

Materials

1. Reagents.

Reagent	Manufacturer	Catalog number
2-Propanol, pro analysi	Merck	1.09634.1011
5-Sulfosalicylic acid hydrate	Sigma	390275-500G
Acrylamide/Bis-acrylamide 40% Acrylamide, 37:1 ratio	Sigma	A6050-100ML
Adenine	Sigma	A2786-5G
Advanced D-MEM	Gibco	12491-023
Agar Bacteriological Grade	MP Biomedicals, INC	150178
Ammonium persulfate for electrophoresis, ≥98%	Sigma,	A3678-100G
β-Mercaptoethanol	Sigma	M7154-25ML
BLOCK-iT™ fluorescent oligo	Invitrogen	2013
Blotting grade Blocker Non-Fat Dry Milk	BioRad	170-6404
Calcium chloride CaCl ₂ •2 H ₂ O	Sigma	21098-500G-F
Chemically Defined Lipid Concentrate (100X), liquid	Gibco	11905-031
Chloroquine diphosphate salt	Sigma	C6628-25G
Cholera Toxin	GENTAUR	100 Inaba 569 B
Cholesterol, Water Soluble powder, cell culture tested	Sigma	C4951-30MG
Cycloheximide	Sigma	C4859-1ML

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Reagent	Manufacturer	Catalog number
D-(+)-Glucose	Sigma	G8270-1KG
Dimethyl sulfoxide (DMSO) ≥ 99.7%, Hybri-Max™, sterile-filtered, hybridoma tested	Sigma	D2650-100ML
Dispase I (neutral protease, grade I)	Roche	04942086 001
DMEM high glucose (with GlutaMAX™ I, 4500 mg/L D-Glucose, without Sodium Pyruvate),	Gibco	61965-059
DMEM low glucose (+1g/L D-Glucose, +L-Glutamine +Pyruvate),	Gibco	31885-049
Dodecylsulfate, Na-salt solution in water 20% SDS Solution	Serva	20767.03
Doxycycline hyclate,	Sigma	D9891-25G
DPBS (1X), liquid - without Calcium, - without Magnesium	Invitrogen	14190-169
EDTA, 0,5M solution Ethylenediaminetetraacetic acid disodium salt solution for molecular biology, 0.5 M	Sigma	E7889-100ML
Epidermal Growth Factor (EGF) human recombinant, expressed in Escherichia coli, lyophilized powder, cell culture tested	Sigma	E9644-.2MG
Ethanol, 96%	Mallinckrodt Baker	8229
Ethidium bromide (EtBr) 1 % solution in water (10mg/ml)	Roth	2218.2
E-Toxate™ Water (H ₂ O) Endotoxin, free	Sigma	2107-10ML
FBS Fetal Bovine Serum	Biochrom AG	S 0115

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Reagent	Manufacturer	Catalog number
GlutaMax I liquid	Gibco	35050-061
Glycine	Sigma	G8898-1KG
GW501516	AXXORA	ALX-420-032-M001
HEPES ≥ 99.5% (titration)	Sigma	H3375-1KG
Hydrocortisone Chromatographic Standard	CALBIOCHEM	386698
Insulin from bovine pancreas	Sigma	I5500-50MG
KBM-2 SingleQuot Kit	Cambrex	CC-4152
Keratinocyte Basal Medium 2 (KBM-2) without calcium	Cambrex	CC-3158
L-165041	Calbiochem	422175
LB-Broth (Luria/Miller)	Roth	X968.1
Lipofectamine™ 2000 Reagent	Invitrogen	11668-019
Magnesium acetate tetrahydrate (CH ₃ COO) ₂ Mg•4H ₂ O for molecular biology, ≥ 99%	Sigma	M5661-250G
MAX Efficiency Stbl2 Chemically Competent Cells	Invitrogen	10268-019
Methanol	Mallinckrodt Baker	8045
Mitomycin C from Streptomyces caespitosus, powder	Sigma	MO503-2MG
Nutrient Mixture F-12 Ham (With sodium bicarbonate, without L-glutamine)	Sigma	N4888-6X500ML
One Shot® TOP10 Chemically Competent E. coli	Invitrogen	C4040-06

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Reagent	Manufacturer	Catalog number
Opti-MEM® I Reduced Serum Medium	Invitrogen	31985-070
Penicillin-Streptomycin	Invitrogen	15140-163
Polybrene (Hexadimethrine bromide)	Sigma	107689-10G
Ponceau S,	Sigma	P 3504-10G
Potassium acetate (CH ₃ COOK) for molecular biology, ≥ 99%	Sigma	P1190-500G
Potassium chloride (KCl) ≥ 99.0%	Sigma	P9333-1KG
PPAR δ blocking peptide	Cayman Chemical	10006247
Restriction Endonucleases	New England BioLabs	all used
RPMI Medium 1640 (with GlutaMAX™ I),	Gibco	361870-044
SeaKem LE Agarose for gel electrophoresis	Cambrex	50004
Sodium chloride (NaCl)	Sigma	S7653-1KG
Sodium phosphate dibasic SigmaUltra, ≥ 99.0%	Sigma	S7907-1KG
Sucrose ≥ 99.5%	Sigma	S9378-1KG
TEMED N,N,N',N'-Tetramethylethylenediamine	BioRad	161-0800
Trichloroacetic acid	Sigma	T9159-500G
Tricine Sample Buffer	BioRad	161-0739
TRIS Trizma base	Sigma	T1503-1KG

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Reagent	Manufacturer	Catalog number
Trizma® hydrochloride solution pH 7.4, 1 M Tris-HCL, 1M Stock Solution, pH 7.40	Sigma	T 2663-1L
Trypsin-EDTA (1X), liquid - 0.05 % Trypsin, 0.53 mM EDTA•4Na	Gibco	25300-096
Tween 20	Serva	37470

2. Kits.

Kit	Manufacturer	Catalog number
BCA Protein Assay Kit	PIERCE	23225
ECL Plus Western Blotting Detection System	Amersham Biosciences	RPN2 132
Jetstar 2.0 MaxiPrep Kit	Genomed	222020
Mini-PROTEAN 3 electrophoresis system	BioRad	165-3301
NE-PER Nuclear and Cytoplasmic Extraction Reagents	PIERCE	78833
NucleoSpin® RNA II Kit	MACHEREY-NAGEL	MN-740955/25
QIAquick Gel Extraction Kit	QIAGEN	28706
QIAprep Spin Miniprep	QIAGEN	27106

3. Laboratory equipment.

Balances		
Instrument	Manufacturer	Type
Ultra fine Balance up to 5,1g	Sartorius	KCBA 100
Fine Balance up to 620g	Sartorius	LC 6215
Balance up to 8500g	Gibertini	Europe 8000
Centrifuges and Rotors		
Instrument	Manufacturer	Type
Centrifuge	Beckman	Avanti J-25
Rotor for Avanti J-25	Beckman	JA 25.50
Rotor for Avanti J-25	Beckman	JLA 10.500
Centrifuge	Hettich	ROTIXA 50RS
Table Centrifuge	Eppendorf	5417C
Cell culture Centrifuge	Beckman	GS-6
Ultracentrifuge	Beckman	Optima LE-80 K
Rotor for Optima LE-80 K	Beckman	SW28
Freezers		
Instrument	Manufacturer	Type
-140°C Ultra Low Freezer	Hereaus	HFC10140 Top-M-21
-80°C Ultra Low Freezer	Hereaus	HFU586 BASIC
-80°C Ultra Low Freezer	SANYO	MDF-U73V
-20°C Freezer	Liebherr	Premium
+4°C Refrigerator	Liebherr	Premium

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Instrument	Manufacturer	Type
Autoclave	H+P Labortechnik AG	Varioklav 75S
Easy-Cast Electrophoresis System	Owl Scientific, Inc	B1A
Easy-Cast Electrophoresis System	Owl Scientific, Inc	B2
Electronic Pipette Set	Eppendorf	Research Pro
Electrophoresis Power Supply	Pharmacia Biotech	EPS 300
Electrophoresis Power Supply	Pharmacia Biotech	EPS 600
Flow Cytometer	Becton Dickinson	FACS Calibur E3583
Fluorometer	Invitrogen	Qubit
Heating Plate	IKA Labortechnik	RCT Basic
Incubator	Jouan	IG150
Incubator Shaker	New Brunswick Scientific	Innova 4000
Inverse Mikroskope	Carl Zeiss	Axiovert 25 CA
Lamin Air	Hereaus	HS12
Light Cycler and Sequence Detection System	Applied Biosystems	ABI PRISM® 7700
Luminescent Image Analyzer	Fujifilm	LAS-1000 CH
Peltier Thermal Cycler	MJ Research	PTC-200
pH-meter	Wissenschaftlich-Technische Werkstätten	pH537
Pipett Boy	Hirschmann Laborgeräte	Pipetus®-akku
Pipette Set	Eppendorf	Reference

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Instrument	Manufacturer	Type
Semi-dry Blotting Chamber	LKB Bromma	2117 Multiphor
Thermomixer	Eppendorf	5436
Thermomixer Compact	Eppendorf	5350 000.013
UV-Photometer	Biometra	GeneRay
Vortex Genie 2	Scientific Industries	G-560E
Water bath	GRANT Instruments	SUB6
Water bath	Memmert, GmbH	WB7

4. Enzymes.

Enzyme	Manufacturer	Catalog number
GoTaq DNA Polymerase	Promega Corporation	M3005
RNaseOUT™ Ribonuclease (RNase) Inhibitor	Invitrogen	10777019
SuperScript™ II Reverse Transcriptase	Invitrogen	18064-071
Moloney Murine Leukemia Virus Reverse Transcriptase (MMLV-RT)	EPICENTRE	M6125H
Restriction Endonucleases	New England BioLabs	all used
T4 DNA Ligase	New England BioLabs	M0202S
PfuUltra™ High-fidelity DNA Polymerase	Stratagene	600380

5. Markers.

Marker	Manufacturer	Catalog number
1 Kb DNA Ladder	New England BioLabs	N3232L
100 bp DNA Ladder	New England BioLabs	N3231L
EasyLadder I DNA 100-2000 bp	BioLine	BIO-33046
EasyLadder II DNA 500-5000 bp	BioLine	BIO-33048
HyperLadder I DNA 200-10 000 bp	BioLine	BIO-33026
MagicMark™ XP Western Protein Standard	Invitrogen	LC5602

6. Disposables.

Disposable	Manufacturer
14 ml Polypropylene Round-Bottom Tube	BD Falcon
5 ml Polystyrene Round-Bottom Tube	BD Falcon
Biopsy Punch, 4mm Sterile and pyrogene-free	PFM (KAI Industries Co.Ltd)
CELLSTAR® 6 Well Cell Culture Multiwell Plates (Growth area per well: 9.6cm ²)	Greiner
CELLSTAR® Cell Culture Dish, WITH VENTS, 100 X 20 MM, (57 cm ²)	Greiner
COREX® II Centrifuge Tubes	GENTAUR

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Disposable	Manufacturer
Corning® CellBIND® 6 Well Clear Microplate, Flat Bottom, with Lid, Sterile (6x9.5 cm ²)	Corning
Corning® CellBIND® Surface 100mmx20mm Culture Dish (55 cm ²)	Corning
Cryo.s™ Cryogenic Storage Vials,	Greiner
Disposable Filter Unit 0,45µm	Whatman
Disposable scalpel No.10	PFM (Feather safety Razor Co.LTD)
Disposable scalpel No.11	PFM (Feather safety Razor Co.LTD)
Gel Blotting Paper GB002	Whatman
Polyallomer Konikal centrifuge tubes	Beckman
Protran BA 85 Nitrocellulose 0,45µM	Schleicher&Schuell
Reagent and centrifuge Tube 15 ml, 120x17mm, PP	SARSTEDT
Reagent and centrifuge Tube 50 ml, 114x28mm, PP	SARSTEDT
Safe- Lock Tubes	Eppendorf
Safe Seal Tip	Biozym
Serological pipets	Corning
Standard tissue culture flask with filter 205 x 120 x 45 mm (150cm ²)	TPP
Standard tissue culture flask with filter 150 x 85 x 35 mm (75cm ²)	TPP
Stericup® and Steritop™ Durapore low binding membrane (PVDF) 73 mm/.45 µm,250 ml reciver	MILLIPORE
Stericup® and Steritop™ Millipore Express PLUS Membrane (PES) 73 mm/.22 µm 500 ml reciver	Millipore
Transfer Pipettes Single-use Pasteur pipettes,3,5 ml	SARSTEDT

7. Primary human material.

All work with patient materials presented in the thesis was carried out in accordance with the Helsinki declaration. All patients undergoing biopsy gave prior written consent.

Primary keratinocytes

Patients sampled for PPAR δ expression study were undergoing in-patient treatment for active disease at the time of biopsy and had PASI (psoriasis area and severity index) scores of 12 or higher, indicating moderate-to-severe disease. Healthy donors skin samples were obtained from routine surgery performed in Department of Dermatology, Charité University Hospital Berlin.

Primary fibroblast

Adult fibroblast were derived from mamma reduction surgery performed in Clinic for General Surgery, Charité University Hospital Berlin.