Stem Cell Therapy for Vascular Disease

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China-UK Forum in Biomedical Innovation
17-21 March 2008
*Background

*Stem cells

*Cardiovascular Disease

*Stem cell repairing/therapy
Stem Cells and Vascular Diseases

Proteomics
Manuel Mayr

Cell biology
Lingfang Zeng

Animal Model
Yanhua Hu

Clinic/human
Stefan Kiechl
Proteomics workflow

Sample Prep

Sample labelling

2-D Separation

Image acquisition

Image analysis

Mass spectrometry

Automated spot digestion

Automated MALDI spotting

Automated spot picking
Adult and embryonic stem cell culture and differentiation into vascular cells

Stem cells → SMCs

Stem cells → Endothelial cells
Mouse models established in our laboratory

- Spontaneous atherosclerosis
- Endothelial injury of artery
- Vein graft atherosclerosis
- Heart infarction
- Heart transplantation
A HANDBOOK OF MOUSE MODELS OF CARDIOVASCULAR DISEASE

With Movies For Surgical Procedures

Editor QINGBO XU

WILEY

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Human/clinic studies, e.g. Bruneck Study
- Brief introduction about our work
- Background about stem/progenitor cells
- Human studies
- Mechanistic studies on stem cell differentiation
Derivation of the ES cell lines.
The ES cell lines were generated from the blastocysts. These cells were isolated by immunosurgery and plated on the MEF feeder layer. ES cells can be propagated in the undifferentiated state while being cultured on top of the MEF feeder layer. When they are cultivated in suspension as 3D cell aggregates (EBs), the ES cells differentiate into specialized cells.
Progenitor cell mobilisation and homing

Stem cell pool in blood

Adipose
Aorta
BM
Spleen
Liver
Intestine
SMP
EPC
AS

Xu Q. Circ. Res. 2008
*Background

*Stem cells

*Cardiovascular Disease

*Stem cell therapy
Endothelial cells

Stem cells

Smooth muscle cells
Atherosclerosis - a vascular blocking disease
Treated with a balloon catheter, i.e. angioplasty
Treated with a balloon catheter, i.e. angioplasty
Restenosis Following Balloon Angioplasty

IEL

SMC Hyperplasia
"Restenosis"
Endothelial cells

Endothelial cell lost

Repairing with stem cells
*Background

*Stem cells

*Cardiovascular Disease

*Stem cell therapy: Mechanism of cell differentiation
Attached “progenitors”

A B C
Wild-type 2 months ApoE-/- 2 months ApoE-/- 18 months

D E
ApoE-/- 18 months
What is the mechanism for stem cell differentiation into vascular cells?
Flow Shear Induces Stem Cell Differentiation: Model & Hypothesis

Shear stress → VEGF receptor → Akt → HDAC3, eNOS

¬ p53, NO, p21, HoxA → Endothelial markers

PDGF-BB → Receptor-ββ → PI3K → HDAC7, HDAC7 splicing → MEF2C → Smooth muscle markers

Flow Shear Induces Stem Cell Differentiation toward Endothelial Cells

In vitro

In vivo matrigel

Flow Shear Induces Stem Cell Differentiation toward Endothelial Cells

Flow Shear Induces Stem Cell Differentiation: Impact of HDAC3
Flow Shear Induces Stem Cell Differentiation: Model & Hypothesis

Shear stress

- VEGF receptor
- Akt
- HDAC3
- eNOS
- p53
- NO
- p21
- HoxA
- Endothelial markers

PDGF-BB
- Receptor-ββ
- PI3K
- FAK
- HDAC7
- HDAC7 splicing
- MEF2C

Collagen IV
- Smooth muscle markers

HDAC7 and SMC differentiation

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<th>0</th>
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<th>3</th>
<th>5</th>
<th>7</th>
<th>9</th>
<th>mSMC</th>
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- **105KD**
  - HDAC7
- **42KD**
  - SMA
- **34KD**
  - Calponin
- **50KD**
  - α-tubulin

**HDAC7**

**SMA**

**Calponin**

**Merge**
HDAC7 and SMC differentiation
HDAC7 and SMC differentiation

Adenovirus-HDAC7-1

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<td>Calponin</td>
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<td>α-tubulin</td>
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</tbody>
</table>

siRNA-HDAC7-1

SM22 → Luc

PBS

PDGF

ctlsi HDAC7si

RLU

ctlsi HDAC7si
Conclusions
HDAC3 is crucial for stem cell differentiation into endothelial cells, while HDAC7 is for SMCs.
Can progenitor cells differentiate to endothelial cells in vivo in response to flow shear?

Shearing stem cells in vivo

Isolation of sca-1+ progenitors

Apply the cells to denuded artery

Endothelial phenotype
Flow shear Induces Stem Cell Differentiation: Endothelial cell therapy

X-gal  β-gal  CD31

Flow Shear Induces Stem Cell Differentiation: Sca-1+ cell therapy

Xiao et al. ATVB 2006
Replace blocked vessels with a normal vessel, e.g. saphenous vein.
Normal vessel

Vein graft atheroma
Stem cell differentiation

- Stem cells
- Endothelial cells
- SMCs
Mouse model of vein bypass graft atherosclerosis
QuickTime™ and a YUV420 codec decompressor are needed to see this picture.
Ongoing work: replacing diseased artery with stem cell-derived vessel
Summary

- Two types of stem cells: embryonic and adult cells

- VEGF plays a crucial role of stem/progenitor cells homing to the damaged vessels

- Signal pathways for endothelial differentiation involving in HDAC3-p53/p21

- Stem cell-derived endothelial cells can repairing damaged vessels in vivo
Innovation & Collaboration

New drug discovery to direct stem cell differentiation into endothelial cells, e.g. HDAC7 inhibitor or HDAC3 enhancer

Biomaterials for vascular production

Stem cell therapy in patients