A Western blot

B ORO staining
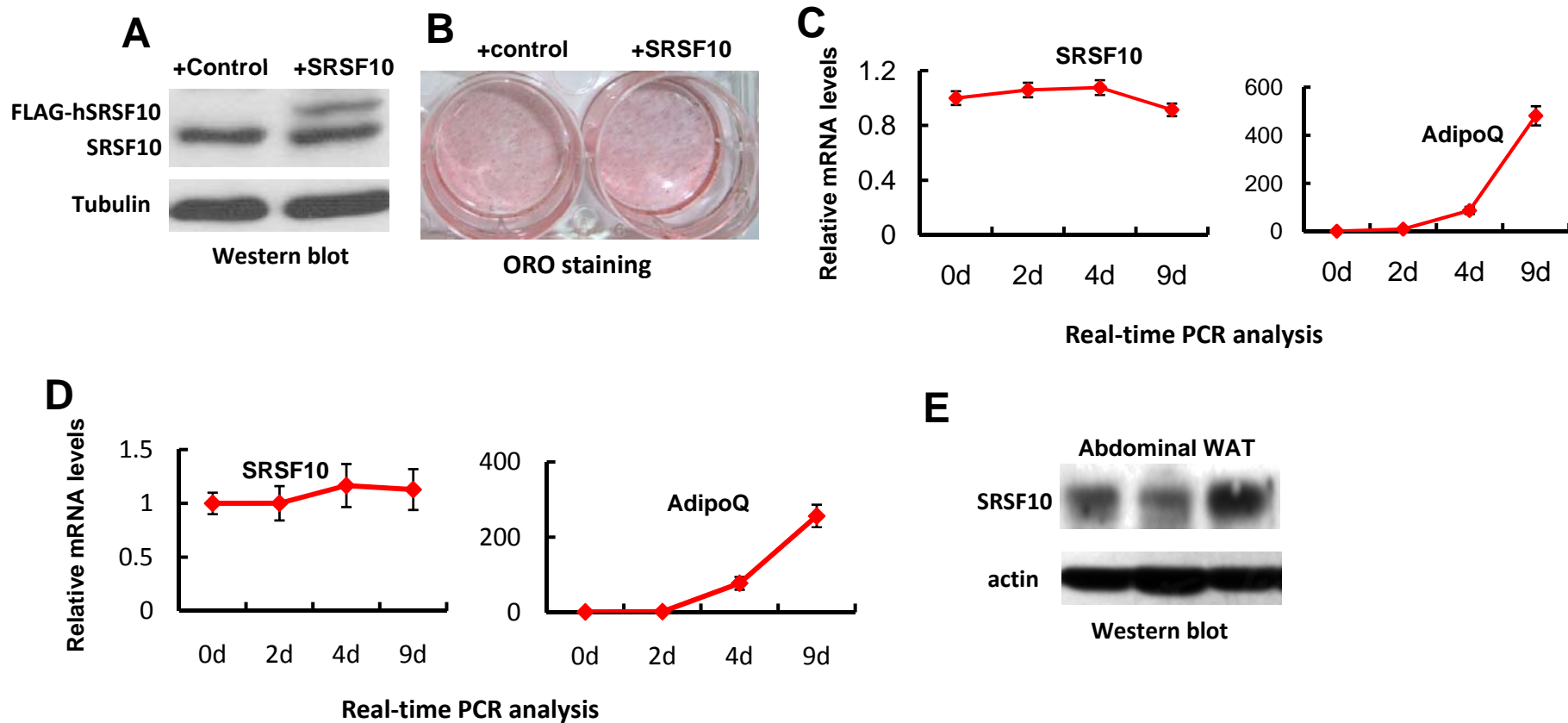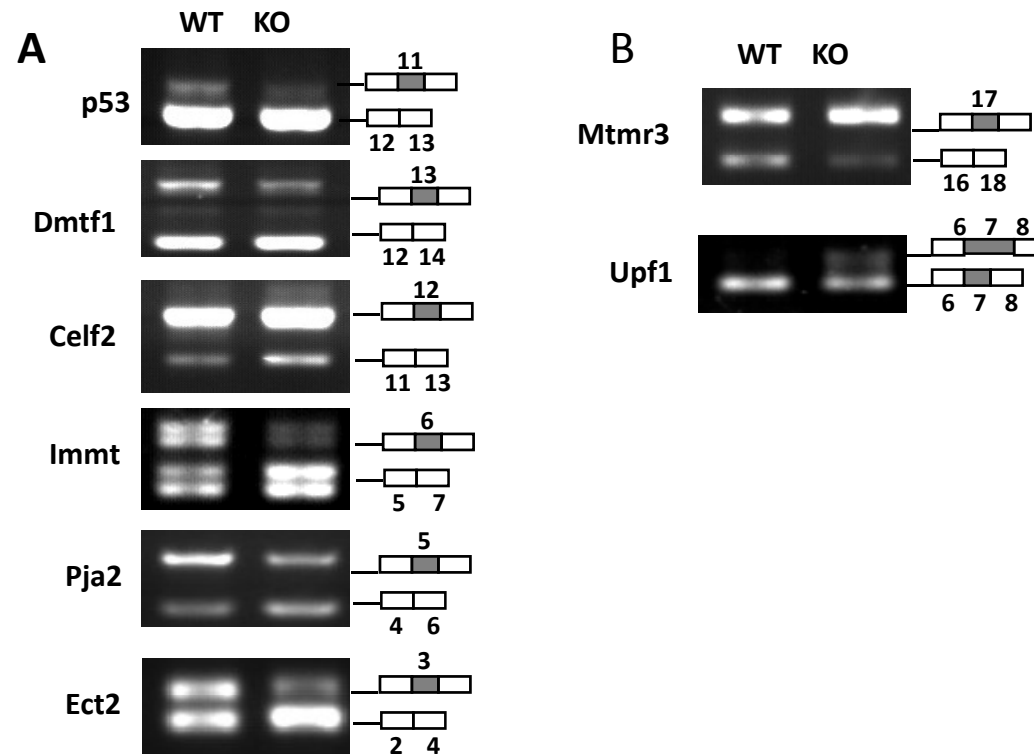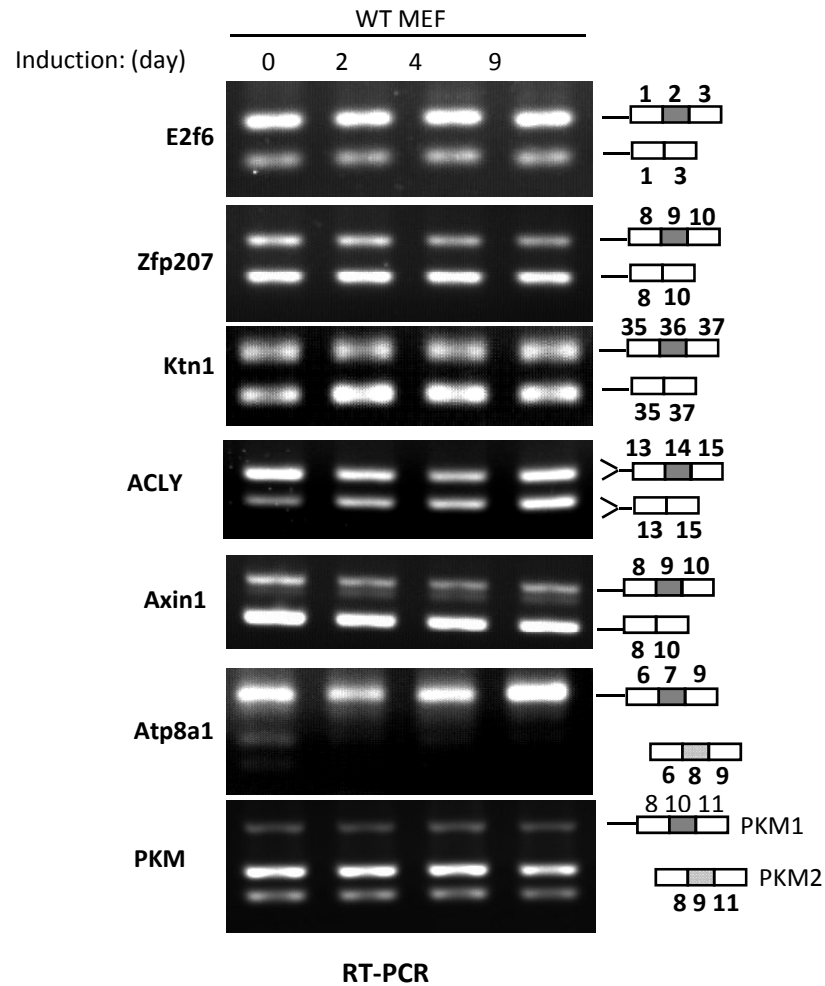
C Real-time PCR analysis

Supplemental Fig. S1. Ectopic expression of human SRSF10 could partially rescue the defective adipogenesis of the shSRSF10 /C3H10T1/2 cells. (A) shSRSF10 C3H10T1/2 cells were infected with retroviruses encoding either human SRSF10 (hSRSF10) or green fluorescent protein (GFP). As comparison, shCtrl C3H10T1/2 cells were also infected with GFP-containing retroviruses. The ectopic expression of FLAG tagged human SRSF10 was confirmed by immunoblotting using SRSF10 antibody. Position of FLAG-hSRSF10 is indicated by an arrow. (B) Hormone-induced adipogenesis of shCtrl-GFP, shSRSF10-hSRSF10 and shSRSF10-GFP C3H10T1/2 cells, as assessed by ORO staining. (C) RNAs were extracted from shCtrl-GFP, shSRSF10-hSRSF10 or shSRSF10-GFP C3H10T1/2 cells at indicated days. Relative mRNA of PPARr (left) or adipoQ (right) was measured as described in Figure 2B.
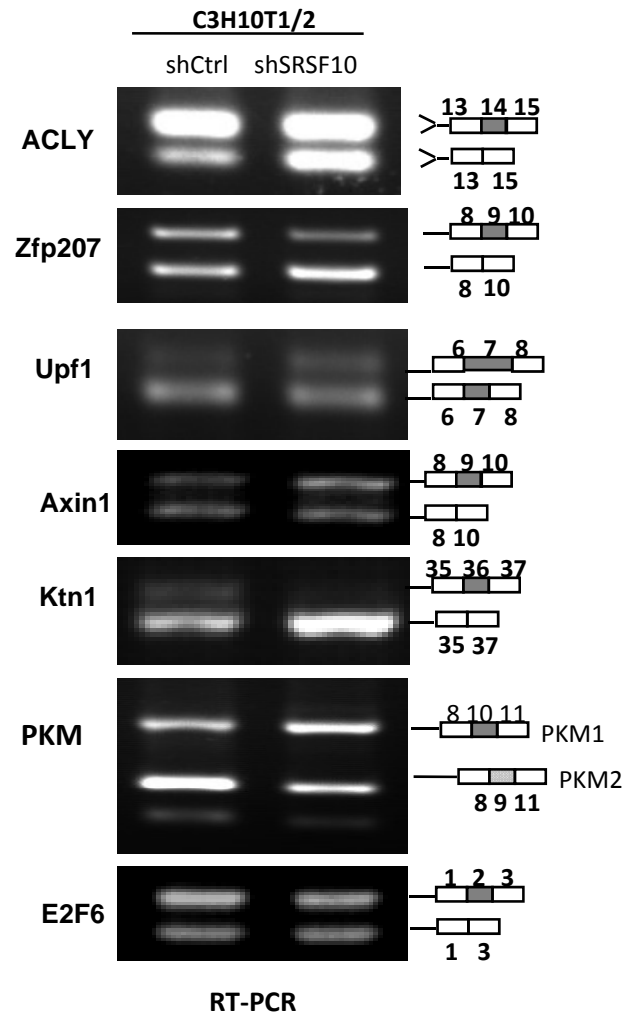
Supplemental Fig. S2. Wild-type amount of SRSF10 is enough to drive C3H10T1/2 cells to differentiate. (A) C3H10T1/2 cells were infected with retroviruses encoding either human SRSF10 (hSRSF10) or empty vector. The ectopic expression of FLAG tagged human SRSF10 was confirmed by immunoblotting. (B) Hormone-induced adipogenesis of cells described in A, as assessed by ORO staining. (C) RNAs were extracted from wild-type MEF cells at indicated differentiation days. Relative mRNA of SRSF10 (left) or adipoQ (right) was measured as described in Figure 2B. (D) RNAs were extracted from wild-type C3H10T1/2 cells at indicated differentiation days. Relative mRNA of SRSF10 (left) or adipoQ (right) was measured as described in Figure 2B. (E) SRSF10 was expressed in abdominal WAT of adult mice. Abdominal WAT was isolated from three adult B6 mice, respectively and subjected to western blot using indicated antibodies.

Supplemental Fig. S3. SRSF10 depletion causes both exon inclusion and exclusion *in vivo*. (A) Examples of SRSF10-dependent exon inclusion. Indicated transcripts containing alternatively spliced exons were analyzed by RT-PCR using RNAs extracted from WT and KO MEF cells, respectively. RNA products are indicated schematically on the left. (B) Examples of SRSF10-dependent exon exclusion were analyzed as described in (A).

Supplemental Fig. S4. Expression pattern of SRSF10- regulated splicing events during adipogenesis. Wild type primary MEF cells were induced to differentiate by using hormone cocktail. RNA samples were collected at indicated times and analyzed by RT-PCR. RNA products are indicated schematically on the right.

Supplemental Fig. S5. Splicing changes of SRSF10-validated genes in KO MEF cells were also observed in C3H10T1/2 cells upon SRSF10 knockdown. Total RNA was extracted from shCtrl and shSRSF10 C3H10T1/2 cells, respectively and subjected to RT-PCR analysis as described in Figure 3.

Supplemental Fig. S6. Changes in PKM splice variants did not alter adipogenesis. (A) C3H10T1/2 cells were infected with retroviruses expressing either PKM2-specific shRNAs (shPKM2-1/2/3) or control shRNA (shCtrl) and then selected for puromycin resistance. RNA samples were collected from the selected cells and PKM2 knockdown efficiency was determined by RT-PCR following Pst I digestion.(B) shPKM2-1/2/3 and shCtrl C3H10T1/2 cells were induced to differentiate by using hormone cocktail. 6 days post induction, cells were stained by ORO.

Supplemental Table S1. Oligo Sequence Information

For plasmid construction

Lipin1-mini-F-HindIII	CCCaagcttAACTCTTCCTAATGATGTACCACC
Lipin1-mini-R-XhoI	CGGctcgagCAGCCTGTGGCAATTCACC
Lipin1-CMV14-F-EcoRI	CGgaattcATGAATTACGTGGGGCAGC
Lipin1-CMV14-R-KpnI	CGGggtaccGCTGAGGCTGAATGCATG
Lipin1-E7-F-HindIII	cccAAGCTTCAGCCTGGTAGATTGCCA
Lipin1-E7-R-BamHI	cgGGATCCCTTTCCGAAGCATGGAAGTGG
Lipin1-E8-F-HindIII	ctcAAGCTTTCCTTCAGGCTCCCGGC
Lipin1-E8-R-BamHI	cgGGATCCCTTTGCAGCCTGTGGCAATTC

For shRNA overexpressing constructs

shCtrl	GATCCTTCTCCGAACGTGTCACGTTTCAAGAGA ACGTGACACGTTCGGAGAATTTTTTGCTAGCG
shSRSF10	GATCCGGAAGACACCAAATCAAATGATTCAAGA GATCATTTGATTTGGTGTCTTCCTTTTTTGCTAG CG
shPKM2-1	GATCCGCTTGCAGCTATTCGAGGAATTCAAGAG ATTCCTCGAATAGCTGCAAGTTTTTTGCTAGCG
shPKM2-2	GATCCGCCATTATCGTGCTCACCAATTCAAGAG ATTGGTGAGCACGATAATGGTTTTTTGCTAGCG
shPKM2-3	GATCCGCCATCTACCACTTGCAGCTATTCGATTC AAGAGATCGAATAGCTGCAAGTGGTAGATGGTT TTTTGCTAGCG

For site directed mutagenesis

Lipin1-mini-ΔE6-F	TAAACTTAAGCTTTGGTCCCCCAGCC
Lipin1-mini-ΔI6-F	GAGGGTTTTGAAGTCAGCTATAATTTCGTTTAAC TGACTGTGAC
Lipin1-mini-ΔE7-F	GTGCCTCCAGCAGCTCTCTAGCTCTTG
Lipin1-mini-ΔI7-F	CAATCACTTGCAATCAAAGGTTTTTTATT
Lipin1-mini-ΔE8-F	CTTCAGGCTCCCGCTCGAGTCTAGAG
Lipin1-mini-ΔE8-SRSF10 -F	GTCCTTCAGGCTCCCGAAAGACAAAAAAGACAA AAAAGACAAACTCGAGTCTAGAGGGC
Lipin1-mini-ΔE8-NTC-F	GTCCTTCAGGCTCCCGTCCCCCTCACCTGGCCGA GGGAGTTCTCTCGAGTCTAGAGGGC

For minigene analysis

Lipin1-F1	ATACGACTCACTATAGGGAGACC
Lipin1-R1	GCCAGAGCATTTCCAGGTTA
Lipin1-F2	TCCCAGTTCGGACAGAGAAT
Lipin1-R2	GCTGGCAACTAGAAGGCAC

For RT-PCR and qPCR

Zfp207-F	TTCCTCCAATGACTCAAG
Zfp207-R	CTGTAGACTGTGTATAAGC
Ktn1-F	GGAGCAAGAAGAAAGTAAATGG
Ktn1-R	TGCAACTCAGTCAACAGATC
Immt-F	CTGCCTGTGGCACAGAGCC
Immt-R	CAGTATGTTGGAGTGTG
Lipin1-F	TCCCAGTTCGGACAGAGAAT
Lipin1-R	GCCAGAGCATTTCCAGGTTA
Axin1-F	ATGGGTCTTCTGGGTTGA
Axin1-R	CTGCCTTTCTTTGTGATTTT
PKM-F	CACACAGATGCTGGAGAG
PKM-R	GCCAAGTTTACACGAAGG
Atp8a1-F	AAGGCTGATAATGCTGTGAAC
Atp8a1-R	TCGTAGAGATGTCGGTTTGG
CEBPa-F	GAACAGCAACGAGTACCGGGTA
CEBPa-R	CCCATGGCCTTGACCAAGGAG
aP2-F	GATGCCTTTGTGGGAACCTG
aP2-R	TCCTGTCGTCTGCGGTGATT
AdipoQ-F	GGAACTTGTGCAGGTTGGAT
AdipoQ-R	GCTTCTCCAGGCTCTCCTTT
PPARγ-F	CTCCTGTTGACCCAGAGCAT
PPARγ-R	AATGCGAGTGGTCTTCCATC
36B4-F	TAAAGACTGGAGACAAGGTGGGAG
36B4-R	AGAAAGCGAGAGTGCAGGGC

Supplemental Table S2：List of significantly changed splicing events regulated by SRSF10

ENSMUSG00000053070	9230110C19Rik	Cassette exon	−0.66	9.22E+18
ENSMUSG00000028560	Usp1	Cassette exon	−0.66	9.22E+18
ENSMUSG00000020917	Acly	Cassette exon	0.66	9.22E+18
ENSMUSG00000020070	Rufy2	A3SS	−0.65	125.15
ENSMUSG00000028560	Usp1	Cassette exon	−0.63	9.22E+18
ENSMUSG00000057406	Whsc1	Cassette exon	0.57	13763.43
ENSMUSG00000004980	Hnrnpa2b1	A5SS	−0.57	126.72
ENSMUSG00000025893	Kbtbd3	Cassette exon	−0.53	285.04
ENSMUSG00000031488	Rab11fip1	Cassette exon	0.51	10.69
ENSMUSG00000038344	Txlng	RI	0.51	3.58
ENSMUSG00000063889	Crem	Cassette exon	−0.5	19.3
ENSMUSG00000022884	Eif4a2	Cassette exon	−0.49	152.55
ENSMUSG00000062822	4833420G17Rik	Cassette exon	0.49	260.54
ENSMUSG00000029062	Cdk11b	Cassette exon	0.49	35.16
ENSMUSG00000025185	Loxl4	A3SS	0.49	11.03
ENSMUSG00000074129,I	Rpl13a	RI	−0.49	11.11
ENSMUSG00000024073	Birc6	A5SS	−0.48	13.95
ENSMUSG00000051149,I	Adnp	Cassette exon	−0.47	3.02E+10
ENSMUSG00000057469	E2f6	Cassette exon	0.47	38.92
ENSMUSG00000030811	Fbxl19	A5SS	−0.47	32.58
ENSMUSG00000021236	Entpd5	Cassette exon	−0.46	14.44
ENSMUSG00000021843	Ktn1	Cassette exon	−0.46	11.45
ENSMUSG00000028229	Fam82b	Cassette exon	0.46	13.43
ENSMUSG00000020593	Lipin1	Cassette exon	−0.46	76.64
ENSMUSG00000022425	Enpp2	Cassette exon	−0.45	12.13
ENSMUSG00000024943	Smc5	A5SS	0.44	13.05
ENSMUSG00000030272	Camk1	RI	0.44	12.34
ENSMUSG00000009585	Apobec3	Cassette exon	−0.43	34.9
ENSMUSG00000032570	Atp2c1	Cassette exon	−0.43	14.42
ENSMUSG00000026455	Klhl12	A3SS	−0.43	11.19
ENSMUSG00000022364	Wdr67	Cassette exon	−0.42	56.61
ENSMUSG00000004980	Hnrnpa2b1	Cassette exon	−0.41	9.22E+18
ENSMUSG00000030516	Tjp1	A5SS	0.4	20.56
ENSMUSG00000032103	Pus3	RI	0.4	64
ENSMUSG00000058301	Upf1	Cassette exon	0.4	20.56
ENSMUSG00000000326	Comt	Cassette exon	−0.39	9.22E+18
ENSMUSG00000032580	Rbm5	Cassette exon	−0.39	13780.65
ENSMUSG00000020250	Txnrd1	Cassette exon	0.39	34.56
ENSMUSG00000052337	Immt	Cassette exon	−0.37	9.22E+18
ENSMUSG00000032410	Xrn1	A5SS	−0.37	42.59
ENSMUSG00000027342	Pcna	A5SS	−0.36	9.22E+18
ENSMUSG00000049295	Zfp219	Cassette exon	−0.35	36.38
ENSMUSG00000052915	Msl1	Cassette exon	0.35	85.12
ENSMUSG00000040669	Phc1	Cassette exon	0.35	64.36
ENSMUSG00000027684	Mecom	A3SS	−0.35	1.03E+13
ENSMUSG00000063802	Hspbp1	A3SS	−0.35	56.6
ENSMUSG00000028999	Rint1	Cassette exon	−0.34	293.11
ENSMUSG00000035863	Palm	Cassette exon	0.34	756.6
ENSMUSG00000059552	p53	Cassette exon	0.34	23.73
ENSMUSG00000002409	Dyrk1b	A3SS	0.34	22.59
ENSMUSG00000036564	Ndrg4	Cassette exon	0.33	11.35
ENSMUSG00000040325	Vprbp	Cassette exon	0.33	11.15
ENSMUSG00000064105	Cnnm2	Cassette exon	−0.32	14.06

ENSMUSG00000063802	Hspbp1	A3SS	-0.32	13.02
ENSMUSG00000029141	Slc4a1ap	Cassette exon	-0.31	29.31
ENSMUSG00000084504,I	Colec12	Cassette exon	0.31	82.34
ENSMUSG00000003680,I	Taf6l	Cassette exon	0.31	39.5
ENSMUSG00000035401	2210018M11Rik	A3SS	-0.31	11.11
ENSMUSG00000005506	Celf1	Cassette exon	-0.3	4.20E+14
ENSMUSG00000021843	Ktn1	Cassette exon	0.3	32.14
ENSMUSG00000034518	Hmgxb4	Cassette exon	0.3	6.84
ENSMUSG00000027639	Samhd1	Cassette exon	0.3	38132.25
ENSMUSG00000027699	Ect2	Cassette exon	0.3	1.49E+14
ENSMUSG00000042508	Dmtf1	Cassette exon	-0.29	8.13
ENSMUSG00000006418	Rnf114	Cassette exon	0.29	53.98
ENSMUSG00000022892	App	Cassette exon	-0.28	1.00E+12
ENSMUSG00000037697	Ddhd1	Cassette exon	-0.28	28.97
ENSMUSG00000062901	Klhl24	Cassette exon	-0.28	18.83
ENSMUSG00000002984	Tomm40	RI	-0.28	24.58
ENSMUSG00000028552	Eps15	Cassette exon	-0.27	2.02E+11
ENSMUSG00000063179	Pstk	Cassette exon	-0.27	51.44
ENSMUSG00000037685	ATP8a1	MXE	-0.27	3.7
ENSMUSG00000037536	Fbxo34	Cassette exon	0.26	33.36
ENSMUSG00000066037	Hnrnpr	Cassette exon	0.26	45.67
ENSMUSG00000024182	Axin1	Cassette exon	-0.26	43.67
ENSMUSG00000026305	Lrrfip1	Cassette exon	0.25	31.29
ENSMUSG00000019795	Pcmt1	A5SS	0.25	37.01
ENSMUSG00000042605	Atxn2	Cassette exon	-0.24	8216535.25
ENSMUSG00000033819	Ppp1r16a	Cassette exon	-0.24	15.82
ENSMUSG00000025278	Flnb	Cassette exon	0.24	78.87
ENSMUSG00000024083	Pja2	Cassette exon	0.24	429.16
ENSMUSG00000041231	Ublcp1	A3SS	-0.24	2467.96
ENSMUSG00000024483	Ankhd1	A3SS	-0.24	322.76
ENSMUSG00000027210	Meis2	Cassette exon	-0.23	21.65
ENSMUSG00000027593,I	Raly	Cassette exon	-0.23	9.22E+18
ENSMUSG00000007880	Arid1a	A5SS	-0.23	14.91
ENSMUSG00000038866	Zcchc2	RI	0.23	76.05
ENSMUSG00000054426	A930005H10Rik	Cassette exon	0.22	88.27
ENSMUSG00000029106	Add1	Cassette exon	0.22	618.2
ENSMUSG00000029505	Ep400	Cassette exon	0.22	343.75
ENSMUSG00000078713	Tomm5	Cassette exon	0.22	9.22E+18
ENSMUSG00000034354	Mtmr3	Cassette exon	-0.22	3.73E+07
ENSMUSG00000020831	0610010K14Rik	A5SS	0.22	180.54
ENSMUSG00000007850	Hnrnph1	RI	0.22	33.13
ENSMUSG00000017421	Zfp207	Cassette exon	-0.21	9.22E+18
ENSMUSG00000068580	Zfyve19	Cassette exon	-0.21	33.87
ENSMUSG00000006498	Ptbp1	Cassette exon	0.21	24947.64
ENSMUSG00000032294	PKM	MXE	0.21	27.82
ENSMUSG00000002107	Celf2	Cassette exon	-0.2	4.33E+07
ENSMUSG00000024571,I	Txnl4a	Cassette exon	0.2	18.93