will be covered by an intervening layer of

connective tissue below the outer epithelium.[15]

However, when collagen membranes are not completely covered by soft tissue and are exposed to the oral cavity, they resorb at an increased rate. The differences in the degradation rates of collagen membranes upon exposure may be dependent on the crosslinking level of the collagen in a given membrane, as was recently shown in an in-vitro study by Sela and colleagues.[16] When premature exposure of a collagen membrane to the oral environment occurs, oral bacteria adhere to the membrane surface.[17] Bacteria of the oral flora produce proteinases with the ability to degrade collagen membranes leading to premature loss of the membrane and exposure of the underlying tissue.[14] In respect to guided bone regeneration, this exposure can cause loss of the grafted material and reduced gains from the attempted ridge preservation procedure.[18]

As a type of collagen membrane, Bio-Gide is a non-cross-linked porcine derived collagen membrane containing both type I and III collagen. It contains a bilayer structure with a cell occlusive outer surface and a fibrous surface. Studies have shown that these qualities of the Bio-Gide membrane contribute to early vascularization and bone formation.[19, 20] The overlying cell occlusive layer allows for fibroblast attachment leading to favorable soft tissue healing. The underlying fibrous layer facing the grafted site also functions a guide for angiogenic and osteoblastic cells.[21] Bio-Gide has been shown to protect the initial blood clot and then degrade over time. Bio-Gide is indicated for use in extraction sockets, ridge augmentation, periodontal defects and sinus floor elevation.

Ridge preservation using freeze-dried bone allograft along with a collagen membrane clinically improves ridge height and width when compared to non-grafted

extraction sites alone. In 2003, Iasella and colleagues examined trephined cores of grafted extraction sites prior to implant placement. They found that the quantity of bone in sites treated with a bone graft and membrane was greater than in sites not treated with a membrane or graft. They also found when using a collagen membrane along with freeze dried bone allograft for extraction sites, that there was greater preservation of ridge height as well as ridge width.[22]

Amnion Chorion Membranes

A new class membranes, sourced from human amniotic tissue, have recently become available for use in guided tissue and guided bone regeneration procedures. Amnion layer contains collagen types III, IV, and V1[23] and the chorion layer contains collagen Types I, III, IV, V, and VI.[24] Allograft membranes that only, utilized the amnion layer of the amniotic sac, have been used successfully since the early 1900s in skin wound applications[25], and subsequently used in numerous applications including ophthalmologic surgery[26], and vaginal repair[27], and specific to periodontal and oral maxillofacial surgery, in vestibuloplasty[28], periodontal surgery[29], nerve repair[30], and as interpositional material to prevent adhesions in temporomandibular joint arthroplasty.[31] Amniotic tissue is considered to be an immunologically privileged tissue. In being immunologically privileged, it will not cause an immune reaction or contribute to a graft versus host rejection.[32] Amnion tissue displays an anti-inflammatory effect evidenced by reduced numbers of inflammatory cells at the healing site. Further, amnion tissue exhibits antibacterial properties by precluding bacteria from entering the grafted site which results in lower post-surgical infection rates.[33] Additionally, growth factors present in amnion include epidermal growth factor, transforming growth factor-β,

transforming growth factor-α, basic fibroblast growth factor, and keratinocyte growth factor[34], which promote and accelerate epithelialization.[35] Amnion contains metalloproteinase-1, which reduces the degradation of collagen and results in the promotion of epithelial wound healing[35]. Collectively, the properties in dental surgery have shown to decrease patient pain[36] and more generally, amnion's ability to promote accelerated healing.[37]

The tissue used in this class of membrane are obtained from mothers who donate their placenta following elective cesarean section in compliance with Food and Drug Administration regulations and tissue standards of the American Association of Tissue Banks. The amnion chorion membrane (ACM), used in the study, is composed of both the amnion and chorion layers which comprise the amniotic sac. Prior to the availability of amnion chorion membrane (ACM) in 2010, only amnion membranes were commercially available, primarily for use in ophthalmologic surgery. The amnion chorion tissues are prepared using a proprietary process (Purion®) that cleanses and preserves the biological properties of the allograft. The dehydrated ACM is then packaged, terminally sterilized (SAL 10 -6), and stored at ambient temperature. A comparison of three commercially available amnion only membranes, demonstrated ACM is 4-5x thicker than amnion only membranes, contain 20x more growth factors, and the chorion layer, on average, contributes 82% of the growth factors found in ACM.[38] ACM contains a vast array of extracellular matrix (ECM) proteins that enhance wound healing. These ECM proteins include fibronectin and laminin. The extracellular matrix proteins positively influence cell differentiation, migration, adhesion and growth adjacent to ACM.[24] Of particular importance is laminin-5. This ECM protein possess a high affinity for

adhesion of gingival epithelial cells[39] and aids the migration of direct-attachment tooth (DAT) cells.[40] Immunohistochemical analysis showed ACM contains high concentrations of laminin 5, especially in the chorion layer, which is not present in the bovine collagen membrane (Bio-Gide®).[41] The absence of this critical ECM protein may explain why in the treatment of periodontal defects using guided tissue regeneration, sites covered with bovine collagen membrane (Bio-Gide®) had on average 2 mm more post-operative gingival recession at six months, compared to amnion membrane treated sites.[42] Moreover, the high concentration of laminin-5 within ACM supports why the use of ACM has been shown to hasten gingival flap re-attachment following surgery.[43] Bioactivity of the biological factors within ACM, following Purion® tissue processing, has been confirmed by endogenous growth factor production, by human dermal fibroblasts, after exposure to amnion chorion extracts.[44] These soluble growth factors have shown to stimulate the proliferation and migration of human microvascular endothelial cells, and caused these cells to produce and release angiogenic growth factors[45]. (REF). In two different in-vitro tests, these soluble growth factors were shown to have a chemotactic effect on the migration of mesenchymal stem cells and human umbilical vein endothelial cells.[45,46] Neovascularization of amnion chorion membrane implanted subcutaneously in-vivo displayed continual increase of microvessel formation at all-time points, suggesting an active intra-implant neovascular process.[45] Results from two murine pre-clinical models showed amnion-chorion membrane effectively recruited circulating hematopoietic progenitor cells and bone marrow progenitor cells to the site of implantation, with parabiosis modeling confirming the circulatory origin of recruited cells.[47]

The FDA classifies ACM as minimally manipulated allograft for use as a wound covering, and today, these Purion processed allografts are used in numerous medical applications including wound care, surgical, orthopedic, spinal, sports medicine, and ophthalmic surgery, as well as dental applications. In the treatment of chronic diabetic lower extremity ulcers, a prospective, randomized, controlled, multicenter clinical study, showed that after 4 weeks, 85% of ACM patients achieved complete healing compared to only 35% of patients treated with a bi-layered bioengineered skin substitute, increasing to 95% and 45% respectively at 6 weeks.[48] In robot assisted laparoscopic radical prostatectomy, only 50% of the control group recovered erectile function following surgery, compared to 95% when ACM was used.[49] In dental surgery, a retrospective study was conducted, consisting of 64 patients (range 34-92) involving 78 sites with moderate to severe chronic periodontitis.[50] There was no exclusion criteria and the group included 14 smokers, two on oral bisphosphonates, and one on immunomodulative medication. On the day of surgery, average probing depth (PD) was 8.5 mm, and clinical attachment loss (CAL) was 9 mm. At 12 months, the average PD was 3.5 mm, and CAL was 4.7 mm, demonstrating that the combination of BioXclude and FDBA decreased average PD by 5 mm and reduced CAL by 4.7 mm. There were no complications from surgery and all sites exhibited excellent early healing.

ACM are easily managed in the oral cavity. The membrane material, upon removal from its packaging, can be folded in half, does not require trimming, and is placed directly at the surgical site without having to be rehydrated. ACM is self-adhering and does not require sutures. When adapting the amnion-chorion membrane, it may fold over itself, and lay over adjacent exposed roots.[51] This feature allows the membrane to

adapt closely to the grafted site and further attributes to its ease of use in dental surgery.

Comparison of BioGide and BioXclude Membranes

Due to differing characteristics between bovine collagen and amnion chorion membranes, we hypothesize that accelerated wound closure, less patient discomfort following surgery, and greater alveolar ridge preservation would occur in sites grafted with FDBA and ACM compared to sites grafted with FDBA and a collagen membrane.

CHAPTER III: MATERIALS AND METHODS

70 patients requiring dental extractions and subsequent site preservation for future dental implants will be enrolled in the study. Please see Appendix A for a flow diagram of the study. The findings of their comprehensive periodontal evaluation such as probing depths (PD), clinical attachment levels (CAL), and recession will have been recorded on the Navy Periodontal Chart Form - NAVMED 6660/2 (Appendix B) by the subject's provider.

A study investigator will initiate the consent process as described within this protocol after being contacted by a provider whose patient is treatment planned for an extraction with site preservation in preparation for an implant and has expressed possible interest in study participation.

Standard Clinical Sequence (with introduction of study and consent):

1. Patient is referred for a periodontal evaluation.
2. A treatment plan including a dental extraction with site preservation and eventual implant therapy is developed for the patient by their provider.
3. The provider asks their patient if he/she would like to learn about the study being conducted at WRNMMC and will be provided a one page brief about the study.
4. If the patient is not interested in the study the provider pursues the established treatment plan.
5. If the patient expresses interest in participation, an investigator is asked to discuss the study and consent process with the patient.

6. If the patient does not consent to be in the study, therapy will continue as planned by the patient's surgeon.

7. If the patient consents, the following process ensues.

Following Consent:

1. Impressions using an irreversible hydrocolloid material (alginate) will be made of the arch in which the extraction will be taking place using stock impression trays. The impressions will be poured with dental stone for models, also called casts (Figure 4).

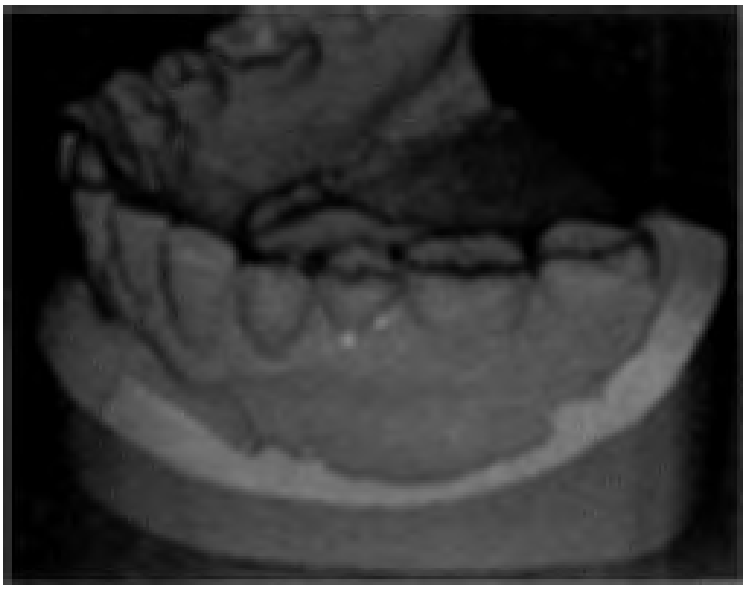


Figure 4: Stone Model example.

2. These stone models will be utilized to aid in the future implant placement process and will be used to fabricate a vacuum-formed retainer and a customized plastic stent to allow standardized measurements of the surgical site.

    a. Vacuum-formed retainer

        i. Prior to fabrication of the acrylic stent, a vacuum-formed

retainer will be made from 0.020 inch clear plastic as per the manufacturers guidelines

ii. The vacuum-formed retainer will be trimmed and delivered to the patient following the extraction

iii. The vacuum-formed retainer will function to:

1. Provide esthetics for the edentulous site from the extraction
2. Maintain space for future implant placement and crown delivery.
3. Maintain space to allow for stent utilization

b. Laboratory acrylic stent:

i. An acrylic stent for making measurements at the extraction site will be fabricated.

ii. Preserving the soft tissue margins on the cast, the tooth to be extracted will be removed from the cast using a slow speed laboratory handpiece (Figures 5 and 6).

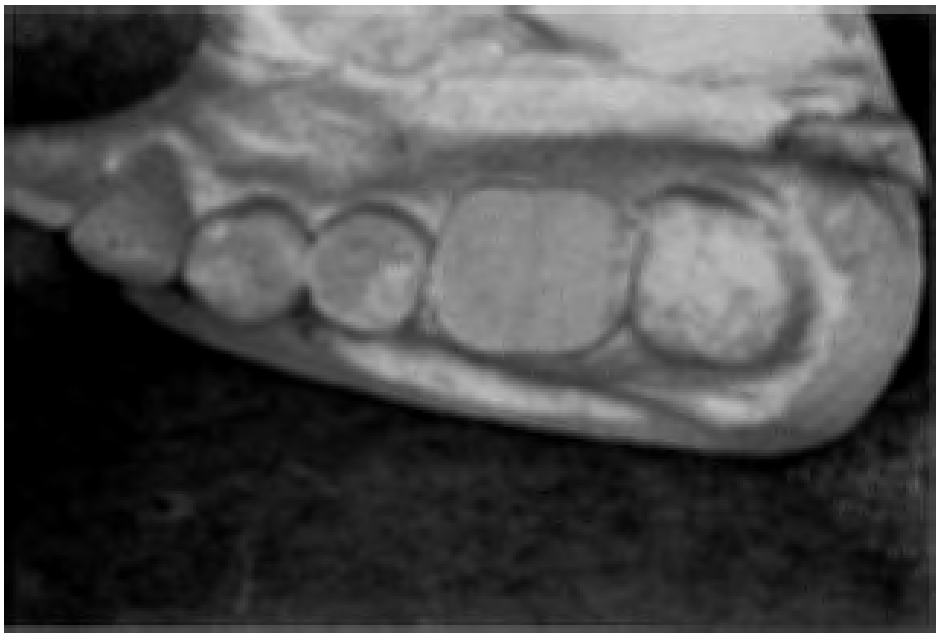


Figure 5: Cast after removing the tooth planned for extraction (#19) at the height of contour, occlusal (top-down) view.

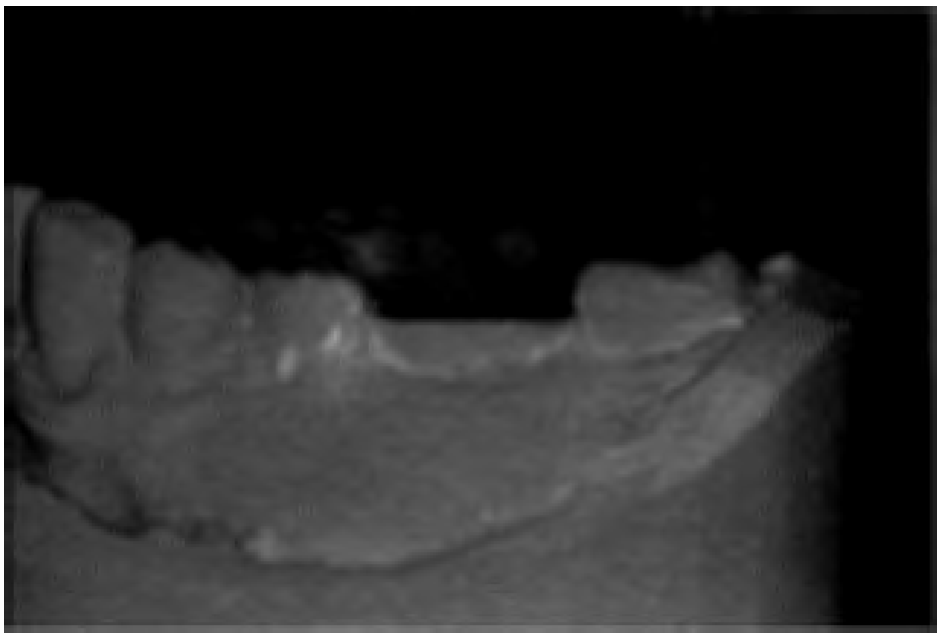


Figure 6: Cast after removing the tooth planned for extraction (#19) at the height of contour, buccal (side) view.

iii. Moldable light curable acrylic material will be adapted to the cast over the site of the removed tooth and the adjacent teeth.

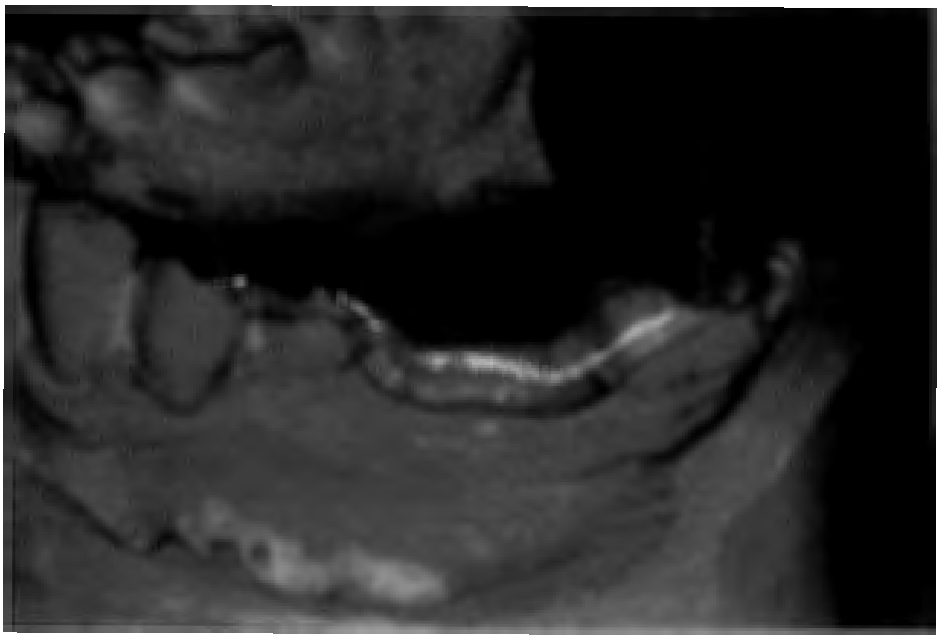


Figure 7: Moldable acrylic applied and cured.

iv. The acrylic will be trimmed to end just above the height of contour of the crowns of the teeth in order to visualize the gingiva.

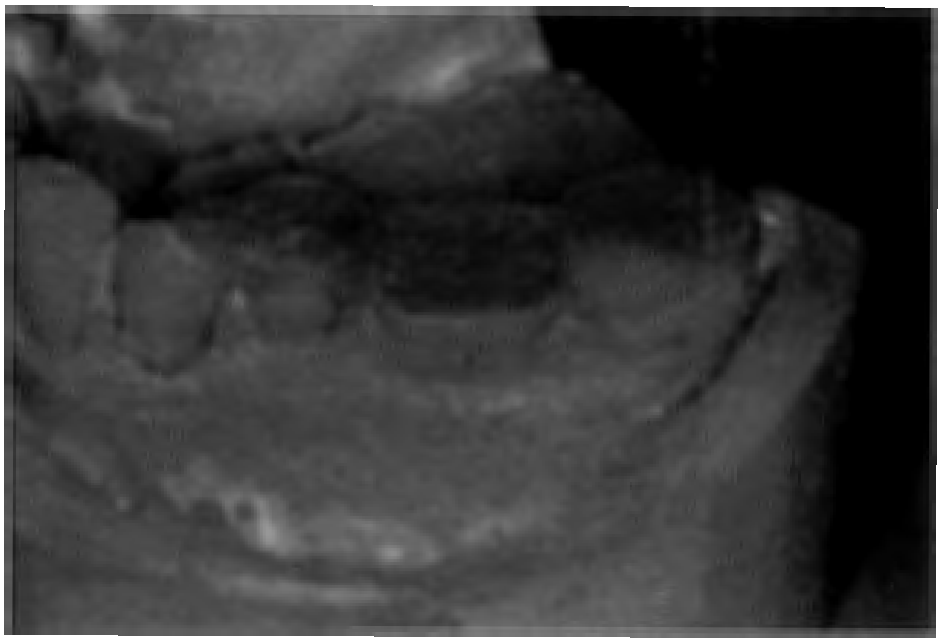


Figure 8: Acrylic trimmed.

v. A fissure bur (1169 bur) will be used to cut grooves in the stent. These grooves accommodate the periodontal probe and allow the investigator to probe the same location and with the same angulation after the extraction and at 6 months.

1. Measurement slots on the acrylic stent to measure bone height around the extraction site and at 6 months before implant placement will include:

    a. Distal-Buccal

    b. Distal-Lingual (or Palatal)

    c. Mesial-Buccal

    d. Mesial-Lingual (or Palatal)

    e. Mid-Facial

    f. Mid-Lingual (or Palatal)

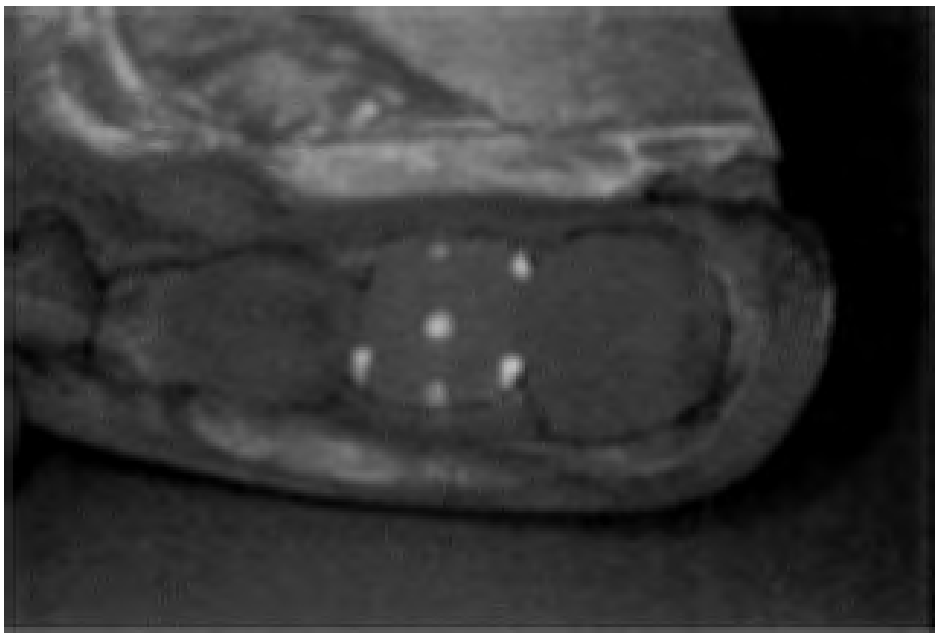


Figure 9: Measuring grooves created.

vi. Following use the stent will be cleaned and disinfected with Dispatch spray and stored in a ziplock plastic bag labeled with the subject's study number. The bag will be locked in a secured drawer maintained by the primary examiner; and then retrieved for measurements at 6 months.

c. Also at baseline, buccal-lingual measurements of the cast will be made at the direct mid-point of the future extraction site 4mm apical to the cementoenamel junctions of the adjacent teeth.

Randomization Procedure:

1. Subjects will be stratified by location of the tooth in the mouth (i.e. maxillary anterior, maxillary posterior, mandibular anterior or mandibular posterior) and randomized to either Bio-Gide or BioXclude in a 1:1 ratio. Both treatments will appear in blocks of 2 or blocks of 4, in a random order. A computer program will randomly sequence each subject's study enrollment numbers (1-70) as in the example below.

    a. For each of the 4 jaw locations, a random sequence table will be generated by the research coordinator following IRB approval in order to maintain blinding of investigators.

2. For each location, seventy envelopes marked 1 -70 will contain either a card stating Bio-Gide or BioXclude. Thereafter, the random sequence table will be placed in a sealed envelope that will not be opened until all data has been collected. Sealed envelopes (1-70) will be stored by the principal investigator in a locked drawer.

3. When each participant goes to surgery the investigator will provide the surgical team the envelope corresponding to that subject's enrollment number and tooth location. The surgical team will open the envelope and remove a card which will state which membrane material to place following extraction.

    a. One of the cards below (Figure 8) will be sealed in each envelope (1-70).

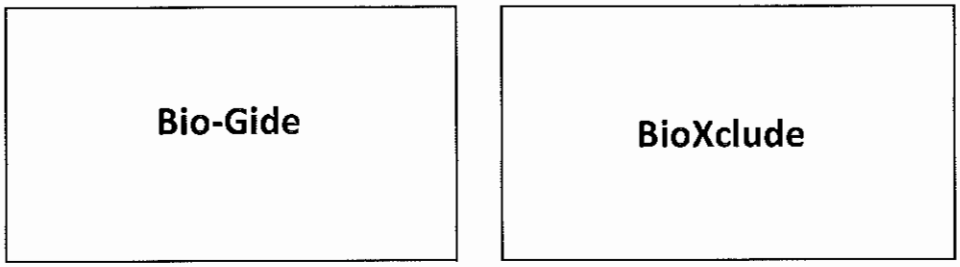


Figure 10: Membrane cards.

4. The study investigators who make the post extraction and pre-implant placement clinical measurements at 6 months will be blinded to which membrane material a given participant received.

Surgical Procedure:

Females of child bearing age will be asked to complete a HCG urinalysis prior to the surgical procedure. If the results of the HCG test are positive, the subject will be exited from the study.

Prior to surgical procedure, in line with standard procedure at the Periodontics Department participants will be offered the option of having the surgery performed using: 1. only local anesthesia, or 2. a combination of oral anxiolysis with Triazolam and local anesthesia, or 3. a combination of IV moderate sedation with Versed and Fentanyl and local anesthesia. The use of sedation will not affect the surgical procedure.

1. The surgical provider will be either a board certified staff periodontist or a 2nd or 3rd year periodontal resident. All surgical providers will be briefed in the protocol. All surgeries will follow the same steps listed below.
    a. Surgical set-up is standardized for all surgeries done at the Naval Postgraduate Dental School Periodontics Department.
    b. Both the experimental (BioXclude) and control (Bio-Gide) materials will be available to the surgeon. The membrane material used will be determined when the sealed envelope is opened by a surgical team member following the dental extraction.
    c. Surgical Procedure Steps:
        i. Administration of oral anxiolysis or IV moderate sedation if patient desired and indicated
        ii. Administration of topical and local anesthetic with any combination of 2% Lidocaine with 1:100K epinephrine, 4% Articane with 1:100K epinephrine, and 0.5% Marcaine with 1:200K epinephrine
        iii. Sulcular incision around the tooth to be extracted
        iv. Periotomes utilized when required to minimize trauma to the surrounding bone, teeth elevated, and atraumatically extracted with forceps was performed. When required, teeth were sectioned within the socket to preserve all socket walls.
        v. The extraction socket was then curetted to remove all soft tissue from the extraction site.

vi. The adjacent soft tissue will be undermined to allow for proper membrane placement and no vertical incisions will be utilized.

vii. Characterization of the defect by a study investigator.

1. Presence or absence of a buccal dehiscence recorded as:
    a. >50% of the apical-coronal dimension of the extraction site
    b. <50% of the apical-coronal dimension of the extraction site
2. Measurement utilizing slots on the acryclic stent to the alveolar crest of the extraction site will include:
    a. Distal-Buccal
    b. Distal-Lingual (or Palatal)
    c. Mesial-Lingual (or Palatal)
    d. Mesial-Buccal
    e. Mid-Lingual (or Palatal)
    f. Mid-Buccal

ii. FDBA will be hydrated with sterile saline as per the manufacturer's instructions and placed into the extraction site

viii. Membrane material (determined from the sealed envelope)

ix. For BioXclude, the untrimmed membrane will be placed dry UP or DOWN, allowed to hydrate, passively covering the graft material, such that the membrane extends at least 3 mm beyond the alveolar crest buccally and lingually, and may fold over itself and lay over

adjacent exposed root surfaces. For Bio-Gide, membrane will be trimmed to size and shape of the extraction socket.

x. The membranes will be secured using a non-resorbable monofilament suture (ie. Gore-tex)

xi. Following suturing, the remaining exposed membrane over the extraction site will be measured horizontally in a mesial-distal and a buccal-palatal/lingual direction over the midpoint of the alveolar crest using a periodontal probe to function as a base-line measurement for soft tissue healing until closure.

xii. Vacuum-formed retainers will be given to the patients

1. Patients will be instructed to wear the vacuum-formed retainers during the day unless when eating and nightly

Post-operative Care (measurements and impressions shown):

1. All participants receive the following post-operative regimen:
    a. Pain medication consisting of any of the following alone or in combination:
        i. Ibuprofen 800 mg , Take 1 tab PO q6-8h for moderate pain or
        ii. Hydrocodone/Acetaminophen 5/325 mg, Take 1-2 tab PO q6h prn severe/breakthrough pain or
        iii. Oxycodone/Acetaminophen 5/325mg, Take 1-2 tab PO q6h prn severe/breakthrough pain
    b. Pain medication for patients who cannot take NSAIDS will be prescribed any of the following alone or in combinations:

i. Acetaminophen 325 mg, Take 1-2 tabs PO q4h for moderate pain

ii. Oxycodone 5mg, Take 1 tab PO q4h prn severe/breakthrough pain

c. Antibiotics consisting of either of the following:

i. Amoxicilin 500mg, Take 1 tab PO q8h for 10 days

ii. Clindamycin 300 mg, Take 1 tab PO q8h for 10 days

d. Bio-Gide patients, 12% Chlorhexidine, 1 bottle, Rinse and spit bid with 1 TBSP as directed on the bottle. BioXclude patients will be instructed to avoid using Chlorhexidine-based oral rinses and not given any following surgery. Patients should rinse with water, gently rolling their head side to side and let the water passively leave the mouth.

2. All patients are provided with the standard post-operative instructions (See Appendix C for an example of the standard postoperative care instruction form).

3. All patients will receive VAS forms. Every time patients feel the need to take pain medication, they will record the type of medication (if taken), time, and log their perceived pain level in the VAS form.

4. Patients are recalled at 1 week to assess post-operative healing and remove plaque/deposits from the adjacent teeth. Degree of soft tissue closure over membrane will be measured.

5. Patients recalled at 2 weeks post-operative to assess healing, remove

plaque from adjacent teeth, and remove sutures at the surgical site. Degree of soft tissue closure over membrane will be measured.

6. Patients recalled approximately at weeks 4, 6, 8, 12, and 16 to assess healing, remove plaque, and reinforce oral hygiene. Degree of soft tissue closure over membrane will be measured. At 4 weeks closure has probably occurred.
7. Impressions for dental stone models will be made in the same manner they were made at baseline at 4 and 12 weeks post-extraction.
8. Patients recalled approximately at 6 months following the surgical procedure to continue with implant therapy.
    a. Prior to implant placement, impressions of the extraction site and study models were created in the same manner as they were at baseline.
    b. After full thickness flap reflection and prior to creation of an osteotomy for implant placement, all measurements were completed using the same acrylic stent by a blinded investigator. Measurements will also be completed on the study model as previously described.
    c. Implants will be placed base on their manufacturer's guidelines.
        i. Standard post-operative care will be provided.
9. Patient will be exited from the study and followed by the periodontist and primary care dentist for subsequent maintenance therapy and implant restoration.

Analysis of Data:

1. Measurement of horizontal alveolar ridge dimensional changes using the stone

models made from impressions made at baseline, 4 weeks, 12 weeks and at time of implant placement at six months.

2. Measurement of vertical changes in alveolar ridge height at 6 locations at the extraction site using a customized acrylic stent immediately following the extraction and at 6 months right before implant placement.
3. Measurement of soft tissue closure over the membranes. This will be measured in a mesial distal and buccal-lingual (or palatal) direction over the midpoint of the alveolar crest using a periodontal probe at 1 week, 2 weeks, and 4 weeks or until the site has fully healed (when all soft tissue margins have approximated with each other).
4. Clinical attachment levels and probing depths of the teeth adjacent to the extraction site before the extraction and at 6 months before implant placement will be compared.
5. Compare postoperative discomfort data collected in the VAS forms.
6. Statistical analysis will assess pre and post-test differences.

Human Subjects Justification

The use of human subjects is warranted because patients will be receiving standard of care therapy for their dental extractions with site preservation. All patients participating in the study will be receiving the same surgical care they would receive if they were not included in the study except for the randomization of the membrane and the measurements made with the stent and on jaw models in order to assess degree of ridge preservation with the two membranes.

Possible risks of participation in this study are no different than the risks of

receiving surgical periodontal treatment outside the parameters of the study. These risks include infection, abscess, bleeding, and post-treatment pain. Other adverse effects include poor bone fill of the extraction site or failure of the bone allograft to integrate in the grafted site. In the event that adverse effects occur in conjunction with the surgical procedure, subjects will be able to call the PI and receive follow-up treatment at the NPDS Periodontics Clinic.

CHAPTER IV: RESULTS

Currently this research protocol has been prepared to be submitted to the IRB. Following results of correspondence with Snoasis, the makers of BioXclude, the protocol will be submitted.

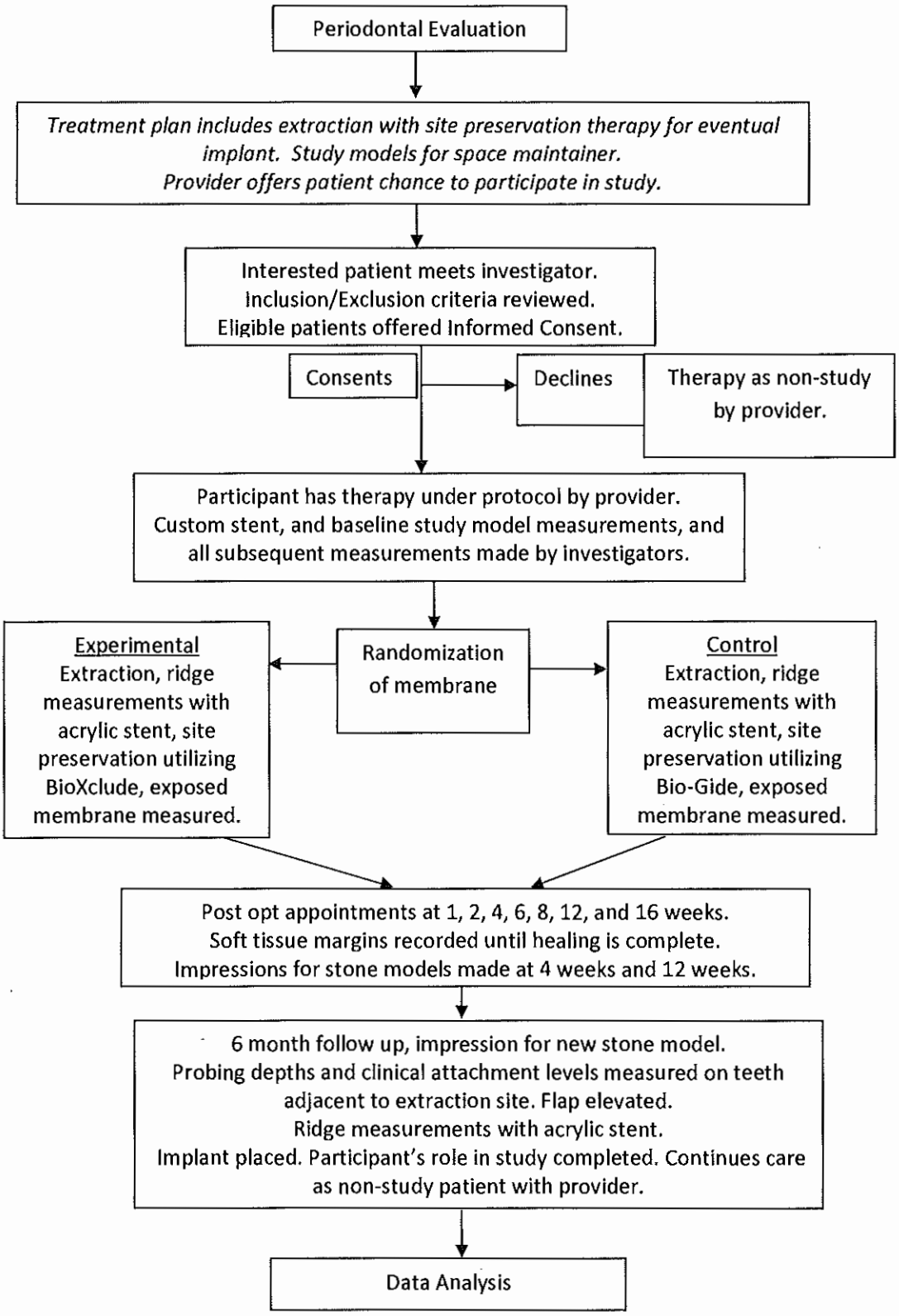
CHAPTER V: DISCUSSION

Research will commence following IRB approval.

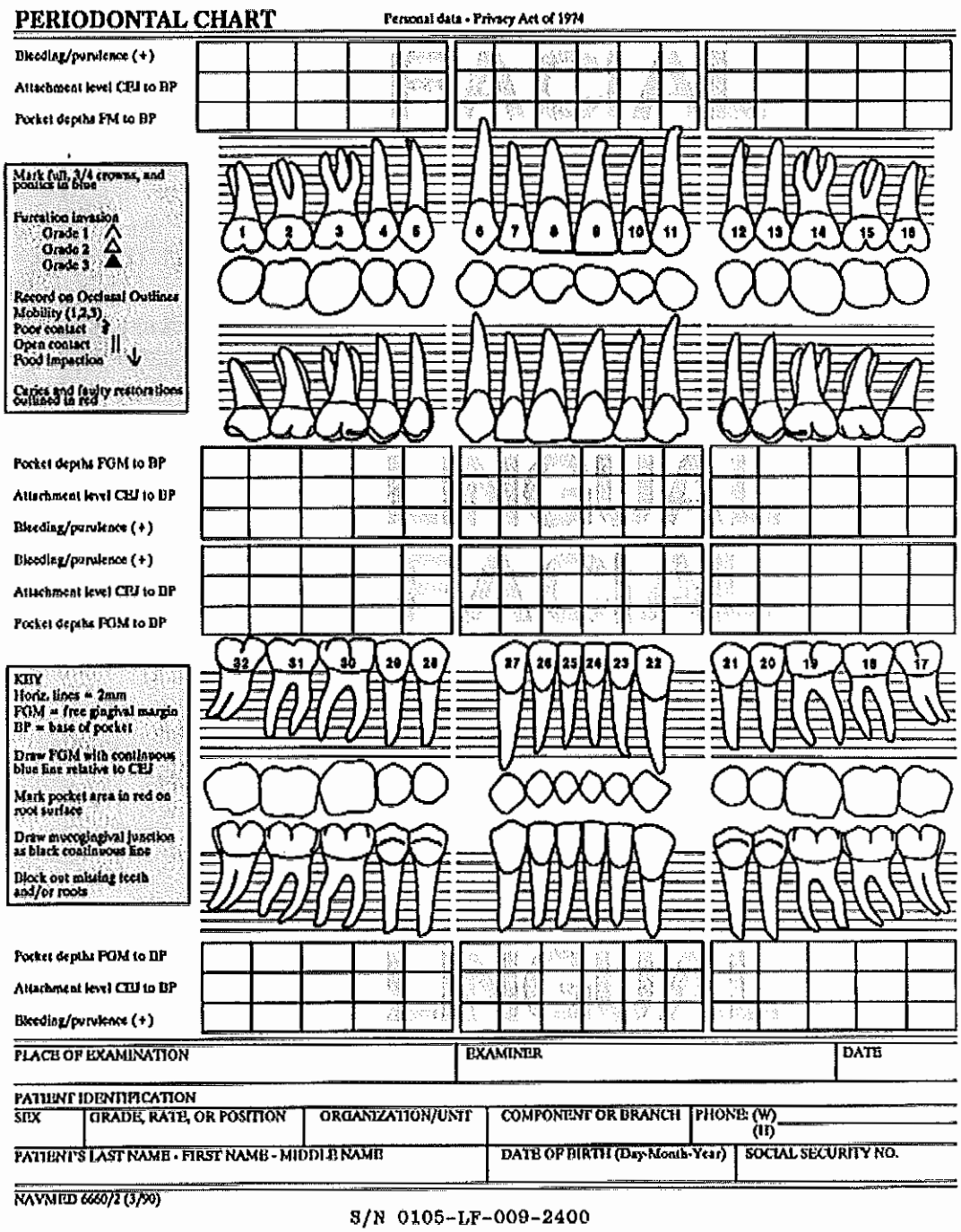
CHAPTER VI: CONLUSIONS

Research will commence following IRB approval.

APPENDIX A: FLOW DIAGRAM OF STUDY DESIGN



APPENDIX B: COMPREHENSIVE PERIODONTAL CHARTING FORM



APPENDIX C: EXAMPLE OF NPDS PERIODONTICS DEPARTMENT POST-OPERATIVE INSTRUCTIONS

PERIODONTICS DEPARTMENT
NAVAL POSTGRADUATE DENTAL SCHOOL
Bethesda, Maryland

For best healing and a minimum of complications, please read and follow these instructions carefully

You may have been given one or more of these medications:

PAIN MEDICATIONS:	_____	Motrin 800 mg:	*1 tablet every 8 hours.* Do not double up on dosage.
	_____	Norco 5/325 mg:	*1 tablet every 6 hours for pain control.* It can be taken in addition to Ibuprofen. This medicine can make you drowsy. Therefore, do not drive or operate machinery while taking this drug. Additionally, do not take with alcoholic beverages; the alcohol will make you sleepier, but will not decrease your comfort.
ANTIBIOTICS:	_____	Doxycyclin 100 mg:	*2 tablets the day of surgery, then 1 tablet every day for 30 days.*
	_____	Amoxicillin 500 mg:	*1 tablet four times a day for 7 to 10 days.*
	_____	Clindamycin 300 mg	*1 tablet four times a day for 7 to 10 days.*
RINSES:	_____	Peridex (Periogard	*1 bottle, rinse twice a day as directed on the bottle, starting the day following surgery.* Do not brush or floss at the surgical site unless instructed to do so.
ANTI-INFLAMMATION:	_____	Medrol Dose Pack:	Take as directed on the package, starting today. Be sure and take the full first row of tablets (first six tablets) today.

The following are a list of post-operative considerations during healing:

BLEEDING:	There may be slight bleeding from the surgical for 1-2 days after surgery. Your saliva may appear slightly reddish. This is common. If you notice an increase in bleeding please contact us.
SUTURES/STITCHES:	You may have sutures placed in your mouth. They may have to be removed in the future. Please leave the sutures alone as much as possible. Early removal or the loss of sutures may impair healing.
DRESSINGS:	There may be a gauze type of dressing placed over the surgical area. It is there for your comfort. If it falls out before your first Post-Operative appointment and you are comfortable, it is fine to leave it out. If the surgical site is uncomfortable and you would like the dressing replaced please contact me.
DIET:	It is very important to maintain a soft diet for at least a week. Chew as much as possible on the side opposite the surgery. This is not the time to start a diet. Please maintain your calorie and fluid intake as at pre-surgical levels. You will not heal well if you are dehydrated or undernourished. Please do not drink using a straw.
ORAL HYGIENE:	It is very important not to brush or floss the surgical site until given express instructions. Normal brushing and flossing procedures can traumatize the tissue and impair healing. You may brush and floss those areas not affected by the surgery. To keep bacteria under control a prescription mouth rinse has been written for you. Initially, use the mouthwash as a rinse. Later you may be instructed to use a cotton-tipped applicator, dipped in the mouthwash, to swab along the gum line of the surgery site. Use a capful (15ml) of the mouthwash twice a day, morning and bedtime, after brushing/flossing your non-surgically treated teeth. You may notice a mild tooth staining as a result of the mouthwash. This is not permanent; the stain will be removed with scaling/polishing at your follow-up appointments. Please do not use a Water-Pik or other irrigator unless instructed to do so.
PHYSICAL ACTIVITY:	Avoid strenuous physical activity (to include running and heavy lifting) for 72 hours. Additionally, no vigorous spitting, rinsing, or spending (pulling). Forceful movements at the site of surgery will negatively affect healing.
SWELLING:	You may experience some swelling. This is common and usually peaks at 2-3 days after surgery. Thereafter you should expect to see a return to normal. To decrease swelling you can apply ice to the site for the first 3-4 hours after surgery. Please call if the swelling appears to increase after the third day, or if you are concerned.
SMOKING	Smoking is deleterious to healing. We advise you to stop smoking for as long as possible after surgery. Stopping smoking will improve potential healing and also improve your overall periodontal health.
FOR SINUS LIFT SURGE PROCEDURES	You may also have received nasal decongestant tablets and spray. Please use these medications as directed on the package. In addition, avoid blowing your nose. If you need to sneeze, please sneeze with your mouth open. Please inform your doctor if you develop sinus congestion that is not minimized with your medications or if you notice any bleeding or discharge from your nose.

If you have any problems or questions, please do not hesitate to call me at 301-295-0077. If there is an emergency you may page your doctor through an automated system. Instructions will be given after dialing 1-800-755-8888. The PIN# for your doctor is_____

Your follow up appointment is scheduled for: _____ _____

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