Supporting Information

Materials and Methods

Animal model and Cell cultures. To generate mutant males (*Mecp2*$^{-/y}$, KO) and their control (*Mecp2*$^{+/y}$, WT) littermates, heterozygous females (*Mecp2*$^{tm1.1Jae/+}$) were mated with wild-type males (*Mecp2*$^{+/y}$). The offspring from the breeding were genotyped by PCR as previously described (1).

Cortical and cerebellar neurons were isolated from postnatal day (P) 0 cortices and P6 cerebella respectively and were cultured as previously described (1-3). All animal experiments were done in accordance with the protocols approved by the Animal Research Committee of the University of California, Los Angeles.

SOLiD sequencing and data analysis. In the 1st sequencing experiment, total RNA of cerebellum (CB) tissues was isolated from WT and KO littermates at postnatal 6-week (pre- or early-symptomatic stage) using Trizol (Invitrogen). Four pairs of WT and KO CB were pooled for constructing sequencing libraries as previously described (4). Briefly, RNA was ligated overnight with the 'A' adapters, reversed transcribed, and amplified before size selection (18-30 nt inserts). After amplification using emulsion PCR, libraries were deposited on slides and sequenced by the SOLiD v2 sequencing system (Applied Biosystems). WT and KO samples were distinguished by adding unique barcode-attached amplification primers (provided by the SOLiD SREK kit). After the usual sequencing reactions, a second set of reactions decoded the barcode sequence. Sequencing results were obtained as 'good' and 'best' beads in colorspace (.csfasta format; a total of 3,660,124 reads in WT and 2,789,136 reads in KO were obtained) by the SOLiD sequence analysis software. The sequencing reads were aligned to all 590 annotated mouse miRNA precursor sequences (miRBASE version 15, retrieved from ftp://mirbase.org/pub/mirbase/) by the SHRiMP program (version 1.1.0) with default parameters (5) (**Table S1**). Mapped reads were loaded into a MySQL database and overlapping alignments were analyzed for read counting using custom Python scripts. We have focused on high confidence alignments by selecting those having 10 or more overlapping sequencing reads aligned to annotated miRNA precursors. The resulting miRNA expression profiles identified 315 miRNA genes (specifying 494 mature miRNAs or their anti-sense sequences) in WT cerebella. Mature miRNAs with more than 1.5-fold difference in read counts between WT and KO were identified as dys-regulated miRNAs (**Table S2**). Sequences of other small non-coding RNAs were retrieved from the Rfam database release 10 (rRNAs, snRNAs and snoRNAs, ftp://ftp.sanger.ac.uk/pub/databases/Rfam/) and the genomic transfer RNA database (tRNA, http://lowelab.ucsc.edu/GtRNAdb/download.html)(6, 7). Read count of non-coding RNAs in WT and KO cerebella (6-week) were normalized to the total read number of Rfam ncRNAs (89,901 reads in WT; 88,926 reads in KO) or tRNAs (24,839 reads in WT; 13,568 reads in KO). Rfam ncRNAs and tRNAs with

more than 1.5-fold difference in read counts between WT and KO were identified as dys-regulated small RNAs (**Table S4**).

In the 2nd sequencing experiment, we have isolated the small RNA population from two litters of WT and KO littermates (one control and two mutant mice in each litter, six animals in total) at postnatal 8-week (symptomatic stage) using PureLink miRNA isolation kit (Invitrogen). We subsequently prepared small RNA libraries for each animal and sequenced individual library separately according to small RNA sequencing procedures of the SOLiD total RNA-seq kit protocol (Applied Biosystems, or ABI). Analysis of small RNA raw datasets (csfasta files) was done with ABI SOLiD's small RNA Analysis Tool V0.50 (RNA2MAP). All raw reads are aligned to filter sequences that include rRNA, tRNA, adaptor, polyA, polyG, polyT and polyC sequences. Alignment is based only on the first 25 bases of the read with 2 mismatches allowed. The remaining reads that have no alignment to the filter sequences are subsequently mapped to annotated mouse miRNA precursor sequences. Alignment is based on first 18 bases of the read with 3 mismatches allowed. Once an alignment is found, the algorithm attempts to find the adaptor location. A test read is created by combining bases from the genome and adaptor sequence and compared against the full length read, a step that tries all possible starting points of the adaptor. The one that minimizes the number of mismatches is used and the small RNA size is determined. The reads that align give rise to the statistics in the "Mapping to miRBASE column (**Table S1**). Remaining reads (reads that do not map to filter sequences or miRBASE sequences) are then aligned to the reference genome (mm9). Finally, the total mappable reads is obtained by adding the total mappable reads from both miRBASE and mm9 alignment (**Table S1**). By combining all sequencing results, expression of 253 (specifying 370 mature miRNAs or their anti-sense sequences) miRNA genes were detected in all 6 small RNA libraries of cerebella. To identify dys-regulated miRNAs in KO cerebella at the symptomatic stage, fold change (FC) between WT and KO for each detected mature miRNAs were calculated in two complementary approaches. In the first approach, the read count in each KO was divided with that of its WT littermate, thus evaluating the data by taking into account the potential difference between litters. In the second approach, we calculated the fold change for each miRNAs by dividing the read count in KO with an averaged read count in WT from two litters. In each approach, a total of 4 FC values were obtained and subsequently averaged for each expressed mature miRNAs. Mature miRNAs with at least a 1.5-fold change in read counts between WT and KO (and 3 out 4 FC values show same trend) in both approaches were identified as dys-regulated miRNAs (**Table S3**).

TaqMan real-time PCR quantification of microRNA. Total RNA was isolated using Trizol and quantified by Nanodrop spectrophotometer (Agilent). 50 ng total RNA was used for reverse transcription of individual miRNA (TaqMan® MicroRNA Reverse Transcription Kit, Applied Biosystems) and TaqMan real-time PCR (Applied Biosystems). MicroRNA reverse transcription and TaqMan PCR (TaqMan® Universal PCR Master Mix, No UNG, Applied Biosystems) were performed in 96-well format using an iCycler real-

time PCR instrument (Bio-Rad). Reactions were done in triplicates and standard curves were generated for several miRNAs (e.g. miR-124a) using serial dilutions of cerebellar total RNAs to evaluate the dynamic range of TaqMan qPCR assays. Fold change was calculated between *Mecp2*$^{+/y}$ and *Mecp2*$^{-/y}$ cerebellum using $2^{-\Delta\Delta Ct}$ method after normalized to the level of endogeneous U6 spliceosomal RNA.

Chromatin immunoprecipitation (ChIP) and Mecp2 ChIP-chip. To immunoprecipitate chromatin bound by Mecp2 using a previously validated polycolonal antibodies (8), cerebella isolated from four pairs of *Mecp2*$^{+/y}$ and *Mecp2*$^{-/y}$ littermates (postnatal 6-week) were homogenized and cross-linked with 1% formaldehyde for 10 minutes at room temperature followed by exposure to 0.125 M glycine to stop the cross-linking reaction. After two washes with ice-cold PBS, cells were collected as pellets and stored at -80˚C before use. Nuclei were extracted and lysed sequentially with lysis buffer 1 (LB1, 50 mM Hepe2-KOH, pH7.5, 140 mM NaCl, 1 mM EDTA, 10% glycerol, 0.5% NP40, 0.25% Triton X-100), lysis buffer 2 (LB2, 10 mM Tris-HCl, pH8.0, 200 mM NaCl, 1 mM EDTA, 0.5 mM EGTA), and lysis buffer 3 (LB3, 10 mM Tris-HCl, pH8.0, 100 mM NaCl, 1 mM EDTA, 0.5 mM EGTA, 0.1% Na-Deoxycholate, 0.5% N-lauroylsarcosine). Chromatin was sonicated using a microtip (Branson sonifier 450) until the DNA fragments were reduced to 200-1000 bp in length. 10 µg antibodies were pre-incubated with 100 µl Dynal protein-G beads (Invitrogen) for at least 6 hours. Immunoprecipitation was performed overnight at 4˚C with antibody conjugated protein-G beads. DNA/protein complexes were washed with RIPA buffer (50 mM Hepes-KOH, pH7.6, 500 mM LiCl, 1 mM EDTA, 1% NP-40, 0.7% Na-Deoxycholate) for 5 times and eluted from beads and reverse cross-linked at 65 ˚C overnight. The DNA was digested with RNase A and proteinase K sequentially, followed by phenol/chloroform extraction and ethanol precipitation.

For microarray analyses, we amplified input and immunoprecipitated DNA using whole genome amplification kits (WGA, Sigma). For hybridization with customized NimbleGen 385K DNA tiling microarray (covering approximately -3.25kb to +0.75kb genomic regions relative to TSS of ~17,000 annotated protein-coding Refseq genes and -4.0kb to +4.0kb genomic regions flanking of 469 annotated mouse miRNA genes; ~100-bp per probe). For hybridization with NimbleGen microarrays, 4 µg of amplified input and ChIP DNA was sent to NimbleGen. Sample labeling, hybridization, data extraction and peak detection were performed according to standard procedures by NimbleGen Systems. Two separate hybridizations using biological independent samples (pooled from four pairs of WT and KO cerebella, postnatal 6-week) were performed and yielded similar results. We combined and averaged the signals from replicate arrays.

For identification of probes associated with significant level of Mecp2 occupancy, a non-parametric one-sided Kolmogorov-Smirno (KS) test was used. Briefly, from the scaled log2-ratio data, a fixed-length window (750bp) is placed around each consecutive probe and the one-sided KS test is applied to determine whether the probes are drawn from a significantly more positive distribution of intensity log-ratios than those in the rest of the array. The resulting score for each probe is the $-\log_{10}$ P-

value from the windowed KS test around that probe. Using NimbleScan software (NimbleGen), peak data files are generated from the P-value data files. NimbleScan software detects peaks by searching for at least 2 probes above a P-value minimum cutoff (-log10) of 2. Peaks within 500bp of each other are merged.

ChIP-qPCR was performed in an iCycler (Bio-Rad) using iQ SYBR Green Supermix (Bio-Rad). Reactions were done in triplicates and standard curves were calculated on serial dilutions of input genomic DNA. For quantification of relative level of MeCP2 occupancy, we calculated the percentage of immunoprecipitated DNA over input DNA. Primer sequences used in real-time PCR are listed below:

Mmu-miR-10a: forward 5'- GCCTTTGATCTTCTTCTCAGGTTG -3',

reverse 5'- GGTGTGAAGGGGGTGTAATAAAAG -3';

Mmu-miR-34c: forward 5'- TACCTGGTTAAGTGGGCTGAGTTC -3',

reverse 5'- CTGGCTTTAGGATCTCCATTTCAG -3';

Mmu-miR-137: forward 5'- AAGGATGAGTTGGTCTACCTGGAG -3',

reverse 5'- AGTGCAAATAGCTCTCCATCAAGG -3';

Methylated DNA immunoprecipitation (MeDIP) and MeDIP-chip. MeDIP assays were carried out as previously described (9). Briefly, cerebellar genomic DNA (6-week) was digested with proteinase K and RNase A sequentially, and purified by phenol/chloroform extraction. Purified genomic DNA was sonicated and heat-denatured (95 ˚C, 10 min). An aliquot of sonicated genomic DNA was saved as input. Fragmented genomic DNA was immunoprecipitated with a monoclonal antibody against 5-methylcytidine (Eurogentec) at 4 ˚C overnight in a final volume of 500 µl of IP buffer (10 mM sodium phosphate (pH 7.0), 140 mM NaCl, 0.05% Triton X-100). We incubated the DNA-antibody mixture with 30 µl protein G Dynabeads (Invitrogen) for 2 h at 4 ˚C and washed it three times with 1 ml IP buffer. We then treated the beads with proteinase K for at least 3 h at 55 ˚C and purified the methylated DNA by phenol-chloroform extraction followed by ethanol precipitation. For real-time PCR analysis, 10 ng of input genomic DNA and 1/30 of the immunoprecipitated (IP) methylated DNA was used for each PCR reaction. MeDIP-qPCR was performed in an iCycler (Bio-Rad) using iQ SYBR Green Supermix (Bio-Rad). Reactions were done in triplicates and standard curves were calculated on serial dilutions of input genomic DNA. ChIP-qPCR primers were used in MeDIP-qPCR to quantify the relative DNA methylation level within the regions occupied by MeCP2. For microarray analyses, we amplified input and immunoprecipitated DNA using whole genome amplification kits (WGA, Sigma). Labeling, hybridization, array processing and data extraction were done following procedures described for ChIP-chip.

Sequenom EpiTYPER DNA Methylation Analysis. We carried out bisulphite conversion of 1 µg genomic DNA using EZ DNA methylation-Gold Kit (Zymo) as previously described (9). Sequenom MassARRAY® MALDI-TOF mass spectrometry (MS) analysis of bisulphite converted DNA was performed in triplicate (EpiTYPER DNA Methylation Analysis). Primers were designed using Sequenom's EpiDesigner tool (http://www.epidesigner.com). The results were similar to those from MeDIP-qPCR.

Luciferase reporter assay and transfections. For luciferase report assays, we cloned *Nrxn3* and *Bdnf* 3' UTR sequences into the pISO fly luciferase construct (10). MiRNA mimics duplexes or 2'-O-methyl miRNA inhibitors (Dharmacon) and pISO luciferase vector were co-transfected into mouse (cortical or cerebellar) neurons using Lipofectamine 2000 (Invitrogen) together with a TK renilla luciferase reporter (Promega). Approximately 24 h after transfection, cells were lysed for dual luciferase assays (Promega). The following primers were used for cloning of 3' UTRs:

Nrxn3: forward 5'- GAGCTCtactctcacagagggcaatgtgtc -3'

reverse 5'- TCTAGAataaacatgcaccatcacacaggg -3'

Bdnf: forward 5'- GAGCTCgtcctagagaaagtcccggtatcc -3'

reverse 5'- TCTAGAgagatacatcatgggcagtggag-3'

BDNF ELISA and electroporation of cortical neurons. A BDNF ELISA kit (Promega) was used to measure BDNF protein levels as per the manufacturer's instructions. All solutions were made exactly as recommended by the kit instructions. Cell lysate was treated with acid to increase the amount of free BDNF. A SPECTRAmax microplate reader (Molecular Devices) was used to measure signal intensity from the wells. A standard curve was generated from the BDNF standard wells on each plate, using the SoftMax Pro 4.6 software. The amount of BDNF per well was calculated by based on the standard curve. To measure the effect of dys-regulated miRNAs (miR-30a/d, miR-381 and miR-495) on endogenous BDNF expression, freshly isolated cortical neurons were electroporated (Lonza/Amaxa, program#: G-013) with 40 pmol miRNA duplex mimics or 2'O-Methyl miRNA inhibitors (final concentration: 20 µM in medium, Dharmacon) and analyzed 6 days post-transfection. To validate the activity-dependent induction of BDNF, the cortical neurons were depolarized in presence of 50mM KCl for 24 h before harvested for ELISA analysis. MiRNA mimics or inhibitor for a *C. elegans* specific miRNA (Dharmacon), cel-miR-67, was used as a control in all ELISA assays.

Lentivirus-mediated knockdown of Mecp2 in cortical neurons. A lentiviral vector encoding shRNA specific for mouse Mecp2 (NM_010788, Sigma MISSION^{TM} TRC shRNA set) was used for silencing Mecp2 expression (TRCN0000039081, targeting CDS, 5'- CCGGGCTGGAAAGTATGATGTATATCTCGA

GATATACATCATACTTTCCAGCTTTTTG-3'). A control shRNA vector (SHC002, The MISSION^{TM} Non-Target shRNA Control Vector, Sigma), which contains a shRNA sequence that does not target human and mouse genes (>4 base pair mismatches to any known human or mouse gene), was used as a negative control for Mecp2 knockdown shRNAs. Knockdown efficiency of endogenous Mecp2 in cortical neurons was monitored by immunoblotting using polyclonal antibodies specific for Mecp2 (Millipore, 07-013).

Lentiviral vector was co-transfected with other helper vectors into HEK-293T cells as previously described (9). Packaged lentiviral particles were harvested at 48h and 72h post transfection. Harvested lentiviral particles were then combined and concentrated. For Bdnf ELISA assays, electroporated postnatal (P0) cortical neurons were transduced with control or Mecp2 shRNA expressing lentiviruses in presence of 4 μg ml^{-1} polybrene overnight and analyzed 6 days post-infection.

Statistical analyses. Paired two-tailed t-test was used for all pair-wise comparisons between WT and KO littermates. A one-way analysis of variance (ANOVA) with Dunnett's post test was used for calculating the significance between a single control and multiple treatment groups in luciferase and BDNF ELISA assays. A hypergeometric test was used to test the significance of relative enrichment of a subset of genes. Statistical analysis was performed using Prism4 (GraphPad) and R.

References for Supporting Information

1. Tao J*, et al.* (2009) Phosphorylation of MeCP2 at Serine 80 regulates its chromatin association and neurological function. *Proc Natl Acad Sci U S A* 106(12):4882-4887.
2. Martinowich K*, et al.* (2003) DNA methylation-related chromatin remodeling in activity-dependent BDNF gene regulation. *Science* 302(5646):890-893.
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5. Rumble SM*, et al.* (2009) SHRiMP: accurate mapping of short color-space reads. *PLoS Comput Biol* 5(5):e1000386.
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9. Wu H*, et al.* (2010) Dnmt3a-dependent nonpromoter DNA methylation facilitates transcription of neurogenic genes. *Science* 329(5990):444-448.
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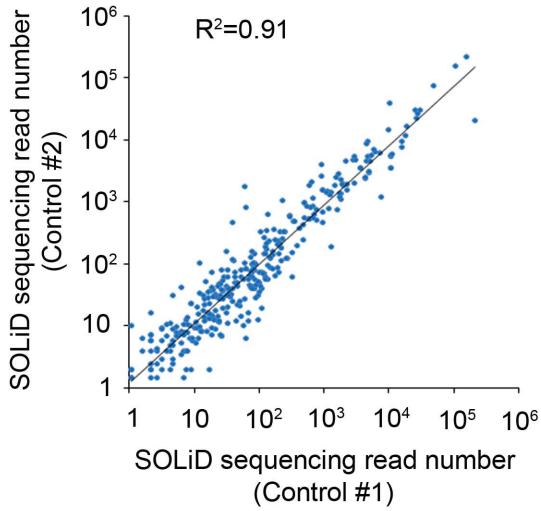


Fig. S1. SOLiD RNA-seq assays generate highly reproducible miRNA expression profiles. Shown is the scatterplot of two SOLiD small RNA sequencing experiments of microRNAs in control cerebellum tissues (postnatal 8-week).

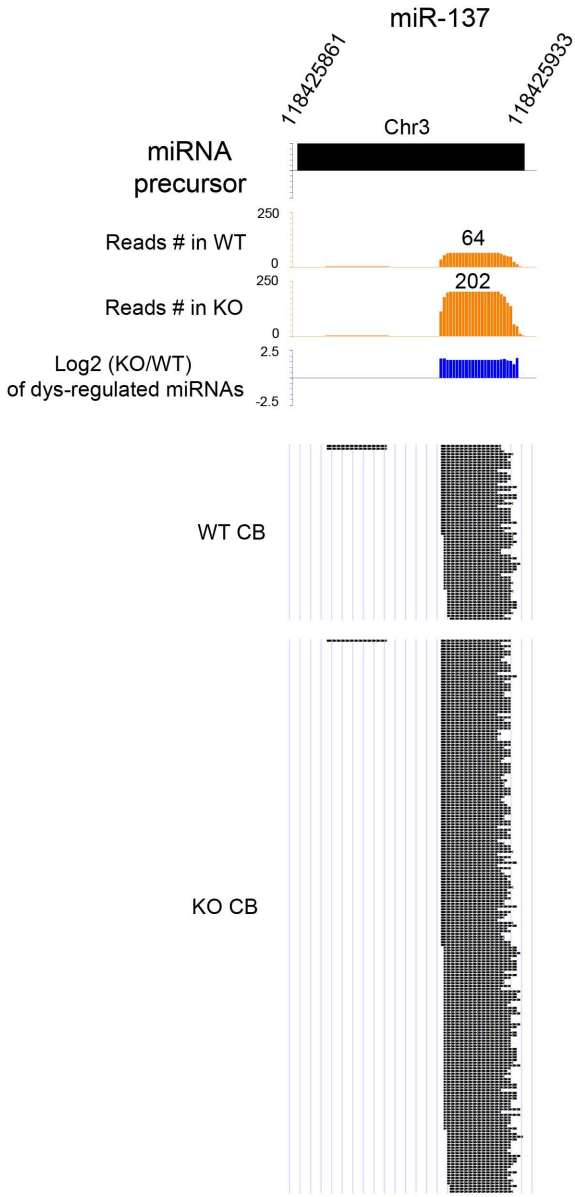


Fig. S2. Representative genomic mapping of SOLiD sequencing reads to an annotated mouse miRNA gene locus (mmu-miR-137, mm8).

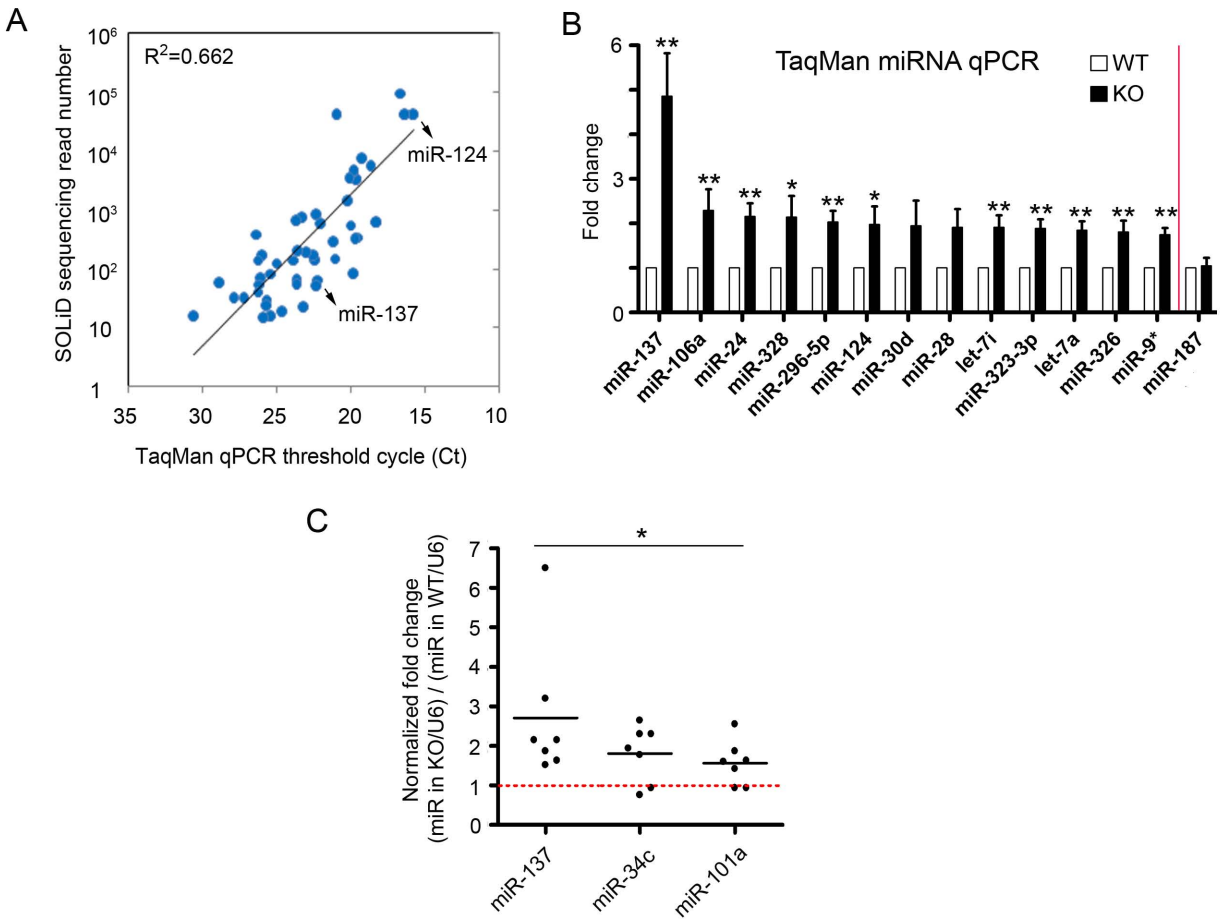
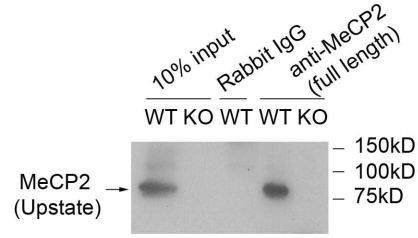


Fig. S3. TaqMan miRNA qPCR validation of SOLiD small RNA sequencing results.
(A) MiRNA expression levels measured by the SOLiD sequencing correlate well with TaqMan qPCR assays. Note that miR-124 is a highly transcribed neuronal specific miRNA.
(B) TaqMan miRNA qPCR analysis of 14 miRNAs in WT and KO cerebella at (pre-/early-symptomatic stage, 6-wk). Note that miR-187 was used as a negative control. MiRNA expression was normalized to levels of the ubiquitously expressed U6 snRNA. Error bars represent standard error of mean, s.e.m. (n=4 pairs, *<0.1, **<0.05; paired two-tailed Student's *t*-test).
(C) TaqMan miRNA qPCR analysis of 3 miRNAs in WT and KO cerebella at (symptomatic stage, 8-wk). Note that miR-137 was also up-regulated at 6-wk. MiRNA expression was normalized to levels of the ubiquitously expressed U6 snRNA. *<0.05, paired two-tailed Student's *t*-test.



Fg. S4. Validation of the antibody used for ChIP-chip of Mecp2.
Polyclonal antibodies used in ChIP-chip assays are qualified to immunoprecipitate endogeneous MeCP2 under ChIP conditions (formaldehyde cross-linking). Commercially available polyclonal antibodies to C-terminus of MeCP2 (Millipore/Upstate) was used for immunblotting of immunoprecipitated MeCP2 (~75kD). Non-specific rabbit IgG was used as a negative control. Immunoprecipitation was performed in both wild-type (WT) and MeCP2 mutant (KO) cerebellum.

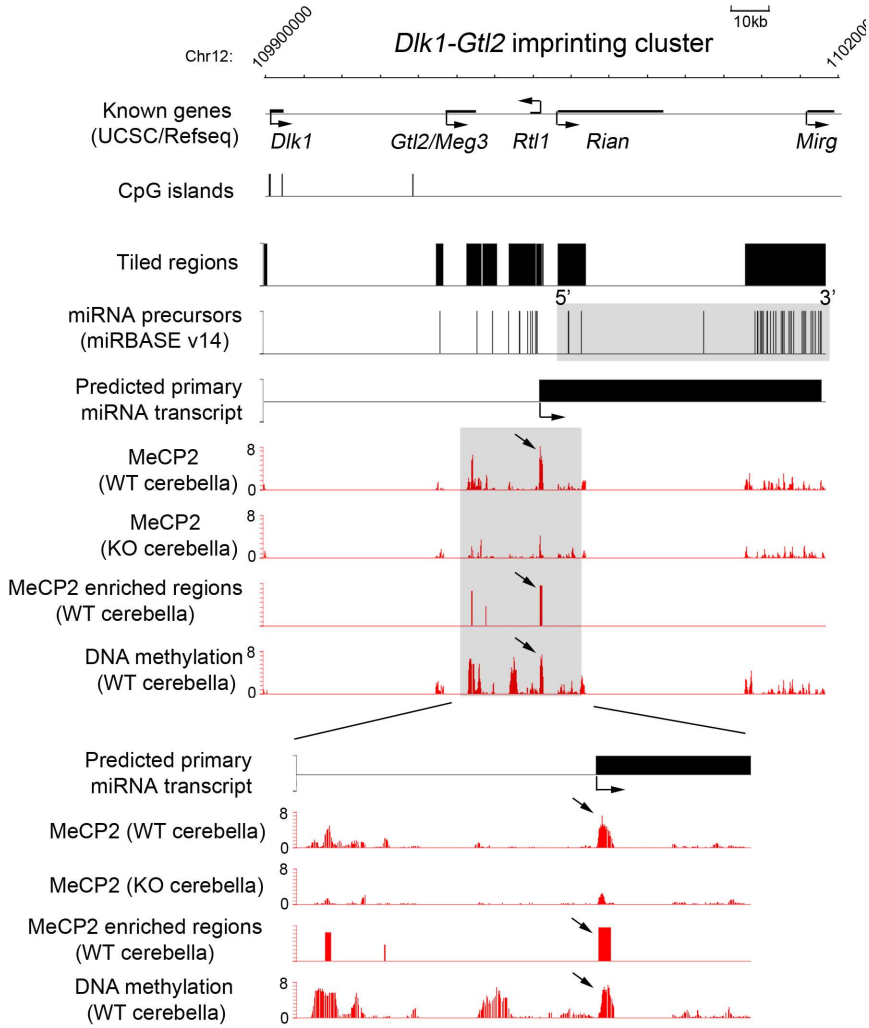


Fig. S5. MeCP2 bound and DNA methylated regions within the Dlk1-Gtl2 imprinting domain are independently validated promoter tiling microarrays.
MeCP2 occupancy and DNA methylation (-log10 (*P*-value)) within the Dlk1-Gtl2 imprinting domain were also determined by NimbleGen promoter arrays. Genomic positions for the large polycistronic cluster of miRNA are highlighted.

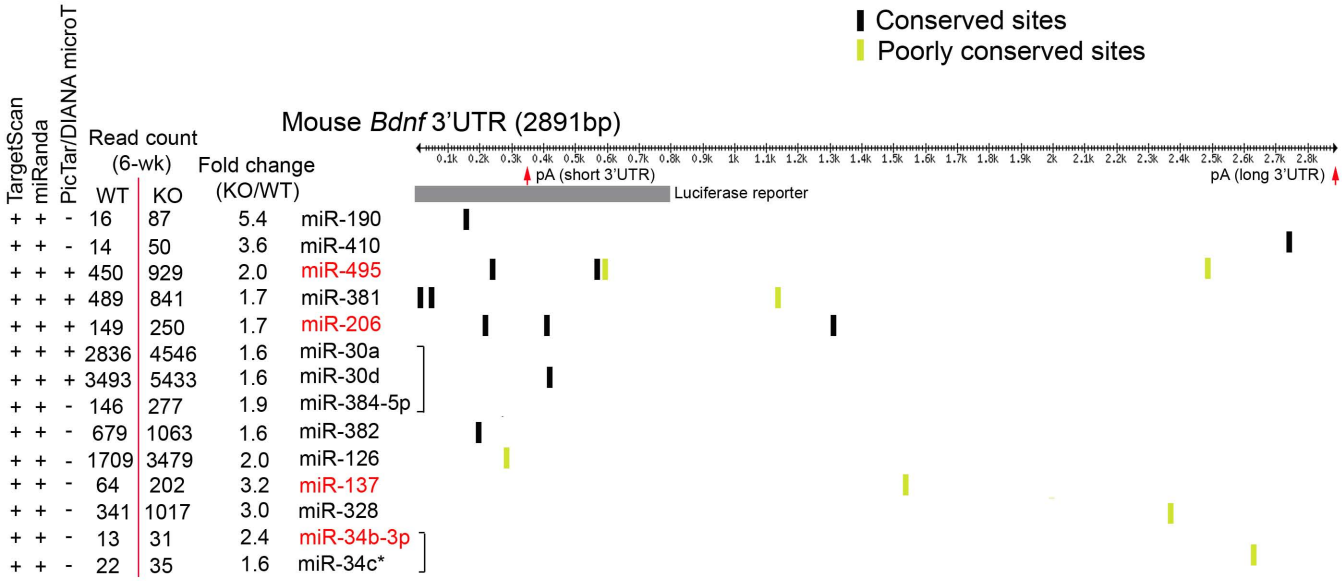


Fig. S6. Summary of SOLiD sequencing read counts for all aberrantly up-regulated miRNAs in KO cerebella that are predicted to target the full-length mouse *Bdnf* 3' UTR. Two alternative polyadenylation (pA) sites are indicated by red arrows. Also shown are aberrantly up-regulated miRNAs at the pre-/early-symptomatic stage that are predicted to target *Bdnf* 3' UTR by at least two independent algorithm (TargetScan, miRanda or PicTar/DIANA microT). MiRNAs up-regulated at both pre-/early-symptomatic (6-wk) and symptomatic stage (8-wk) are highlighted in red.

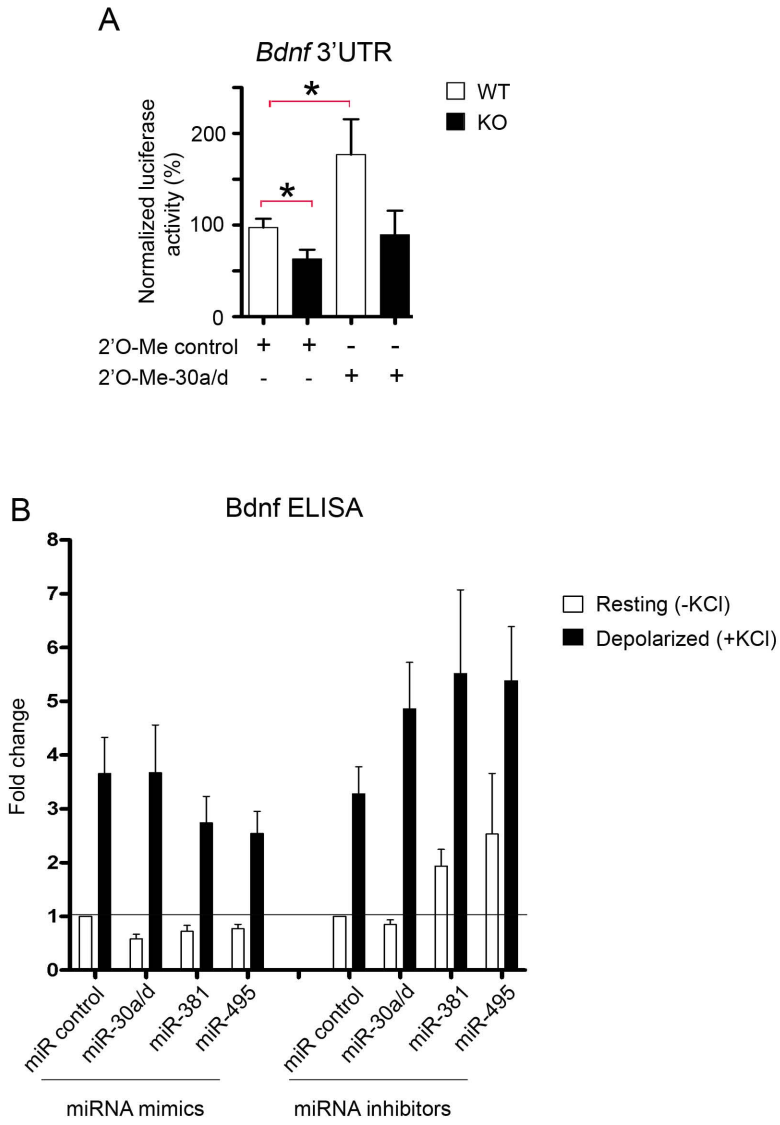


Fig. S7. Effect of dys-regulated miRNAs on the Bdnf 3'UTR and endogenous Bdnf expression level. (A) Luciferase assays in WT and KO postnatal cerebellar neurons transfected with control (100nM) or miR-30a/d (100nM) 2'O-Methyl oligonucleotide inhibitors. Error bars represents s.e.m. (*, $P<0.05$, $n\geq6$). (B) Cultured postnatal cortical neurons (wild-type) were electroporated with miRNA mimics or inhibitors for control, miR-30a/d, miR-381 and miR-495. The Bdnf protein levels in both resting and depolarized neurons were determined by ELISA assays. Error bars represents s.e.m.. of four independent experiments.

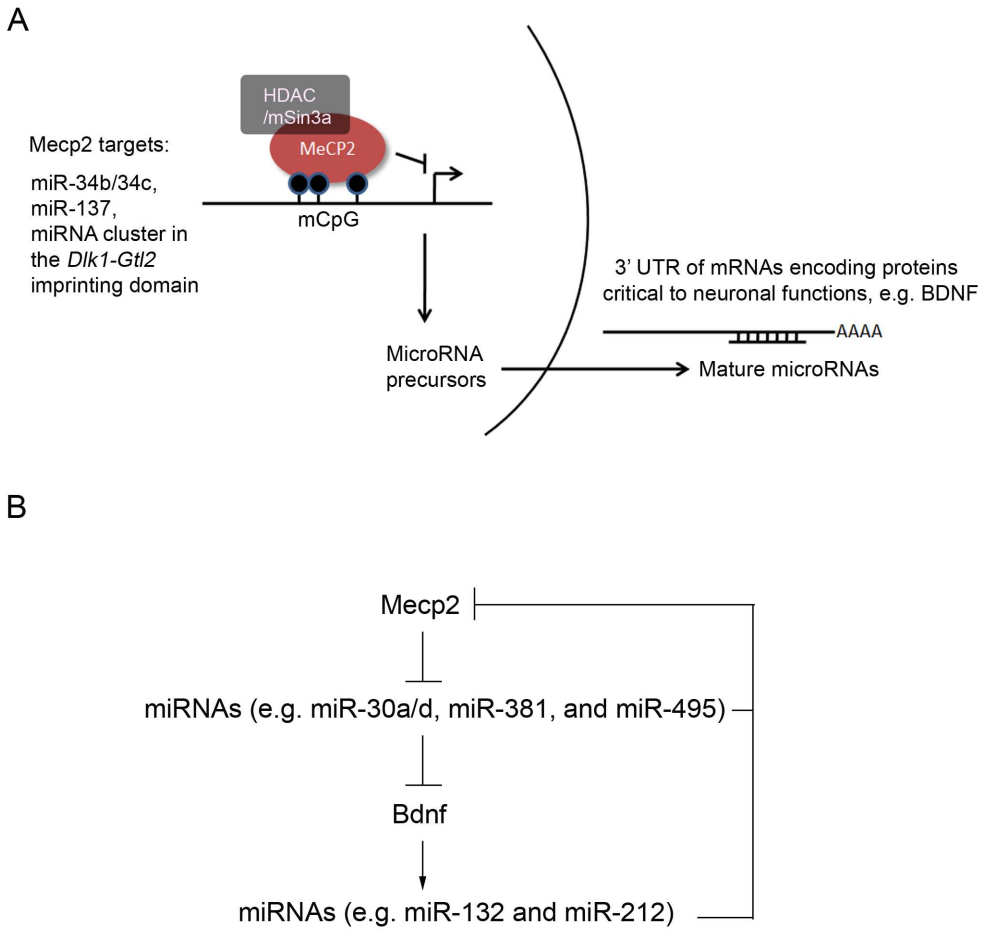


Fig. S8. Proposed models of Mecp2-dependent regulation of microRNAs (miRNAs).
(A) Mecp2 binding at DNA methylated promoters may promote transcriptional repression of a specific set of miRNAs by recruiting transcriptional co-repressor complexes (e.g. HDAC/mSin3a). In Mecp2-null brains, these miRNA targets could become aberrantly up-regulated, thereby leading to down-regulation of their targeted mRNAs (e.g. Bdnf).
(B) Mecp2 may also indirectly promote transcription of miRNAs by regulating Bdnf levels and/or neuronal activity. Note that both directly and indirectly regulated miRNAs by Mecp2 may negatively regulate Mecp2 levels through direct targeting the Mecp2 3' UTR, thereby forming a regulatory feedback loop between Mecp2 and these miRNAs.

Table S1. Summary of SOLiD small RNA sequencing results of wild-type (WT) and *Mecp2*-null (KO) cerebella samples (postnatal 6 or 8 week old)

Library	Read Length (bp)	Total number of reads	Mapping to miRBase (against miRNA precursor sequences downloaded from ftp://mirbase.org/pub/mirbase/)	Mapping to mm9 (against 23 chromosomes, no random or haplotype sequences)	Total Mappable (reads mapped to miRNAs and mm9 genome)
WT (pooled, 6-wk)	25	3,660,124	583,466 (15.94%)	N/A	583,466
KO (pooled, 6-wk)	25	2,789,136	615,915 (22.08%)	N/A	615,915
WT (litter1, 8-wk)	50	33,608,586	1,689,951 (5.03%)	363,035 (1.08%)	2,052,986
KO#1 (litter1, 8-wk)	50	34,823,537	2,051,745 (5.89%)	305,892 (0.88%)	2,357,637
KO#2 (litter1, 8-wk)	50	34,888,098	2,140,833 (6.14%)	356,674 (1.02%)	2,497,507
WT (litter2, 8-wk)	50	38,433,920	1,604,644 (4.18%)	294,404 (0.77%)	1,899,048
KO#1 (litter2, 8-wk)	50	33,039,277	1,463,075 (4.43%)	292,006 (0.88%)	1,755,081
KO#2 (litter2, 8-wk)	50	36,680,731	2,327,842 (6.35%)	305,435 (0.83%)	2,633,277

Table S2. The list of mautre miRNAs dys-regulated in postnatal 6-week *Mecp2*-null cerebella (pre-/early-symptomatic stages)

Note:

1. Column 3-6 represents the genomic coordinates of aggregated reads mapped to annotated miRNA precursors.
2. Total RNA samples derived from four pairs of WT and KO littermates were pooled and sequenced together.

Up-regulated miRNAs (6-wk, sequenced as pooled samples):

miRNA precursor	mature miRNA	chr	start	end	strand	WT_read#	KO_read#	log2(KO/WT)
mmu-mir-190	mmu-mir-190	chr9	67084504	67084527	-	16	87	2.44
mmu-mir-448	mmu-mir-448	chrX	143592825	143592849	+	15	75	2.32
mmu-mir-410	mmu-mir-410	chr12	110981975	110981998	+	14	50	1.84
mmu-mir-340	mmu-mir-340-5p	chr11	49883222	49883247	+	91	322	1.82
mmu-mir-300	mmu-mir-300*	chr12	110962529	110962555	+	10	33	1.72
mmu-mir-137	mmu-mir-137	chr3	118136820	118136845	+	64	202	1.66
mmu-mir-328	mmu-mir-328	chr8	107832274	107832300	-	341	1017	1.58
mmu-mir-1839	mmu-mir-1839-5p	chr7	88674806	88674831	+	42	119	1.50
mmu-mir-1946b	mmu-mir-1946b	chr9	21417992	21418019	-	15	39	1.38
mmu-mir-29a	mmu-mir-29a*	chr6	31012706	31012731	-	85	218	1.36
mmu-mir-125b-2	mmu-mir-125b*	chr16	77646561	77646587	+	47	117	1.32
mmu-mir-26b	mmu-mir-26b	chr1	74440899	74440924	+	548	1320	1.27
mmu-mir-322	mmu-mir-322*	chrX	50407478	50407503	-	10	24	1.26
mmu-mir-369	mmu-mir-369-3p	chr12	110981676	110981699	+	15	36	1.26
mmu-mir-34b	mmu-mir-34b-3p	chr9	50911676	50911699	-	13	31	1.25
mmu-mir-7a-1	mmu-mir-7a	chr13	58494155	58494186	-	380	883	1.22
mmu-mir-543	mmu-mir-543	chr12	110955513	110955540	+	162	369	1.19
mmu-mir-24-1	mmu-mir-24	chr13	63402559	63402584	+	7624	17053	1.16
mmu-mir-24-2	mmu-mir-24	chr8	86732774	86732801	+	7624	17053	1.16
mmu-mir-320	mmu-mir-320	chr14	70843364	70843391	+	333	708	1.09
mmu-mir-874	mmu-mir-874	chr13	58124491	58124516	-	153	322	1.07
mmu-mir-1944	mmu-mir-1944	chr15	79913323	79913349	-	31	65	1.07
mmu-mir-374	mmu-mir-374	chrX	100768421	100768444	-	13	27	1.05
mmu-mir-495	mmu-mir-495	chr12	110957006	110957027	+	450	929	1.05
mmu-mir-126	mmu-mir-126-5p	chr2	26446886	26446911	+	1709	3479	1.03
mmu-mir-33	mmu-mir-33*	chr15	82028598	82028621	+	52	105	1.01
mmu-let-7i	mmu-let-7i	chr10	122422695	122422718	-	85	169	0.99
mmu-mir-150	mmu-mir-150	chr7	52377133	52377158	+	291	572	0.97
mmu-mir-744	mmu-mir-744*	chr11	65548241	65548264	-	19	37	0.96
mmu-mir-93	mmu-mir-93*	chr5	138606759	138606784	-	33	64	0.96

mmu-mir-346	mmu-mir-346	chr14	35707810	35707836	+	53	102	0.94
mmu-mir-326	mmu-mir-326	chr7	106700839	106700864	+	169	323	0.93
mmu-mir-384	mmu-mir-384-5p	chrX	102539667	102539692	-	146	277	0.92
mmu-mir-434	mmu-mir-434-3p	chr12	110832775	110832800	+	300	559	0.90
mmu-mir-374	mmu-mir-374*	chrX	100768448	100768471	-	36	67	0.90
mmu-mir-299	mmu-mir-299	chr12	110948883	110948910	+	39	72	0.88
mmu-mir-331	mmu-mir-331-3p	chr10	93426523	93426547	-	193	354	0.88
mmu-mir-674	mmu-mir-674	chr2	117010887	117010913	+	167	306	0.87
mmu-mir-9-3	mmu-mir-9*	chr7	86650204	86650231	+	590	1080	0.87
mmu-mir-700	mmu-mir-700	chr4	134972471	134972496	-	116	212	0.87
mmu-mir-666	mmu-mir-666-5p	chr12	110955314	110955339	+	11	20	0.86
mmu-mir-140	mmu-mir-140*	chr8	110075189	110075214	+	1350	2449	0.86
mmu-let-7b	mmu-let-7b*	chr15	85537805	85537833	+	21	38	0.86
mmu-mir-497	mmu-mir-497	chr11	70048233	70048259	+	709	1279	0.85
mmu-mir-188	mmu-mir-188-5p	chrX	6825152	6825176	-	40	72	0.85
mmu-mir-28	mmu-mir-28	chr16	24827955	24827979	+	19	34	0.84
mmu-mir-124-3	mmu-mir-124	chr2	180628787	180628813	+	41767	74427	0.83
mmu-mir-124-2	mmu-mir-124	chr3	17695722	17695750	+	41771	74429	0.83
mmu-let-7e	mmu-let-7e	chr17	17967331	17967356	+	594	1058	0.83
mmu-mir-9-1	mmu-mir-9*	chr3	88019572	88019601	+	617	1091	0.82
mmu-mir-9-2	mmu-mir-9*	chr13	83878461	83878490	+	617	1091	0.82
mmu-mir-124-1	mmu-mir-124	chr14	65209545	65209573	+	42280	74760	0.82
mmu-mir-873	mmu-mir-873	chr4	36615584	36615609	-	25	44	0.82
mmu-mir-136	mmu-mir-136*	chr12	110833576	110833599	+	302	525	0.80
mmu-mir-381	mmu-mir-381	chr12	110965080	110965106	+	489	841	0.78
mmu-mir-455	mmu-mir-455	chr4	62917938	62917965	+	74	127	0.78
mmu-mir-323	mmu-mir-323-3p	chr12	110950768	110950792	+	123	210	0.77
mmu-mir-484	mmu-mir-484	chr16	14159724	14159749	+	1060	1806	0.77
mmu-mir-3470a	mmu-mir-3470a	chr6	83040345	83040375	-	30	51	0.77
mmu-mir-206	mmu-mir-206	chr1	20669137	20669162	+	149	250	0.75
mmu-mir-362	mmu-mir-362-5p	chrX	6819142	6819167	-	52	87	0.74
mmu-mir-15a	mmu-mir-15a*	chr14	62250872	62250895	-	18	30	0.74
mmu-mir-203	mmu-mir-203	chr12	113369139	113369163	+	33	55	0.74
mmu-mir-350	mmu-mir-350	chr1	178702468	178702493	-	54	89	0.72
mmu-mir-154	mmu-mir-154*	chr12	110976685	110976709	+	82	135	0.72
mmu-mir-217	mmu-mir-217	chr11	28663761	28663786	+	14	23	0.72
mmu-mir-666	mmu-mir-666-3p	chr12	110955351	110955377	+	752	1229	0.71
mmu-mir-149	mmu-mir-149	chr1	94746959	94746983	+	169	275	0.70

mmu-let-7f-1	mmu-let-7f	chr13	48633253	48633278	-	1041	1686	0.70
mmu-let-7f-2	mmu-let-7f	chrX	148346897	148346922	+	1091	1757	0.69
mmu-mir-544	mmu-mir-544	chr12	110967582	110967606	+	82	132	0.69
mmu-mir-30a	mmu-mir-30a	chr1	23279114	23279140	+	2836	4546	0.68
mmu-mir-34c	mmu-mir-34c*	chr9	50911145	50911169	-	22	35	0.67
mmu-mir-146a	mmu-mir-146a	chr11	43187932	43187957	-	24	38	0.66
mmu-let-7a-2	mmu-let-7a	chr9	41344815	41344841	+	4477	7024	0.65
mmu-mir-382	mmu-mir-382	chr12	110971992	110972018	+	679	1063	0.65
mmu-mir-296	mmu-mir-296-5p	chr2	174092592	174092613	-	23	36	0.65
mmu-mir-423	mmu-mir-423-5p	chr11	76891621	76891645	-	16	25	0.64
mmu-mir-872	mmu-mir-872	chr4	94331859	94331884	+	75	117	0.64
mmu-mir-30d	mmu-mir-30d	chr15	68172813	68172839	-	3493	5433	0.64
mmu-mir-181d	mmu-mir-181d	chr8	86702654	86702679	-	1036	1605	0.63
mmu-mir-143	mmu-mir-143	chr18	61808849	61808873	-	743	1151	0.63
mmu-mir-106a	mmu-mir-106a	chrX	50095714	50095739	-	138	213	0.63
mmu-let-7a-1	mmu-let-7a	chr13	48633582	48633628	-	4717	7278	0.63
mmu-mir-377	mmu-mir-377	chr12	110978761	110978786	+	26	40	0.62
mmu-mir-20a	mmu-mir-20a	chr14	115443406	115443430	+	54	83	0.62
mmu-mir-106b	mmu-mir-106b	chr5	138607009	138607034	-	142	218	0.62
mmu-mir-380	mmu-mir-380-3p	chr12	110950053	110950074	+	38	58	0.61
mmu-mir-341	mmu-mir-341	chr12	110849766	110849794	+	210	320	0.61
mmu-mir-299	mmu-mir-299*	chr12	110948851	110948880	+	46	70	0.61
mmu-mir-690	mmu-mir-690	chr16	28600020	28600070	-	23	35	0.61
mmu-let-7b	mmu-let-7b	chr15	85537756	85537801	+	5628	8535	0.60

Down-regulated miRNAs (6-wk, sequenced as pooled samples):

miRNA precursor	mature miRNA	chr	start	end	strand	WT_read#	KO_read#	log2(KO/WT)
mmu-mir-290	mmu-mir-290-5p	chr7	3218641	3218663	+	15	2	-2.91
mmu-mir-26b	mmu-mir-26b*	chr1	74440935	74440955	+	10	2	-2.32
mmu-mir-302c	mmu-mir-302c	chr3	127248325	127248347	+	35	8	-2.13
mmu-mir-222	mmu-mir-222	chrX	18724023	18724050	-	672	185	-1.86
mmu-mir-302a	mmu-mir-302a	chr3	127248458	127248482	+	91	28	-1.70
mmu-mir-488	mmu-mir-488	chr1	160435821	160435843	+	16	5	-1.68
mmu-mir-671	mmu-mir-671-5p	chr5	24097951	24097976	+	55	18	-1.61
mmu-mir-676	mmu-mir-676*	chrX	97576454	97576476	+	13	5	-1.38
mmu-mir-16-2	mmu-mir-16	chr3	68813841	68813866	+	3378	1319	-1.36
mmu-mir-92a-2	mmu-mir-92a	chrX	50095028	50095050	-	56	22	-1.35

mmu-mir-16-1	mmu-mir-16	chr14	62250768	62250794	-	3454	1362	-1.34
mmu-mir-144	mmu-mir-144	chr11	77886550	77886573	+	20	8	-1.32
mmu-mir-214	mmu-mir-214	chr1	164153569	164153593	+	30	12	-1.32
mmu-mir-330	mmu-mir-330	chr7	19766836	19766860	+	39	16	-1.29
mmu-mir-302d	mmu-mir-302d	chr3	127248584	127248608	+	153	63	-1.28
mmu-mir-22	mmu-mir-22*	chr11	75277237	75277261	+	308	129	-1.26
mmu-mir-652	mmu-mir-652	chrX	139173604	139173628	+	94	40	-1.23
mmu-mir-221	mmu-mir-221	chrX	18723429	18723454	-	851	373	-1.19
mmu-mir-770	mmu-mir-770-5p	chr12	110801921	110801944	+	22	10	-1.14
mmu-mir-302b	mmu-mir-302b	chr3	127248194	127248218	+	120	55	-1.13
mmu-mir-92a-1	mmu-mir-92a	chr14	115443699	115443723	+	65	30	-1.12
mmu-mir-10b	mmu-mir-10b	chr2	74564132	74564155	+	33	17	-0.96
mmu-mir-132	mmu-mir-132	chr11	74987226	74987250	+	1449	757	-0.94
mmu-mir-324	mmu-mir-324-3p	chr11	69825598	69825625	+	53	28	-0.92
mmu-mir-20b	mmu-mir-20b	chrX	50095333	50095357	-	62	33	-0.91
mmu-mir-431	mmu-mir-431	chr12	110828670	110828695	+	48	26	-0.88
mmu-mir-151	mmu-mir-151-3p	chr15	73085244	73085269	-	59	32	-0.88
mmu-mir-466j	mmu-mir-466j	chr10	60423529	60423573	+	31	17	-0.87
mmu-mir-25	mmu-mir-25	chr5	138606556	138606580	-	203	113	-0.85
mmu-mir-212	mmu-mir-212	chr11	74986945	74986971	+	69	40	-0.79
mmu-mir-140	mmu-mir-140	chr8	110075150	110075174	+	372	221	-0.75
mmu-mir-15b	mmu-mir-15b	chr3	68813698	68813722	+	138	82	-0.75
mmu-mir-709	mmu-mir-709	chr8	86610058	86610086	+	37	22	-0.75
mmu-mir-28	mmu-mir-28*	chr16	24827995	24828017	+	10	6	-0.74
mmu-mir-148a	mmu-mir-148a	chr6	51219824	51219848	-	71	43	-0.72
mmu-mir-574	mmu-mir-574-3p	chr5	65361604	65361629	+	75	46	-0.71
mmu-mir-1959	mmu-mir-1959	chr5	119975652	119975676	-	13	8	-0.70
mmu-mir-199b	mmu-mir-199b*	chr2	32174006	32174031	+	359	222	-0.69
mmu-mir-142	mmu-mir-142-3p	chr11	87570406	87570430	+	16	10	-0.68
mmu-mir-466f-4	mmu-mir-466f	chr13	71245988	71246015	+	24	15	-0.68
mmu-mir-466d	mmu-mir-466d-3p	chr2	10433644	10433689	+	11	7	-0.65
mmu-mir-29c	mmu-mir-29c	chr1	196863794	196863821	+	95636	63680	-0.59
mmu-let-7a-1	mmu-let-7a*	chr13	48633552	48633577	-	261	174	-0.58
mmu-mir-297a-5	mmu-mir-297a	chr2	10393893	10393920	+	18	12	-0.58
mmu-mir-676	mmu-mir-676	chrX	97576491	97576514	+	24	16	-0.58

Table S3. The list of mautre miRNAs dys-regulated in postnatal 8-week *Mecp2*-null cerebella (at symptomatic stages)

Note:

1. Column 3-6 represents the genomic coordinates of aggregated reads mapped to annotated miRNA precursors.
2. Column 7-12 represents normalized read count (read per million reads) for each mature miRNA of the indicated sample. A total of six small RNA samples were derived from two litters (#1 and #2, one WT and two KO in each litter) and sequenced individually.
3. Column 13 represents the mean of four log2 ratios of (read count in each KO/ averaged read count in WT of litter #1 and #2)

Up-regulated miRNAs (8-wk, sequenced as individual samples):

miRNA precusor	mature miRNA	chr	start	end	strand	WT#1	KO#1_1	KO#1_2	WT#2	KO#2_1	KO#2_2	log2(KO/WT)
mmu-mir-99b	mmu-mir-99b	chr17	17967159	17967180	+	4	125	17	4	3	6	3.22
mmu-mir-206	mmu-mir-206	chr1	20669137	20669158	+	1	6	3	2	14	4	2.19
mmu-mir-34c	mmu-mir-34c	chr9	50911177	50911204	-	12	93	274	102	380	109	1.90
mmu-mir-379	mmu-mir-379	chr12	110947276	110947296	+	1	3	3	1	6	4	1.89
mmu-mir-299	mmu-mir-299*	chr12	110948855	110948876	+	1	1	6	1	3	7	1.72
mmu-mir-874	mmu-mir-874	chr13	58124495	58124518	-	15	84	118	53	152	57	1.60
mmu-mir-30b	mmu-mir-30b*	chr15	68168993	68169015	-	2	3	8	2	8	4	1.51
mmu-mir-101a	mmu-mir-101a	chr4	101019558	101019584	-	43	31	210	73	161	214	1.42
mmu-mir-674	mmu-mir-674	chr2	117010887	117010910	+	18	25	90	37	86	89	1.42
mmu-mir-103-1	mmu-mir-103	chr11	35595946	35595978	+	10396	31995	58085	39819	110117	62482	1.39
mmu-mir-103-2	mmu-mir-103	chr2	131113836	131113870	+	10442	32110	58306	39932	110293	62658	1.39
mmu-mir-672	mmu-mir-672	chrX	101311568	101311590	-	1	10	6	10	31	10	1.34
mmu-mir-532	mmu-mir-532-5p	chrX	6825582	6825605	-	31	64	135	162	497	257	1.30
mmu-mir-455	mmu-mir-455	chr4	62917937	62917959	+	2	5	5	4	8	8	1.27
mmu-mir-30e	mmu-mir-30e*	chr4	120445223	120445245	-	1	2	1	1	3	3	1.25
mmu-mir-210	mmu-mir-210	chr7	148407307	148407328	-	1	1	3	2	3	6	1.22
mmu-mir-199a-1	mmu-mir-199a-5p	chr9	21300978	21301004	-	25	14	100	30	70	66	1.19
mmu-mir-199a-2	mmu-mir-199a-5p	chr1	164147976	164148002	+	25	14	101	31	71	66	1.17
mmu-mir-29b-1	mmu-mir-29b*	chr6	31013065	31013089	-	14	35	24	31	69	63	1.10
mmu-mir-16-1	mmu-mir-16	chr14	62250772	62250795	-	31	27	76	44	64	150	1.09
mmu-mir-199b	mmu-mir-199b*	chr2	32174006	32174030	+	170	227	781	362	705	507	1.06
mmu-mir-127	mmu-mir-127*	chr12	110831064	110831092	+	81	74	203	101	204	267	1.04
mmu-mir-665	mmu-mir-665	chr12	110824579	110824601	+	3	7	9	9	17	17	1.03
mmu-mir-543	mmu-mir-543	chr12	110955513	110955539	+	38	455	542	474	581	513	1.03
mmu-mir-137	mmu-mir-137	chr3	118136820	118136843	+	149	266	328	122	217	285	1.01
mmu-mir-34a	mmu-mir-34a	chr4	149442581	149442611	+	922	2219	2816	2600	3436	5645	1.00
mmu-mir-146b	mmu-mir-146b	chr19	46417281	46417304	+	14	14	26	22	53	48	0.96
mmu-mir-377	mmu-mir-377	chr12	110978761	110978785	+	19	50	72	39	45	60	0.95
mmu-mir-7a-1	mmu-mir-7a*	chr13	58494161	58494186	-	231	1595	1407	1079	1134	899	0.94

mmu-mir-16-2	mmu-mir-16	chr3	68813841	68813863	+	27	16	51	37	56	123	0.94
mmu-mir-15a	mmu-mir-15a	chr14	62250908	62250934	-	27	97	105	56	59	54	0.94
mmu-mir-27a	mmu-mir-27a	chr8	86732627	86732651	+	2	4	6	2	2	3	0.89
mmu-mir-744	mmu-mir-744*	chr11	65548245	65548266	-	2	8	7	6	3	12	0.89
mmu-mir-495	mmu-mir-495	chr12	110957006	110957028	+	6	31	49	41	49	43	0.86
mmu-mir-708	mmu-mir-708*	chr7	103398007	103398029	+	3	8	12	9	11	10	0.80
mmu-mir-19b-1	mmu-mir-19b	chr14	115443580	115443605	+	334	614	1001	491	568	668	0.79
mmu-mir-34b	mmu-mir-34b-3p	chr9	50911679	50911701	-	4	2	9	4	8	7	0.78
mmu-mir-19b-2	mmu-mir-19b	chrX	50095168	50095192	-	348	624	1012	498	575	681	0.77
mmu-mir-1839	mmu-mir-1839-3p	chr7	88674845	88674869	+	5	14	16	31	53	38	0.75
mmu-mir-181b-2	mmu-mir-181b	chr2	38709365	38709394	+	1674	3379	2840	2869	4429	4578	0.74
mmu-mir-211	mmu-mir-211	chr7	71350718	71350740	+	3	5	5	2	1	7	0.73
mmu-mir-338	mmu-mir-338-5p	chr11	119876130	119876154	-	41	117	89	115	178	129	0.72
mmu-mir-19a	mmu-mir-19a	chr14	115443271	115443294	+	85	120	218	80	89	113	0.71
mmu-mir-340	mmu-mir-340-3p	chr11	49883264	49883293	+	61	634	1139	1780	3418	789	0.70
mmu-mir-181b-1	mmu-mir-181b	chr1	139863227	139863256	+	1511	2804	2254	2482	3943	3928	0.70
mmu-mir-24-2	mmu-mir-24-2*	chr8	86732739	86732761	+	4	13	14	11	13	11	0.69
mmu-mir-488	mmu-mir-488*	chr1	160435783	160435805	+	2	2	8	4	5	4	0.69
mmu-mir-15a	mmu-mir-15a*	chr14	62250874	62250912	-	24	48	51	47	60	69	0.68
mmu-mir-199a-1	mmu-mir-199a-3p	chr9	21300943	21300965	-	16	23	36	16	26	18	0.67
mmu-mir-199a-2	mmu-mir-199a-3p	chr1	164148014	164148036	+	16	23	36	16	26	18	0.67
mmu-mir-199b	mmu-mir-199b	chr2	32174044	32174066	+	16	23	36	16	26	18	0.67
mmu-mir-666	mmu-mir-666-5p	chr12	110955314	110955341	+	9	15	22	23	34	29	0.66
mmu-mir-29a	mmu-mir-29a*	chr6	31012708	31012733	-	84	101	169	104	184	137	0.65
mmu-mir-382	mmu-mir-382*	chr12	110972026	110972050	+	14	34	32	28	28	37	0.64
mmu-let-7e	mmu-let-7e	chr17	17967331	17967354	+	131	181	180	340	547	558	0.64
mmu-mir-434	mmu-mir-434-5p	chr12	110832738	110832760	+	18	61	53	73	108	62	0.63
mmu-mir-425	mmu-mir-425*	chr9	108471162	108471186	+	7	13	3	4	8	10	0.63
mmu-mir-107	mmu-mir-107	chr19	34895186	34895227	-	4727	6968	9436	9401	15867	10939	0.61

Down-regulated miRNAs (8-wk, sequenced as individual samples):

miRNA precusor	mature miRNA	chr	start	end	strand	WT#1	KO#1_1	KO#1_2	WT#2	KO#2_1	KO#2_2	log2(KO/WT)
mmu-mir-331	mmu-mir-331-3p	chr10	93426528	93426549	-	25	7	1	7	2	8	-1.82
mmu-mir-10a	mmu-mir-10a	chr11	96178501	96178522	+	9	1	3	2	2	1	-1.70
mmu-mir-365-2	mmu-mir-365	chr11	79539970	79539992	+	88	26	17	29	17	24	-1.48
mmu-mir-365-1	mmu-mir-365	chr16	13453989	13454011	+	88	27	17	29	17	24	-1.46
mmu-mir-326	mmu-mir-326	chr7	106700839	106700863	+	163	38	18	84	72	63	-1.37
mmu-mir-143	mmu-mir-143	chr18	61808853	61808875	-	26	9	7	7	8	6	-1.19
mmu-mir-378	mmu-mir-378*	chr18	61557527	61557551	-	103	38	16	43	30	48	-1.15
mmu-mir-382	mmu-mir-382	chr12	110971992	110972018	+	196	70	32	86	62	93	-1.14
mmu-let-7f-1	mmu-let-7f	chr13	48633250	48633280	-	10694	3449	5392	3603	1	5530	-0.99
mmu-mir-361	mmu-mir-361	chrX	110188472	110188498	-	955	420	170	460	243	607	-0.97
mmu-mir-329	mmu-mir-329	chr12	110951752	110951775	+	123	45	47	57	36	57	-0.96
mmu-mir-140	mmu-mir-140	chr8	110075149	110075174	+	68	28	28	56	34	37	-0.96
mmu-mir-340	mmu-mir-340-5p	chr11	49883222	49883244	+	136	67	41	57	17	76	-0.94
mmu-let-7a-1	mmu-let-7a*	chr13	48633555	48633579	-	24	11	10	12	7	10	-0.92
mmu-let-7c-2	mmu-let-7c-2*	chr15	85537095	85537117	+	23	11	10	12	7	11	-0.83
mmu-mir-328	mmu-mir-328	chr8	107832277	107832302	-	128	91	38	216	131	138	-0.79
mmu-mir-193b	mmu-mir-193b	chr16	13449664	13449688	+	23	19	15	45	19	25	-0.78
mmu-mir-708	mmu-mir-708	chr7	103397961	103397987	+	127	85	63	148	95	87	-0.73
mmu-mir-125a	mmu-mir-125a-5p	chr17	17967782	17967814	+	27614	18887	12841	27130	20355	15513	-0.70
mmu-mir-98	mmu-mir-98	chrX	148347773	148347796	+	202	109	115	146	86	131	-0.66
mmu-mir-29c	mmu-mir-29c	chr1	196863781	196863822	+	1717	1866	923	1367	659	546	-0.63

Table S4. The list of other non-coding small RNAs (ncRNAs) dys-regulated in postnatal 6-week *Mecp2*-null cerebella

ncRNA_name	WT_read# (rpm)	KO_ read# (rpm)	Log2(KO/WT)
chr5.trna1609-AspGTC	443	1695	1.94
SNORD45	745	2350	1.66
chr3.trna293-HisGTG	2174	6265	1.53
chr13.trna1041-LysCTT	523	1474	1.49
chr8.trna886-GlyGCC	1892	5012	1.41
chr10.trna856-AspGTC	25283	63016	1.32
chr13.trna113-GlnTTG	684	1695	1.31
chr6.trna157-CysGCA	12400	28597	1.21
chr11.trna1432-CysGCA	11554	26386	1.19
chr11.trna1433-CysGCA	11554	26386	1.19
chr9.trna593-CysGCA	11554	26386	1.19
chr11.trna791-CysGCA	11554	26386	1.19
chr11.trna1442-CysGCA	12078	27565	1.19
chr17.trna458-CysGCA	12078	27565	1.19
chr13.trna69-AlaAGC	8052	18278	1.18
chr11.trna1823-IleAAT	1369	3096	1.18
chr13.trna956-IleAAT	1369	3096	1.18
chr13.trna968-IleAAT	1369	3096	1.18
chr13.trna989-IleAAT	1369	3096	1.18
chr13.trna990-IleAAT	1369	3096	1.18
chr6.trna162-CysGCA	684	1548	1.18
chr13.trna100-IleAAT	1369	3096	1.18
chr11.trna393-IleAAT	1369	3096	1.18
chr12.trna470-IleAAT	1369	3096	1.18
chr13.trna92-IleAAT	1369	3096	1.18
chr13.trna68-AlaTGC	8092	18278	1.18
chr13.trna66-AlaAGC	7971	17836	1.16
SNORD57	1824	4060	1.15
snoU97	122	270	1.14
chr13.trna988-PheGAA	403	884	1.14
chr11.trna1444-CysGCA	3140	6412	1.03
chr6.trna175-CysGCA	3301	6707	1.02
chr19.trna108-PheGAA	443	884	1.00
chr10.trna961-PheGAA	483	958	0.99
chr14.trna457-PheGAA	483	958	0.99

chr5.trna1315-PheGAA	483	958	0.99
chr13.trna60-PheGAA	483	958	0.99
chr19.trna106-PheGAA	483	958	0.99
chr7.trna967-GluCTC	1530	2948	0.95
chr11.trna550-ThrCGT	805	1548	0.94
chr6.trna1005-CysGCA	2738	5233	0.93
SNORD38	178	337	0.92
chr10.trna1316-LeuTAA	2214	4127	0.90
SNORD127	189	337	0.84
SNORD22	334	585	0.81
SNORD60	2681	4656	0.80
SNORD91	3849	6477	0.75
chr15.trna913-MetCAT	1932	3243	0.75
chr8.trna783-MetCAT	1932	3243	0.75
chr6.trna1023-CysGCA	443	737	0.73
chr11.trna1493-GlnTTG	2174	3611	0.73
chr3.trna753-GlnCTG	2416	3906	0.69
SNORD44	701	1113	0.67
chr3.trna756-GlnCTG	2577	4054	0.65
chr11.trna1817-GlnCTG	2577	4054	0.65
chr3.trna297-GlnCTG	2577	4054	0.65
chr13.trna64-GlnCTG	2577	4054	0.65
chr9.trna961-GluTTC	8374	13119	0.65
chr13.trna105-GluTTC	8374	13119	0.65
chr14.trna347-GluTTC	8374	13119	0.65
chr7.trna337-GluTTC	8374	13119	0.65
chr13.trna982-GlnCTG	2496	3906	0.65
chr9.trna342-GlnCTG	2496	3906	0.65
chr1.trna1547-GluTTC	8414	12824	0.61
chr14.trna359-GluTTC	8414	12824	0.61
chrX.trna637-AlaTGC	3825	5823	0.61
SNORD115	43136	65639	0.61
chr13.trna979-AspGTC	49760	75472	0.60
chr15.trna355-MetCAT	684	1032	0.59
chr3.trna309-GlnCTG	2456	3685	0.59
chr11.trna2022-AlaTGC	1329	1990	0.58
chr3.trna754-GluTTC	1208	811	-0.58
chr3.trna750-AsnGTT	2093	1400	-0.58

chr11.trna1911-LeuCAA	3865	2580	-0.58
snR78	979	652	-0.59
chr13.trna998-LeuCAA	3865	2506	-0.63
chr13.trna65-LeuCAA	3865	2506	-0.63
chr13.trna85-LeuCAA	3865	2506	-0.63
snoU6-77	278	180	-0.63
chr8.trna168-LeuCAG	3664	2358	-0.64
SNORD52	512	326	-0.65
SNORD110	1190	753	-0.66
RSV_RNA	6852	4318	-0.67
snoR38	1824	1147	-0.67
SNORD88	2770	1732	-0.68
SNORD75	7775	4847	-0.68
SNORD90	434	270	-0.68
SCARNA4	5517	3430	-0.69
SNORD24	1224	742	-0.72
SNORD113	53170	30925	-0.78
SNORD101	679	394	-0.79
SNORD103	5873	3351	-0.81
U2	1513	843	-0.84
SNORD111	222	124	-0.85
SNORD71	790	427	-0.89
SNORD96	645	349	-0.89
chr7.trna861-LeuTAG	3019	1548	-0.96
RNaseP_nuc	2291	1102	-1.06
chr17.trna113-GlyCCC	100125	47317	-1.08
chr6.trna317-GlyCCC	100125	47317	-1.08
SNORD121A	1613	675	-1.26
SNORD73	979	405	-1.27
SNORD69	356	146	-1.28
chr11.trna945-ArgCCG	2134	737	-1.53
SNORA7	990	304	-1.71
SNORD63	13726	3531	-1.96
snoU2-25	1301	304	-2.10
SNORD41	20345	4206	-2.27

Table S5. The list of miRNA transcript units whose proximal (1-kb) or distal (5-kb) promoter regions are directly targeted by MeCP2 in postnatal cerebella

Note:

1. MiRNA TSS (column 4) and strand (column 5) indicates the predicted transcription start sites (TSSs) of primary miRNA transcripts and their transcription orientation, respectively (Marson et al. , 2008).
2. Column 1-3 represent Mecp2 bound regions (peaks) that are within 5kb genomic regions flanking TSSs of predicted miRNA transcripts.
3. Up-regulated and down-regulated miRNAs in KO cerebella at 6-week are highlighted in red and green, respectively.
4. Mature miRNA consistently dys-regulated at both 6-week and 8-week are underlined.
5. Mecp2 binding sites that are within predicted miRNA proximal promoters (-1kb to +1kb relative to TSS) are highlighted in bold.

PEAK_Chr	PEAK_START	PEAK_END	miRNA TSS	strand	Primary miRNA transcripts
chr9	50854658	50854917	50855975	-	mmu-mir-34c/mmu-mir-34b
chr9	**50856963**	**50857515**	50855975	-	mmu-mir-34c/mmu-mir-34b
chr9	50860174	50860778	50855975	-	mmu-mir-34c/mmu-mir-34b
chr9	106009049	106009221	106006787	+	mmu-mir-135a-1
chr9	**112080066**	**112080415**	112079730	-	mmu-mir-128b
chr9	112081666	112082915	112079730	-	mmu-mir-128b
chr8	**87072122**	**87072430**	87071400	-	mmu-mir-181d/mmu-mir-181c
chr8	87072122	87072430	87075322	-	mmu-mir-181d/mmu-mir-181c
chr7	18338786	18339344	18335227	+	mmu-mir-330
chr7	**18338786**	**18339344**	18339655	+	mmu-mir-330
chr7	44993472	44993821	44989813	+	mmu-mir-150
chr7	64029627	64030076	64032877	+	mmu-mir-211
chr7	96043287	96043844	96046435	+	mmu-mir-708
chr7	99406318	99407171	99409568	+	mmu-mir-326
chr7	99407769	99408318	99409568	+	mmu-mir-326
chr7	**101294682**	**101294831**	101295518	+	mmu-mir-139
chr5	24094893	24095242	24099289	+	mmu-mir-671
chr5	138398958	138399207	138401650	-	mmu-mir-25/mmu-mir-93/mmu-mir-106 (removed from other analysis)
chr5	139628870	139629219	139626575	-	mmu-mir-339
chr4	40909644	40910158	40911203	+	mmu-mir-207
chr4	40909853	40910002	40911203	+	mmu-mir-207
chr4	120330456	120331105	120329210	-	mmu-mir-30c-1/mmu-mir-30e

chr3	17983112	17983844	17986735	+	mmu-mir-124-2
chr3	88296498	88296973	88293150	+	mmu-mir-9-1
chr3	88296498	88296973	88300421	+	mmu-mir-9-1
chr3	**118423690**	**118424950**	118425726	+	mmu-mir-137
chr3	118427238	118428285	118425726	+	mmu-mir-137
chr3	127538252	127538401	127537000	+	mmu-mir-302b/mmu-mir-302c/mmu-mir-302a/mmu-mir-302d/mmu-mir-367
chr19	22203263	22203814	22206213	+	mmu-mir-204
chr18	61528119	61528191	61525800	-	mmu-mir-378
chr18	61771246	61771767	61774675	-	mmu-mir-143
chr18	61771246	61771767	61774500	-	mmu-mir-145
chr16	13362868	13363386	13360250	+	mmu-mir-365-1/mmu-mir-193b
chr16	24307390	24307639	24308838	+	mmu-mir-28
chr16	24311990	24312647	24308838	+	mmu-mir-28
chr16	93255794	93256148	93258000	+	mmu-mir-802
chr16	**93258662**	**93259713**	93258000	+	mmu-mir-802
chr15	73248197	73248456	73250468	-	mmu-mir-151
chr15	73252297	73252746	73250468	-	mmu-mir-151
chr15	81972908	81973260	81974458	+	mmu-mir-33
chr15	**103100732**	**103100981**	103100932	+	mmu-mir-148b
chr14	113924383	113925008	113921800	+	mmu-mir-17/mmu-mir-18a/mmu-mir-19a/mmu-mir-20a
chr13	114152046	114152238	114155325	+	mmu-mir-449b/mmu-mir-449a
chr13	**114154008**	**114154961**	114155325	+	mmu-mir-449b/mmu-mir-449a
chr13	114159582	114159836	114155325	+	mmu-mir-449b/mmu-mir-449a
chr12	109285806	109286426	109284453	+	mmu-mir-345 mmu-mir-341/mmu-mir-370/mmu-mir-882/mmu-mir-379/mmu-mir-411/mmu-mir-299/mmu-mir-380/mmu-mir-323/mmu-mir-758/mmu-mir-329/mmu-mir-494/mmu-mir-666/mmu-mir-543/mmu-mir-495/mmu-mir-368/mmu-mir-654/mmu-mir-376b/mmu-mir-376a/mmu-mir-300/mmu-mir-381/mmu-mir-487b/mmu-mir-539/mmu-mir-544/mmu-mir-382/mmu-mir-134/mmu-mir-485/mmu-mir-453/mmu-mir-154/mmu-mir-496/mmu-mir-377/mmu-mir-541/mmu-mir-409/mmu-mir-412/mmu-mir-369/mmu-mir-410
chr12	**110044260**	**110045917**	110043900	+	
chr11	**70047944**	**70048138**	70048625	+	mmu-mir-497/mmu-mir-195

chr11	87568072	87568249	87572275	+	mmu-mir-142
chr11	119865144	119865793	119863040	-	mmu-mir-338
chr10	**122390142**	**122390855**	122391350	-	mmu-let-7i
chr1	160194099	160194748	160199016	+	mmu-mir-488
chr1	195213122	195213659	195211200	-	mmu-mir-205

Table S6. The list of transcriptionally detectable miRNAs generated from the polycistronic transcript within the *Dlk1-Gtl2* imprinting domain

Note:

1. Mature miRNAs significantly up-regulated (log2 (KO/WT)>0.58) at the pre-/early-symptomatic stage (6-week) are highlighted in bold.
2. Mature miRNAs consistently dys-regulated (>1.5-fold) at both 6-week and 8-week are underlined.
3. Columns 2-4 represent genomic coordinates of clustered sequencing reads mapped to guide and passenger strand of annotated miRNA precursors.

miRNA_precursor	chr	start	end	strand	WT_read_count	KO_read_count	FC(KO/WT)	log2(KO/WT)
mmu-miR-341	chr12	110849766	110849794	+	210	320	1.52	0.61
mmu-miR-370	chr12	110856477	110856506	+	30	39	1.30	0.38
mmu-miR-370	chr12	110856515	110856541	+	32	34	1.06	0.09
mmu-miR-379	chr12	110947276	110947301	+	212	255	1.20	0.27
mmu-miR-379	chr12	110947313	110947336	+	29	34	1.17	0.23
mmu-miR-411	chr12	110948400	110948425	+	77	104	1.35	0.43
mmu-miR-411	chr12	110948436	110948459	+	67	59	0.88	-0.18
mmu-miR-299	chr12	110948851	110948880	+	46	70	1.52	0.61
mmu-miR-299	chr12	110948883	110948910	+	39	72	1.85	0.88
mmu-miR-380	chr12	110950053	110950074	+	38	58	1.53	0.61
mmu-miR-323	chr12	110950768	110950792	+	123	210	1.71	0.77
mmu-miR-329	chr12	110951714	110951739	+	323	242	0.75	-0.42
mmu-miR-329	chr12	110951752	110951777	+	191	143	0.75	-0.42
mmu-miR-494	chr12	110953578	110953603	+	45	64	1.42	0.51
mmu-miR-666	chr12	110955314	110955339	+	11	20	1.82	0.86
mmu-miR-666	chr12	110955351	110955377	+	752	1229	1.63	0.71
mmu-miR-543	chr12	110955479	110955502	+	31	33	1.06	0.09
mmu-miR-543	chr12	110955513	110955540	+	162	369	2.28	1.19
mmu-miR-495	chr12	110957006	110957027	+	450	929	2.06	1.05
mmu-miR-667	chr12	110958231	110958256	+	16	13	0.81	-0.30
mmu-miR-667	chr12	110958272	110958297	+	99	137	1.38	0.47
mmu-miR-376c	chr12	110960943	110960969	+	76	65	0.86	-0.23
mmu-miR-376c	chr12	110960981	110961005	+	78	115	1.47	0.56
mmu-miR-376b	chr12	110961681	110961706	+	282	276	0.98	-0.03
mmu-miR-376b	chr12	110961718	110961743	+	143	163	1.14	0.19
mmu-miR-376a	chr12	110961997	110962021	+	29	41	1.41	0.50
mmu-miR-376a	chr12	110962034	110962059	+	307	379	1.23	0.30
mmu-miR-300	chr12	110962529	110962555	+	10	33	3.30	1.72
mmu-miR-300	chr12	110962574	110962600	+	516	702	1.36	0.44
mmu-miR-381	chr12	110965040	110965064	+	28	20	0.71	-0.49

mmu-miR-381	chr12	110965080	110965106	+	489	841	1.72	0.78
mmu-miR-487b	chr12	110965593	110965619	+	580	700	1.21	0.27
mmu-miR-539	chr12	110966347	110966370	+	18	23	1.28	0.35
mmu-miR-544	chr12	110967582	110967606	+	82	132	1.61	0.69
mmu-miR-382	chr12	110971992	110972018	+	679	1063	1.57	0.65
mmu-miR-382	chr12	110972028	110972053	+	104	153	1.47	0.56
mmu-miR-134	chr12	110972356	110972380	+	112	104	0.93	-0.11
mmu-miR-668	chr12	110972984	110973008	+	160	178	1.11	0.15
mmu-miR-154	chr12	110976649	110976674	+	192	184	0.96	-0.06
mmu-miR-154	chr12	110976685	110976709	+	82	135	1.65	0.72
mmu-miR-496	chr12	110977376	110977401	+	80	102	1.28	0.35
mmu-miR-377	chr12	110978761	110978786	+	26	40	1.54	0.62
mmu-miR-541	chr12	110980633	110980658	+	638	703	1.10	0.14
mmu-miR-541	chr12	110980675	110980700	+	17	20	1.18	0.23
mmu-miR-409	chr12	110981383	110981408	+	59	80	1.36	0.44
mmu-miR-409	chr12	110981414	110981441	+	196	170	0.87	-0.21
mmu-miR-369	chr12	110981642	110981666	+	118	147	1.25	0.32
mmu-miR-369	chr12	110981676	110981699	+	15	36	2.40	1.26
mmu-miR-410	chr12	110981975	110981998	+	14	50	3.57	1.84