Inner Nuclear Membrane Protein MAN1 and Regulation of R-Smad Signaling

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The Nuclear Envelope

By D. W. Fawcett
The Nuclear Envelope

Artwork by Don Guzy
The Nuclear Lamina is Composed of 10 nm-diameter Filaments

Aebi et al. (1986)
Some Characterized Proteins of the Inner Nuclear Membrane

NPC

LBR
Emerin
Myne-1
Nurim
MAN 1
LAPs

Nuclear lamina composed of A- and B-type lamins

Heterochromatin

Schirmer et al. Science
2003;301:1380-1382 -- 80 transmembrane proteins
Mutations in \textit{LMNA} Encoding A-type Lamins Cause Different Diseases

**Striated Muscle Disease**
- Autosomal Dominant Emery-Dreifuss Muscular Dystrophy
- Dilated Cardiomyopathy with Conduction Defect 1
- Limb Girdle Muscular Dystrophy Type 1B

**Partial Lipodystrophy Syndromes**
- Dunnigan-type Partial Lipodystrophy
- Mandibuloacral Dysplasia (with developmental anomalies)

**Peripheral Neuropathy**
- Charcot-Marie-Tooth Disorder Type 2B1

**“Premature Aging” Syndromes**
- Hutchinson-Gilford Progeria Syndrome
- Atypical Werner Syndrome
Mutations in Integral Inner Nuclear Membrane Proteins Cause Several Diseases

**Emerin**
- Emery-Dreifuss Muscular Dystrophy (X-linked)

**LBR**
- Pelger-Huët Anomaly (Heterozygous)
- HEM/Greenberg Skeletal Dysplasia (Homozygous)

**MAN1**
- Osteopoikilosis, Buschke-Ollendorff Syndrome and “Non-sporadic” Melorheostosis (Heterozygous)
“MAN Antiserum” Recognizes a Nuclear Envelope Antigen
Cloning, cDNA Sequencing
Genomic Analysis of MAN1

MAN1 on Chromosome 12q14
MW ~97 kDa
Lin et al. (2000)
Structure of the LEM (and LEM-like Domain of LAP2) Common to Several Integral Inner Nuclear Membrane Proteins

Laguri et al. (2001)
MAN1 is Localized to the Inner Nuclear Membrane

Lin et al. 2000
Integral Proteins Reach the Inner Nuclear Membrane by Diffusion-retention

Integral proteins synthesized on rough ER can diffuse to INM (size limit ~60 kDa) and be retained by binding to lamina or chromatin. The same proteins can potentially reach the Golgi/PM.
MAN1 Amino-terminal, Nucleoplasmic Domain Confers Inner Nuclear Membrane Targeting

Wu et al. 2002
MAN1 is Immobilized in the Inner Nuclear Membrane Relative to the ER

Wu et al. 2002
MAN1 Yeast 2-Hybrid Screen

Nothing

Smad 2 and Smad3

Lin et al. 2005
MAN1 Binds Smad 2/3 in Vitro

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Binding of MAN1-CT to Smad2/3 Invitro

Lin et al. 2005
MAN1 Binds Smad2 in Vivo

Lin et al. 2005
Smad Structure

The Carboxyl-terminal Part of MAN1 Binds to the MH2 Domain of Smad 2/3

Lin et al. 2005

Smallest Domain Binding Smads AA 730-910
Smads in TGF-β Signaling

MAN1 Inhibits TGF-β Transcription Activation

Lin et al. 2005
Creation of Cell Lines Overexpressing MAN1

Lin et al. 2005
MAN1 Inhibits TGF-β-mediated Cell Proliferation Arrest

Lin et al. 2005
Three-dimensional Structure of the MAN1 Region (aa 666-750)

Caputo et al. 2006
Superimposition of WH Domain of MAN1 on Similar Domain Found by DALI Server

Caputo et al. 2006
Calculation of the Electrostatic Potential at the Surface of MAN1 WH Domain

Caputo et al. 2006
Interaction of the Carboxyl-terminal Nucleoplasmic Region of MAN1 with DNA

Caputo et al. 2006
 Modeling of the Complex Between the WH Domain of MAN1 and DNA

Caputo et al. 2006
MAN1 in r-Smad Signaling

Goal: 3-dimensional structure complexed with R-Smad MH2
MAN1 Inhibits Smad1-mediated Signaling in Xenopus

- Osada et al. (2003) XMAN1, an inner nuclear membrane protein, antagonizes BMP signaling by interacting with Smad1 in Xenopus embryos. Development 130:1783-1794.
Other Results Showing MAN1 Inhibits Smad1/2/3 Signaling in Mammals


Acknowledgements MAN1

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