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Excimer laser photoablative filtration surgery: histology and ultrastructure in 4 human cadaver eyes

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Abstract

The purpose of this study was to verify the feasibility of ab externo layer-by-layer excimer laser photoablative removal of limbal tissue down to the trabecular meshwork and to assess the damage caused by this procedure to the neighbouring structures. Excimer laser photoablation (193 nm) can remove layers of corneal tissue effectively with little or no damage to the adjacent areas. Previous experimental studies have demonstrated a decrease in outflow resistance after ab-externo photoablative removal of juxtacanalicular tissue. We have performed ab-externo photoablative removal of limbal tissue overlying the trabecular meshwork in four freshly enucleated eyes from our Eye Bank. The beam of an excimer laser (wavelength 193 nm; fluence 180 mJ/Sq.cm) was shaped using a metal mask with a rectangular opening of 1.2 × 2.5 mm. After removing the conjunctiva, photoablation was carried out at maximum surgical microscope magnification (40 ×) until trabecular meshwork appeared at the bottom of the crater. Light microscopy showed that craters had smooth walls and their base reached the Schlemm's canal area; all structures appeared of normal morphology. Transmission electron microscopy showed a thin layer of amorphous material or pseudomembrane on the side walls of the crater; corneoscleral collagen fibers were abruptly interrupted and undistorted. At the bottom of the crater the trabecular meshwork and Schlemm's canal tissues appeared normal.

Introduction

The excimer laser has been shown to be able to remove thin layers of corneoscleral tissue down to the filtering structures of the angle, thus eliminating most of the outflow resistance [1]. This procedure is called 'Photoablative Filtration Surgery' or 'Partial External Trabeculectomy'. Early clinical trials have shown favorable short term results [2–4]. We have investigated by histology and transmission electron microscopy the morphology of ab externo excimer laser photoablative removal of the limbal tissue everlying the trabecular meshwork.

Materials and methods

Four freshly enucleated human eyes from our Eye Bank, not suitable for corneal transplantation, aged from 60 to 78 years were used. The procedures were performed with an Argon Fluoride excimer laser emitting at 193 nm, with 180 mJ/Sq.cm fluence, at a repetition rate of 10 Hz, model ExciMed UV200LA by SUMMIT Technology, Waltham, Massachusetts, USA. The excimer laser beam was directed at a right angle to the tangential of the globe surface at the limbus. The beam was shaped in a rectangular fashion using a custom-made metal mask with a central window measuring 1.2 × 2.5 mm. After removing the conjunctiva, photoablation was carried out over the Schlemm's canal area until the trabecular meshwork was reached. Between 1700 and 2500 laser pulses were necessary. After photoablation all specimens were immediately immersed in 2.5% glutaraldeide and 2% osmium tetroxyde, then dehydrated in absolute alcohol and embedded in epoxy resin. One to two micron thin sections were stained with toluidine blue and fucsin for histology; ultrathin sections were stained with

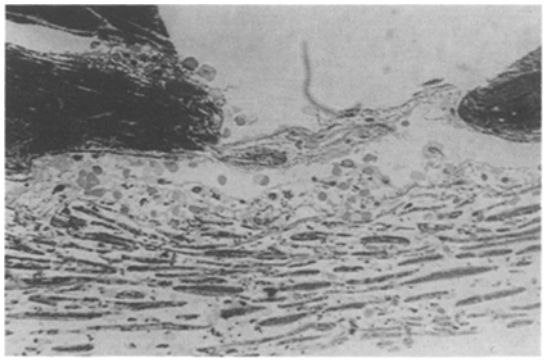

Fig. 1. Light microscopy. The photoablative crater has its bottom centered over the Schlemm's canal area; the walls are smooth and there is no visible damage to the trabecular meshwork.

3% uranile acetate and lead citrate for transmission electron microscopy. We used an electron microscope Siemens Elmiscop 101.

Results

With light microscopy the section of photoablative craters showed that the photoablative removal of corneoscleral tissue reached the trabecular meshwork (Fig. 1). The juxtacanalicular tissue was part of the base of the crater itself; only its outer part was removed, probably involving 10–15 micron of trabecular tissue. The remaining trabecular meshwork was intact and undamaged, maintaining some separation between the anterior chamber and the subconjunctival space. In all specimens the base of the crater was well centered on the Schlemm's canal area; in two cases the outer wall of the canal itself was unroofed. The walls of the craters were steep and smooth particularly throughout the corneoscleral portion of the excision, where tissue is more dense; the architecture of corneal lamellae was normal. Transmission electron microscopy confirms the smoothness of the crater walls (Fig. 2); they appear covered by an electron-dense pseudomembrane. This is an electron dense layer probably made by the aggregation of photoablation products [5]. The collagen fibers are regularly distributed all along the photoablated zone even immediately adjacent to the pseudomembrane. Cells and fibers appear cut precisely without any apparent thermal or mechanical damage.

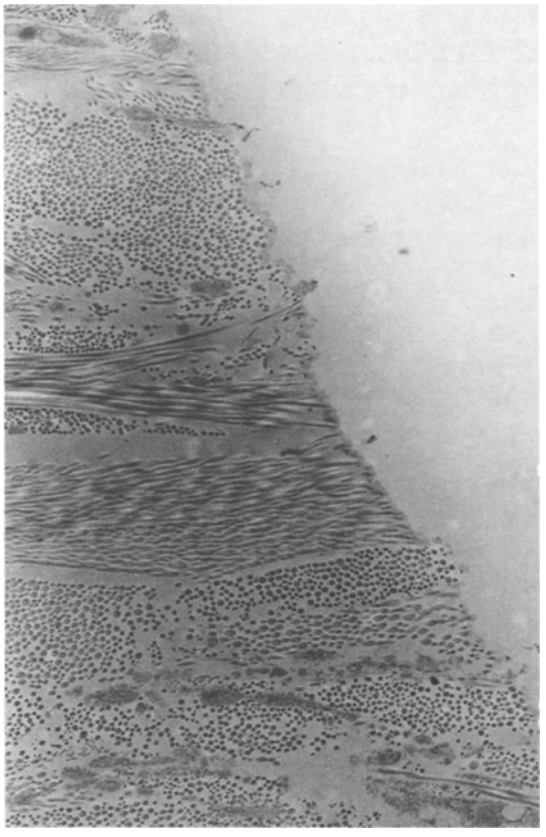

Fig. 2. Transmission Electron Microscopy (2100 ×). The wall of the crater is smooth and collagen fibers preserve their normal distribution and orientation. A pseudomembrane lining the walls is visible.

Conclusions

The excimer laser is capable of removing microscopic layers of tissue with little evidence of damage to the neighbouring structures [6–8]. Photoablative filtration is being evaluated clinically for primary-open angle glaucoma; to date it seems to produce consistent lowering of IOP for up to one year in most studies [2–4]. The mechanism by which the intraocular pressure is lowered is not clear yet; early investigations suggested that an increase in outflow was caused by the selective removal of corneoscleral tissue and outer wall of Schlemm's canal, or the juxtacanalicular part of the trabecular meshwork [1]. Our results demonstrate clearly the ability of this laser to obtain a crater precisely centered over the Schlemm's canal area in four cadaver eyes. The walls of excimer laser excisions were cut smoothly and damage was minimal or undectectable.

Whether this will result in better long-term control of intra ocular pressure remains to be established. This is an important difference from the conventional filtration surgery where manual manipulation and crushing by surgical instruments may cause more tissue disruption. Also our findings demonstrate that experimental ab externo photoablative filtration is not a 'trabeculectomy' nor a full thickness procedure. We speculate that the remaining thin layer of trabecular tissue might act as a microporous passive filter.

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