

Supplemental Materials

Molecular Biology of the Cell

Barnes et al.

Supplemental materials

Tropomyosin isoforms differentially affect muscle contractility in the head and body regions in the nematode *Caenorhabditis elegans*

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Supplemental Figure S1

Supplemental Tables S1-S3

CAAGATCGT	CGAGCTTGA	AGAGGAGIT	CCGCGTCGT	GGTAA	AACTTGA	AATCCCT	TGAAC	TTCCGAG	<i>C.elegans</i> exon 7a	
CAAATCGT	CGAGCTTGA	AGAGGAGIT	CCGCGTCGT	GGTAA	AACTTGA	AATCTCT	TGAAC	TTCCGAG	<i>C.briggsae</i> exon 7a	
CAAATCGT	CGAGCTTGA	AGAGGAGIT	CCGCGTCGT	CGAAA	AACTTGA	AATCTCT	TGAAC	TTCCGAG	<i>C.remanei</i> exon 7a	
CAAGATCGT	CGAACT	TGAGGAGGAG	CTCCG	TGTCGT	CGAAA	CAACTTGA	AATC	ACTTGAGGT	TTCCGAG	<i>C.elegans</i> exon 7b
CAAGATCGT	CGAGCTTGA	AGAGGAGIT	CCGCGTCGT	CGAAA	CAACTTGA	AATCCCT	TGAGGT	TTCCGAG	<i>C.briggsae</i> exon 7b	
CAAGATCGT	CGAGCTTGA	AGAGGAGIT	CCGCGTCGT	CGAAA	CAACTTGA	AATCTCT	TGAAG	TTCCGAG	<i>C.remanei</i> exon 7b	

GA	AAAGGCACT	TGA	BAAGGAGGAC	ATCT	TTGCC	GAGCAGAT	TCGT	CAG	TC	GATT	T	CAGACT	GAAGGAG	<i>C.elegans</i> exon 7a
GAGA	AAAGGCACT	TGA	BAAGGAGGAC	ATCT	TCGCT	GAGCAGAT	CCGT	CAG	TC	GATT	T	CAGACT	GAAGGAG	<i>C.briggsae</i> exon 7a
GA	AAAGGCACT	TGA	BAAGGAGGAT	TATCT	TTGCT	GAGCAGAT	TCGT	CAG	TC	GATT	T	CAGACT	GAAGGAG	<i>C.remanei</i> exon 7a
GAGA	AAAGGCACT	TGA	BAAGGAGGAC	ATCT	TCGCT	GAGCAGAT	CCGT	CAG	TC	GATT	T	CAGACT	GAAGGAG	<i>C.elegans</i> exon 7b
GAGA	AAAGGCACT	TGA	BAAGGAGGAC	ATCT	TCGCT	GAGCAGAT	CCGT	CAG	TC	GATT	T	CAGACT	GAAGGAG	<i>C.briggsae</i> exon 7b
GAGA	AAAGGCACT	TGA	BAAGGAGGAC	ATCT	TCGCT	GAGCAGAT	CCGT	CAG	TC	GATT	T	CAGACT	GAAGGAG	<i>C.remanei</i> exon 7b

Supplemental Fig. S1. Nucleotide sequence alignment of *lev-11* exon 7s (E7a and E7b) from *C. elegans*, *C. briggsae*, and *C. remanei*. Conserved residues in more than half of examined sequences are shaded in black.

Supplemental Table S1. Worm strains

Strain	Transgene	Genotype
KH2372	E7a-mCherry E7b-EGFP	<i>ybEx2372 [eef-1A.1::LEV-11E6E7aE8-GGS6-mCherry eef-1A.1::LEV-11E6E7bE8-GGS6-EGFP]</i>
KH2386	E7a-mCherry E7b-EGFP	<i>ybIs2381 [myo-3::LEV-11E6E7aE8-GGS6-mCherry myo-3::LEV-11E6E7bE8-GGS6-EGFP pBSKII(-)] X</i>
ON269	GFP-LEV-11A	<i>ktEx187 [myo-3::GFP::LEV-11A cDNA]</i>
ON335	GFP-LEV-11O	<i>ktEx238 [myo-3::GFP::LEV-11O cDNA]</i>
ZZ12	-	<i>lev-11(x12) I</i>
VC20516	-	A whole genome-sequenced strain containing <i>lev-11(gk334531) I</i> from the Million Mutation Project
ON315	-	<i>lev-11(gk334531) I</i> (outcrossed 6 times)

Supplemental Table S2. Sequences of the primers used to detect endogenous LEV-11 mRNAs in the RT-PCR assays and molecular cloning.

Position	Direction	Sequence
Exon 1 ¹	Forward	5'- <u>CACCATGGACGCCATTAAGAAGAAGATG</u> -3'
Exon 1 ²	Forward	5'- <u>AGGAGATATA</u> <u>CCATGG</u> CTAG <u>CATGGACGCCATTAAGAAGAAGATG</u> - 3'
Exon 3b ¹	Forward	5'- <u>CACCATGT</u> CGAAGGTAAACAAGGAGG-3'
Exon 7a	Forward	5'-CAGATTCGTCAGCTCGATTTC-3'
Exon 7a	Reverse	5'-GCAAAGATGTCCTCCTTCTCA-3'
Exon 7b	Forward	5'-CAACGTGAGGACTCGTACGAG-3'
Exon 7b	Reverse	5'-CAGTCTGGATGAGACGGTG-3'
Exon 9c	Reverse	5'-TTAATATCCGGAGAGCTCTTGGAAG-3'
Exon 9c ²	Reverse	5'- <u>GTTAGCAGCCGGATCT</u> TAATATCCGGAGAGCTCTTGGAAG-3'

¹Underlines indicate CACC sequences for directional TOPO cloning.

²Dashed underlines indicate overlapping sequences with pET-3d for In-fusion cloning. Bold characters indicate extra codons for N-terminal Ala-Ser extension.

Supplemental Table S3. Sequences of primers used in plasmid construction.

Primers used to amplify genomic fragments.

Amplified fragments	Sequence
<i>lev-11</i> exon 6 – exon 8	5' - <u>AAAAAGCAGGCT</u> <u>cca</u> CCATGGTTGAAGCTGATCT-3' 5' -TATACAAAGTTGCTTCGAGTCTGTCGACCTCCT-3'

Underlines indicate parts of *attB* sequences for BP cloning or *Xba* I recognition site (exon 6 Reverse). Lower case indicates nucleotides for introducing Kozak's consensus.

Primers used to construct GFP-LEV-11 expression vectors.

LEV-11A or LEV-11O cDNA	5' -ACAAGCTTTGGAATTCATGGACGCCATTAAGAAGAAGATG-3' 5' -TGTATGGCCGGCTAGCGGATGTTTAATATCCGGAGAGCTC-3'
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Underlines indicate overlapping sequences with pPD118.20 for In-fusion cloning.

Primers used for mutagenesis.

Mutation	Sequence
<i>lev-11</i> exon 7a – termination codon	5' -GATTCGTCAGCTCtAaTTCAGACT-3' 5' -AGTCTGAAtTaGAGCTGACGAATC-3'
<i>lev-11</i> exon 7b – termination codon	5' -CGCACCGTCTaATCCAGAC-3' 5' -GTCTGGATtAGACGGTGCG-3'
LEV-11A(E234K) protein	5' - CGTCTCATCCAGACTGAAGaAGGCTGAGACCCGTGCCGAATTCG-3' 5' -
LEV-11O(E196K) protein	CGAATTCGGCACGGGTCTCAGCCTtCTTCAGTCTGGATGAGACG-3' 5' -CAAGATCGTGGAGCTTGAAGAGaAGTTGCGCGTCGTTGG-3' 5' -CCAACGACGCGCAACTtCTCTTCAAGCTCCACGATCTTG-3'

Lower case indicates nucleotides for disrupting potential splice sites (exon 4b), introducing termination codons (exons 5 and 7), disrupting a termination codon (exon 9b), introducing missense mutations in LEV-11A(E234K) and LEV-11O(E196K).
