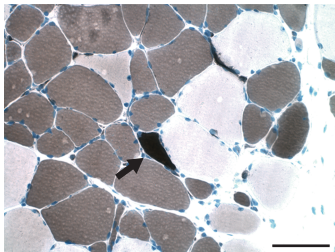
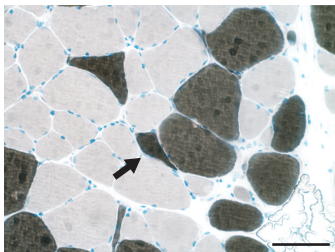


Supplemental Figure 1

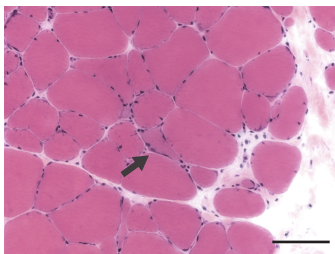
ATPase
pH4.3



ATPase
pH9.4



H&E



Supplemental Figure 2

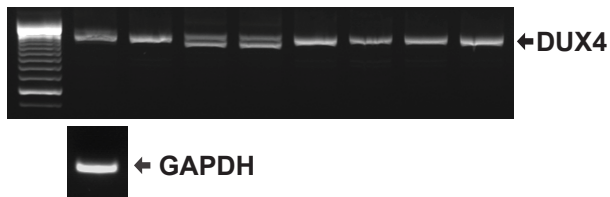
SETMAR

Patient PAPFQYTPDHVVGPG**T**DIDPTQITFFGCICV
Homo sapiens PAPFQYTPDHVVGPG**A**DIDPTQITFFGCICV
Mus musculus PKPFQYTPDHVAGPG**A**DIDPTQITFFGCACI
Canis lupus familiaris PEPFQYTPDHVAGPG**T**DVDPTQITFFGCICL
Anolis carolinensis APAFQYSPDHVAGKE**G**KPDPSEISFFPGCSCH
Xenopus tropicalis LPAFQYTPELIAGPG**A**EQDPSEVTIQGCDRC
Danio rerio LSYFQYVPENVQGPG**C**DLDPNAVTLPGCSCR

FAM105B

Patient YNTEEFITVYPTDPP**E**DWPVVTLIAEDDRHY
Homo sapiens YNTEEFITVYPTDPP**K**DWPVVTLIAEDDRHY
Mus musculus YNTEEFITVYPTDPP**K**DWPMVTLIAEDDRHY
Canis lupus familiaris YSTEEFITVYPTDPP**M**DWPVVTLIAEDDRHY
Anolis carolinensis YSTDEFIAFYPNPE**E**HWPVVTLITEDDRHY
Xenopus tropicalis CGTDEFITYYPND-**K**TNWPTVTLITEDDRHY
Danio rerio TDTEEFVTHYPDDHK**H**EWPCVCIVTEDDRHY

Supplemental Figure 3



Supplemental figure 1

H&E and ATPase staining of matching sections of biopsied muscle from affected subject II-5. Some small angular fibers (arrow) are observed in the area with less endomysial fibrosis. Some of the small angular fibers are type 2C. Scale bar = 100 μ m.

Supplemental figure 2

Two of three variants that co-segregate with the phenotype are in the *SETMAR* and *FAM105B* genes. Amino acids p.Ala63 in *SETMAR* and p.Lys325 in *FAM105B* are not well conserved, suggesting that these are rare but benign polymorphisms.

Supplemental figure 3

DUX4 expression of biopsied muscle from subject II-5. Full length *DUX4* expression was detected by RT-PCR in all 8 replicates from a single biopsy sample. Eight replicates were run because the *DUX4* signal may vary between PCR reactions due to its very low expression levels. PCR for *GAPDH* is used as a control for mRNA integrity and cDNA synthesis. The PCR in the lanes 3 and 4 suggests the presence of a rare short isoform as well as the long isoform [1].

Supplemental Table 1

		chromosome	Affected			Unaffected			
			III-11	II-5	III-10	II-6	II-2	II-1	III-8
PRDM2	NM_001007257:c.A2440G:p.T814A	1		+	+	-	-	-	+
KBTBD10	NM_006063:c.G1735T:p.A579S	2		+	+	-	+	-	-
SCRN3	NM_024583:c.G352A:p.E118K	2		+	+	-	-	+	-
FRZB	NM_001463:c.T272G:p.M91R	2		+	+	-	-	+	-
SETMAR	NM_001243723:c.G187A:p.A63T	3	+	+	+	-	-	-	-
STAB1	NM_015136:c.G2869A:p.G957S	3		+	+	-	-	+	+
DZIP3	NM_014648:c.C3514T:p.Q1172X	3		+	+	-	-	+	-
DIRC2	NM_032839:c.T140G:p.V47G	3	-	+	+	-	-	-	-
CPEB2	NM_001177383:c.C779A:p.P260Q	3		+	+	-	+	-	-
CHST2	NM_004267:c.C364T:p.L122F	3		+	+	-	-	+	-
IDUA	NM_000203:c.C1345A:p.H449N	4		+	+	-	-	-	+
FAM105B	NM_138348:c.A973G:p.K325E	5	+	+	+	-	-	-	-
FOXD1	NM_004472:c.G332A:p.G111D	5		+	+	-	+	-	-
SMCHD1	NM_015295:c.823_825del:p.K275del	18	+	+	+	-	-	-	-
LRRC4B	NM_001080457:c.G1220T:p.G407V	19		+	+	-	-	+	-
BFSP1	NM_001161705:c.G1036C:p.V346L	20		+	+	-	+	-	-
NFS1	NM_021100:c.A299C:p.E100A	20		+	+	-	+	-	-
RLIM	NM_183353:c.T1411C:p.S471P	X	-	+	+	-	-	-	-

Eighteen variants within the linkage peaks were found to co-segregate with the phenotype of II-5, II-6 and III-10. Among these, three variants, in *SETMAR*, *FAM105B* and *SMCHD1*, co-segregated with the phenotype in all available subjects.

Supplemental Table 2

SETMAR p.Ala63Thr

SIFT	0.06	Tolerated
PolyPhen-2	0.523217	Benign

FAM105B p.Lys325Glu

SIFT	0.71	Tolerated
PolyPhen-2	0	Benign

The variants in *SETMAR* and *FAM105B* result in p.Ala63Thr and p.Lys325Glu alterations, respectively. *In silico* analysis using SIFT (<http://sift.jcvi.org/>) and PolyPhen-2 (<http://genetics.bwh.harvard.edu/pph2/>) predicts that these alterations are tolerated, i.e., benign.

Supplemental reference

- [1] Snider L, Geng LN, Lemmers RJ, et al. Facioscapulohumeral dystrophy: incomplete suppression of a retrotransposed gene. *PLoS Genet* 2010;6:e1001181.