CLINICAL PROTOCOL TITLE: **A Phase I/II Prospective, Randomized, Multicenter, Double-Masked, Vehicle-Controlled Clinical Trial to Evaluate the Safety and Efficacy of Corneal Collagen Cross-Linking of Keratoprosthesis Carrier Tissue in High-Risk Keratoprosthesis Implantation**

Version	Date last Revised	Date of IRB Approval	Notes
3.0	9/10/2018	10/25/2018	Changed minimum time in study from 104 to 52 weeks
3.0	12/11/2018	12/16/2018	Minor corrections to clarify changes made in October, 2018.
3.1	3/13/2019	03/21/2019	Improvements to the subject recruitment and consent process (section 7.1)
4.0	**1/10/2020**		**Changes to eligibility criteria, informed consent procedures, case report forms, manual of procedures, DSMC charter. Adding Adverse Events that are to be excluded from reporting requirements and Monitoring Plan.**

1. Signature Page

Principal Investigator Signature Page

IRB Protocol #:	2019P000428
IND Number:	108,059
Investigational Drug(s):	Riboflavin (vitamin B2) 0.1% solution and dextran 20% solution
Regulatory Sponsor & Coordinating Center:	Joseph Ciolino, MD Massachusetts Eye and Ear Infirmary 243 Charles St. Boston, MA 02114
Funding Sponsor:	United States Department of Defense
DSMC Chair:	Dean Cestari, MD

I have read the protocol and supporting documents and I agree that it contains all necessary details for me and my staff to conduct this study as described. I will conduct this study as outlined herein and will make reasonable efforts to complete the study within the time designated. I will provide all the study personnel under my supervision adequate training to ensure that they are fully aware of the study and its procedures.

Site Investigator Name	Site Investigator Signature
Date	Site Name

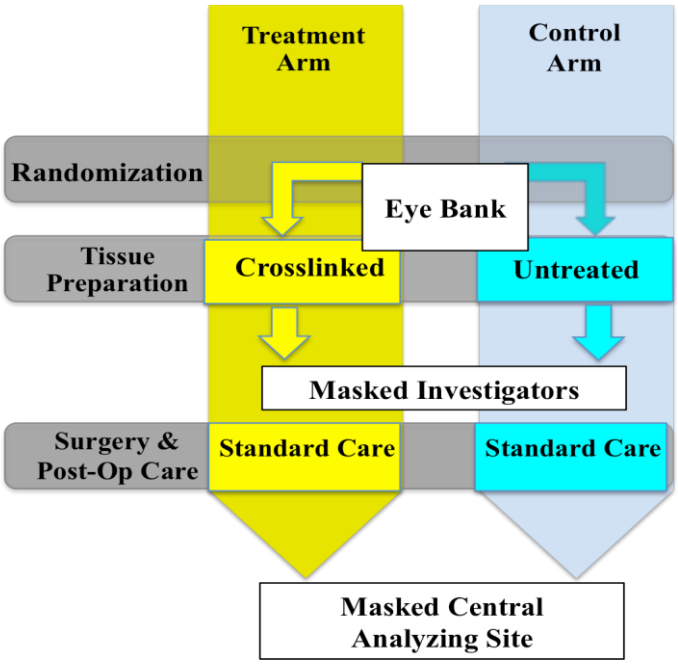
Sponsor representative signature:

Dean Cestari, MD

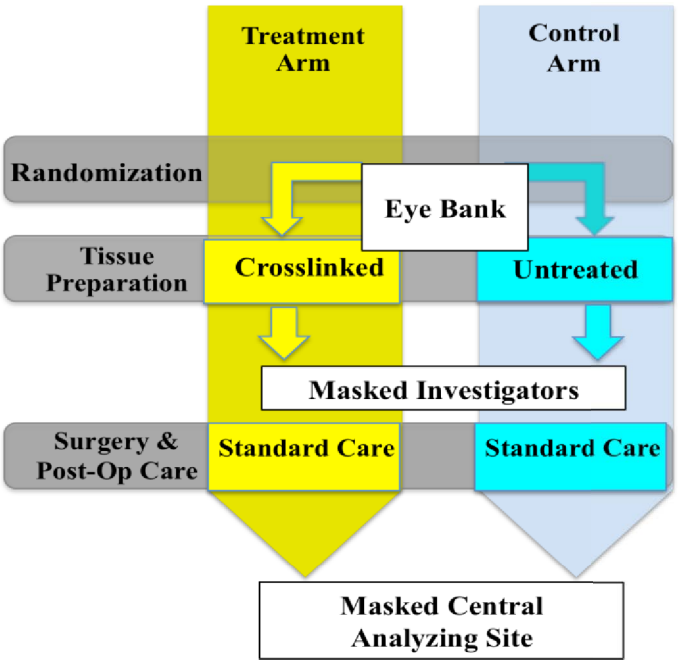
DSMC Chair and Medical Monitor

Name	Signature
Date	Site Name

STUDY DESIGN SCHEMATIC



STUDY DESIGN SCHEMATIC



1. CLINICAL PROTOCOL

1.1 Background

The cornea is an optically clear tissue that can become opaque as the result of a variety of pathological conditions. Most conditions can be treated with a cadaveric cornea transplant. However, under some pathological conditions, such as autoimmune diseases (Steven Johnson Syndrome, Ocular Cicatricial Pemphigoid, Rheumatoid Arthritis), and previous cornea transplant graft failure, repeat corneal transplants have a very poor likelihood of success.

Additionally, due to the changes in survival, munitions, and style of warfare, the proportion of surviving soldiers with serious anterior segment eye trauma has progressively escalated over the last century. According to Veterans Affairs (VA) data, eye trauma accounted for 15% of all battlefield injuries during Operation Iraqi Freedom and Operation Enduring Freedom, resulting in over 186,000 eye injuries in American soldiers between 2000 and 2010. These soldiers and veterans with devastating corneal injuries and autoimmune disorders associated with traumatic and degenerative changes of the cornea also have a very poor likelihood of cornea transplant success.

In these high risk patient populations, a keratoprosthesis (artificial cornea) is used. The Boston keratoprosthesis (Boston KPro) is an artificial cornea developed at the Massachusetts Eye and Ear Infirmary under the leadership of Dr. Claes Dohlman. It was FDA-approved in 1992 for use in subjects with repeated corneal graft failures, ocular chemical burns, autoimmune and cicatrizing diseases, aniridia and ocular herpetic infections. This increasingly popular prosthesis has helped restore vision to thousands of subjects who otherwise had no other viable alternatives. However, despite the modifications made to improve the Boston KPro over the years, some of these keratoprosthesis subjects go on to develop sterile ulcerations and thinning of the cornea tissue which acts as a carrier for the keratoprosthesis. This corneal melting (keratolysis) of the donor cornea still remains a rare yet devastating post-operative complication that leads to exposure to pathogens, perforation, leakage of the aqueous humor and extrusion of the keratoprosthesis.

While bench-top studies and small case series have suggested that corneal melts can be reduced by collagen cross-linking the donor cornea, a prospective, randomized, multicenter, double-masked, vehicle-controlled study is needed to evaluate the safety and efficacy of cross-linking of the keratoprosthesis carrier cornea.

1.2 Rationale

The experimental drug for this trial is riboflavin (vitamin B2) 0.1% solution and dextran 20% solution. The medication includes a formula that is applied topically to the cornea while the tissue is exposed to UV light. The riboflavin reduces the intensity of the UV light that penetrates the cornea by 95%.

Cross-linking is a term that refers to the linking of polymers (long chain) molecules by chemical bonds. It is commonly performed in industry when manufacturing synthetic materials. Biologically, cross-linking has been used to modify or strengthen tissue such as porcine valves used in heart surgery. Corneal cross-linking was first suggested in Germany in the 1990s. Since the cornea is predominantly made of long structural proteins called collagen, it was theorized that cross-linking the collagen should make the cornea stronger and resistant to degradation. The combined use of ultraviolet A light and riboflavin (as a photosensitizer) was shown to be a simple and effective cross-linking technique. Animal studies have shown this method to be safe. In addition, researchers found cross-linked corneas to be highly resistant to enzymatic degradation.

Human clinical trials have been conducted in Europe for the use of riboflavin/UVA corneal cross-linking in subjects with ocular diseases that involve corneal thinning such as keratoconus. Such trials are currently underway in the United States. There are many published reports demonstrating the safety of this method in humans. The Massachusetts Eye and Ear Infirmary has been approved to join one of the multi-center study trials using the Caporossi-Baiocchi-Mazzotta (CBM) VEGA X-linker). The goal of our study is to use the same protocol designed for that study for our study. The exception is that the corneal cross-linking of the donor corneal tissue will be performed ex-vivo prior to the Boston KPro assembly. The Boston KPro assembly will otherwise remain unchanged and the patient will not be exposed to the UVA light.

Unfortunately, there are no good animal models to test our technique with the Boston KPro. Rabbits are poor models since their clinical results have notoriously poor correlations with human clinical outcomes.

Initially, we worked on determining the optimal riboflavin/UVA cross-linking method to increase resistance of the corneas to enzymatic degradation ex-vivo.

The methodology was as follows. The corneal epithelia from the enucleated eyes of mature white New Zealand rabbits were mechanically removed using a 57 blade. The corneas with 2-3mm scleral rims were excised and their irises were removed. They were fitted into Barron® artificial anterior chambers with the endothelium bathed in isotonic saline solution. The VEGA® LED-based UVA emitter was switched on for at least 30 minutes prior to calibration then was calibrated before the experiments. The corneas were then pre-treated with 0.1% riboflavin/20% dextran T500 every 2-3 minutes for 15 minutes followed by UVA light (λ= 370nm, irradiation = 3mW/cm2) at a fixed distance of 54mm from the top of the corneas for 5-minute intervals, while applying riboflavin between irradiations. Either the anterior stroma (A) or the anterior (A) and posterior stroma (P) was irradiated for different durations (7.5A, 7.5A/7.5P, 15A, 15A/15P, 30A, 30A/30P and 60A minutes). The primary control was no riboflavin/no UVA. Secondary controls included riboflavin/no UVA and UVA/no riboflavin. The corneas were trephined into 8.5-mm buttons and incubated in clean glass vials containing 0.3% collagenase A solutions at 37°C and rotating at 150 rotations-per-minute. The time to total dissolution of the corneas was measured. Each group included 4 or more corneas.

Results showed that the primary control dissolved at 4.8h ±0.8 while tissue cross-linked for 15 minutes dissolved after 50.0 +/- 8.1 hours. Compared to the control, there was a marked increase in resistance to degradation for all UVA/riboflavin cross-linked corneas. In general, anterior stromal cross-linking was superior to anterior/posterior stromal cross-linking (whether the irradiation time was split or doubled between the anterior and posterior stromata compared to their matched groups of anteriorly cross-linked corneas). The minimal treatment that yielded the greatest resistance in all groups was 15 minutes of anterior stromal cross-linking (50.0h □8.1; $p<0.0001$). The results were similar for 15, 30, and 60 minutes of cross-linking. Therefore, we identified several methods to increase the corneal resistance to enzymatic degradation using the corneal cross-linking technique.

The UVA emitter emits constant irradiation energy of 3mW/cm2 that requires the UVA light source to be at 54mm from the cross-linked cornea. The irradiated area can be controlled using

the UV spot aperture control. This fixed setup ensures homogenous irradiation of the corneal surface with the desired fixed energy. Unfortunately, this UVA emitter was not designed to irradiate at any energies other than 3mW/cm2. Changing the distance will vary the size of the irradiated area (and possibly the energies unpredictably). The new irradiation area created will not be commensurate with the desired area we set up on the aperture control. In addition, we cannot guarantee that the new area created would provide homogenous irradiation. Therefore, we will use the manufacturer's recommended distance of 54 mm with constant irradiation energy of 3mW/cm2. The VEGA emitter above and Avedro KXL UVA emitter to be used in this study have equivalent light settings and intensity.

We also studied the assembly of the Boston keratoprosthesis device in 3 human cross-linked corneas using the same methodology described above (after 30 minutes of cross-linking). The Boston KPro devices were assembled with cross-linked tissue and untreated control cornea tissue. Dr. Dohlman, Dr. Ciolino and Dr. Chodosh inspected the assembled KPro units and found that grossly and microscopically, the Boston KPro devices fit securely around the cross-linked tissue. There did not appear to be any difference in the Boston KPro assembly between the cross-linked corneas and 3 untreated human control corneas.

Three subjects received KPros in collagen cross-linked corneas in Athens, Greece under the care of Dr. John Kanellopoulos who also practices in New York. The P.I. (Dr. Ciolino) has traveled to Athens and performed a chart review on the three cases and has maintained communication with Dr. Kanellopoulos on the status of the three cases. At last follow-up, all the cases were doing well despite their history of keratolysis and Dr. Kanellopoulos has since published a paper on 9 such cases. The technique used in this project is very similar to the procedure used by Dr. Kanellopoulos.

This study's population will include subjects who are both candidates for a Boston KPro and had either a history of corneal melting (keratolysis) or have high risk for corneal melting. Boston KPro candidates include subjects with visual loss due to corneal scarring that are not candidates for a traditional corneal transplant. Typically, these subjects have either failed a corneal transplant or have ocular findings that strongly predict a very high likelihood of cornea transplant failure (e.g. subjects with extensive neovascularization of the cornea). Subjects at high risk for corneal melt include, but are not limited to, those with underlying autoimmune diseases such as ocular cicatricial pemphigoid, Stevens Johnson Syndrome, rheumatoid arthritis, lupus, and others. We are aiming to enroll 84 subjects, male or female, ages greater than 18, who are

otherwise healthy enough to undergo an elective procedure such as a corneal transplant. More information on the specific inclusion and exclusion criteria can be found in section 4 below.

Accumulating experimental data and the fact that collagen crosslinked cornea tissue resists enzymatic digestion suggest that crosslinked cornea tissue may resist the sterile cornea ulcers that develop in keratoprosthesis subjects. This approach would be a significant step forward in addressing the need for an improved treatment modality in this desperate patient population.

Riboflavin and UV light are used and marketed outside most of the United States for the treatment of corneal ectasias (keratoconus), but not for this purpose of preventing corneal melts. To our knowledge, the drugs and devices have not been withdrawn from any research or market due to reasons related to safety or efficacy.

2. STUDY OBJECTIVES

2.1 Primary Objective

This is a phase I/II prospective, randomized, multicenter, double-masked, vehicle-controlled clinical trial evaluating the safety and efficacy of corneal collagen cross-linking the keratoprosthesis carrier tissue in subjects who are candidates for high-risk keratoprosthesis implantation because of a history of corneal melts, sterile corneal ulcers, or autoimmune diseases (e.g., Stevens-Johnson syndrome, ocular cicatricial pemphigoid).

Objective 1:

Determine the safety of using a collagen cross-linked cornea as a carrier for the Boston Keratoprosthesis in subjects who are at high risk for corneal melts and are not candidates for a standard corneal transplant.

Objective 2:

Determine the efficacy of using a collagen cross-linked cornea as a carrier for the Boston Keratoprosthesis in preventing corneal melts and increasing the retention of the keratoprosthesis. This will be tested using the outcome measures listed in section 10.

2.2 Secondary Objective(s):

N/A

3. STUDY DESIGN

3.1 Study Design Description

The study is a multi-center, parallel, randomized, double-blinded study with a subsequent follow-up period of 2 years (week 52) and standard of care follow up every 6 months for an additional 3 years or until study closure to collect event data. Once a patient has been identified as a potential study participant who informed consent for this study will be obtained and eligibility will be confirmedWhen a screened participant meets the inclusion and exclusion criteria to be enrolled in the study, the investigator will request a cadaveric cornea from CorneaGen (previously KeraLink International) the central eye bank for this study. The eye bank will assign the subject the next available subject (randomization) number, in ascending sequential order according to the block schedule until all assignments have been used. The eye bank will then prepare the cadaveric cornea according to one of following two randomization groups in equal allocations:

<u>Group 1: Active Treatment Arm</u>

De-epithelialized corneas will be cross-linked with riboflavin 0.1% with dextran 20% AND ultraviolet A light.

<u>Group 2: Control Treatment Arm</u>

- De-epithelialized corneas will be exposed to only riboflavin 0.1% with dextran 20% (NO ultraviolet A light).

As detailed in section 5, both the treatment arm and control arm corneas will be labeled with their unique randomized identification number in a sealed bottle of OptiZol® solution, as per the standard of care for cornea donor preservation. The tissue bank will follow their standard operating procedures for the packaging and shipment of the tissue. The masked corneas will be sent to the masked investigators, who will perform the assembly and implantation of the Boston Keratoprosthesis. The procedure will be unchanged from a typical Keratoprosthesis assembly and insertion (see study procedures in section 7).

Subjects will be followed up post-operatively by the site investigators as standard for this procedure. This will include visits at Day 1, Weeks 1, 4, 16, 24, 36, and 52, . Subjects following Boston KPro surgery are typically seen every 6months after one year follow-up. However, some may need to be seen more regularly if they require more frequent monitoring for conditions such as glaucoma. The investigators will monitor for systemic and ocular adverse events at all follow-up visits. All adverse events will be reported to the principal investigator and Medical Monitor; and then to the MEEI HSC, sub-sites, DOD, and FDA per the specified guidelines.

4.2 Allocation/Randomization to Treatment

Eighty-four subjects will be randomized equally to treatment or control group. Randomization will be stratified by 16 centers, as well as two pre-operative diagnostic categories: autoimmune disease vs. prior corneal melt without autoimmune disease. Thus there will be a total of 32 strata. Stratification will ensure balance of the stratification factors between the two arms of the study. A number of blocks of varying size will be determined (e.g. 5 blocks of size 4, 6 blocks of size 2) and randomization assignments will be prepared accordingly. Since we have 32 strata, relatively small block sizes will be planned to minimize the chance of large departures from the desired allocation ratio that could occur due to incomplete blocks by the time of completion of patient recruitment. Although the ability of the clinical staff to predict future treatment assignments is increased due to smaller block sizes, we do not think that this will be a major issue for the present study.

The study statistician at Mass Eye and Ear will prepare the computer-generated randomization schedules and treatment strata prior to commencement of the study. Codes will be used to label different treatment assignments so study site personnel will be blinded to treatment allocation. The randomization schedule will be determined in 4 steps: 1) arrange the initial treatment assignments arbitrarily, 2) generate via computer a list of pseudorandom numbers, 3) link the initial treatment codes with the pseudorandom numbers, 4) order the pseudorandom numbers from smallest to largest with associated treatment codes to determine the final order of treatment assignments. The final treatment order and strata will be communicated by the MEE unblinded study staff directly to CorneaGen prior to study start up.

4.2.1 Masking Procedures

When a participant is enrolled into the study, the Coordinator at each site will request a cadaveric cornea from CorneaGen for each eligible subject. The eye bank will use the

randomization schedules according to the stratum of the patient (center and autoimmune disease status) and assign the subject the next treatment available. Each consecutive patient who falls into the same stratum will be assigned the next treatment assignment on this sheet until all assignments have been used. Whenever a patient is enrolled who falls into a stratum for which no sheets have been started or for which all previous sheets have been completed, the next sheet will be used according to the appropriate stratum, and so on. After the tissue is prepared, it will be sent to the site in a bottle labeled with the subject's identification number.

4.2.2 Breaking the Mask

In the event of an emergency, the investigator can call CorneaGen to unmask the study treatment for an individual subject.

5. SUBJECT SELECTION

5.1 Subject Inclusion Criteria

The eligibility criteria must be met at the screening visit, and relevant medical and non-medical conditions will be taken into consideration when deciding whether this protocol is suitable for a particular subject. Subjects will be screened using the following inclusion and exclusion criteria, which are designed to select subjects for whom the protocol treatment is considered appropriate:

- Willing and able to provide written informed consent
- Willing and able to comply with study assessments for the full duration of the study
- Age ≥ 18 years
- Candidate for a Boston Keratoprosthesis / Cornea transplant
- Subjects with an eye at risk for a cornea sterile ulcer which includes:
    - Autoimmune diseases (mucus membrane pemphigoid, Stevens-Johnson syndrome, systemic lupus erythematosis, and rheumatoid arthritis)

    OR
    - History of previous sterile cornea ulceration

5.2 Subject Exclusion Criteria

- Age < 18 years
- Inability to provide written informed consent and comply with study assessments for the full duration of the study
- No or minimal tear production with low tear lake with evidence of keratinization of the bulbar conjunctiva based on clinical assessment
- Ocular surface infection within 30 days prior to study entry
- Confirmed or suspected ocular or periocular malignancy based on pathological or clinical assessment
- Inability to wear a contact lens due to lid abnormalities or shortened fornix as evidenced by clinical assessment
- Pregnancy (positive urine pregnancy test) or lactating
- Participation in another simultaneous interventional medical investigation or trial

- Review of patient medical record
- Patient self-reported

6. STUDY DRUG(S)/DEVICE(S)

6.1 Study Drug/Device Information

Riboflavin Solution and Ultraviolet A Light System.

The experimental drug for this trial is Riboflavin solution (0.1% riboflavin and 20% dextran supplied in a sterile, single-dose container). Avedro Inc. (Baltimore, MD) is supplying both the 0.1% riboflavin and 20% dextran ophthalmic solution and the UV light source (KXL system) that are manufactured, tested and released according to current good manufacturing practices (cGMP) and the FDA's quality system regulations. The riboflavin being used is the same solution that is currently under investigation in the United States in multiple clinical trials. In this trial, the medication is applied topically to the cornea while the tissue in the active group is exposed to UV light thus reducing the intensity of the UV light by 95%. Tissue in the control group will have the medication applied without exposure to UV light.

Active Treatment Arm

The donor cornea will be placed on an artificial anterior chamber maintainer (a device which holds the cornea in place while bathing the cornea endothelium in saline [balanced salt solution]). The donor corneal epithelium will be removed mechanically. Riboflavin solution (0.1% riboflavin and 20% dextran supplied in a sterile, single-dose container) will be applied to the cornea every 2-3 minutes for 15 minutes prior to the beginning of the UV light application. The riboflavin solution will be supplied as a 3-mL solution by Avedro, Inc. and is the same solution that is currently under investigation in the United States in multiple clinical trials. The UV source will be from the KXL® System (Avedro, Inc.). The machine is first calibrated to ensure irradiation energies of 3.0 mW/cm2. A wavelength of 365 nm will be used to direct 5.4 J/cm^2 using a beam diameter of 9.5mm to treat the de-epithelialized corneal surface for 30 minutes. Every 5 minutes, the UV light will be used while another drop of riboflavin is applied. Once the cross-linking is completed, the cornea will be rinsed profusely with sterile normal saline to remove any riboflavin remaining on the surface. As is the standard of care, the cornea

will be placed in a sterile container containing OptiZol® solution, sealed, and labeled with the subject identification number.

Control Treatment Arm

Subjects in the control arm will receive corneas that have NOT been cross-linked, but have undergone procedures to ensure that the control tissue looks the same as the treated tissue. These steps will be performed in order to reduce the likelihood that the masked investigators would become aware of the tissue assignment. The corneas in the control arm will be placed on an anterior chamber maintainer and de-epithelialized in the same manner as the tissues in the treatment arm. The control arm tissue will then be topically administered riboflavin every 2-3 minutes for 15 minutes given that the riboflavin can cause the tissue to develop a slight yellow tint even after thorough rinsing. The control arm corneas will NOT be exposed to UV light. Importantly, corneas treated with riboflavin without UV light have demonstrated the same resistance to enzymatic digestion as corneas which have not undergone cross-linking corneas. Like the corneas in the treatment arm, the control arm corneas will be rinsed profusely with normal saline, placed in a sterile container containing OptiZol® solution, sealed, and labeled with the subject identification number.

6.2 Study Drug/Device Compliance/Adherence

Subjects will not be administering the study drug.

6.3 Study Drug Supplies

Avedro Inc. will be providing the Riboflavin solution directly to CorneaGen.

6.4 Study Drug/Device Storage and Accountability

All logs will be maintained by CorneaGen.

6.5 Other Medications

Study site Principal Investigators must closely monitor and confirm with subjects that Ocular Con-Meds are being used correctly.

6.6 Administration

Subjects may continue to receive all medications and standard treatments administered for their conditions at the discretion of their treating physician. All concomitant medications will be reviewed at each study visit.

6.7 Concomitant Medication

Antibiotics:

First line recommended antibiotic prophylaxis consists of polytrim and vancomycin (14 mg/ml with or without preservatives).

Polytrim should be given as one drop 4 times a day until full graft epithelialization at which point the antibiotic will be decreased to once a day, or as necessary with regard to each patient's needs per the discretion of the study investigator.

In monocular subjects only, fortified vancomycin 14 mg/mL topical drops will be used once a day for the study duration based on the drug availability and upon the discretion of the site principal investigator.

Steroids:

All subjects will receive topical 1% prednisolone acetate drops 4x/day for 6 weeks, 3x/day for 6 weeks, 2x/day for 6 weeks, then 1x/ day for the remainder of the first 6 post-operative months. After 6 months, the subjects can receive pred mild, lotemox, FML or another low dose steroid at least once a day.

Subjects who develop sterile vitritis or another inflammatory condition may require higher doses of steroids and the regimen will be recorded.

Subjects who develop elevated intraocular pressure secondary to steroids should be managed, if possible, by initiating anti-hypertensive therapy as per standard ‘glaucoma’ care regimens. If this proves inadequate and the intraocular pressure management becomes difficult, the dose (frequency and potency) of the topical corticosteroid will be reduced, especially if there is a

history of steroid-responsiveness. If maximally tolerated medical therapy does not sufficiently control intraocular pressure, alternative approaches to intraocular pressure control should be considered.

NSAIDs:

NSAIDs should be excluded unless needed for cystoid macular edema.

Allergies and Contraindications: The medications can be substituted with alternative equivalents if a patient has an allergy or a contraindication to a specific medication noted above.

6.8 Rescue Medication or Therapy

Subjects who do not retain the cross-linked KPro will be withdrawn from the study and the cornea will be replaced with a cadaver cornea or standard KPro at the physician's discretion.

7. BIOSPECIMEN COLLECTION

7.1 Specimen preparation, handling, and shipping

N/A

7.2 Instruction for specimen preparation, handling and storage

N/A

7.3 Specimen shipment

N/A

7.4 Future use of stored specimens

N/A

8. STUDY PROCEDURES

8.1 Screening Process

Potential study participants will be identified by the study investigators at the designated study sites. All qualified patients who:

- meet the inclusion/exclusion criteria in section 4,
- are determined to be candidates for KPro surgery,
- have discussed the risks and potential benefits of that surgery with a study licensed physician, and
- state their intent to have that surgery will be asked if they are interested in hearing more about the study.

If the patient expresses interest in the study, the licensed physician/investigator will review the study design and treatment regimen with him or her, and will go over the information in the consent form. If the patient states that he or she is interested in participating in the study, a screening visit will be scheduled, and the patient will be given a copy of the consent to take home and read. Patients are encouraged to go over the consent form with other health care providers.

Those subjects who volunteer to participate (after signing the written informed consent) will be enrolled consecutively during routine visits to the study sites. The study eye will be identified at the screening visit. Only one eye of each eligible patient will be enrolled into the study. Standard subject reimbursement ($75) is provided at visit one (screening visit) and the final visit (visit week 52) to cover parking, meals and other incidentals associated with the patient's participation in the study. Visit one includes the consent process and the final visit will include a number of imaging steps not required for clinical care. All non-standard assessments will be covered by the study funds.

Candidate subjects that live at a great distance from the study site and are unable to attend a screening visit and provide signed informed before the day of the scheduled surgery may be

allowed to provide verbal consent for the purposed of screening of medical record and randomization (the ordering of the tissue from the tissue bank). In this event the subject in question will be consented following the "Remote Consent" procedures described below.

8.2 Consent Process:

8.2.1 Remote Consent

Candidate subjects that live at a great distance from the study site and/or are unable to attend a screening visit and provide signed informed before the day of the scheduled surgery may be allowed to provide remote consent for the purpose of screening of medical record and randomization (the ordering of the cornea tissue from the tissue bank). In order for the subject to be eligible for remote consent the following process has to be followed.

1. When a potential study subject is identified by study staff the MD investigator will review the study and the consent form with the candidate subject at their next visit. If the candidate subject expresses interest in the study but needs additional time to consider participation the study MD will give the candidate subject a copy of the consent form and ask that the candidate subject call the study staff if they are interested in participating in the research study.
2. When the candidate subject calls study staff and confirm their interest in participating in the study, the study staff will schedule a time for the candidate subject to speak to the MD investigator. Study staff will also mail the candidate subject a new copy of the study Informed Consent Form.
3. At the time of the scheduled call the MD investigator will review the consent form with the subject over the phone. MD investigator will answer all subject questions.
4. If the candidate subject agrees to participate in the research study the MD investigator will ask the candidate subject to sign, date and time the consent form. Once the consent form is signed the MD investigator will ask the study subject to mail the signed consent form to the study staff.
5. After the phone call is compete the MD investigator will complete and sign the "Informed Consent Checklist" to affirm that consenting procedures were followed.
6. When the study staff received the signed consent form over the mail the MD investigator will sign the consent form.

7. Study subjects that have provided remote consent will be re-consented in person by the MD investigator at their next scheduled visit (day of surgery). MD investigator will complete a new "Informed Consent Checklist" to record this in person consent. Study subjects will be given a copy of both of the signed consent form.
8. Both consent forms (remote & in person) will be filed in the subject study binder and a note will be made in the patient's medical record to note their participation in the study.

8.2.2 Written Consent

At the start of the screening visit, the MD investigator will go over the consent form in person with the potential study subject, answering any questions that the patient may have. The MD Investigator will specifically review the study procedures, visit schedule, risk and benefits, alternative treatments and rights to withdraw and ask questions with all potential subjects before signing the consent form. If the patient agrees to participate in the trial, the consent form will be signed in front of the MD investigator who will also sign and date the consent form. A copy of the signed consent form will be given to the patient and a note will be made in the patient's medical record to note their participation in the study. The study-specific screening evaluations will then be performed.

A note will be made on the study record that the informed consent was signed by the participant. The informed consent process will follow the guidelines set by the relevant site's IRB. Every patient has a right to withdraw at any time from the study without affecting their care or relationship with the treating physician and participating institution. The financial responsibilities of the patient will be discussed

8.3 Screening Procedures:

Screening evaluations and eligibility will be determined by the MD investigator at the screening visit within 60 days of the scheduled Boston KPro surgery (Day 0). The following evaluations and procedures will be performed for all subjects during the screening period unless otherwise specified below:

- Written informed study consent
- Record current ocular and systemic medications
- Record significant medical/surgical history in the past 5 years

- Record demographic data, including date of birth, sex, and race/ethnicity
- Review of systems
    - Constitutional, Ear/Nose/Throat, Respiratory, Cardiovascular, Gastrointestinal, Genital/Urinary, Integmentary, Musc/Skeletal, Endocrine, Heme/Lymph, Allergic/Immun, Neurologic, Psychiatric, All Others
- Urine pregnancy test for women of childbearing potential, if appropriate (May be repeated the day of surgery if surgery is greater than 30 days from screening.)
- Ocular assessments (study eye)
    - Best spectacle-corrected visual acuity (BCVA)
    - Slitlamp examination
    - Intraocular pressure
    - Funduscopy or ultrasound (B-scan) evaluation of the posterior segment of the eye if there is no adequate view of the retina
    - Slit Lamp Photography

Historical results for best corrected VA, intraocular pressure, slit lamp examination, slit lamp photography and funduscopy can be used if completed within 60 days prior to screening visit

8.4 Day 0: Drug or Device Procedures

Study drug will only be administered by CorneaGen prior to shipment to MEEI as described in section 5.1. Implantation of the Boston Keratoprosthesis unit may occur up to 60 days after the screening visit. When applicable, the pregnancy test will be repeated on day 0 or within 30 days of the scheduled surgery.

<u>Keratoprosthesis Implantation Procedure</u>

Subjects will undergo keratoprosthesis implantation under general or retrobulbar/peribulbar anesthesia. Massed corneal donor tissues will be removed from the Optisol GS storage media. The donor cornea will be trephined from the endothelial surface. Trephine size will be selected according to recipient corneal size (range 7.0–9.0 mm), and the donor–recipient disparity will be 0.25–0.50 mm. In addition, a central 3-mm opening will be punched into the donor cornea, as standard for the Boston Keratoprosthesis. The remainder of the cornea donor tissue is placed on

top of the keratoprosthesis front plate. A back plate is then placed on top of the cornea donor tissue and secured in place to complete the keratoprosthesis unit.

As a unit, keratoprosthesis and donor tissue are then transferred to the patient's eye and sutured into position using interrupted sutures, not unlike a traditional cornea transplant. A 10-0 or 9-0 nylon suture material will be used. The technique of suturing will be interrupted since the risk of suture-related complications with running sutures is higher in vascularized host beds, which are likely present in the majority of keratoprosthesis candidates. Moreover, running sutures are often used to decrease post-operative astigmatism, but it is not relevant in the case of a keratoprosthesis since there is no astigmatism induced by the corneal sutures. The keratoprosthesis optic corrects for any corneal astigmatism. The keratoprosthesis implantation may be combined with other procedures such as cataract extraction, intraocular lens implantation, glaucoma procedures, and vitrectomy procedures as necessary with regard to each patient's needs. At the conclusion of the case, the bandage contact lens) that accompanies the Boston Keratoprosthesis device will be placed on the eye.

Post-Operative Care

All subjects will receive the standard treatment with corticosteroid and antibiotic eye drops. All subjects will receive topical 1% prednisolone acetate drops 4x/day for 6 weeks, 3x/day for 6 weeks, 2x/day for 6 weeks, then 1x/ day for the remainder of the first 6 post-operative months. After 6 months, the subjects can receive pred mild, lotemox, FML or another low dose steroid at least once a day.

Subjects who develop sterile vitritis or another inflammatory condition may require higher doses of steroids and the regimen will be recorded.

Subjects who develop elevated intraocular pressure secondary to steroids should be managed, if possible, by initiating anti-hypertensive therapy as per standard 'glaucoma' care regimens. If this proves inadequate and the intraocular pressure management becomes difficult, the dose (frequency and potency) of the topical corticosteroid will be reduced, especially if there is a history of steroid-responsiveness. If maximally tolerated medical therapy does not sufficiently control intraocular pressure, alternative approaches to intraocular pressure control should be considered.

In addition to standard treatment with corticosteroids, subjects will receive topical antibiotics four times a day until full graft epithelialization at which point the antibiotic will be decreased to once a day, or as necessary with regard to each patient's needs. In monocular subjects only, fortified vancomycin 14 mg/mL topical drops will be used once a day based on the drug availability and upon the discretion of the site principal investigator.

There may be specific features in a given patient that will lead the investigator to use a different post-operative regimen; therefore we will record the use of all medications at each visit.

A bandage contact lens (Kontur Kontact Lens Co, Inc.) will be worn continuously on the study eye. The diameter, base curve, brand, and replacement schedule will depend on the patient's needs. The presence and type of contact lens used will be recorded at each visit.

All subjects will be instructed in the signs and symptoms of infection or vitritis (decrease in vision; redness, pain or irritation persisting for more than a few hours; painful sensitivity to strong light) are urged to contact their ophthalmologist immediately should they develop such symptoms.

8.5 Standard of Care Procedures

The following procedures will be performed as standard of care and included in study analysis:

- At screening:
    - Informed consent (must be obtained before screening starts and before surgery) Record current ocular and systemic medications
    - Record significant medical/surgical history in the past 5 years
    - Record demographic data, including date of birth, sex, and race/ethnicity
    - Review of Systems
        - Constitutional, Ear/Nose/Throat, Respiratory, Cardiovascular, Gastrointestinal, Genital/Urinary, Integmentary, Musc/Skeletal, Endocrine, Heme/Lymph, Allergic/Immun, Neurologic, Psychiatric, All Others
    - Best corrected VA assessment at 4 meters (with refraction)
    - Intraocular pressure (measured by digital palpation)
    - Slit lamp examination

- Slit lamp photography
- Funduscopy (Ultrasound B-Scan will be performed, if necessary).
  Historical results for best corrected VA, intraocular pressure, slit lamp examine, slit lamp photography and funduscopy can be used if completed within 60 days prior to screening visit

- At day 0:
  - Keratoprosthesis implantation

- At day 1:

Day 1 visits will be performed by MEE clinical staff (physicians) that are not study investigators. Post-operative appointments with physicians are standard of care treatments for patients that have surgery. However study investigators that perform the surgery often do not hold clinical appointments every day of the week and therefore are unable to see the patients for the Day 1 visit. In order to ensure subject safety study patients are scheduled with available MEE physicians. The following procedures are performed as standard of care. Study staff will collect the data from these exams from the subject's medical record.

- Best corrected VA assessment at 4 meters (with refraction)
- Intraocular pressure (measured by digital palpation)
- Slit lamp examination
- Funduscopy/ Ultrasound B-Scan (if necessary)

- At weeks 1, 4, 16, 24, 36, and 52:
  - Best corrected VA assessment at 4 meters (with refraction)
  - Intraocular pressure (measured by digital palpation)
  - Slit lamp examination
  - Funduscopy/ Ultrasound B-Scan (if necessary)
  - Slit Lamp Photography
  - Cornea thickness as measured by AS-OCT

- At all times following day 0:
    - Topical steroids
    - Topical antibiotics
    - Bandage contact lens

8.6 Follow-up Procedures

Additionally, to determine the safety and efficacy of using riboflavin-ultraviolet light induced collagen cross-linked cornea tissue in promoting retention, follow-up visits will be scheduled at Day 1 and 7 (±3 days), week 4 (±3 days), week 16 (±2 weeks), week 24 (±3 weeks), week 36 (±3 weeks), and week 52 (±4 weeks). Following week 52 visit, standard of care visits will occur every six months. The study will continue to collect data from these standard of care visits (post week 52 visit) in order to monitor subject safety for an additional 3 years or until study closure. Any procedures/exams performed during these visits will be standard of care and the MD will decide which procedures/exams are needed. Study staff will collect data for any procedures/exams that may be conducted as they pertain to the study (changes in medical history and medication prescription; BCVA, slit-lamp exam, IOP, funduscopy/B-Scan, OCT images, slit lamp photography and review of potential adverse events).

A description of imaging procedures follow in this section and a table of follow-up procedures organized by time-point can be found in section 7.7 below.

Cornea Thickness as measured by AS-OCT

Because corneal thinning will be used as a secondary endpoint and is an indication of corneal melts, cross- sections of the cornea will be obtained using anterior segment AS-OCT. This non-contact high-resolution imaging modality provides cross-sectional images of the cornea and anterior segment with a resolution of approximately 5-18 μm. To evaluate the cornea, cross-sectional OCT images will be obtained at 0, 45, 90, 135, 180, 225, 270, 315 and 360 degrees.

Slit Lamp Photography

Because it is possible that slit lamp findings may be predictive of keratoprosthesis loss or corneal thinning, digital corneal photography will be done using a slit-lamp with a digital camera attachment and a flash-through-the-slit illumination system. By fine focusing on the surface of the cornea prior to corneal transplant surgery at the screening visit, and at Week(s) 1, 4, 16, 24, and 52, the photographer will capture the entire cornea using diffuse illumination and 10x magnification. If lids are drooping, the photographer will attempt to gently remove them from area of focus with a cotton swab.

8.7 Unscheduled Visits

Subjects are typically seen every 3-4 months after one year follow-up, however some subjects may need to be seen more regularly if they require more frequent monitoring for conditions such as glaucoma. These exams will be standard of care visits and any procedures/exams performed during these visits will be standard of care and will be decided by the MD investigator. Study staff will collect data for any procedures/exams that may be conducted as they pertain to the study (changes in medical history and medication prescription, BCVA, slit-lamp exam, IOP, funduscopy/B-Scan, OCT images, slit lamp photography and review of potential adverse events). Study staff will not collect data for any procedures/exams that are not study related and are not listed above.

8.8 Early Termination

Subjects have a right to withdraw from the study at any time. Additionally, the subject may be withdrawn from the study for any reasons: if it is in the best interest of the subject, intercurrent illness, adverse events, or worsening condition. The site investigators or Medical Monitor may request the withdrawal of a subject because of protocol violations, administrative reasons, or any other valid and ethical reasons.

Reasons for subject termination may include, but are not limited to, the following:

- Investigator determination that it is not in the best interest of the subject to continue participation
- Serious adverse events

- Any other safety concerns

In the event of an adverse event in the study eye that is considered by the investigator or Research Monitor to be severe in intensity, the donor tissue will be removed and replaced with fresh cadaveric human cornea donor tissue with or without a keratoprosthesis, discontinuing the patient from further participation in the study.

8.9 Schedule of Activities (Study Table)

	Screen	Day 0	Day 1	Wk. 1	Wk. 4	Wk. 16	Wk. 24	Wk. 36	Wk. 52
Visit Window	-60 days		+3 days	± 3 days	±3 days	± 2 weeks	± 3 weeks	± 3 weeks	± 4 weeks
Informed consent (study and surgical)	X	KPro Surgery with Crosslinked Donor Tissue							
Demographic data	X								
Medical/surgical history/chart review	X								
Review of Systems	X		X	X	X	X	X	X	X
BCVA ***	X		X	X	X	X	X	X	X
Slit-lamp exam***	X		X	X	X	X	X	X	X
IOP ***	X		X	X	X	X	X	X	X
Funduscopy/ Ultrasound B-scan (if necessary)***	X		X	X	X	X	X	X	X
Contact Lens Review			X	X	X	X	X	X	X
Pregnancy Test*	X								
Cornea thickness as measured by AS-OCT				X	X	X	X	X	X
Slit Lamp Photography***	X			X	X	X	X	X	X
Review Adverse Events			X	X	X	X	X	X	X
Concomitant Medication Check	X		X	X	X	X	X	X	X

Subjects will be instructed to strictly follow the study visit schedule.
* An additional urine pregnancy test needs to be performed just prior to surgery if there is greater than 30 days between screening visit and surgery.
**All subjects will be followed for at least 2 years. Following week 52 visit, standard of care visits will occur every 6 months for an additional 3 years or until study closure.
*** Historical results for best corrected VA, intraocular pressure, slit lamp examine, slit lamp photography and funduscopy can be used if completed within 60 days prior to screening visit

9. SAFETY AND EFFECTIVENESS ASSESSMENTS

9.1 Safety Assessments

The safety of the study intervention will be evaluated at every visit following surgery and will be defined by the incidence of related adverse events. Specifically, we will evaluate:

- **Systemic safety**: Incidence and severity of systemic adverse events during the study (based on physical examination, subject self-reporting, and changes in vital sign).
- **Ocular safety**: Incidence and severity of ocular adverse events during the study (ophthalmic examination, adverse events spontaneously reported). Ocular adverse events of special interest will include Infections Keratitis, Positive fungal rim culture and Retinal Detachment

All adverse events will be reviewed by the site investigator within 24 hours of notification and reported on the following schedule:

- Possibly, Probably, or Definitely Related **Expected AE** – Report to IRB on annual basis (continuing review)
- Possibly, Probably, or Definitely Related **Expected Serious AE** – Report to IRB on annual basis (continuing review)
- Possibly, Probably, or Definitely Related **Unexpected AE** – Report to IRB within 5 business days of discovery of event.
- Possibly, Probably, or Definitely Related **Unanticipated Problem** – Report to IRB within 5 business days discovery of event (24 hours for death of data loss)
- Possibly, Probably, or Definitely Related **Unexpected Serious AE** – Report to IRB within 5 business days of discovery of event

The Medical Monitor (DSMC Chair), Dean Cestari, MD is required to review all unanticipated problems involving risks to subjects or others, serious adverse events, and subject deaths. The monitor must comment on the outcomes of the event, the relationship of the event to participation in the study, and must indicate whether he concurs with the details of the report provided by the principal investigator. The Medical Monitor, is responsible to oversee the safety

of the research and report observations/findings to the IRB or a designated institutional official. The Medical Monitor will review all unanticipated problems involving risks to subjects or others associated with the protocol and provide an independent report of the event to the IRB. The Medical Monitor may discuss the research protocol with the investigators; shall have authority to stop a research protocol in progress, remove individual human subjects from a research protocol, and take whatever steps are necessary to protect the safety and well-being of human subjects until the IRB can assess the monitor's report; and shall have the responsibility to promptly report their observations and findings to the IRB or other designated official and the HRPO.

- Reports for events determined by either the investigator or Medical Monitor to be possibly or definitely related to participation in the study must be promptly forwarded to the USAMRMC ORP HRPO
- Reports of events resulting in death must be promptly forwarded to the USAMRMC ORP HRPO

9.2 Surgery Related Adverse Events

A surgical related adverse event is any adverse event that is related to or possibly related to the Boston Keratoprosthesis surgical procedure. Any surgical related, normal and expected postoperative complaints or symptoms (adverse events) that occur within 30 days of surgery (week 4 visit) are not required to be reported as adverse events, unless the event involves a clinically significant change in severity or duration of symptoms or requires clinical intervention that is different from ordinary postoperative care. Examples include:

- Eye redness
- Epithelial defects
- Decreased vision
- Anterior chamber cell
- High eye pressure (up to 28)
- Eye pain

Other expected postoperative care complaints and symptoms that will are not required to be reported as an adverse event include: headache, incisional pain, nausea, vomiting, low grade fever, dizziness, irritability, nervousness, temporary sleep problems like insomnia or sleepiness, constipation, confusion and similar events, tenderness, throat soreness secondary to intubation,

or mild to moderate swelling and/or bruising around the implant site, back pain due to lying on the table during the procedure.

9.3 Effectiveness Assessments

The efficacy of corneal collagen cross-linking of keratoprosthesis carrier tissue in high-risk keratoprosthesis implantation will be judged using the following parameters and time points:

Primary endpoints or outcome measure(s)

- Time from surgery to device loss or replacement

Secondary endpoints or outcome measure(s)

- Twelve-month retention rate
- Incidence of delayed epithelial healing at day 30 (week 4 visit)
- Cornea thickness metrics measured by AS-OCT; at week(s) 1, 4, 16, 24, 36, and 52.
- Time from surgery to retroprosthetic membrane treatment (laser or surgical interventions)
- Time from surgery to occurrence of vitritis (sterile or infectious)
- Time from surgery to occurrence of ocular surgery to address melt, including partial graft.

10. ADVERSE EVENT RECORDING AND REPORTING

10.1 Recording Requirements

Participants are informed of the anticipated risks of the trial during the consent process and continually asked to report any unusual ocular or systemic problems that occur during their participation in this trial to the site investigators or study staff. In addition, participants are seen regularly in site clinics as part of their standard post-surgical treatment. During each clinical examination participants will be queried about occurrence of adverse events. Slit lamp examination and ocular assessments will determine change in corneal thickness and or presence of corneal ulcers. Lastly, site investigators and study staff will be asked to report unanticipated problems or adverse events within 48 business days of discovery to the coordinating center (MEE) to allow for proper reporting.

All observed or volunteered adverse events (serious or non-serious) and abnormal test findings, regardless of study group or suspected causal relationship to the study drug will be recorded in the subjects' case histories (source data, case report form). For all adverse events, sufficient information will be pursued and/or obtained so as to permit 1) an adequate determination of the outcome of the event (i.e., whether the event should be classified as a serious adverse event) and; 2) an assessment of the causal relationship between the adverse event and the study drug.

Adverse events thought to be associated with the study drug will be followed until the event (or its sequel) or the abnormal test finding resolves or stabilizes at a level acceptable to the Sponsor-Investigator and the study Medical Monitor.

Study staff will alert the treating physician immediately of any participant complaints or problems that are expressed to them. The site investigator and/or site study staff will report any unexpected and serious adverse events to the coordinating center (MEE) within 48 hours of discovery regardless of the assumed relation to the study intervention. The site investigator and the study staff will follow the reporting guidelines below.

- **Causality and Severity Assessment**

The Sponsor-Investigator of the IND application and the Medical Monitor (DSMC chair) will promptly review documented adverse events to determine 1) if there is a reasonable possibility that the adverse event was caused by the study drug(s); and 2) if the adverse event meets the criteria for a serious adverse event.

If the Sponsor-Investigator's and Medical Monitor's final determination of causality is "unknown and of questionable relationship to the study drug", the adverse event will be classified as associated with the use of the study drug for reporting purposes. If the Sponsor-Investigator's and Medical Monitor's final determination of causality is "unknown but not related to the study drug", this determination and the rationale for the determination will be documented in the respective subject's case history (source data or case report form).

10.2 Reporting Procedures

- **Reporting of Adverse Events to FDA**
  - **Written IND Safety Reports (if applicable)**
  - **Telephoned IND Safety Reports – Fatal or life-threatening suspected adverse reactions**
- **Reporting Adverse Events to the Partner Human Research Committee**

The Partners Human Research Committee (PHRC) policy for "REPORTING ADVERSE EVENTS AND UNANTICIPATED PROBLEMS" will be followed.

- **Reporting Adverse Events to External Parties**

Adverse events will be reported to the USArmy's Human Research Protections Office (HRPO) and the US Food and Drug Administration per their guidelines. Certain adverse events, specifically infections with positive donor rim cultures, that are related to the cornea donor tissue will be reported to CorneaGen within 48 hours of discovery.

10.3 Termination, of Subjects due to Adverse Events

Termination from the study will happen if any of the following occurs:

- Subject's wish to withdraw for any reason.

- Investigator or Research Monitor determination that it is not in the best interest of the subject to continue participation
- KPro removal

In the event an adverse event in the study eye is severe enough to require the donor study tissue to be removed and replaced with fresh cadaveric human cornea donor tissue with or without a keratoprosthesis, the patient will be terminated from further participation in the study. However, prior to termination the patient will be followed for 30 days after replacement surgery to collect any additional event data.

11. STATISTICAL METHODS/DATA ANALYSIS

11.1 Primary endpoint(s) or outcome measure(s)

- Time from surgery to device loss or replacement

11.2 Secondary endpoints or outcome measure(s)

- Twelve-month retention rate
- Incidence of delayed epithelial healing at day 30 (week 4 visit)
- Cornea thickness metrics measured by AS-OCT; at week(s) 1, 4, 16, 24, and 52.
- Time from surgery to retroprosthetic membrane treatment (laser or surgical interventions)
- Time from surgery to occurrence of vitritis (sterile or infectious)
- Time from surgery to occurrence of ocular surgery to address melt, including partial graft.

11.3 Sample Size Determination

To determine the number of subjects to enroll, we performed sample size calculations based on a review of keratoprosthesis retention rates from prior studies and conservative estimates of efficacy in the treatment arm. For the control arm, we used twelve-month retention rates ranging from 50 to 60%, which has been reported in autoimmune eyes at high risk for keratoprosthesis loss. Therefore, we anticipate the twelve month retention rate for the untreated (not cross-linked) cornea will be 50-60%.To estimate the retention rate in the treatment arm, we reviewed the retention rates from the two published case reports that describe cross-linking of the keratoprosthesis carrier cornea in high-risk eyes (Kanellopoulos et. al. & Robert). While the two case reports describe a 100% retention rate in a total of 12 eyes at one year, the exact retention rate for a multicenter study is not known. Using a two-sided log rank test for equality of survival curves, we performed sample size calculations so that we could detect a 30% difference in retention rate between the treatment group and the control group and specified the probability of type 1 error of 0.05, a study power of 80%, a follow up period of twelve-months, and a 4% loss to follow up. In order to detect a difference between the control arm and the treatment arm using

these considerations, the calculations resulted in sample size estimates between 82 and 88 subjects (Table 2). For practical purposes, we also took into consideration the time available for the study and the number of high risk keratoprosthesis procedures performed at each study site. Using the mid-range estimates, we aim to enroll 84 subjects into this study.

Eighty-four subjects will be randomized equally to treatment or control group. Randomization will be stratified by 16 centers, as well as two pre-operative diagnostic categories: autoimmune disease vs. prior corneal melt without autoimmune disease. Eye with a history of autoimmune disease may have a different risk of a new melt than eye that have a history of a prior corneal melt. Thus there will be a total of 32 strata. Stratification will ensure balance of the stratification factors between the two arms of the study. A number of blocks of varying size will be determined (e.g. 5 blocks of size 4, 6 blocks of size 2) and randomization assignments will be prepared accordingly. Since we have 32 strata, relatively small block sizes will be planned to minimize the chance of large departures from the desired allocation ratio that could occur due to incomplete blocks by the time of completion of patient recruitment. Although the ability of the clinical staff to predict future treatment assignments is increased due to smaller block sizes, we do not think that this will be a major issue for the present study.

Retention Rate for Control Group	Retention Rate for Treatment Group	Total Sample Size
0.50	0.80	88
0.55	0.85	84
0.60	0.90	82

11.4 Analysis Population

N/A

11.5 Effectiveness Analysis

We will conduct an initial analysis to examine if distributions of baseline characteristics are balanced between the two treatment groups in our randomized study. Standard descriptive statistics will be reported, median (min-max) for numerical variables and frequency count (%) for categorical variables. Comparisons of numerical characteristics between treatment groups

will be conducted using the Mann-Whitney Wilcoxon test. Comparisons of categorical characteristics will be conducted using Chi-square, or Fisher's exact test as appropriate.

Primary Analysis

The primary efficacy analysis will follow an intent-to-treat analysis strategy including all randomized eyes. The major goal of this study is to determine the efficacy of using a collagen cross-linked cornea as a carrier for the Boston Keratoprosthesis in preventing corneal melts and increasing the retention of the keratoprosthesis. The primary endpoint of the efficacy analysis is time from surgery to device loss or replacement. Kaplan-Meier plot of event free survival by treatment groups will be provided and log-rank test will be used to examine if survival curves are significant different between groups. In addition, we will use a proportional hazards regression model to test the effectiveness of using a collagen cross-linked cornea as a carrier for the Boston Keratoprosthesis in preventing corneal melts and increasing the retention of the keratoprosthesis. We will obtain an estimate (95% CI) of the hazard ratio of device loss comparing treatment with control. This framework allows for the control of covariates, which may be unbalanced by chance despite the randomization of treatment assignment. If Kaplan-Meier plots of event free survival by study time, or related plots of log (-log) (survival) indicate violations of the proportional hazards assumption, then weighted log-rank test will be used according to strategies described by Peckova and Fleming.

Secondary Analysis

We plan to conduct a number of pre-specified analyses of secondary study endpoints. Analyses of secondary "time-to-event" type endpoints (time to retroprosthetic membrane treatment and time to occurrence of vitritis) will use the Kaplan-Meier approach with log-rank test and Cox proportional hazards regression, similar to the primary analysis for time to device loss. Comparison of cornea thickness metrics at week 1, 4, 16, 24, and 52 for treatment versus control group will be performed using the Mann-Whitney Wilcoxon test at each time point. Furthermore, mixed model will be used to examine the effect of time, treatment and interaction between them on cornea thickness. Comparison of twelve-month retention rate and incidence of delayed epithelial healing at Day 30 (week 4 visit) between groups will be performed using Chi-square

test, or Fisher's exact test as appropriate. In addition, the occurrence of any adverse events will be described and compared between treatment arms of the study.

11.6 Safety Analysis

Any adverse events, ocular or systemic, from all subjects will be utilized to summarize safety data for this study. The severity of each adverse event and the relationship of the event to the study medication will be assessed. If adverse events occur, the first concern will be the safety of the study participants. Any serious adverse event occurring during the study will be immediately reported to the principal investigator, and then to the PHRC, sub-sites, DOD, and FDA per the specified guidelines. The Research Monitor shall have the authority to stop the protocol, remove subjects from the protocol, and take any necessary steps to protect the safety and well-being of subjects until the IRB can assess the monitor's report. The monitor shall promptly report their observations and finding to the IRB or other designated official.

11.7 Interim Analysis

N/A

12. DATA AND SAFETY MONITORING

12.1 Data and Safety Monitoring Plan

A Data Safety and Monitoring Committee (DSMC) will be formed prior to trial initiation. The committee members will consist of experts in corneal disease, and the study biostatistician. In addition, Dr. Dean Cestari will function as the Medical Monitor. The Medical Monitor will provide oversight and be responsible for reporting the findings of the DSMC to the PHRC.

The DSMC will meet in an open session with select investigators prior to enrollment opening to review the protocol, objectives of the DSMC, and the plans for the DSMC throughout the course of the study. Following this open session, the DSMC will convene on an annual basis to review the data derived from the trial in a masked fashion.

The annual meeting of the DSMC will allow members to review the ongoing progress of the study and safety of the participants. From this information, the DSMC will make recommendations to the Principal Investigator and the site IRBs whether continuation of the trial is recommended.

The DSMC will review masked cumulative participant data in order to assess the integrity of the study. Specifically the DSMC will be asked to evaluate the data integrity, participant safety and scientific validity. Site summaries of patient recruitment, retention rates, data quality and completeness, protocol deviations, unanticipated events and adverse events will be shared with the DSMC to allow for a complete assessment. Data will be shared in a masked fashion with interventions coded as either A or B. If for any reason the DSMC requires unmasking of data to further evaluate safety parameters, it will be provided to them. The DSMC also can request an interim analysis if deemed necessary.

Following each meeting, the DSMC Chair, on behalf of the DSMC, will provide a recommendation to the Principal Investigator and IRB on the continuation of the study. The Committee can request continuation, modification, suspension or termination of the trial. The DSMC Chair may discuss the protocol with investigators, interview subjects, and consult with others outside the study about the research to help him reach his determination of continuation, modification, suspension, or termination of the trial.

13. DATA HANDLING, RECORD-KEEPING AND MONITORING

13.1 Data Recording, Record-Keeping and Monitoring

David Zurakowski, PhD, Biostatistician will serve as the trial statistician. He will lead the data analysis for this study as well as prepare the appropriate documentation for the Data Safety Monitoring Committee meetings.

A paper (hard copy) Case Report Form (CRF) will be completed for each subject enrolled into the clinical study. The site investigators will review, approve and sign/date each completed CRF; the investigators' signature serving as attestation of the investigator's responsibility for ensuring that all clinical data entered on the CRF are complete, accurate and authentic.

All data from CRF's will be added to the study's electronic data capture system, StudyTrax. StudyTrax, a web-based electronic data capturing system, is a HIPAA and 21 CFR Part 11 compliant product that is hosted by Partners Healthcare and provides numerous academic medical centers with an electronic data capturing system to run multi-center clinical trials and other human subject research. StudyTrax will be used to collect all study data on subjects, including clinical assessments, subject medications, adverse events, as well as OCT images and Corneal Photographs. Data entered in StudyTrax are exportable to SPSS, Excel and SAS allowing for real-time data queries of enrollment, adverse event reporting.

The site investigators and research staff participating in the study are familiar with the protocol and study procedures. After each visit the CRFs will be checked by site research staff to confirm that all study procedures were completed as outlined in the protocol. Staff will also ensure that informed consents were signed at the time of enrollment and that all eligibility criteria have been met.

Source Data are the clinical findings and observations, laboratory and test data, and other information contained in Source Documents. Source Documents are the original records (and certified copies of original records); including, but not limited to, hospital medical records, physician or office charts, physician or nursing notes, subject diaries or evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, x-rays, etc. StudyTrax will serve as the Source Data in addition to the Case Report Forms. When applicable,

information recorded on the CRF shall match the Source Data recorded on the Source Documents.

All study data and regulatory documentation will be monitored as outlined in the study Monitoring Plan.

13.2 Data Management

13.2.1. Identifiers

Each site will be given a unique site identification (ID) number. This number will precede each subject specific identification number to create a unique code for each participant. This code will then be included in the subject's electronic file within StudyTrax and ascribed to all data within the system to maintain subject confidentiality.

13.2.2. Confidentiality

All enrolling sites will maintain their site-specific study subject files within StudyTrax. Only appropriate study personnel at each site will be given access to StudyTrax with a unique username and password. In addition, the program allows for multiple levels of access providing added security.

The subject identification process described above allows MEEI personnel the ability to review files for data integrity and safety while maintaining the confidentiality of the study subjects.

Corneal photographs and OCT images from each site will be shared with coordinating center via secure email. All images will contain no identifiers except for the unique subject ID. The trial Image Analyst, will have access to each image through DropBox (Partners Healthcare account) allowing for confidential and masked retrieval of all study images.

Representatives from the US Army Medical Research and Materiel Command (USAMRMC) are eligible to review study records. No sensitive information is being collected for this study.

13.2.3. Disposition of Data

All subject data will be stored within StudyTrax. MEE will be the primary administrator for this study in StudyTrax. StudyTrax is hosted within a secure network. The data within StudyTrax

will be extracted at the completion of the study. This data will be held in a secure network file at MEE for at least two years following a marketing application submission to the FDA or longer, if required by the PHRC.

13.2.4. Sharing Study Results

Throughout a subject's participation in this study the investigators will be encouraged to share appropriate clinical information collected during study participation with subject's other medical care providers.

13.3 Laboratory Evaluations

A urine pregnancy test will be performed at screening (or 30 days prior to surgery). The urine collected will solely be used to evaluate levels of HCG and will not be used for other evaluations or stored for further use.

14. STUDY DISCONTINUATION CRITERIA

14.1 DSMC Discontinuation of the Clinical Research Study

If the DSMC, DSMC Chair and Medical Monitor determines that the trial needs to be suspended temporarily or permanently, the site investigators will be notified and instructed to inform their IRBs. The PI will inform the DOD and FDA and determine the best way to notify participants of the termination of the study to ensure patient safety. This plan will be communicated to all site investigators for notification of all their participants of the termination or suspension of the study and reasons why. Any additional measures to ensure participant safety will be taken.

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Unfortunately, there are no good animal models to test our technique with the Boston KPro. Rabbits are poor models since their clinical results have notoriously poor correlations with human clinical outcomes.

Initially, we worked on determining the optimal riboflavin/UVA cross-linking method to increase resistance of the corneas to enzymatic degradation ex-vivo.

The methodology was as follows. The corneal epithelia from the enucleated eyes of mature white New Zealand rabbits were mechanically removed using a 57 blade. The corneas with 2-3mm scleral rims were excised and their irises were removed. They were fitted into Barron® artificial anterior chambers with the endothelium bathed in isotonic saline solution. The VEGA® LED-based UVA emitter was switched on for at least 30 minutes prior to calibration then was calibrated before the experiments. The corneas were then pre-treated with 0.1% riboflavin/20% dextran T500 every 2-3 minutes for 15 minutes followed by UVA light (λ= 370nm, irradiation = 3mW/cm2) at a fixed distance of 54mm from the top of the corneas for 5-minute intervals, while applying riboflavin between irradiations. Either the anterior stroma (A) or the anterior (A) and posterior stroma (P) was irradiated for different durations (7.5A, 7.5A/7.5P, 15A, 15A/15P, 30A, 30A/30P and 60A minutes). The primary control was no riboflavin/no UVA. Secondary controls included riboflavin/no UVA and UVA/no riboflavin. The corneas were trephined into 8.5-mm buttons and incubated in clean glass vials containing 0.3% collagenase A solutions at 37°C and rotating at 150 rotations-per-minute. The time to total dissolution of the corneas was measured. Each group included 4 or more corneas.

Results showed that the primary control dissolved at 4.8h ±0.8 while tissue cross-linked for 15 minutes dissolved after 50.0 +/- 8.1 hours. Compared to the control, there was a marked increase in resistance to degradation for all UVA/riboflavin cross-linked corneas. In general, anterior stromal cross-linking was superior to anterior/posterior stromal cross-linking (whether the irradiation time was split or doubled between the anterior and posterior stromata compared to their matched groups of anteriorly cross-linked corneas). The minimal treatment that yielded the greatest resistance in all groups was 15 minutes of anterior stromal cross-linking (50.0h □8.1; $p<0.0001$). The results were similar for 15, 30, and 60 minutes of cross-linking. Therefore, we identified several methods to increase the corneal resistance to enzymatic degradation using the corneal cross-linking technique.

The UVA emitter emits constant irradiation energy of 3mW/cm2 that requires the UVA light source to be at 54mm from the cross-linked cornea. The irradiated area can be controlled using