

**Supplementary Table 1.** sgRNA and oligonucleotide sequences.

Purpose	Species	Gene	Sequence 5' → 3'	Amplicon (bp)	Application
CRISPR sgRNA	Human	MymK	sgRNA1: CTCACAGCTACAGAAGATGA	N/A	Fig. 1d
			sgRNA2: AAAGAAGAAGCGTAGCATCA		
	Human	MymX	sgRNA1: GGCTCCCAGGACATGCGAG		Fig. 4a
			sgRNA2: ACCTCTCCCTCCTCTCCAGG		
	Mouse	MymK	sgRNA1: TAGCGATGCTCACTGTGCGGG		Extended Data Fig. 5a
			sgRNA2: GCGTCCTTACCATCGCTGTG		
			sgRNA3: AGACAAACCAGGCCCATCAC		
	Mouse	MymX	sgRNA1: GCTGCTGCCTGTTGCCCGCC		Extended Data Fig. 11a
			sgRNA2: GAGGCCTCTCCAGAATCCGG		
			sgRNA3: CCTCTGGGAGTGGTCCACTC		
	Lizard	MymK	sgRNA1: CACGCCAAACATCACCAAGG		Extended Data Fig. 5f
			sgRNA2: TTTGACGGTGATCATGAGGA		
<i>Ciona</i>	MymK	sgRNA2.6: gCAATGGAGTTGTGCAGAGG	Fig. 2h		
		sgRNA4.68: gCAGCTCCAAGTGCAAACGA			
Genotype Analysis	Human	MymK	F: CTTCCTTCCCAGCCATCCAG R: GGGCTAGTGAGCAGGGACTA	492	Fig. 1d
	Human	MymX	F: AACTGAAGGGAGGGGAACT R: TGGAGGACAGAGGGCAATA	599	Fig. 4a
	Mouse	MymK	F: GCCTTTACCACCTTCTCCCC R: CCCACCTCACACCTTCCTTC	6,278	Extended Data Fig. 5a
	Mouse	MymX	F: AGTTCAGGCTTCAGGTCAGAG R: GCTAGGGGAGTGGGAACTGT	743	Extended Data Fig. 11a
	Lizard	MymK	F: CTTCATGTATGTTGGAAGGAGGCT R: GTTGAAGTGTAGACGCTTTTCTCTG	395	Extended Data Fig. 5f, g
	Lamprey	MymX	F: ATTGGAGGCACAGATGGTGG R: GGGGTTTCACGACAAGCAAC	498	Fig. 3e
	<i>Ciona</i>	MymK	F: CGCGATCACAAATGACGAAAC R: CCCGCAATTACAACATGCTAG	1400	Extended Data Fig. 8a
RT-PCR	Lamprey	18s	F:CGCTTTGGTGACTCTGGATAA R: GCCTGCCATCGTAAGTTGATA	100	Fig. 4e, f
	Lamprey	Myh1	F: GTGAAGCAGAAGCTGGAGAA R: ACGAGTTTCACCTTGGACTTT	100	Fig. 4f
	Lamprey	MymX	F: TTCTCTCCTGCGTGCTGTG R: GCGTTCTTGTTCATCGCCATC	106	Fig. 4e, f
	Lamprey	MymK	F: AGAATTCGAGGAGCCGCAG R: AGGAACGCCCTCAGTTGGAT	174	Fig. 6c
	Lamprey	MymK	F1: CGGGGTGCGTACATAGACAG	275	Extended

Analyze mRNA isoforms	Lamprey	MymK	F2: GCACGTGGTTCCTCACCAA	130	Data Fig. 15
	Lamprey	MymK	F3: GAGTGAGCGAACTGAAACACG	187	
	Lamprey	MymK	F4: TGGTCTGCGTGGCACG	131	
	Lamprey	MymK	R: TAGACCATGGCCTCGATGTG	N/A	
	Lamprey	MymK	F: AGTTAATTAAGGATCCGCCACC <u>ATGGGCTCCATCCTGGTC</u> R: CTGGCGGCCGCTCGAG <u>TCACACGCAGCAGCAGA</u>	701	Fig. 6b