Universiteit
Leiden
The Netherlands

Elucidation of the migratory behaviour of the corneal endothelium

Miron, A.

Citation
Miron, A. (2023, March 9). *Elucidation of the migratory behaviour of the corneal endothelium*. Retrieved from https://hdl.handle.net/1887/3570514

Version:	Publisher's Version
License:	Licence agreement concerning inclusion of doctoral thesis in the Institutional Repository of the University of Leiden
Downloaded from:	https://hdl.handle.net/1887/3570514

Note: To cite this publication please use the final published version (if applicable).

Elucidation of the migratory behaviour of the corneal endothelium

Alina Miron

Elucidation of the migratory behaviour of corneal endothelium

Alina Miron
Leiden University Medical Center, Leiden, The Netherlands
Netherlands Institute for Innovative Ocular Surgery, Rotterdam, The Netherlands

Cover image and design by:	Iulia Ghenea, IE Clothing
Cover:	The cover image embraces the innovative mindset nourished by the traditional flame. Always in motion, curiosity leads to research and innovation. Instilled by folklore and traditional culture, it exudes a wealth of knowledge that will provide a major impulse to scientific research.
Scientific photography:	Netherlands Institute for Innovative Ocular Surgery, Rotterdam, The Netherlands
Layout and printed by:	ProefschriftMaken (www.proefschriftmaken.nl)
ISBN:	9789464692105

Copyright © 2023, Alina Miron
Copyright of the published material in chapters 2-7 and 9 lies with the publisher of the journal listed at the beginning of each chapter. All rights reserved. It is not permitted to reprint, reproduce or utilize (parts of) this thesis in any form by electronic, mechanical, or other means now known or hereafter invented, including photocopying and recording in any information storage or retrieval system without prior written consent of the author.

Financial support
Cover design and printing of this thesis was generously provided by: Rotterdamse Stichting Blindenbelangen, Stichting Leids Oogheelkundig Ondersteuningsfonds (LOOF)
Publication of this thesis was partially supported by: European Union's Horizon 2020 research and innovation programme (grant number 667400 – ARREST BLINDNESS Consortium);

Elucidation of the migratory behaviour of the corneal endothelium

Proefschrift

ter verkrijging van
de graad van doctor aan de Universiteit Leiden,
op gezag van rector magnificus prof.dr.ir. H. Bijl,
volgens besluit van het college voor promoties
te verdedigen op donderdag 9 maart 2023
klokke 15.00 uur

door

Alina Miron
geboren te Iași, România
in 1982

Promotor:

Prof. Dr. M.J. Jager

Co-promotors:

Dr. G.R.J. Melles, Netherlands Institute for Innovative Ocular Surgery, The Netherlands

Dr. S. Oellerich, Netherlands Institute for Innovative Ocular Surgery, The Netherlands

Leden Promotiecommissie

Prof. Dr. F.H.J. Claas

Dr. Y.Y. Cheng

Dr. N. Legali, Linköping University, Zweden

Prof. Dr. C. Cursiefen, Uniklinik Köln, Duitsland

For my Family

TABLE OF CONTENTS

Preface		9
Chapter 1	Introduction and outline of the thesis	13
Part 1	**DMEK Graft Analysis before and after Transplantation**	31
Chapter 2	*In vivo* endothelial cell density decline in the early postoperative phase after Descemet membrane endothelial keratoplasty (DMEK)	33
Chapter 3	Endothelial cell viability after DMEK graft preparation	43
Part 2	**Regenerative strategies for the treatment of Fuchs endothelial dystrophy**	59
Chapter 4	Corneal endothelial wound healing: understanding the regenerative capacity of the innermost layer of the cornea	61
Chapter 5	Asymmetrical endothelial cell migration from in vitro Quarter-Descemet membrane endothelial keratoplasty grafts	83
Chapter 6	Improving endothelial explant tissue culture by a novel thermoresponsive cell culture system	95
Chapter 7	In vitro endothelial cell migration from limbal edge-modified Quarter-DMEK grafts	105
Chapter 8	Early and late-onset cell migration from peripheral corneal endothelium	119
Chapter 9	Preclinical testing of small diameter Descemet membrane endothelial keratoplasty grafts to increase tissue availability	137
Chapter 10	Summary, Discussion, and Future Perspectives	151
Chapter 11	Nederlandse samenvatting (Dutch summary)	177
Appendices	List of publications Curriculum Vitae Acknowledgements	205

Preface

PREFACE

The human eye has millions of functional cells, and is considered the second most complex organ in the body after the brain. The eyes dominate emotional communication, governing interactions between the brain and the heart and capturing a lifetime of an individual's views. Although, we can interpret emotions (e.g., anger, disgust, fear, happy, sad, surprise, and to a lesser extent contempt, embarrassment, interest, pain, and shame) by analyzing the expression of the face and eyes, our view is modified by social learning and culture. The fine work of the renowned French photographer Rehahn illustrates ethnic culture, landscapes and portraits with emotions. His most praised works include a close-up shot of a 103-year-old Rengao woman who reveals - through her warm and sparkling eyes - satisfaction and happiness, attesting to a rich life story. Moreover, the eyes can offer a unique glimpse into the body's health and by regular monitoring, an eye doctor may be able to spot systemic medical conditions potentially leading to early diagnostic and treatments. The retinal blood flow and vessels can, for instance, signal a risk for stroke or high blood pressure, and a yellow sclera may be a sign of hepatitis.

The eye is protected from germs, dust, harmful objects, and to some extent, from the damaging ultraviolet wavelengths in sunlight by the eyelids and the cornea. The cornea also acts as the eye's outermost refractive surface focusing the light that will reach the brain as electrical signals which are then translated further into images. In order for a person to see well, all five main layers of the cornea must be free of any cloudy or opaque areas. The cornea can recover from minor injuries on its own, however, deep injuries will take longer to heal and might also cause pain, blurred vision, extreme sensitivity to light, and in some cases even corneal scarring. Also, corneal dystrophies can affect one or more parts of the cornea through accumulation of foreign material that will cause the cornea to lose its transparency, potentially leading to loss of vision. To restore vision in such cases, a cornea transplant (i.e., keratoplasty) is performed in which (a part of) the defective cornea is replaced with healthy corneal tissue from a deceased donor. In cases in which the inner most layer of the cornea (i.e., endothelium) is affected, it is mostly restored by performing a Descemet Membrane Endothelial Keratoplasty (DMEK) procedure. However, the global shortage of available corneal donor grafts and a rise in the ageing population cause a shortness in potential transplants. Thus, a considerable clinical interest exists for developing tissue-engineered constructs and new cell therapies using cultivated cells.

More than 9 years have passed since I became fascinated by this clear window of the eye. The story begins in Rotterdam where I was doing a six-month internship at the Netherlands Institute for Innovative Ocular Surgery (NIIOS) center located in the vicinity of the Erasmus Bridge. Before completing a 2-year program and being awarded the degree Professional Doctorate in engineering (PDEng) at the Delft University of Technology, I was referred to set up a protocol for culturing a cell type with a rare nesting behavior. But applying these results into clinical practice is another challenge and if you fast-forward the time, you will still find me trying to explain paradigms that show-up while digging for answers. For a successful implementation of regenerative therapies for the corneal endothelium, we need to understand the dynamic cellular changes that occur in vivo in both normal and diseased tissue. Only after this, can effective solutions be proposed to reduce the global shortage of donor corneas.

In this thesis, I will focus on the *in vivo* and *in vitro* behavior of corneal endothelial cells before and after endothelial keratoplasty. Regenerative strategies for the treatment of Fuchs endothelial corneal dystrophy, the most common corneal disorder requiring transplantation, will be tackled from a dual perspective, i.e., regeneration without allogeneic corneal endothelial cell transplantation and targeted activation of endogenous self-repair mechanisms.