

Table S1. Quantification of effect of MOs, mutants and drug treatments

Figure number and panel	Assay	Genotype/Treatment and quantification of phenotype					
		<i>smo</i> ^{+/-} incross	<i>shha</i> ^{+/-} incross				
Fig. 1A left	MyHC IHC, 10s <i>myod</i> ISH <i>hsp90α</i> ISH, 5s	a14/50 [28%] r16/50 [32%]	r9/41 [22%] r13/50 [26%]				
Fig. 1A right	<i>myf5</i> ISH, 6s <i>myod</i> ISH, 6s	r28/90 [31%] r33/100 [33%]	r22/90 [24%] r27/100 [27%]				
Fig. 1B	<i>myog</i> ISH, 5s	a10/42 [24%]	r14/66 [21%]				
Fig. 1C	Myod IHC, 6s	n56/56 (wt) [100%]	a105/105 [100%] (in adaxial cells)	<i>smo</i> ^{+/-} incross a39/145 [27%] (in adaxial cells)	<i>shha</i> ^{+/-} incross r55/226 [24%] (in adaxial cells)]		
Fig. 2A	<i>myog</i> ISH, 5s	n20/20 [100%]	a46/55 [84%] r9/55 [16%]				
Fig. 2B left	<i>myog</i> ISH, MyHC IHC 5s	n20/20 n[100%]	a46/60 [77%] r14/60 [23%]				
Fig. 2B right	MyHC IHC <i>prdm1</i> ISH	n40/40 [100%]	r31/31 [100%] (MyHC) n31/31 [100%] (<i>prdm1</i>)				
Fig. 2C left	<i>eng2a</i> ISH, 15s slow MyHC IHC	n16/21 [76%] n16/21 [76%]	n22/28 [79%] n22/28 [79%]	<i>sib</i> + <i>myf5</i> MO n22/30 [73%] n22/30 [73%]	<i>shha</i> ^{-/-} a5/21 [24%] r5/21 [24%]	<i>shha</i> ^{-/-} + <i>myf5</i> MO a6/28 [21%] r6/28 [21%]	<i>shha</i> ^{-/-} + <i>myod</i> MO a8/30 [27%] a8/30 [27%]
Fig. 2C top right	slow MyHC IHC, 15s	n47/60 [78%]	n36/49 [74%]	<i>shha</i> ^{-/-} r13/60 [22%]	<i>shha</i> ^{-/-} + <i>myog</i> MO a6/49 [12%] w7/49 [14%]		
Fig. 2C bottom right	<i>myod</i> ISH, 10s <i>mrf4</i> ISH, 10s	n41/57 [72%] n41/57 [72%]			r16/57 [28%] a16/57 [28%]		
Fig. 3A left	<i>cdkn1c</i> , ISH, 5s	n30/30 [100%]	r14/50 [28%]	<i>cyA</i> a50/50 [100%]			
Fig. 3A middle	<i>cdkn1c</i> ISH, 11s	n30/30 [100%]	a46/46 [100%]	<i>con</i> (wt) ur0/43 [0%]	<i>shh</i> mRNA ur33/37 [89%]		
Fig. 3A right	<i>cdkn1c</i> ISH, 24 hpf	n37/50 [74%]	a13/50 [26%]	<i>sib</i> <i>shha</i> ^{-/-}	<i>sib</i> + <i>cdkn1c</i> MO <i>shha</i> ^{-/-} + <i>cdkn1c</i> MO		
Fig. 3B	<i>myog</i> , ISH, 5s	n26/37 [70%]	r11/37 [30%]	<i>con</i> (<i>shha</i> ^{-/-}) r12/22 [55%] n2/22 [9%]	<i>shha</i> ^{-/-} + <i>cdkn1c</i> MO a8/22 [36%]		
Fig. 3C top	Myod IHC, 8s	r0/86 [0%]	r93/106 [88%]				
Fig. 3C bottom	MyHC IHC 15s	r0/15 [0%]	r10/17 [59%]				
Fig. 3D	Myod, IHC, TB	r0/50 [100%]	r55/62 [89%]				
Fig. 4A	<i>cdkn1c</i> ISH, 12s	n50/50 [100%]	r14/60 [23%] wr46/60 [77%]	<i>con</i> (<i>myf5</i> ^{hu2022/+} incross) <i>myf5</i> ^{hu2022/+} incross + <i>myod</i> MO			
Fig. 4B	<i>cdkn1c</i> ISH, 15s	n30/30 [100%]	r41/42 [98%] n1/42 [2%]	<i>con</i> (wt) <i>myf5</i> MO	<i>myf5</i> ^{hu2022/+} incross n50/50 [100%]		

Fig. 4C	<i>cdkn1c</i> ISH, 24 hpf	con (wt) n30/30 [100%]	<i>myf5</i> MO + <i>myod</i> MO r59/60 [98%] n1/60 [2%]						
Fig. 4D	<i>cdkn1c</i> ISH, 15s	con (wt) n26/26 [100%]	wt + <i>shh</i> RNA e22/23 [96%] n1/23 [4%]	<i>myod</i> MO	<i>myod</i> MO + <i>shh</i> RNA e41/43 [95%] n2/43 [5%]	<i>myf5</i> MO + <i>myod</i> MO r36/38 [95%] n2/38 [5%]	<i>myf5</i> MO + <i>myod</i> MO + <i>shh</i> RNA er51/63 [81%] e2/63 [3%] r7/63 [11%] n3/63 [5%]		
Fig. 4E	<i>smyhc1</i> ISH, 24 hpf	n48/48 [100%]	ur37/43 [86%] n6/43 [14%]	wr13/13 [100%]	ur11/11 [100%]	r54/59 [91%] wr1/59 [2%] n4/59 [7%]	r44/52 [85%] ur6/52 [11%] n2/52 [4%]		
Fig. 5A	slow MyHC IHC, 24hpf	con (<i>smo</i> ^{-/-}) 171/12	<i>smo</i> ^{-/-} + <i>cdkn1</i> DNA	con (wt + <i>cyA</i>)	<i>cyA</i> + <i>myod</i> MO	<i>cyA</i> + <i>cdkn1</i> DNA	<i>cyA</i> + <i>myod</i> MO + <i>cdkn1</i> DNA	<i>smo</i> ^{-/-} + <i>cdkn1</i> MO 82/22	
Fig. 5B	slow MyHC and GFP IHC, 18som	15/3	114/4						
Fig. 5C bottom left	MyHC IHC, 12som	17/38	158/40	69/135	0/70	195/95	6/86		
Fig. 5C bottom right	MyHC IHC, 12som	0/38 [0%]	23/40 [58%]	2/135 [1%]	0/70 [0%]	65/95 [68%]	2/86 [2%]		
Fig. 6A	<i>myhz1</i> ISH, 24 hpf	<i>myf5</i> ^{hu2022/+} (<i>incross</i>) n20/20 [100%]	<i>myf5</i> ^{hu2022} sib + <i>myod</i> MO r19/25 [20%]	<i>myf5</i> ^{hu2022} sib + <i>cdkn1c</i> MO r34/41 [83%]	<i>myf5</i> ^{hu2022} + <i>myod</i> MO sr6/25 [24%]	<i>myf5</i> ^{hu2022} <i>myod</i> MO + <i>cdkn1c</i> MO a7/41 [17%]			
Fig. 6B	MyHC IHC, 24 hpf	con (wt) n40/40 [100%]	<i>cdkn1c</i> MO w55/60 [92%] n5/60 [8%]						
Fig. 6C	Myod, IHC, 24 hpf	n10/10 [100%]	r10/10 [100%]						
	Myogenin IHC, 24 hpf	n10/10 [100%]	r7/10 [70%] n3/10 [30%]						
Fig. 6D	<i>mylz2</i> ISH, 3 dpf	con MO a0/60 [0%] w3/60 [5%] n57/60 [95%]	<i>cdkn1c</i> MO (9ng) a19/20 [95%] w1/20 [5%] n0/20 [0%]	<i>myod</i> MO (2ng) a18/20 [90%] w2/20 [10%] n0/20 [0%]	<i>myod</i> MO (0.5ng) a2/20 [10%] w16/20 [80%] n2/20 [10%]	<i>cdkn1c</i> MO (3ng); <i>myod</i> MO (0.5ng) a15/20 [75%] w5/20 [25%] n0/20 [0%]	<i>cdkn1c</i> MO (3ng) a0/70 [0%] w10/70 [14.2%] n60/70 [85.8%]	<i>myod</i> MO (0.25ng) a7/40 [18%] w7/40 [18%] n26/40 [65%]	<i>cdkn1c</i> MO(3ng) <i>myod</i> MO (0.25ng) a29/50 [58%] w17/50 [34%] n4/50 [8%]
Fig. S1 left	BrdU IHC, 5s <i>myod</i> ISH, 5s	n41/41 [100%]	n15/15 [100%]						
Fig. S1 right	BrdU IHC, 5s <i>hsp90a</i> ISH, 5s	con (wt) n34/34 [100%]	<i>cdkn1c</i> MO n62/62 [100%]	<i>cyA</i> n31/31 [100%]					
Fig. S3 left	<i>myod</i> ISH, 8s	con (<i>shha</i> ^{-/-}) r0/55 [0%]	<i>shha</i> ^{-/-} + <i>cdkn1c</i> 5' MO r0/66 [0%]	<i>shha</i> ^{-/-} + <i>cdkn1c</i> ATG MO r0/39 [0%]					
Fig. S3 right	<i>myog</i> ISH, 8s	r0/66 [0%]	r19/44 [43%]						
Fig. S5	MyHC IHC, 15s	con (wt) n50/50 [100%]	<i>myod</i> RNA ur29/44 [66%]						
	<i>myog</i> ISH, 15s	n30/30 [100%]	ur19/22 [86%]						
	MyHC IHC, <i>prdm1</i> ISH, 15s	n50/50 [100%]	ur35/46 [76%]						
	<i>cdkn1c</i> ISH, 15s	n50/50 [100%]	ur25/38 [66%]						

IHC, immunohistochemistry; ISH, in situ hybridization; n, normal; wr, weakly reduced; r, reduced; sr, strongly reduced; a, ablated; ur up-regulated; wt, wild type.

Fig. 1A left. Fraction of embryos showing reduced (r) or absent (a) accumulation of MyHC or *myod* mRNA in adaxial cells.

Fig. 1A right. Fraction of embryos showing reduced (r) adaxial *myf5* and *myod* mRNAs in a double ISH assay.

Fig. 1B. Fraction of embryos showing reduced (r) or absent (a) expression of *myogenin* mRNA in adaxial cells.

Fig. 1C. Fraction of embryos showing normal (n), reduced (r) or absent (a) accumulation of Myod in adaxial cells.

Fig. 2A. Fraction of embryos showing normal (n), absent (a), or reduced (r) *myogenin* mRNA. A similar fraction is shown in Fig. 2B left, except these embryos were also stained by pan MyHC IHC.

Fig. 2B right. Fraction of embryos with normal (n) or reduced (r) *prdm1* mRNA or MyHC accumulation.

Fig. 2C left. Fraction of embryos showing normal (n), reduced (r) or absent (a) *eng2a* (lacking in *shha*^{-/-}) and slow MyHC.

Fig. 2C top right. Fraction of embryos showing normal (n), reduced (r) or absent (a) slow MyHC IHC.

Fig. 2C bottom right. Fraction of embryos showing normal (n), reduced (r), or absent (a) *mrf4* expression or adaxial *myod* accumulation.

Fig. 3A left. Fraction of embryos showing normal (n), reduced (r) or absent (a) *cdkn1c* expression in adaxial cells.

Fig. 3A middle. Fraction of embryos showing normal (n), absent (a) or up-regulated (ur) levels of *cdkn1c* transcript in adaxials.

Fig. 3A right. Fraction of embryos from a *smo*^{p641/a} incross showing normal (n) or absent (a) *cdkn1c* mRNA in tail somites.

Fig. 3B. Fraction of embryos showing normal (n), reduced (r) or absent (a) *myogenin* mRNA in adaxial cells.

Fig. 3C top. Fraction of embryos with reduced Myod protein level in adaxial cells.

Fig. 3C bottom. Fraction of embryos with reduced (r) somitic MyHC protein level.

Fig. 3D. Fraction of embryos showing normal (n) or reduced (r) Myod accumulation.

Fig. 4A,B,C. Fraction of embryos showing normal (n), reduced (r) or weakly reduced (wr) *cdkn1c* mRNA.

Fig. 4D. Fraction of embryos in which *cdkn1c* mRNA was normal (n), expanded laterally (e), reduced (r) or expanded laterally in PSM but reduced in the somite area (er).

Fig. 4E. Fraction of embryos showing normal (n), up-regulated (ur), weakly reduced (wr) or reduced (r) *smyhc1* mRNA expression.

Fig. 5A,B,C bottom left. Number of fibres / Number of embryos scored.

Fig. 5C bottom right. Number of rescued embryos / Number of embryos scored.

Fig. 6A. Fraction of embryos showing normal (n), reduced (r), strongly reduced (sr) or absent (a) *myhz1* mRNA.

Fig. 6B. Fraction of embryos showing normal (n) or weak reduction (wr) of pan-MyHC immunoreactivity.

Fig. 6C. Fraction of embryos imaged by confocal microscopy showing normal (n) or reduced (r) Myod or Myogenin immunoreactivity.

Fig. 6D. Fraction of embryos with ablated (a), weak reduction (wr) or normal (n) *mylz2* mRNA in hyoid muscle.

Fig. S1. Fraction of embryos with normal (n) BrdU immunoreactivity, including absence in adaxial cells.

Fig. S3 left. Fraction of embryos with reduced (r) adaxial *myod* mRNA.

Fig. S3 right. Fraction of embryos with reduced (r) adaxial *myogenin* mRNA.

Fig. S5. Fraction of embryos showing normal (n) or up-regulated (ur) levels of MyHC, *myogenin*, *prdm1* or *cdkn1c* mRNA accumulation.