

miR-1, miR-10b, miR-155 and miR-191 are novel regulators of BDNF

Cellular and Molecular Life Sciences

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ELECTRONIC SUPPLEMENTARY MATERIAL

Online resource 1 List of PCR and qPCR primers used in the study.

Online resource 2 Conservation of miR binding sites and flanking sequences in BDNF 3'UTR. Light blue background: conserved nucleotides within sequence complementary to miR seed; grey background: conservation within 20 nucleotides flanking miR seed sequence.

Online resource 3 **A.** List of broadly conserved microRNA families. **B.** List of miRs from broadly conserved miR families. **C.** Potential miR binding sites within *Mus musculus* BDNF 3'UTR predicted with different bioinformatics tools. In all analyses, only predictions for miRs from broadly conserved miR families expressed in *Mus musculus* are shown. In PITA analysis, sites with $\Delta\Delta G < -10$ and $\Delta\Delta G < 0$ are indicated in columns D and E, respectively. In miRanda analysis only sites of conserved miRs with good mirSVR scores are shown. TargetScan ((Lewis et al. 2005), www.targetscan.org), PITA ((Kertesz et al. 2007), <http://genie.weizmann.ac.il/pubs/mir07/index.html>), miRanda ((John et al. 2004), www.microrna.org), PicTar ((Krek et al. 2005), <http://pictar.mdc-berlin.de/>). *mmu* – *Mus musculus*.

Online resource 4 **(a)** BDNF mRNA expression in different cell lines and mouse hippocampus, relative to β -actin. **N=3-5** **(b)** Conservation between miR-1 and miR-206 in mouse (*mmu*) and human (*hsa*). miR seed sequence is underlined and conserved nucleotides are shown in bold. **(c)** Overview of the induction of myogenic differentiation and timeline of RNA/protein isolation and luciferase assays. Images of undifferentiated myoblasts and differentiated myotubes of C2C12 mouse skeletal muscle cell line. **(d)** BDNF protein levels in C2C12 myoblasts and myotubes normalized to total protein content. **N=3.** **(e)** Expression of miR-1 and miR-206 in C2C12 myoblasts and myotubes normalized to sno202. **N=2.** Error bars denote mean $\pm$ SEM; * $p < 0.05$.

Online resource 5 miR expression in different cells, normalized to miR-191 (human cells) or sno-202 (mouse cells). **N=3-5.** Values are presented as mean $\pm$ SEM. n.e – not expressed; n.t – not tested.

Online resource 6 miR binding sites within SV40 late polyA sequence predicted with PITA. Only predictions for miRs from broadly conserved miR families with $\Delta\Delta G < -5$ are shown. *mmu* – *Mus musculus*.