Analysis of the LEMD3 gene in individuals affected with melorheostosis

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Genome search in three families
Clinical Report

Melorheostosis in a Family With Autosomal Dominant Osteopoikilosis: Report of a Third Family

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Fig. 3. Radiological appearance of the left foot of the proband at the age of 5 years. a: Typical sclerotic areas can be seen in the first metatarsal head, the proximal phalanx of the hallux, the fifth metatarsal, the phalanges of the fifth toe, and the base of the fourth metatarsal. There is shortening of the fifth ray. b: In the calcaneum, there is also a dense sclerotic lesion (arrow).

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Family with melorheostosis
Osteopoikilosis

- Benign condition
- Autosomal dominant
- Hyperostotic spots
- Isolated or in association with other skin/bone lesions
Buschke-Ollendorff syndrome

BOS = osteopoikilosis + connective tissue nevi (elastic type)

Widely disseminated, multiple, skin-colored to yellow, small papules (few mm in diameter)

Localized, asymmetrically distributed, larger lesions (yellow plaques)

Light micrograph – Van Gieson – x100

elastic-type nevus
Melorheostosis

- joint contractures
- curving or shortening of limb(s)
- chronic pain, swelling of joints
- skin, subcutaneous tissue or muscle involvement
- irregular linear areas of increased radiodensity along the major axis of the tubular bones
- areas of osteophytic periosteal excrescences (dripping candle wax)
- ectopic bone formation
Loss-of-function mutations in \textit{LEMD3} result in osteopoikilosis, Buschke-Ollendorff syndrome and melorheostosis

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LEMD3: integral protein of the inner nuclear membrane

Adapted from Gruenbaum Y et al. Nature Rev Mol Cell Biol 6,21,2005
LEMD3 function

Modulation of the pathway
- Antagonists
- Pseudoreceptors
- Scaffold/anchoring proteins
- Trafficking/degradation proteins
- Oligomerization (I-Smads)

Nuclear membrane proteins

Transcriptional regulators

LEMD3 mutations

Smad interacting part

▲ Nonsense mutation  ♦ Frameshift mutation  ♦ Splice site mutation with frameshift
Analysis of LEMD3 in a larger series of patients

- group A: patients with osteopoikilosis, short stature and learning problems
  
n=3

- group B: patients with osteopoikilosis with(out) BOS skin lesions
  
n=15

- group C: patients with melorheostosis belonging to a family with OP/BOS
  
n=5

- group D: patients with melorheostosis (sporadic occurrence)
  
n=23
## LEMD3 analysis

<table>
<thead>
<tr>
<th>group</th>
<th>phenotype</th>
<th>nucleotide and residue changes</th>
<th>reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>osteopoikilosis, melorheostosis, short stature, MR</td>
<td>Not tested</td>
<td>Jurenka and Van Allen 1995</td>
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<td></td>
<td>osteopoikilosis, short stature, MR, ectopic kidney</td>
<td>microdeletion</td>
<td>Hellemans et al. 2004</td>
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<tr>
<td></td>
<td>osteopoikilosis, short stature, MR</td>
<td>microdeletion</td>
<td>Hellemans et al. 2006</td>
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<td>osteopoikilosis</td>
<td>c.2134dupT; p.Met712fsX</td>
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<td>Hellemans et al. 2006</td>
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<td>c.1914dupA; p.Leu638fsX</td>
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<td>Hellemans et al. 2006</td>
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<td>Hellemans et al. 2006</td>
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<td>c.1813delA; p.Ile605fsX</td>
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<td>osteopoikilosis</td>
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<td>Hellemans et al. 2006</td>
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<td>B</td>
<td>BOS</td>
<td>c.1609C&gt;T; p.Arg537X</td>
<td>Hellemans et al. 2004, Debeer et al. 2003</td>
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<td>BOS</td>
<td>c.830dupA; p.Lys277X</td>
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<td>osteopoikilosis</td>
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<tr>
<td>C</td>
<td>melorheostosis (BOS in other relatives)</td>
<td>c.1913T&gt;A; p.Leu638X</td>
<td>Hellemans et al. 2006</td>
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<td>melorheostosis (BOS in other relatives)</td>
<td>normal</td>
<td>Hellemans et al. 2006 + unpublished</td>
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</table>

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Group C – familial melorheostosis

p.Val759fsX
Group D – sporadic melorheostosis

p.Leu638fsX
normal LEMD3 gene
Germline LEMD3 mutations

Fertilization

MUTATION
Germline LEMD3 mutations

- Green: normal LEMD3 gene
- Red: abnormal LEMD3 gene
Germline LEMD3 mutations

Other genes mutated?

normal LEMD3 gene  abnormal LEMD3 gene
Somatic mutations

- Somatic *LEMD3* mutations responsible for the spotty, localized character of the lesions?
  - skin biopsy from:
    - elastic type nevus (BOS patient)
    - hard sclerodermic-like lesion (melorheostosis patient in BOS family)
- No LOH or allelic imbalance
- No second hit in *LEMD3*
- Only cDNA of wild type *LEMD3*
Sporadic melorheostosis

- normal LEMD3 gene
Sporadic melorheostosis

Other genes mutated?

normal LEMD3 gene  abnormal LEMD3 gene
Somatic mutations

- Somatic *LEMD3* mutations in melorheostosis patients without an identifiable germline mutation?
  - biopsies from skin and bone of 2 patients
- No somatic LOH or mutation in *LEMD3*
Conclusions (1)

- Heterozygous loss-of-function mutations in LEMD3 result in osteopoikilosis and Buschke-Ollendorff syndrome
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- Individuals with melorheostosis and osteopoikilosis (usually belonging to a family) have heterozygous LEMD3 mutations in the germline.
Conclusions (1)

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- Individuals with melorheostosis (sporadic and isolated) do not have germline mutations in LEMD3
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- Somatic defects in LEMD3 have not yet been identified in melorheostosis lesions.
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- Somatic defects in LEMD3 have not yet been identified in melorheostosis lesions.

- The cause of sporadic, isolated melorheostosis remains to be unravelled.
Microdeletions encompassing the LEMD3 gene result in a syndromic condition with osteopoikilosis, mental retardation and short stature

LEMD3
short stature gene
mental retardation gene
Heterozygous knock-out mouse: development of melorheostosis?
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