Repositioning of Small Molecules for Inherited Connective Tissue Diseases

Michael Briggs
Professor of Skeletal Genetics
Newcastle University
Talk outline

1. Inherited connective tissue disorders; *skeletal dysplasias*
2. Genotype vs. common disease mechanisms
3. ER stress as a common mechanism with therapeutic potential
4. Repurposing of drugs for rare connective tissue diseases
5. Biomarkers are essential
6. Future perspectives
Heritable connective tissue disorders have been acknowledged as a distinct group of rare diseases for over 50 years and there are more than 500 unique genetic disorders that affect a broad range of tissues including skin, cartilage, bone, tendon and skeletal muscle.

1966; 3rd Edition
Heritable Disorders of Connective Tissues

- Marfan Syndrome
- Homocystinuria
- The Ehlers-Danlos Syndrome
- Osteogenesis Imperfecta
- Alkaptonuria
- Pseudoxanthoma elasticum
- The mucopolysaccharidosis
- Other genetic disorders of connective tissues
  > genetic disorders of the osseous skeleton

Victor McKusick
2004; 1st Edition
Connective Tissue and Its Heritable Disorders: Molecular, Genetic and Medical Aspects

• Osteogenesis Imperfecta
• The Ehlers Danlos Syndrome
• Cutis laxa and premature ageing
• Pseudoxanthoma Elasticum
• The Marfan Syndrome and others
• The homocystinurias
• Menkes disease and others
• Epidermolysis bullosa
• Prolidase Deficiency
• α1-antitrypsin
• Rickets and osteomalacia
• Osteopetrosis
• Alkaptonuria
• FOP
• Disorders of lysosomal enzymes
• Skeletal dysplasias
• Disorders of Keratinization
• Alport Syndrome
• Miscellaneous disorders.........
Genetic Skeletal Diseases

- Extensive clinical and genetic heterogeneity
- Short-limbed dwarfism and osteoarthritis
- Over 450 unique phenotypes
- ~1/4000 incidence
- GSDs are difficult to diagnose
- There are currently no treatments
- Significant burden in pain and disability leads to poor quality of life and high healthcare costs
1. FGFR3 group
2. Type 2 collagen group
3. Type 11 collagen group
4. Sulphation disorders group
5. Perlecan group
6. Filamin group
7. Short-rib dysplasia (SRP) (with or without polydactyly) group
8. Multiple epiphyseal dysplasias and pseudoachondroplasia group
9. Metaphyseal dysplasias
10. Spondylometaphyseal dysplasias (SMD)
11. Spondylo-epi(-meta)physeal dysplasias (SE(M)D)
12. Severe spondylo dysplastic dysplasias
13. Moderate spondylo dysplastic dysplasias (brachyolmias)
14. Acromelic dysplasias
15. Acromesomelic dysplasias
16. Mesomelic and rhizo-mesomelic dysplasias
17. Bent bones dysplasias
18. Slender bone dysplasias
19. Dysplasias with multiple joint dislocations
20. Chondrodysplasia punctata (CDP) group
21. Neonatal osteosclerotic dysplasias
22. Increased bone density group (without modification of bone shape)
23. Increased bone density group with metaphyseal and/or diaphyseal involvement
24. Decreased bone density group
25. Defective mineralization group
26. Lysosomal Storage Diseases with Skeletal Involvement (Dysostosis Multiplex Group)
27. Osteolysis group
28. Disorganized development of skeletal components group
29. Cleidocranial dysplasia group
30. Craniosynostosis syndromes and other cranial ossification disorders
31. Dysostoses with predominant craniofacial involvement
32. Dysostoses with predominant vertebral and costal involvement
33. Patellar dysostoses
34. Brachydactylies (with or without extraskeletal manifestations)
35. Limb hypoplasia–reduction defects group
36. Polydactyly–Syndactyly–Triphalangism group
37. Defects in joint formation and synostoses
Classification of genetic skeletal diseases

Nosology: 2010 revision:

- **456 conditions** placed in **40 groups** defined by molecular, biochemical and/or radiographic criteria.

- **316 conditions** are associated with mutations in one or more of **226 different genes**.

  - dominant vs. recessive
  - null vs. haploinsufficiency vs. hypermorphic vs. hypomorphic vs. antimorphic

  - Allelic series vs. Genetic heterogeneity

  - Genetic modifiers of phenotypic severity
Medicines in Development By Disease and Phase

Some medicines are listed in more than one category.

- Autoimmune Disorders: 18
- Blood Disorders: 7
- Cancer: 105
- Cancer, Blood: 65
- Cancer-Related: 10
- Cancer, Skin: 15
- Cardiovascular Diseases: 4
- Digestive Disorders: 14
- Eye Disorders: 16
- Genetic Disorders: 85
- Growth Disorders: 7
- Infectious Diseases: 28
- Neurological Disorders: 32
- Respiratory Disorders: 20
- Skin Conditions: 2
- Transplantation: 14
- Other: 35

Legend:
- Application Submitted
- Phase III
- Phase II
- Phase I
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<tr>
<th>Product Name</th>
<th>Sponsor</th>
<th>Official FDA Designation</th>
<th>Development Status</th>
<th>Website</th>
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<tr>
<td>BMN-111</td>
<td>BioMarin Pharmaceutical</td>
<td>treatment of achondroplasia</td>
<td>Phase I</td>
<td><a href="http://www.bmrn.com">www.bmrn.com</a></td>
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<td>(modified recombinant human C-type natriuretic peptide)</td>
<td>San Rafael, CA</td>
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<td>macimorelin acetate (AEZS-130)</td>
<td>AEterna Zentaris</td>
<td>diagnosis of growth hormone deficiency</td>
<td>Phase III</td>
<td><a href="http://www.aezsinc.com">www.aezsinc.com</a></td>
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<td>MOD-4023 (hGH-CTP)</td>
<td>PROLOR Biotech</td>
<td>treatment of growth hormone deficiency</td>
<td>Phase III</td>
<td><a href="http://www.prolor-biotech.com">www.prolor-biotech.com</a></td>
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<td>octreotide (oral)</td>
<td>Chiasma</td>
<td>for the oral treatment of acromegaly</td>
<td>Phase I</td>
<td><a href="http://www.chiasmapharma.com">www.chiasmapharma.com</a></td>
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<td></td>
<td>Jerusalem, Israel</td>
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<td>Signifor® LAR pasireotide</td>
<td>Novartis Pharmaceuticals</td>
<td>treatment of acromegaly</td>
<td>Phase III</td>
<td><a href="http://www.novartis.com">www.novartis.com</a></td>
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<td>East Hanover, NJ</td>
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<td>testosterone undecanoate (oral)</td>
<td>SOV Therapeutics</td>
<td>treatment of constitutional delay in growth and puberty in adolescent boys (14-17 yrs of age)</td>
<td>Phase II</td>
<td><a href="http://www.sovtherapeutics.com">www.sovtherapeutics.com</a></td>
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<td>Morrisville, NC</td>
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Genotype vs. common disease mechanisms
Shared disease mechanisms in genetic skeletal diseases

**EuroGrow 2007-10**

- **Prx1 Nf1**
  - Decreased rate of VEGF-mediated vascular invasion leading to increased hypertrophic zone
  - ER stress & UPR classical and novel pathways
  - Increased and/or spatially dysregulated apoptosis
  - Numerous pathways affected

- **Matn3**
  - Delay in hypertrophy and/or reduction in hypertrophic zone
  - Stabilisation of HIF complex and strong up regulation of VEGF leading to reduced hypertrophic zone*

- **Col10a1**
  - Changes in expression of Type II collagen
  - Aggrecan
  - Mmp9
  - Mmp13

- **Dtd**
  - Disrupted Ihh signalling
  - Numerous pathways affected

- **Evc**
  - Role in primary cilia

- **Fgfr3**
  - Numerous pathways affected

- **Col10-Vhl**
  - Stabilisation of HIF complex and strong up regulation of VEGF leading to reduced hypertrophic zone*

* Denotes mutations.
SYBIL comprises 18 Partners with a budget of 12 million Euros over 5 years.

Systems biology for the functional validation of genetic determinants of skeletal diseases
Functional validation of genetic determinants of skeletal diseases and the age related factors contributing to these conditions.

Develop and validate a portfolio of disease models:-
• >200 cellular models
• 25 novel zebrafish models
• 20 novel mouse models

Large-scale phenotyping of cellular and animal models using state-of-the-art - Omics technologies.

‘-Omics Knowledge Factory’ to generate the relevant Omics profiles essential for:
• defining disease pathways
• identifying and validating new biomarkers
• discovering potential therapeutic targets

Systems Biology to integrate the extensive high-throughput and data-dense information that will be generated by the ‘Omics Knowledge Factory’.
Disruptions to ER homeostasis can occur due to:

- viral infection
- chronic inflammation
- hypoxia
- accumulation of misfolded mutant proteins

And induces a stress response called **ER-stress**.
Chronic ER-stress leads to cell death and is associated with an ever-growing list of diseases such as:-

- neurodegenerative
- Cerebro-vascular
- musculoskeletal
- cardiac traits

In this regard, these clinically diverse diseases are unified by the ER-stress

This renders ER-stress an attractive target for novel therapeutic approaches in a broad range of diseases
ER stress as a common mechanism with therapeutic potential
Chondrocytes have an irregular shape and are unable to align correctly.
Candidate pathway; the unfolded protein response (UPR)

Cell protection

Apoptosis

Trends in Cell Biology
Chemical chaperone treatment reduces intracellular accumulation of mutant collagen IV and ameliorates the cellular phenotype of a COL4A2 mutation that causes haemorrhagic stroke

Lydia S. Murray1, Yinhui Lu2, Aislynn Taggart1, Nicole Van Regemorter3, Catheline Vilain3, Marc Abramowicz2, Karl E. Kadler2 and Tom Van Agtmael1,3

1Institute of Cardiovascular and Medical Sciences, College of Medical, Veterinary and Life Sciences, University of Glasgow, Glasgow G12 8QQ, UK 2Wellcome Trust Centre for Cell-Matrix Research, Faculty of Life Sciences, University of Manchester, Manchester M13 9PT, UK 3Department of Medical Genetics, Hopital Erasme – Université Libre de Bruxelles, Brussels, Belgium

Endoplasmic Reticulum Stress As a Therapeutic Target in Cardiovascular Disease

Tetsuo Minamino, Issai Komuro, Masafumi Kitakaze

An unfolded protein response is the initial cellular response to the expression of mutant matrilin-3 in a mouse model of multiple epiphyseal dysplasia

Seema Nandall1, M. Helen Rajpar1, Peter A. Bell, Christopher Clowes2, Leo A. H. Zeef, Benjamin Gardner, David J. Thornton, Raymond P. Boot-Handford, Michael D. Briggs

Pathogenic uromodulin mutations result in premature intracellular polymerization

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Epidermolysis bullosa simplex: a paradigm for disorders of tissue fragility

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The co-chaperone and reductase ERdj5 facilitates rod opsin biogenesis and quality control

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Received June 11, 2014; Revised and Accepted July 18, 2014

Review series
Repurposing of drugs for rare connective tissue diseases

Model systems

Disease mechanisms

Readouts

misfolding – secretion – cell proliferation
Chemical chaperone treatment reduces intracellular accumulation of mutant collagen IV and ameliorates the cellular phenotype of a COL4A2 mutation that causes haemorrhagic stroke

Lydia S. Murray¹, Yinhui Lu², Aislynn Taggart¹, Nicole Van Regemorter³, Catheline Vilain³, Marc Abramowicz³, Karl E. Kadler² and Tom Van Agtmaal¹,*

Reduced protein retention

![Cellular phenotype imaging](image)

Reduced expression of ER stress genes
An unfolded protein response is the initial cellular response to the expression of mutant matrilin-3 in a mouse model of multiple epiphyseal dysplasia

Seema Nundlall • M. Helen Rajpar • Peter A. Bell • Christopher Clowes • Leo A. H. Zeeff • Benjamin Gardner • David J. Thornton • Raymond P. Boot-Handford • Michael D. Briggs
• Urea cycle disorders (neonatal-onset deficiency)
• Cystic fibrosis
• Cancers
• Dementia
• Spinal Muscular Atrophy – FDA orphan drug status
Despite sharing similar disease mechanisms small molecules can still be genotype-specific.

Assay of ‘ER stress’ in a cell culture model.
Statin treatment rescues FGFR3 skeletal dysplasia phenotypes

Akihiro Yamashita¹, Miho Morioka¹, Hiromi Kishi¹, Takeshi Kimura¹,², Yasuhito Yahara¹, Minoru Okada¹, Kaori Fujita¹, Hideaki Sawai³, Shiro Ikegawa⁴ & Noriyuki Tsumaki¹,⁵

\( Fgfr3^{Ac} \)

Vehicle  Rosuvastatin
“The identification and validation of new biomarkers for genetic skeletal diseases will aid in disease prognosis and patient stratification”
Genes and mutations

Disease mechanisms

Cell and pre-clinical mouse models

High-throughput screening

Corrector molecules

Biomarkers
Kasia Pirog
Peter Bell
Beth Gibson
Ella Dennis
Rob Jackson
Sarah Edwards

Ray Boot-Handford
(Manchester)

Michael Wright
(Northern Genetic Service)