

Cover Page



Universiteit Leiden



The handle <http://hdl.handle.net/1887/20426> holds various files of this Leiden University dissertation.

**Author:** Commandeur, Suzan

**Title:** Organotypic in vitro models of human cutaneous squamous cell carcinoma

**Issue Date:** 2013-01-16

Organotypic *in vitro* models of  
human cutaneous squamous cell carcinoma

Suzan Commandeur

Organotypic *in vitro* models of human cutaneous squamous cell carcinoma  
© Suzan Commandeur, 2013

All rights reserved. No part of this thesis may be reproduced, stored or transmitted in any way without prior written permission of the copyright owner.

ISBN:	9789088915505
Cover design:	Suzan Commandeur
Photo on front cover:	Application of test compound on <i>in vitro</i> human skin model. © Aat Mulder
Lay out:	Suzan Commandeur
Printed by:	Proefschriftmaken.nl    Uitgeverij BOXPress
Published by:	Uitgeverij BOXPress, 's-Hertogenbosch

The research presented in this thesis was performed at the Department of Dermatology of the Leiden University Medical Center, The Netherlands.

The research presented in this thesis was financially supported by ZonMw, the Netherlands Organisation for Health Research and Development.

Financial support for the publication of this thesis was kindly provided by Stichting Stimuleringsfonds Alternatieven voor Dierproeven, Stichting Proefdiervrij, Animal Free Research, Aeon Astron Europe B.V., GlaxoSmithKline B.V., LEO Pharma B.V., Galderma B.V., Louis Widmer Nederland B.V., Astellas B.V., Fagron B.V. and Merz GmbH.

Organotypic *in vitro* skin models of  
human cutaneous squamous cell carcinoma

Proefschrift

ter verkrijging van  
de graad van Doctor aan de Universiteit Leiden,  
op gezag van de Rector Magnificus Prof. Mr. P.F. van der Heijden,  
volgens besluit van het College voor Promoties  
te verdedigen op woensdag 16 januari 2013  
klokke 16.15 uur

door

**Suzan Commandeur**  
geboren te Delft  
in 1983

## **Promotiecommissie**

Promotor	Prof. Dr. R. Willemze
Co-promotores	Dr. A. El Ghalbzouri Dr. C.P. Tensen
Overige leden	Prof. Dr. S. Gibbs <i>VU University Medical Centre (VUmc), Amsterdam</i>
	Prof. Dr. J.A. Bouwstra <i>Leiden Amsterdam Centre for Drug Research (LACDR), Leiden</i>
	Prof. Dr. L.H.F. Mullenders

Voor Jaap



## Contents

	List of abbreviations	9
<b>Chapter 1</b>	General introduction	11
<b>Chapter 2</b>	An in vitro three-dimensional model of primary human cutaneous squamous cell carcinoma <i>Exp Dermatol. 2009 Oct;18(10):849-56.</i>	37
<b>Chapter 3</b>	Functional characterization of cancer associated fibroblasts of human cutaneous squamous cell carcinoma <i>Exp Dermatol. 2011 Sep;20(9):737-42.</i>	59
<b>Chapter 4</b>	Replacement of animal-derived collagen matrix by human fibroblast-derived dermal matrix for human skin equivalent products <i>Biomaterials. 2009 Jan;30(1):71-8.</i>	77
<b>Chapter 5</b>	Gene expression analysis of squamous cell carcinoma of immunosuppressed transplant recipients. <i>Submitted</i>	101
<b>Chapter 6</b>	Epidermal growth factor receptor activation and inhibition in 3D in vitro models of normal skin and human cutaneous squamous cell carcinoma in vitro. <i>Cancer Science (accepted)</i>	131
<b>Chapter 7</b>	General discussion	147
<b>Chapter 8</b>	Nederlandse samenvatting	162
	List of publications	170
	Curriculum vitae	172
	Short summary	174



## List of abbreviations

<b>6-4PP</b>	6-4 photoproducts	<b>HRP</b>	horseradish peroxidase
<b>AE</b>	air-exposed	<b>LOH</b>	loss of heterozygosity
<b>AEC</b>	3-amino-9-ethylcarbazole	<b>KEGG</b>	Kyoto encyclopedia of genes and genomes
<b>AK</b>	actinic keratosis	<b>KGF</b>	keratinocyte growth factor
<b>BAF</b>	B allele frequency	<b>MMP</b>	matrix metalloproteinase
<b>BCC</b>	basal cell carcinoma	<b>NF</b>	normal fibroblast
<b>BM</b>	basement membrane	<b>NFKB</b>	nuclear factor κB
<b>BP</b>	biological processes	<b>NHDF</b>	normal human dermal fibroblast
<b>BSA</b>	bovine serum albumin	<b>NHEK</b>	normal human epidermal keratinocyte
<b>CAF</b>	cancer-associated fibroblast	<b>NMF</b>	natural moisturizing factor
<b>CPD</b>	cyclobutane-pyrimidine dimers	<b>NMSC</b>	non-melanoma skin cancer
<b>cnLOH</b>	copy neutral LOH	<b>NS</b>	normal skin
<b>CNV</b>	copy number variation	<b>OTR</b>	organ transplant recipient
<b>DAB</b>	3,3'-diamino- benzidine	<b>PBS</b>	phosphate buffered saline
<b>DED</b>	de-epidermized dermis	<b>PCA</b>	principal component analysis
<b>DEG</b>	differentially expressed gene	<b>PCR</b>	polymerase chain reaction
<b>DEJ</b>	dermal-epidermal junction	<b>PDGF</b>	platelet-derived growth factor
<b>DEP</b>	differentially expressed probe	<b>PGSEA</b>	parametric GSEA
<b>DMEM</b>	Dulbecco's modified Eagle medium	<b>QPCR</b>	quantitative RT-PCR
<b>ECM</b>	extracellular matrix	<b>RANKL</b>	receptor activator of NF-κB ligand
<b>EGF</b>	epidermal growth factor	<b>RSN</b>	robust spline normalization
<b>EGFR</b>	epidermal growth factor receptor	<b>RT-PCR</b>	reversed transcriptase PCR
<b>ELISA</b>	enzyme linked immunosorbent assay	<b>SB</b>	stratum basale
<b>ESC</b>	epidermal stem cell	<b>SC</b>	stratum corneum
<b>FBS</b>	fetal bovine serum	<b>SCC</b>	squamous cell carcinoma
<b>FC</b>	fold change	<b>SNP</b>	single nucleotide polymorphism
<b>FDR</b>	false discovery rate	<b>SS</b>	stratum spinosum
<b>FDM</b>	fibroblast-derived matrix	<b>STR</b>	short tandem repeat
<b>FGF</b>	fibroblast growth factor	<b>TF</b>	transcription factor
<b>GO</b>	gene ontology	<b>TFBS</b>	transcription factor binding site
<b>GSEA</b>	geneset enrichment analysis	<b>TGF-β</b>	transforming growth factor β
<b>GWEA</b>	genome-wide expression analysis	<b>TSG</b>	tumor suppressor gene
<b>HGF</b>	hepatocyte growth factor	<b>UVR</b>	ultraviolet radiation
<b>HPV</b>	human papilloma virus	<b>VST</b>	variance-stabilizing transformation
<b>HSE</b>	human skin equivalent		

