

Supplemental Figure 1. Comparative expression of K_v7 transcripts during skeletal myogenesis in C_2C_{12} cells and HSkM cells. Quantification of K_v7 mRNA expression levels in murine C_2C_{12} (grey columns) and in primary human (HSkMC; black columns) differentiated myotubes relative to myoblasts upon 48 hrs of exposure to differentiation media (DM), by use of quantitative real-time PCR. Each data point is from at least 4 separate experiments in GM- or DM-exposed cells. * = $p \le 0.05$.



Supplemental Figure 2. Effect of shK_v7.4 transfection on the expression levels of K_v7 transcripts in C₂C₁₂ myoblasts. Transcripts levels analysis of all members of K_v7 subfamily analyzed in C₂C₁₂ myoblasts transfected with control (empty columns) and shKv7.4 plasmid (black columns) by use of quantitative PCR. Data are expressed using the 2^{$-\Delta\Delta$ dct} formula. Each data point is from at least 4 separate transfections (scramble- or shK_v7.4- plasmid). * = p≤0.05 versus control-transfected C₂C₁₂ cells.



Supplemental Figure 3. Effect of XE-991 on C₂C₁₂ differentiation. Transcripts levels of Myog and TnT-1 were analyzed by quantitative PCR in C₂C₁₂ exposed to DM in presence of different doses of XE-991. Data are expressed using the $2^{\Lambda-\Delta\Delta ct}$ formula. Each data point is from at least 3 separate experiments. *= p<0.05 versus respective control (vehicle).



Supplemental Figure 4. Effect of K_v7.4 over-expression on the expression of differentiation markers in C₂C₁₂ myoblasts. Transcripts levels of Kv7.4, TnT-1, and Pax3 were analyzed by semi-quantitative PCR in C₂C₁₂ myoblasts transfected with control plasmid (empty columns) and pcDNA3.1 carrying K_v7.4 gene. Data are normalized the OD of each gene of interest relative to that of the reference gene S16. Each data point is from three separate transfections (scramble- or K_v7.4- plasmid). *= p≤0.05 versus respective control.



Supplemental Figure 5. Effect of REST-silencing on the expression of differentiation markers in C_2C_{12} myoblasts. REST, Myog, TnT-1 and K_v7.4 transcript levels were analyzed by qPCR in C_2C_{12} myoblasts transfected with shREST plasmids. Data are expressed using the 2^{-...Act} formula relative to the control condition (scramble plasmid). Each data point is from three separate transfections (scramble- or REST- plasmid). * = p≤0.05 versus respective control.

Supplemental Table I. Primer sequences.

	sense primer (5'-3')	reverse primer (5'-3')
mouse primers		
Troponin 1 (TnT-1)	CTGTGGTGCCTCCTTTGATTC	TGCGGTCTTTTAGTGCAATGAG
Myogenin (Myog)	AGGAGAGAAAGATGGAGTCCAGAG	TAACAAAAGAAGTCACCCCAAGAG
PAX3	GGGCAGAATTACCCACGCA	AGACGGTTCCCTTTGTCGC
REST/NRSF	CATGGCCTTAACCAACGACAT	GGCGGATATGGTTGGGCAG
K _v 7.1	GGACCAGAGACTGGTGATCATC	TTGCTGGGTAGGAAGAGCTCAG
K _v 7.2	GCTTTCCGCATCAAGGGTG	TGCTAACTTTGAGGCCAGG
K _v 7.3	CACCGTCAGAAGCACTTTGAG	CCTTTAGTATTGCTACCACGAGG
K _v 7.4	CCGTTCTGTCAGGATTCTGAAGT	CCGTTCTGTCAGGATTCTGAAGT
K _v 7.5	CTGGCAGCAAGAATTGTGAA	GTGGTCATGGCT TCAATGTG
S16	CTGGAGCCTGTTTTGCTTCTG	TGAGATGGACTGTCGGATGG
(ChiP)		
Reg-1	AAGTTGCAGAGGGTGGA	TCCGGATTCCCAGGACATGA
Reg-2	CAGCAGATCAATGCTGCC	ATGGCCAAACCAGACGCT
Reg-3	TCCCCGATTTGGGAAGGC	GGCTCCAACTTACATGAAGACG
Reg-4	TGCAGAGTGAACAGGGCGA	GGCTCCAACTTACATGAAGACG
human primers		
Troponin 1 (TnT-1)	TGATCCCGCCAAAGATCCC	TCTTCCGCTGCTCGAAATGTA
K _v 7.1	TTGCCCTGAAGGTGCAGCAGAA	TTCTCGGCAGCATAGCACCTCCAT
K _v 7.2	TCCTGGAAATCGTGACTATCGT	TTCCGGGCAAACTTGAGCC
K _v 7.3	CCGCCCAGTCAAGAGAAACAA	CAGGACAATCAGGAACACCAAC
K _v 7.4	TGCTGTCCACTATCCAGGAC	CGATGAAGTCGATGACACAGAA
K _v 7.5	CAGGGGCCAGAGTATTAAGAGC	CTTCTGCACTTTGGTGGGACT
REST/NRSF	GCCGGCTGCGCGAATACAGT	TGAGGTGCGGCCAGTTCAGC
S16	TCGGACGCAAGAAGACAGCGA	AGCGTGCGCGGCTCAATCAT