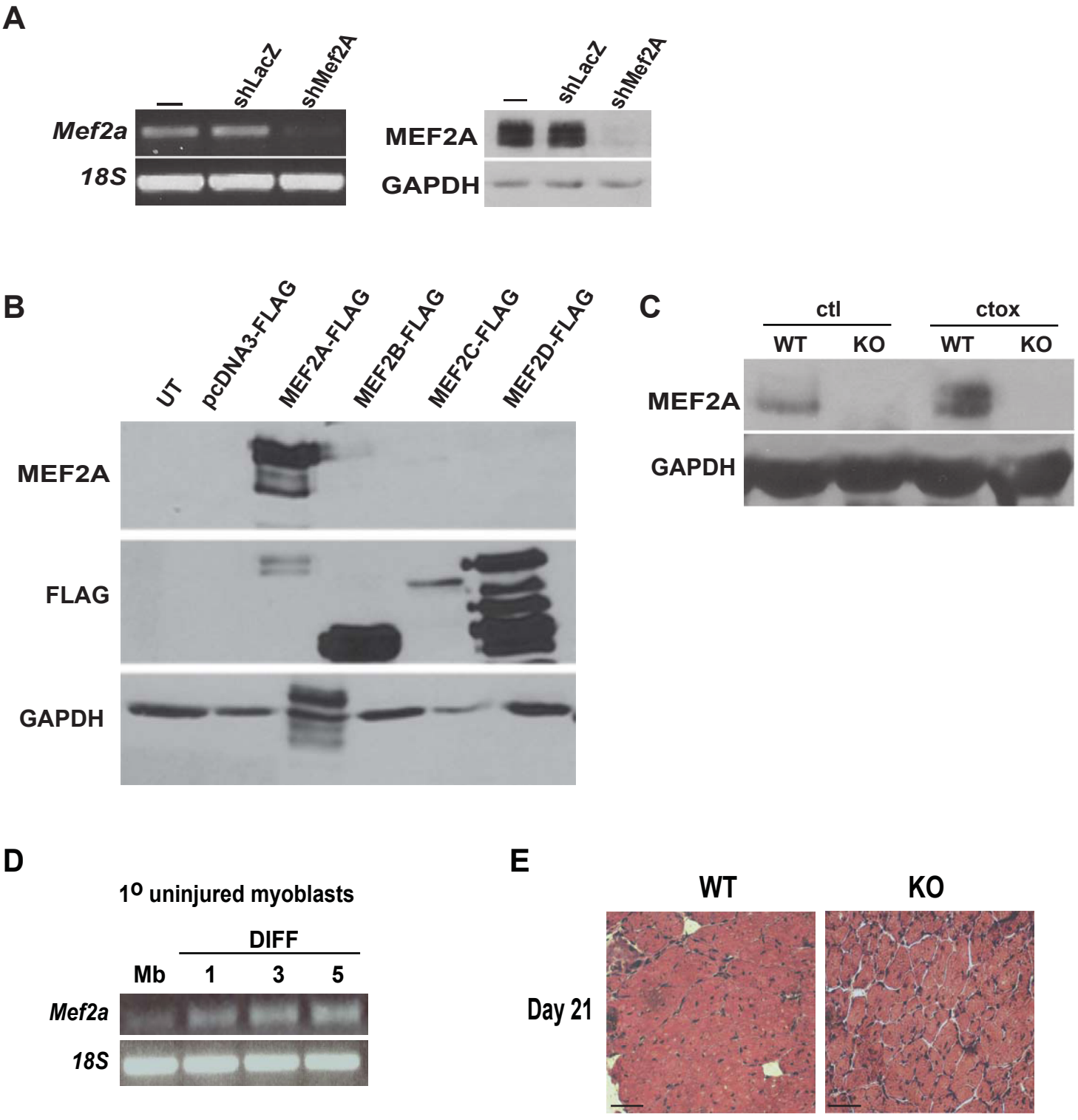

Fig. S1. Antibody and shRNA specificity for MEF2A and MEF2A expression in regeneration. (**A**) RT-PCR (left) and western blot (right) analysis of C2C12 DIFF3, post-transduction with sh*lacZ* or sh*Mef2a*. 18S rRNA and GAPDH are loading controls. (**B**) Immunoblot of MEF2-FLAG expression in COS cells with the MEF2A antibody (C-21, Santa Cruz). GAPDH is the loading control. (**C**) Immunoblot of MEF2A expression in uninjured (ctl) and injured (ctox, day 7) skeletal muscle from WT and Mef2a KO mice. (**D**) RT-PCR analysis of *Mef2a* expression in differentiation using uninjured primary myoblasts. (**E**) Hematoxylin and Eosin staining of transverse TA muscle sections at day 21 post-injury. Data reveals substantially improved myofiber formation in regenerating *Mef2a* KO.

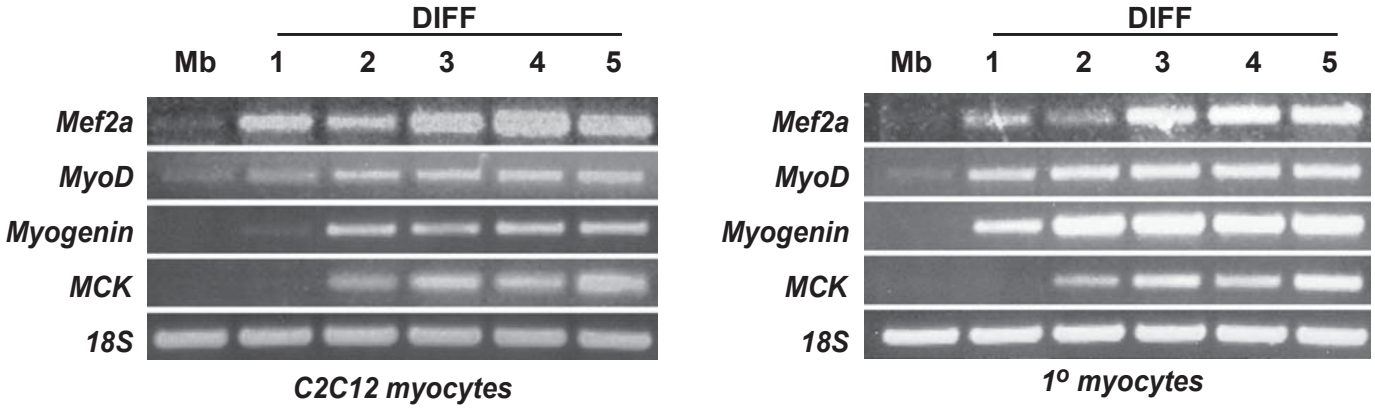

Fig. S2. C2C12 differentiation mimics regenerating primary myocytes. RT-PCR time course of myogenic marker expression during differentiation, myoblasts (Mb) through DIFF 5, in C2C12 cells and primary WT myocytes.

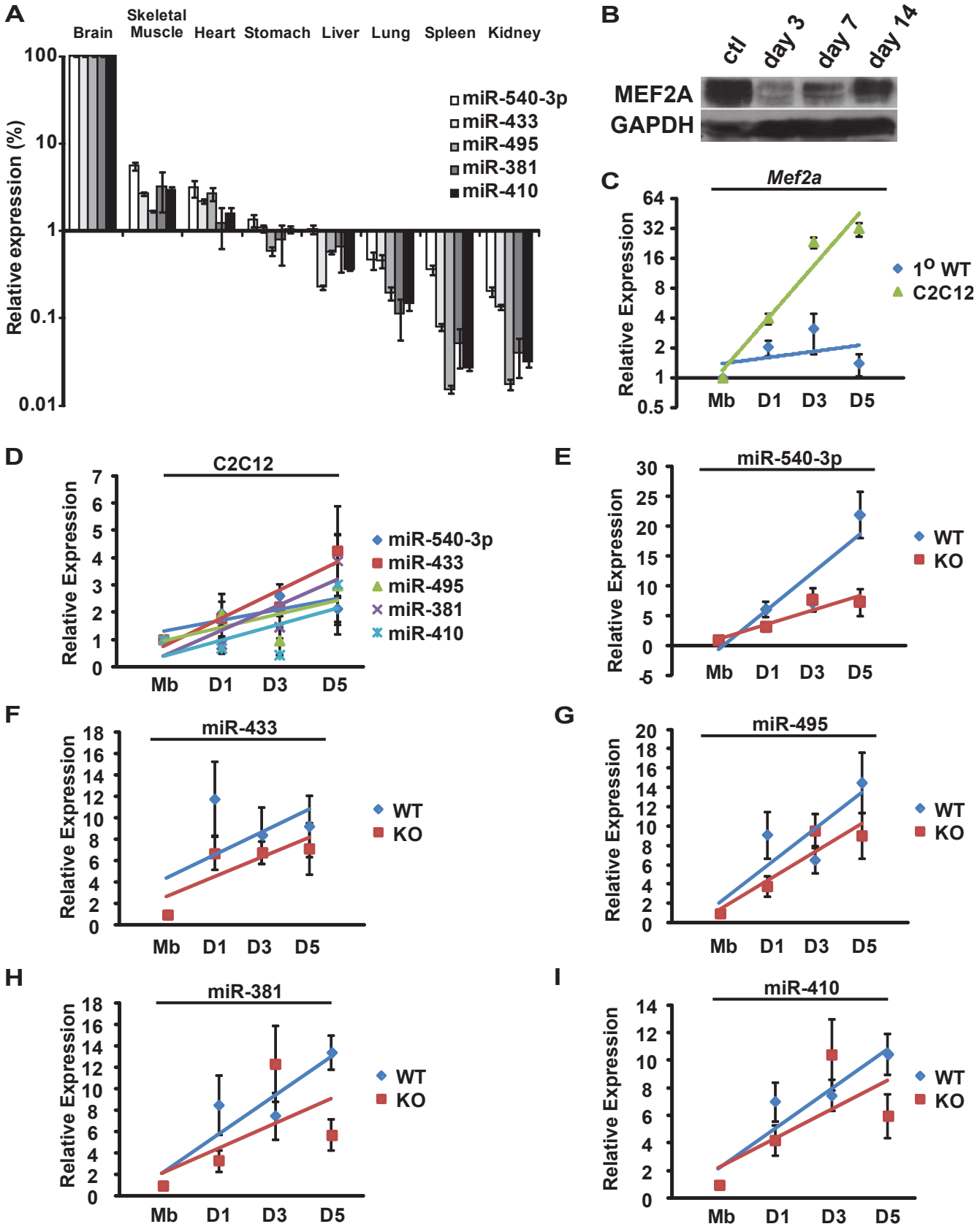

Fig. S3. miR expression patterns mimic MEF2A. (**A**) Expression of selected miRNAs in various tissues are displayed relative to the brain (normalized to 100%) (*n*=3). (**B**) Immunoblot of MEF2A expression in control (ctl) and regenerating muscle (days 3-14). (**C**) Mef2a expression in primary WT myocytes and C2C12 cells during differentiation. (**D**) miR expression in C2C12 cells during differentiation. (**E-I**) Expression of individual miRs in WT and *Mef2a* KO primary myocytes during differentiation. Myoblast (Mb) and DIFF 1-5 (D1-D5) time points are indicated. Changes in expression are displayed as linear trendlines. Error bars represent s.e.m.

RT-PCR

Oligo	Direction	Sequence 5' to 3'
18SrRNA	Forward	CATTCGAACGTCTGCCCTAT
	Reverse	CCTCCAATGGATCCTCGTTA
Mef2a	Forward	ACACGCATAATGGATGAGAGGAACCGAC
	Reverse	CAACGATATCCGAGTTCGTCCTGCTTTC
Mef2b	Forward	GAAAGAAAGCCGCTCTGCACAG
	Reverse	ACCTCTGGCCCCTCCTCCATA
Mef2c	Forward	CAGGGACGAGAGAGAGAAGAAAC
	Reverse	CAATCTTTGCCTGCTGATCATTAG
Mef2d	Forward	CTTTCCTCTCTGGCACTAAGGAC
	Reverse	CCAGTCTATAACTCTGCATCATC
MyoD	Forward	TCGAAGGTCTGCAGGCTCTGC
	Reverse	TGCAGTCGATCTCTCAAAGCACC
Myogenin	Forward	TGGAGCTGTATGAGACATCCC
	Reverse	GAGTTGCATTCACTGGGCAC
MCK	Forward	CTGACCCCTGACCTCTACAAT
	Reverse	CATGGCGGTCCTGGATGAT

qRT-PCR

Oligo	Direction	Sequence 5' to 3'
GAPDH	Forward	TGGCAAAGTGGAGATTGTTGCC
	Reverse	AAGATGGTGATGGGCTTCCCG
Dlk1	Forward	AGTGCGAAACCTGGGTGTC
	Reverse	GCCTCCTTGTTGAAAGTGGTCA
Gtl2	Forward	TGGAATAGGCCAACATCGTCA
	Reverse	AGGCTCTGTGTCCATGTGTCC
pre-miR-127	Forward	TTTGATCACTGTCTCCAGCCTGCTG
	Reverse	GATGATGAGACTTCCGACCAGCCA
sFRP1	Forward	CAACGTGGGCTACAAGAAGAT
	Reverse	GGCCAGTAGAAGCCGAAGAAC
sFRP2	Forward	CCCCTGTCTGTCTCGACGA
	Reverse	CTTCACACACCTTGGGAGCTT
sFRP4	Forward	AGAAGGTCCATACAGTGGGAAG
	Reverse	GTTACTGCGACTGGTGCGA
Axin2	Forward	TGACTCTCCTTCCAGATCCCA
	Reverse	TGCCCACACTAGGCTGACA
5S rRNA stem loop	Forward	GTTGGCTCTGGTGCAGGGTCCGAGGTATTCGCACCAGAGCCAACAAAGCC
miR 540-3p stem loop	Forward	GTTGGCTCTGGTGCAGGGTCCGAGGTATTCGCACCAGAGCCAACACAGAG
miR 433 stem loop	Forward	GTTGGCTCTGGTGCAGGGTCCGAGGTATTCGCACCAGAGCCAACACACCG
miR 495 stem loop	Forward	GTTGGCTCTGGTGCAGGGTCCGAGGTATTCGCACCAGAGCCAACAAGAAG
miR 381 stem loop	Forward	GTTGGCTCTGGTGCAGGGTCCGAGGTATTCGCACCAGAGCCAACACAGAG
miR 410 stem loop	Forward	GTTGGCTCTGGTGCAGGGTCCGAGGTATTCGCACCAGAGCCAACACAGGC
5SrRNA	Forward	GAATACCGGGTGCTGTAGGC
miR 540-3p	Forward	CAAGGGTCACCCTCTGACTCT
miR 433	Forward	ATCATGATGGGCTCCTCGGT
miR 495	Forward	GCCAAACAAACATGGTGCACTT
miR 381	Forward	GCCTATACAAGGGCAAGCTCTC
miR 410	Forward	CCGCCAATATAACACAGATGGCC
ChIP -39 MEF2	Forward	TCTGGGGGGCTCATTTTTCCG
	Reverse	GAGCCGAGGCGAGCTCTATTC

EMSA

Oligo	Direction	Sequence 5' to 3'
-39 MEF2	Sense	AAGGCTGCGCCTTTATATAAACCCCA
	Antisense	AAGGTGGGGTTTATATAAAGGCGCAG
-39 MUT	Sense	AAGGCTGCGCGGGTATATAAACCCCA
	Antisense	AAGGTGGGGTTTATATACCCGCGCAG

Table S1. Oligonucleotide sequences. Oligonucleotides used for RT-PCR/qRT PCR and EMSA analyses. DNA binding sites in EMSA oligonucleotides are underlined.

Gene Symbol	Fold Change
miR-337	-1.7
miR-540	-1.6
miR-665	-1.3
miR-431	-1.2
miR-341	-1.4
miR 370	-1.3
Rian/MBII-343	-2.1
miR-379	-1.7
miR-411	-1.4
miR-299	-1.6
miR-380	-1.4
miR-329	-1.6
miR-494	-1.3
miR-679	-1.6
miR-666	-1.4
miR-543	-1.7
miR-667	-1.4
miR-376c	-1.2
miR-376b	-1.2
miR-376a	-1.1
miR-300	-1.4
miR-381	-1.5
miR-382	-1.4
miR-134	-1.8
miR-668	-1.3
miR-154	-1.6
miR-496	-1.4
miR-377	-1.5
miR-541	1.0
miR-412	-1.7
miR-369	-1.5

Table S2. MicroRNA expression in injured Mef2a KO muscle. Fold changes in gene expression are presented for additional non-coding RNA transcripts in the *Gtl2-Dio3* locus detected by microarray. Expression values represent fold changes in gene expression between adult *Mef2a* KO cardiotoxin-injured (ctox) and WT ctox muscle tissue samples at regeneration day 7. Non-coding transcripts include microRNAs and the Rian/MBII-343 C/D box sno RNAs and are listed from top to bottom according to their 5′ to 3′ order within the *Gtl2-Dio3* locus.