

**Changes in corneal endothelial cell density
after transscleral ab interno glaucoma gel stent implantation**

STUDY SPONSOR:

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REV. A

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Investigator Statement of Compliance

As an Investigator, I agree to: 1) Implement and conduct this study diligently and in strict compliance with this protocol, applicable government laws and regulations, conditions of approval imposed by the reviewing ethics committee or competent authority, the Declaration of Helsinki and Good Clinical Practice / ICH Guidelines. 2) Supervise all testing of the device involving human subjects, 3) Ensure that the requirements for obtaining informed consent are met and 4) Maintain all information supplied by the Sponsor in confidence and, when this information is submitted to an EC or any other group, it will be submitted with a designation that the material is confidential.

Investigator Signature

Date

REVISION HISTORY

Date	Revision
October 15th, 2018	A

Protocol Outline

Protocol Title: "Changes in corneal endothelial cell density after transscleral ab interno glaucoma gel stent implantation"

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Principal Investigator: Prof. Dr. H. Reitsamer

Research Team: Prof. Dr. H. Reitsamer, Doz. Dr. W. Hitzl, Dr. M. Lenzhofer

I. Abstract

STUDY OBJECTIVE	Corneal endothelial cell loss is a known sequela of glaucoma tube shunt implantation. Big tube shunts show a decrease of endothelial cell count postoperatively (-11.5% after Ahmed glaucoma valves and -12.4% after Molteno shunt 2 years postoperatively). The objective of this study is to evaluate the change of endothelial cell count after XEN45 in patients with or without cataract operation.
PATIENT POPULATION	In 140 open angle eyes central endothelial cell counts were recorded preoperatively, before the XEN45 implantation was performed in Dept. Ophthalmology at Paracelsus Medical University Salzburg. XEN45s were already performed in the year 2013-2017 (so up to 5 years) in open angle glaucoma eyes in combination with or without cataract operation. In these eyes central endothelial cell count and central corneal thickness was measured preoperatively and documented in patients records.
STUDY DESIGN	A prospective, observational, monocentric trial to evaluate the course of endothelial cell density after the XEN implant. The study will take place in the Dept. Ophthalmology at Paracelsus Medical University Salzburg/SALK.

Up to 140 consecutive patients with preoperatively recorded endothelial cell counts will be summoned to a consecutive endothelial cell count record and measurement of central corneal thickness

postoperatively. Informed consents will be obtained from patients, who are interested in participating in the study.

Patients will be assessed for endothelial cell density and the position of the XEN45 measured by anterior segment optical coherence tomography. The distance of the XEN45 to cornea, the angle of the tube, and the tube length in the anterior chamber will be recorded with anterior segment optical coherence tomography. Endothelial cell density will be measured on 3 positions: central, superior-nasal (location of XEN45 implantation), infero-temporal (far away for the XEN45 implant).

OUTCOME PARAMETERS	<p>The primary endpoint is the change of central endothelial cell density compared to preoperative data.</p> <p>Secondary objectives are the differences of supero-nasal and infero-temporal endothelial cell density compared to the central endothelial cell density, and pachymetry compared to preoperative data.</p>
EXAMINATION SCHEDULE	Subjects will undergo study visits at the following times: preoperative data (2013-2017) out of patient records, postoperative visit in the year 2018-2019 (1-6 years post XEN45 implantation).
CLINICAL PARAMETERS	<p>The following clinical assessments will be performed at the postoperative examination:</p> <ol style="list-style-type: none">1. Endothelial cell count on different locations of the study eye2. Central corneal thickness3. Anterior segment optical coherence tomography (to determine the position of the XEN45)4. Slit lamp examination of the cornea, anterior segment including gonioscopy (to determine the position of the XEN45), and dilated fundus examination5. Best corrected visual acuity6. Measurement of IOP7. Number and frequency of ocular (glaucoma) medications8. Ocular symptoms and assessment of complications (especially symptoms, which may be a hint for loss of endothelial cell count)

9. Secondary surgical procedures (if applicable including needlings, lasers, cataract operation, bleb revisions, keratoplasty, secondary IOP lowering procedures, other operations)

II. Background and Significance/Preliminary Studies

Glaucoma is a leading cause for irreversible visual function loss worldwide.¹ However, the reduction of intraocular pressure (IOP) is the only proven therapy for glaucoma. To reduce the IOP, most patients are treated with IOP lowering eye drops, but there is a rising group of patients, which has an unmet need for surgical IOP reduction.²⁻⁶ All members of the commonly performed filtering glaucoma surgeries (e.g. trabeculectomy or big tube shunts) are capable to lower the IOP.

In the last years, new minimally invasive glaucoma surgeries (MIGS), have been introduced for glaucoma therapy.⁷⁻⁹ The XEN Glaucoma Gel Microstent (XEN-GGM, Allergan Plc, USA) is a member in the MIGS group.¹⁰ It bypasses aqueous humor from the anterior chamber to the subconjunctival space.¹¹

Corneal endothelial cell loss is a known sequela of glaucoma tube shunt implantation.¹² Big tube shunts show a decrease of endothelial cell count postoperatively (-11.5% after Ahmed glaucoma valves and -12.4% after Molteno shunt 2 years postoperatively).¹³⁻¹⁵ Corneal endothelial cell density further progressively decreased after Ahmed glaucoma valve implantation with time. The distance of the Ahmed tube tip to the cornea is significantly associated with endothelial cell loss in the quadrant of implantation.¹³⁻¹⁵ A dramatic decrease of endothelial cells may cause corneal edema and affects the patient's vision. Even a keratoplasty (transplantation of the cornea or the corneas' inner layers) may be necessary due to a massive decrease of endothelial cell density.

Also after classic filtering surgery (trabeculectomy with and without Mitomycin C), a significant loss of endothelial cells is reported.¹⁶

The Cypass Implant (Alcon), a minimally invasive glaucoma surgery (MIGS) competitor to the XEN45, was withdrawn from global market due to statistically significant endothelial cell loss at 5 years. (see <https://www.alcon.com/news/media-releases/alcon-announces-voluntary-global-market-withdrawal-cypass-micro-stent-surgical>)

Therefore the objective of this study is to evaluate the change of endothelial cell count after XEN45 in combination with or without cataract operation.

III. Study Aims

The present study aims to longitudinally evaluate the change of endothelial cell count after XEN45 in 2 patient groups: XEN solo procedures and combined XEN with cataract operations.

IV. Administrative Organization

In 140 open angle eyes preoperative central endothelial cell counts were recorded preoperatively, before the XEN45 implantation was performed. XEN45s were already performed in the year 2013-2017 (so up to 5 years) in open angle glaucoma eyes in combination with or without cataract operation. In these eyes central endothelial cell count and central corneal thickness was measured preoperatively and documented in patients records.

The study will take place in the Dept. Ophthalmology at Paracelsus Medical University Salzburg/SALK.

V. Study Design

Experimental design of the study

A prospective, observational, monocentric trial to evaluate the course of endothelial cell density after the XEN implant.

Up to 140 consecutive patients with preoperatively recorded endothelial cell counts will be summoned to a consecutive endothelial cell count record and measurement of central corneal thickness postoperatively. Informed consents will be obtained from the patients, who are interested in participating in the study. The study will be approved by the local ethics committee.

Patients will be assessed for endothelial cell density and the position of the XEN45 measured by anterior segment optical coherence tomography. The distance of the XEN45 to cornea, the angle of the tube, and the tube length in the anterior chamber will be recorded with anterior segment optical coherence tomography. Endothelial cell density will be measured on 3 positions: central, superior-nasal (location of XEN45 implantation), infero-temporal (far away for the XEN45 implant).

Subjects will undergo study visits at the following times: preoperative data (2013-2017) out of patient records, postoperative Visit in the year 2018-2019 (1-6 years post XEN45 implantation).

2 patient groups will be analyzed separately: patients, who had a solo XEN procedure and patients, who had a combined XEN and cataract operation.

Study population general description

This study will include subjects presenting with open angle glaucoma and who meet the following inclusion and exclusion criteria. Any questions regarding a patient's eligibility should be discussed with the sponsor prior to enrollment.

An inclusion of both eyes of the same patient is possible.

Inclusion Criteria

1. Open Angle Glaucoma
2. History past XEN45 implantation with or without combined cataract procedure
3. Available preoperative endothelial cell count data not older than 1 year before XEN operation without any operation between the day of data record and XEN45 operation.
4. Age 18 years or older and of legal age of consent
5. Signed written informed consent
6. Availability, willingness, and sufficient cognitive awareness to comply with examination procedures

Exclusion Criteria

1. Patients with secondary IOP lowering procedures (excluding SLT, needlings, bleb revisions, YAG lasers, Argon lasers) after XEN implantation will be excluded from analysis, because the secondary IOP lowering procedures may influence endothelial cell count itself. Cataract operations without complications are no exclusion criteria, but have to be recorded.

Sample size determination and power analysis

No formal sample size calculation was done due to the unknown effect size.

Description of hypothesis for primary endpoint:

Null hypothesis: H0: the postoperative mean number of endothelial cells is lower than the preoperative.

Alternative hypothesis: H1: the postoperative mean number of endothelial is larger or equal than the preoperative

Study outcomes/endpoints

The primary endpoint is the change of central endothelial cell density compared to preoperative data.

Secondary objectives are the changes of supero-nasal and infero-temporal endothelial cell density compared to the central endothelial cell density, and pachymetry compared to preoperative data.

VI. Study Procedures

Slit lamp examination

The visibility of the XEN in slit lamp examination in eyes looking directly to the examiner will be recorded (yes/no). Beside biomicroscopic slit lamp examination of the cornea and anterior segment, signs of inflammation or corneal alterations will be recorded.

Gonioscopy

Gonioscopy and angle assessment of XEN quadrant (superior-nasal) by Shaffer Grade. The position of the XEN will be noted (anterior Schwalbe line = in cornea, in Schwalbe line, anterior to Schlemm's Canal, in Schlemm's Canal, non pigmented Trabecular meshwork, pigmented Trabecular meshwork, posterior pigmented trabecular meshwork, scleral Spur, ciliary body band, Iris), the length of the XEN stent in anterior chamber will be noted, if there is an endothelial/iris touch of the XEN stent will be noted:

FIGURE 1: Shaffer Angle Grading

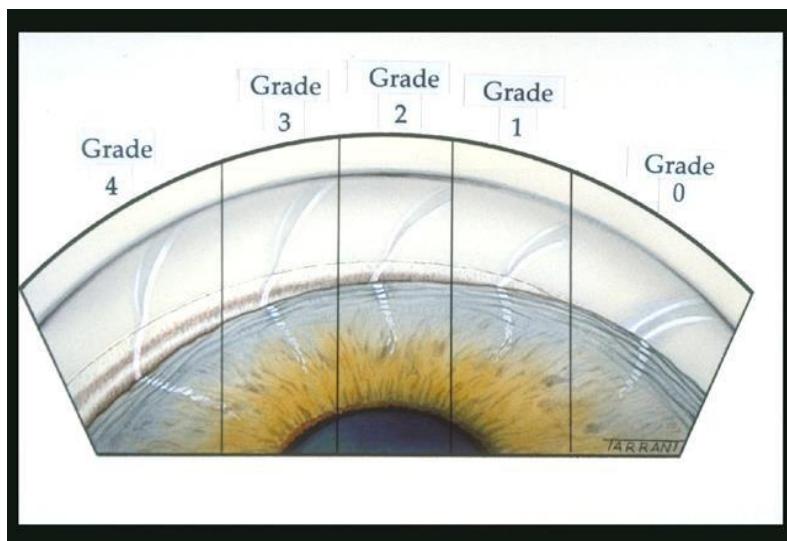


TABLE 1: SHAFFER GRADE FOR ANGLE EVALUATION

Shaffer Grade	Angle
Grade 4	45-35° angle
Grade 3	35-20° angle
Grade 2	20° angle
Grade 1	10° angle or less
Grade 0	0° angle (closed)

Optical coherence tomography

Anterior segment optical coherence tomography (AS-OCT) is used to determine the position of the XEN45 in the anterior chamber. The distance of the XEN45 to cornea, the angle of the tube, and the tube length in the anterior chamber will be recorded with anterior segment optical coherence tomography.

Endothelial cell count

Endothelial cell density will be measured on 3 positions: central, superior-nasal (location of XEN45 implantation), infero-temporal (far away for the XEN45 implant).

Pachymetry

The central corneal thickness has to be measured in the cornea center.

Dilated fundus examination

Per clinical routine.

Measurement of IOP

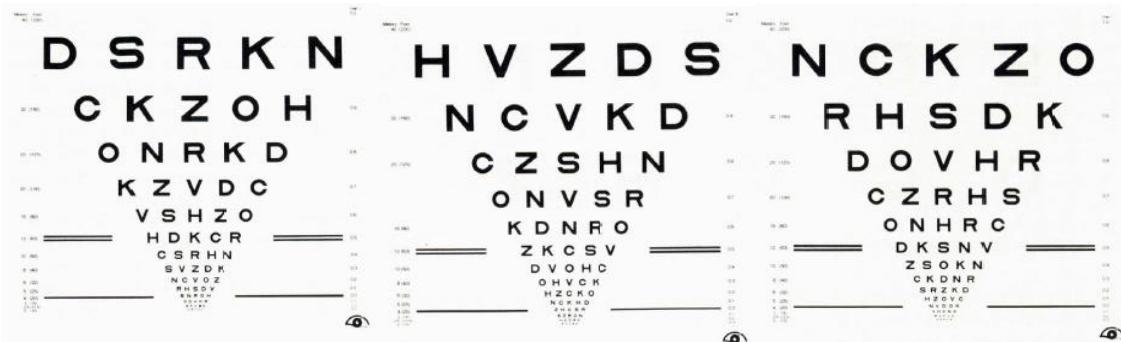
Goldmann applanation tonometry are used to measure IOP. IOP readings are repeated until 2 consecutive or nonconsecutive measurements are obtained differing by 1 mmHg or less, and the average of the 2 readings serves as the IOP measurement for the visit.

Best corrected visual acuity

Before BCVA testing, the investigator performs a manifest refraction. Once the best-corrected manifest refraction was determined, best corrected visual acuity on the study eye was examined with the patients' pupils in their natural state. The pinhole method for measuring visual acuity is not acceptable.

The BCVA has to be tested at a single seating by the same experienced investigator in the same room from a 4 meters distance with standardized low light conditions. A comfortable seating of the patient and to the chart placement at eye level is important while testing visual acuity. The patients have to be told not to lean forward. Breaks during the testing are allowed as needed.¹⁷
¹⁸

FIGURE 2: ETDRS chars



The ETDRS logMAR charts (Lighthouse International, New York, NY) are used for assessment of best corrected distance visual acuity. The chart has five letters per row ranging in size from +1.0 to -0.30 logMAR at 4 meters.

Testing Paradigm for distance BCVA. Subjects are required to identify each letter on the chart until they identified a full row of letters incorrectly, at which point the test was terminated, and the acuity calculated. Vision testing starts with the top of the chart. Patients are elated to guess if they were not sure of the letter. Only a single reading of the chart is allowed. When a letter is read correctly, the examiner circles this letter on a score sheet with a layout identical to that of the chart. If visual acuity is poor, the test distance is reduced to 1 meter. Visual acuity as low as 0,025dec (+1,6logMAR) can be measured.

If patients could not see any letter of EDTRS chart at 1 meter, they are tested for finger count (0,01dec, logMAR +2), hand movement (0,001dec, logMAR +3) or light perception visual acuity (0,0001dec, logMAR +4) at 0.6 meters.¹⁹ Results are recorded on a sheet.

Scoring Snellen for distance BCVA. Snellen line assignment method is used to score results. The smallest line, where a patient has read half or more of the letters correctly, is recorded as BCVA in Snellen (decimal).

Ocular symptoms and assessment of complications

Especially symptoms, which may be a hint for loss of endothelial cell count have to be recorded.

Secondary surgical procedures

If applicable record secondary surgical procedures including needlings, lasers, cataract operation, bleb revisions, keratoplasty, secondary IOP lowering procedures, other operations.

VII. Visit

Subjects will be examined and evaluated according to the following schedule of visit:

Visit	Exam	Visit Window
Preop	preoperative data out of patient chart records	(2013-2017)
PO1	Visit	Year 2018-2019 (1-6 years post XEN45 implantation).

Preoperative Procedures

No subject may be entered into the study who does not meet the inclusion/exclusion criteria. The details of the study must be fully explained and informed consent must be signed prior to any study-specific examinations being performed.

Visit

The visit will request the following information:

Exams	Screening Evaluation
Informed Consent	Yes
Demographic Information	Yes
Ocular History and Medications	Ocular history, including presence of ocular pathology and ocular medications
Glaucoma History and Medications	Including number of IOP Lowering Medications
Secondary surgical procedures	if applicable including needlings, lasers, cataract operation, bleb revisions, keratoplasty, secondary IOP lowering procedures, other operations
Slit Lamp Exam	Biomicroscopic slit lamp examination of the cornea and anterior segment
Endothelial cell count	Endothelial cell density will be measured on 3 positions: central, superior-nasal (location of XEN45 implantation), infero-temporal (far away for the XEN45 implant).
Pachymetry	Will be measured in the cornea center
Anterior segment optical coherence tomography	to determine the position of the XEN45

Anterior segment optical coherence tomography	Best Corrected distance Visual Acuity (BCVA line assignment method): Monocular best-corrected visual acuity should be measured in Snellen line assignment method.
IOP (Medicated)	Intraocular pressure (IOP) using Goldmann
Gonioscopy	to determine the position of the XEN45
Assessment of Adverse Events and Serious Adverse Events	See Section Adverse Event and Serious Adverse Event Reporting

Safety Monitoring Plan

Definition of adverse events, serious adverse events:

The XEN procedure is standard of care. The XEN implant is CE-marked for the implantation in primary open angle glaucoma patients. XEN is used in the treatment of glaucoma worldwide. Complications occurring during the study period will be treated in the discretion of the treating investigator. In every serious adverse event, appropriate measures should be taken to treat/resolve and monitor the subject. Any subjects who are withdrawn from the study due to an adverse event shall still be followed until the outcome is resolved.

Before and after the study visit, patients will be followed up in the clinical routine.

The investigators will classify the adverse events based on the following definitions according to ISO 14155 (2011).

Adverse Event (AE)

Any untoward medical occurrence, unintended disease or injury, or untoward clinical signs (including abnormal laboratory findings) in subjects, users or other persons, whether or not related to the investigational medical device.

Adverse Device Effect (ADE)

An adverse event related to the use of an investigational medical device.

Device deficiency

Inadequacy of a medical device with respect to its identity, quality, durability, reliability, safety or performance. Device deficiencies include malfunctions, use errors, and inadequate labelling.

Serious Adverse Event (SAE)

An adverse event that a) led to death,
 b) led to serious deterioration in the health of the subject, that either resulted in
 1) a life-threatening illness or injury, or

- 2) a permanent impairment of a body structure or a body function, or
- 3) in-patient or prolonged hospitalization, or
- 4) medical or surgical intervention to prevent life-threatening illness or injury or permanent impairment to a body structure or a body function,

c) led to foetal distress, foetal death or a congenital abnormality or birth defect

NOTE: Planned hospitalization for a pre-existing condition, or a procedure required by the CIP, without serious deterioration in health, is not considered a serious adverse event.

Serious Adverse Device Effect (SADE)

Adverse device effect that has resulted in any of the consequences characteristic of a serious adverse event

VIII. Analysis Plan

Statistical methods

Data consistency was checked and data were screened for outliers and normal, Gamma, Log-normal and Tweedie distributions by using quantile plots. Continuous variables were also tested for these distributions by using Kolmogorov-Smirnov test. Depending on data distribution the following methods will be used: In case of parametrically testing, mixed models based on normal, Gamma and Tweedie distributions will be used to analyze data. Independent and unstructured working correlation matrices were used for modeling the corresponding covariance of Endothelial cell number over time. The model based estimator was used to estimate the covariance matrix. The corrected quasi likelihood under independence of model criterion will be used to select the final models. 95% confidence intervals will be computed for means and results will be illustrated by using Whisker plots. In case of nonparametrically testing, Wilcoxon's matched pairs test will be used. All reported tests will be done one-sided, and p-values < 0.05 were considered as statistically significant. All statistical analyses will be performed by use of STATISTICA 13 (Hill, T. & Lewicki, P. Statistics: Methods and Applications. StatSoft, Tulsa, OK) and NCSS 10 Statistical Software (2015). NCSS, LLC. Kaysville, Utah, USA.

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