EC OPHTHALMOLOGY
Research Article

Methodology of Separation and Ablation of Corneal Tissue Using Femtosecond and Excimer Laser

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Abstract

Aim: The aim of this study is to compare and record the effect of femtosecond laser, excimer laser and the microkeratome on the corneal tissue. Which of these used methods in a modern refractive surgery is the most accurate and least intrusive? We investigated the homogeneity of the stromal bed and the side cut.

Methods: The research was conducted on the five pieces of porcine corneal tissue. The laser treatment was carried out on the devices of Zeiss company (femtosecond laser VisuMax: 1100 - 1600 nm and excimer laser MEL 80, operating at a wavelength of 193 nm). Operating conditions were 21°C and 45% humidity. We used three different techniques for separation and ablation of the corneal tissue: Refractive lenticule extraction (ReLEx), femtosecond laser assisted *in situ* keratomileusis (femtoLASIK) and technique with microkeratome. The size of the flap, transition and optical zone were insignificant to the investigation. All records were taken on the electron microscope Tescan VEGA TS 5136XM.

Results: In the ReLEx method, electron microscopy images show greater surface irregularities in both, optical and non-optical portions. The surface is visibly smoother and homogenous in method using higher energy settings. Nevertheless, in both samples we found obvious tissue line between the non-optical and optical zones. From the optical point of view, it may cause undesirable light scattering. We therefore conclude that the ReLEx method is very susceptible to the correct energy settings.

The ASA (Aberration smart ablation) method showed better surface quality than the TSA (Tissue saving ablation) method. However, methods mentioned above show better morphological results than the ReLEx method. Nevertheless, we cannot state that the quality of the stromal bed directly affects the healing, postoperative complications, visual acuity and postoperative corneal biomechanical properties.

Conclusion: For good results of the ReLEx surgery method, the correct setting of laser beam values is essential. However, the quality of corneal stroma bed appears to be still better in older methodologies (when using excimer laser). The microkeratome method appears to be the least desirable for creation of the flap.

Keywords: *ReLEx; Microkeratome; FemtoLASIK; Porcine Eyes; Electron Microscopy*

Abbreviations

TSA: Tissue Saving Ablation; ASA: Aberration Smart Ablation; SEM: Scanning Electron Microscope; ReLEx: Refractive Lenticule Extraction; LASIK: Laser Assisted *In Situ* Keratomileusis

Introduction

The refractive laser procedures have been used since 1983 when Stephen Trokel a scientist at Columbia University in collaboration with Srinivasan performed the first Photorefractive Keratectomy (PRK). It has already been operated on more than 30 million patients

[1] throughout the world. Currently the most modern laser technology in ophthalmology is the use of femtosecond laser. Dr. Juhsaz and Dr. Kurtz at the University of Michigan designed the first ophthalmic femtosecond (FS) laser system in the early 1990s. It has been developed to perform laser assisted *in situ* keratomileusis refractive surgery [2]. It reduced the complication rate due to LASIK flap creation, improved the predictability of flap proportions and improved the quality of the optical surface compared to microkeratome surgery [3].

The aim of the study is to compare and record the effect of femtosecond laser, excimer laser and the microkeratome on the corneal tissue. Which of these methods used in a modern refractive surgery is the most accurate and least intrusive? Can monitoring of the stromal bed help us to understand the postoperative values of visual acuity, healing speed and the number of postoperative complications? We did not find any study, which compares all available methods from the viewpoint of symmetry of corneal stromal bed and side cut [4-6].

Subjects and Methods

The research was conducted on porcine corneal tissue. The separation and ablation of corneal tissue was carried out on these laser devices: femtosecond laser VisuMax (wavelength of 1100 to 1600nm) and excimer laser MEL 80 (wavelength of 193nm). Operating conditions were 21°C and 45% humidity. Dissected corneal tissue was placed in a fixative solution of 5% glutaraldehyde. As the buffering agent to glutaraldehyde, cacodylate buffer was used at a concentration of 0.1M. All records were taken on an electron microscope Tescan VEGA TS 5136XM [4,7]. Images were created at an accelerating voltage of 20kV and a magnification 88 - 3000×. Size of the flap, transition and optical zone were irrelevant to the investigation.

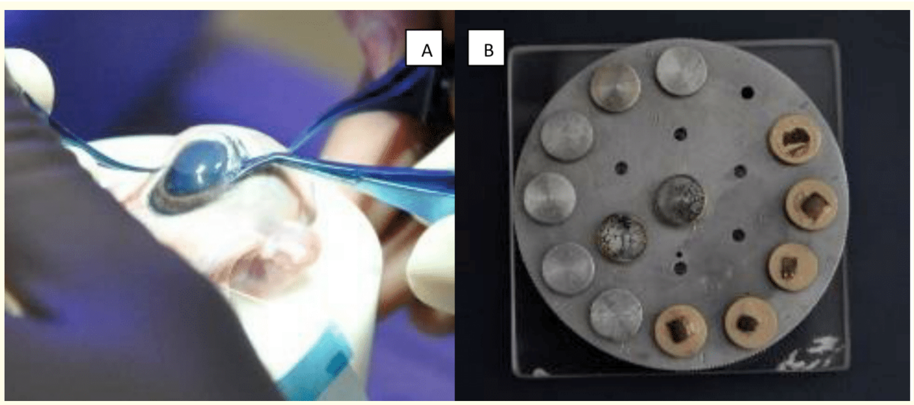

Figure 1: *Cutting off the cornea in limbus and discs with corneal grafts, prepared for SEM (Scanning electron microscope).*

The ReLEx as the first method represents the latest technology for the creation of the corneal flap and diopters removal. All steps of procedure are carried out on one machine. Refractive error is corrected by corneal lenticules removal [8-11]. We corrected minus five diopters.

The second method for refractive error correction was femtoLASIK. We created a corneal lamella on the femtosecond laser; then, the porcine eyes were moved under the excimer laser, where we removed minus three diopters by laser ablation [12-17].

Methodology of separation and ablation of corneal tissue

The porcine eyes were received within two hours after pig`s death. As the corneal tissue samples were fresh, epithelial abrasion was not performed. The first pair of eyes was modified by ReLEx method. This separation and ablative process was conducted with a standard value of the energy (Table 1). For the second eye, we had increased the energy of 4mJ (Table 2). For simpler distinction, we have named the technique "ReLEx HiE".

Variables	Lenticule	Lenticule side	Flap	Flap side
Spot distance [μm]	3	2,5	3	2
Track distance [μm]	3	2,5	3	2
Energy offset	30	30	30	30

Table 1: *ReLEx settings.*

Variables	Lenticule	Lenticule side	Flap	Flap side
Spot distance [μm]	3	2,5	3	2
Track distance [μm]	3	2,5	3	2
Energy offset	34	34	34	34

Table 2: *ReLEx HiE settings.*

For another pair of corneas, we used the femtosecond laser only for the creation of corneal lamella. Furthermore, these eyes have been modified by excimer laser. We used two different ablation profiles. The first eye was modified by the TSA[1] ablation profile and the second eye by the ASA[2] ablation profile [18-23].

After completion of the ablation process, we performed a withdrawal of the corneal tissue as close to perilimbal area as possible. For the last pair of eyes, we used microkeratome for the separation process and chose the softer mode. In this corneal sample, we did not use a laser.

Results

In the ReLEx method, the images from electron microscopy show clearly a visible transition line. From an optical point of view, it may cause undesirable light scattering. Irregularity of the transition line will be reduced using the higher separation energy. From these facts, we can conclude that for this type of method it is very important to have the proper power settings. Consequently, we can also assume that the appropriate adjustment will be the induction of secondary ocular phenomena greatly reduced.

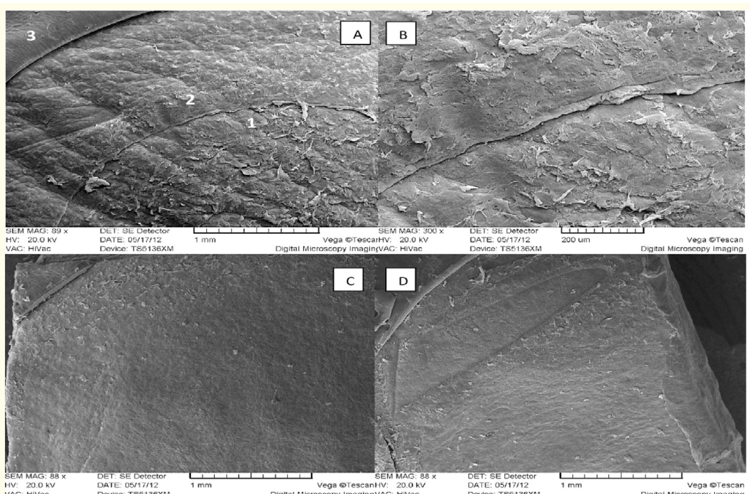

Figure 2: *ReLEx: [A] Overview of optical1, nonoptical2 and epithelial3 part of cornea. [B] ReLEx: The interface between optical and non-optical part. [C] Femtoflap + excimer ASA: overview. [D] Femtoflap + excimer TSA: overview.*

[1]The TSA profile is designed to preserve tissue. Another concern with the TSA profile is that it does not always adequately correct asphericity. The TSA profile was optimized for lower correction cases as derivative from the ASA profile [22].

[2]The ASA profile is an aspherically optimized profile; however, it does consume more of the patient's corneal tissue in the process as compared with the TSA profile. ASA was best suited for higher diopter ranges of correction, as it tends to result in overcorrection in eyes presenting with less than -3.00 D of myopia. The higher asphericity in low diopters results in more tissue ablation [1].

In the second pair of eyes, we used VisuMax only to create the flap. The ablation of diopters was performed on an MEL 80 excimer laser. The corneal surface appeared to be smooth and uniformed without any apparent transition zone between the optical and non-optical parts. The TSA method demonstrated in comparison to a faster course of operation a less smooth surface (we believe that this is due to the fact that the ASA profile removes more corneal tissue); yet both of these ablation methods had better properties of the final corneal surface [13,24].

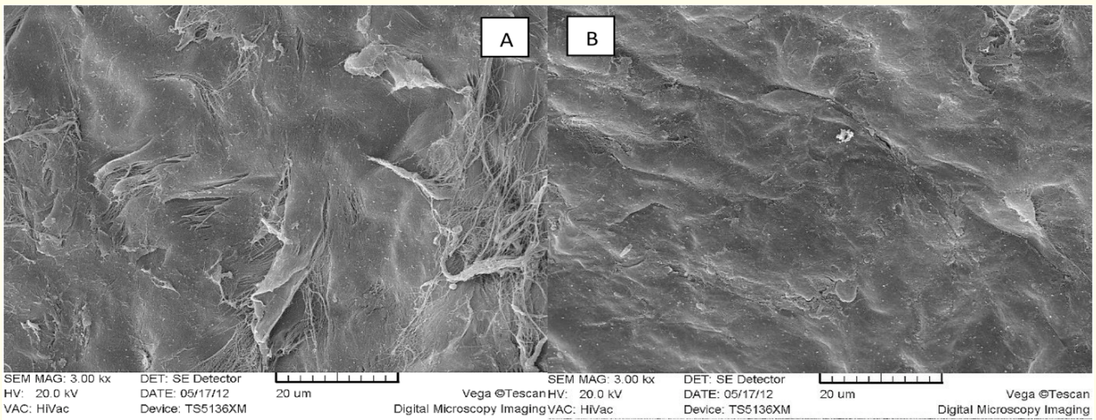

Figure 3: *ReLEx: [A] Femtoflap + excimer ASA: optical part. [B] Femtoflap + excimer TSA: optical part.*

Images of the last pair of eyes treated by using the microkeratome show even a "contusion" of the corneal bed in a place where the microkeratome starts. The remaining portion of the bed had micro slits, which were running from the direction of movement of the microkeratome. Side cut was irregular. This method, related to quality of the corneal bed, appears to be the least desirable [4,6,9,13,25-27].

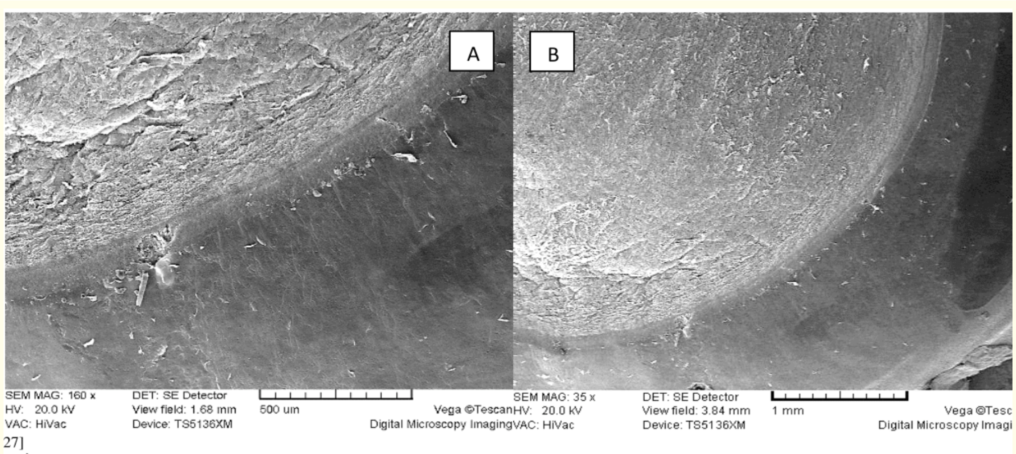

Figure 4: *ReLEx: [A] Microkeratome: edge of flap. [B] Microkeratome: overview.*

Discussion

For good results of the ReLEx surgery method, it is essential to set the laser beam values correctly. However, the quality of the corneal stromal bed appears to be still smoother in older excimer methodologies [28,29]. We can also claim that ablation profile affects the smoothness of stromal bed more than laser frequency value. The current experience shows that the more laser spots we have, the smoother surface we will create [3]. Nevertheless, following previous studies we cannot state that the quality of the stromal bed directly affects the healing, post-operative complications and post-operative corneal biomechanical properties. We have found no connection between symmetry of the stromal bed or side cut on the outcomes of laser eye surgery [24,26,30].

It is interesting that the ReLEx method, in which the quality of the stromal bed was worse, had a better post-operative values: higher visual acuity, faster healing and lower amount of the higher order aberrations [29,30,33]. We think that the clearly visible line between the optical and the non-optical zone can cause the night vision disturbances and front light scatter changes. The current studies, however, refute this fact [34]. Microkeratome method appears to be the least desirable for creation of flap [31]. In this method, we observed higher irregularity of the side cut and the "tissue damage" of the corneal bed.

Financial Interest

None.

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