

Non-CpG methylation by DNMT3B facilitates REST binding and gene silencing in developing mouse hearts

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Supplemental Tables and Captions

Table S1. List of primers or oligos using in qRT-PCR, bisulfite sequencing, EMSA, qChIP and luciferase assays

Assay	Primer sequence (sense 5'-3')	Primer sequence (anti-sense 5'-3')
qRT-PCR		
<i>Gapdh</i>	ACGGCAAATTCAACGGCACAGTCA	TGGGGGCATCGGCAGAAGG
<i>Hcn1</i>	CTTCAGTTCCTGGTTCCCCT	GGAATATTGTTTTCCCCAGGA
<i>Hcn2</i>	CTGTCGGATGGCTCCTATTT	TATTCCTCCAGCACCTCGTT
<i>Hcn3</i>	GGCTGCTGAGTGTGCTGG	GGTGTCAGCCCTTACACTGG
<i>Hcn4</i>	GGCGCCAGTACCAGGAGA	AACATCTTGCCCTTGGTAGCG
<i>Rest</i>	GTGCGAACTCACACAGGAGA	GAGGTTTAGGCCCGTTGTGA
<i>Nppa</i>	ATTGACAGGATTGGAGCCCAGAGT	TGACACACCACAAGGGCTTAGGAT
<i>Nppb</i>	AGGGATACAGGAGCTGCTGG	CCTCGCCTCCCAGGCTTCTG
<i>Dnmt3a</i>	CTGCAAGAGTGTCTGGAGCA	GGAAGCCAAACACCCTTTCC
<i>Dnmt3b</i>	GATGAGGAGAGCCGAGAACG	CAGAGCCCACCCTCAAAGAG
Bisulfite sequencing		
Control region	GGTTGGTGGGTTTTTAGAAGTATTT	AATCATTAACCTACATCCCTTACAC
<i>Hcn2</i> -RE1	GGATGTAGGTTAATGATTTTATTTG	TAACCTAAACCCCTAAACATCTTCC
qCHIP		
<i>Hcn2</i> -RE1	CCCGAAAACCTGTAGCTTGAAG	ACTGGAGAAGGGACAAGCAG
EMSA		
Wild type	TTCAGGACCACGGACAGCTC	
CpG Me+	TTCAGGACCAC ^m GGACAGCTC	
Non CpG Me+1	TTC ^m AGGAC ^m C ^m ACGGACAGCTC	
Non CpG Me+2	TTCAGGACCACGGAC ^m AGC ^m TC ^m	
Non CpG Me+1&2	TTC ^m AGGAC ^m C ^m ACGGAC ^m AGC ^m TC ^m	
Luciferase assay		
<i>Hcn2</i> -RE1 Wildtype	CAATGGTACCCGTGTTTCGGTGGAT	CCCTCTCGAGAAAGGTGGCATTCTGG
	CAA	TC
<i>Hcn2</i> -RE1 Mutant	GGAGCCGCCACAGGTCCTACTTCA	ACTGTAGCTTGAAGTAGGACCTGTGG
	AGCTACAGTTTT	CTCC

Table S2: List of primers used in qRT-PCR verification for DEGs found in RNA-seq

Genes	Primer sequence (sense 5'-3')	Primer sequence (anti-sense 5'-3')	Product (bp)
<i>Abcc8</i>	CAAGGTGTCCTCAACAACGG	CCAGGTGCTATGGTGAATGTG	141
<i>Ache</i>	TGGTCACCATGCAGAGGATCT	AGGTAGCGGTTGAACTTCTCC	115
<i>Acs16</i>	AAGTGACAGAGAGTCAGTGGG	TAGGGCGGAGAGCCTTCAT	177
<i>Acta1</i>	CCCAAAGCTAACCGGGAGAAG	GACAGCACCGCCTGGATAG	89
<i>Aebp1</i>	TTGGAAACGCTGGATCGGTTA	CTTGACCTTGCCAGGCATTT	103
<i>Afp</i>	ACCTTCCTGTCTCAGTCATTCT	CCTGACATCCAGGTAGATTTC	93
<i>Ankrd1</i>	ATGCCCAAATCTGGGTTTACA	TCACCTCGGTCAATTTTGAAGT	140
<i>Ap3b2</i>	TGGCCTGTAAGAACATAGAGGT	AGATGGACAGTAGAGCGAGGT	93
<i>Aplp1</i>	CACCAGGAGCGTATGGACC	ACACCTCGGAACCGATCAGA	137
<i>Atp2b2</i>	CAGAGCCGAATTGAGCAGGAG	CCCCTGTGAGTGAGCTTTCAT	184
<i>Bdh1</i>	TTCCCCTTCTCCGAAGAGC	CCCAGAGGGTGCATCTCATAG	151
<i>Bex2</i>	TCCAAAGTGGAACAAGGCGT	GCACGTAGTAGTCTCCAGCTTC	142
<i>Bmp6</i>	AACGCCCTGTCCAATGACG	ACTCTTGCGGTTCAAGGAGTG	138
<i>Bsn</i>	AGGCATCAATAACGGAATCACTC	GTCGTGGGCTACTTTGTCCAA	124
<i>Cacna1a</i>	TTGAGGCTGGAATTAAGATCGTG	CTCAGTGTCCGTAGGTCAAAC	148
<i>Cacna2d2</i>	CCGCCTCTGTTGCTTTTACC	ACGCCTCCAAAAATCCGCA	137
<i>Cadm3</i>	GGGGCCAATCTTTCCAGG	TGCAGAGATGAGTCTTCATGGT	116
<i>Cadps</i>	CTGTACGTGTTTCGTGATGCG	GTCTCCCCATTGAGAAAAGCC	143
<i>Ccdc64</i>	CCTAGTGGCGATGAAGGATTC	CTCCTCGTTGCGTTCCAGAA	174
<i>Ccnd2</i>	GAGTGGGAAGTGGTAGTGTTG	CGCACAGAGCGATGAAGGT	154
<i>Cdkn1a</i>	GAAAGAAGCGGAAGATCCTCC	GGGCCTCAGGGATTGTTTGG	124
<i>Celf3</i>	CCCATCTTCGAGCAGTTTGGT	CGAGCACAGTACGTCAGGAAA	101
<i>Ckmt1</i>	ACCCAGCAGTCTATGCAC	GCCGCTCTTGGATCACAGG	187
<i>Clic5</i>	GTGAAGGCTGGGATCGACG	CCAGGTTGTGTAGATCGGCT	142
<i>Clu</i>	CCATCTGCAACTAGCTGTGAG	TCCGTTTTCTTCGGAAGTAAGAC	120
<i>Clvs1</i>	AAAAAGCTCGTCTGGAAGTGA	ACGTAAAAACCCAATGTCTGGT	104
<i>Cpeb1</i>	GGACTCCCGTTCTAGCAGC	GGAAATGCGAAGGCTTGAAATCA	97
<i>Crmp1</i>	ACGCCGATGTCTACCTAGAAG	CATCAATGCCACCGGGAATGA	120
<i>Ctgf</i>	GGGCCTCTTCTGCGATTTC	ATCCAGGCAAGTGCATTGGTA	151
<i>Diras2</i>	CAAGCTCAACCACAACGTGAA	CTTGCCGTCGATCTGGAGG	88
<i>Dlgap3</i>	GAGAGTCCTAGCCGCATCC	GGCCCGTTATAGTCTCGCTT	101
<i>Dpp6</i>	GGGGAGTAATCCTCCACAGAG	GACAGACTGGTATCTTCCGCT	126

<i>Drd2</i>	ACCTGTCCTGGTACGATGATG	GCATGGCATAGTAGTTGTAGTGG	105
<i>Edn1</i>	GAGCGCGTCGTACCGTATG	ACTGACATCTAACTGCCTGGT	103
<i>Elavl2</i>	ACACAGCCAATGGTCCAACC	ATCTCGCCAATGCTCCCCAAA	157
<i>Elavl3</i>	ATGGTCACTCAGATACTGGGG	TTCTGGGGTAGGTAGTTGACG	146
<i>Enox1</i>	GCTGGATTTGAGAGCGTTGC	GGTGGTATCTAGGGCTATGCT	96
<i>Fam155a</i>	CCAGGGGTGCTTGGATGTG	TTTCTGAGCCCGCTCTGAATC	107
<i>Fbxl16</i>	CTTGCCTCGAAACGGTCTG	CCGGGATAGTGGTGTAGCC	157
<i>Fgf12</i>	CAGGCCGTGCATGGTTTCTA	TCGTGTAGTGATGGTTCTCTGT	139
<i>Fhl2</i>	ATGACTGAACGCTTTGACTGC	CGATGGGTGTTCCACACTCC	142
<i>Fn1</i>	AGGCCCCTAGACTTTAAGCAA	CTGAGCAGCATCCAAGTTCAT	159
<i>Fosl2</i>	CACGCCGAGTCCTACTCCA	GTGGGCTGTACCATCCACTG	137
<i>Gabrb3</i>	CACGCTTGACAATCGAGTGG	GCGGATCATGCGGTTTTTCAC	109
<i>Gdap1l1</i>	CACCACAGATTCCATGATCCC	TCTTCAGGTAGCTCACGTCATC	185
<i>Gnao1</i>	TTGGGCGTGGAGTATGGTG	GCAGAGAACGGTTCAGTGTCTT	95
<i>Gnptg</i>	AACACATTCGGGCTGAATAACC	GGACTCCACTAGGCTAAAGCAC	132
<i>Golga7b</i>	CCACTGAGGTCCACAATCTTC	CTCTGGCGGGAATTTGGTCTG	122
<i>Gpr19</i>	CTGATGGGATTGCATGAGGAG	GAACAACCACAAAGCGCCAA	117
<i>Gprin1</i>	TGGGTCTGTATCGGTGAGAAT	GGTCCTGTACTGTTGGAAGATG	102
<i>Hcn2</i>	CCGGCGTCAACAAGTTCTC	TGCCACGGAATGATAATGA	180
<i>Hcn3</i>	CAGCCGACGGTCAACAAGTT	GTAAAACCGGAAGTCGCTGTAG	126
<i>Hrc</i>	TGGTTGCACACTTGTCTCCTT	ATGCCTGCATTGTTGTTCCAG	119
<i>Igsf9b</i>	TCAGCACCCGAGGTCTTGT	GATTACGTCGCATCGCAGAAC	107
<i>Kcnb1</i>	AGAAACACACAGCAATAGCGT	GTACTCCCGTGGAGACTCTTG	130
<i>Kcnh7</i>	CATGTGGCACCGCAAAACAC	TAGCACAGTTCTGCACTCTGG	103
<i>Kcnk3</i>	ACGGAGGCAAGGTGTTCTG	ACGACACGAAACCGATGAGC	189
<i>L1cam</i>	AAAGGTGCAAGGGTGACATTC	TCCCCACGTTCTGTAGGT	104
<i>Lad1</i>	GACACCGCGAGCTGTATCTG	CAAGGTCTAAGAACTCCAGGTGA	150
<i>Lin28a</i>	GGCATCTGTAAGTGGTTCAACG	CCCTCCTTGAGGCTTCGGA	146
<i>Lpar3</i>	CACCATGCGGAAGATGATCTG	GAGCCGTTTTTATTGCACACC	156
<i>Lppr2</i>	CCCTTGCTTCGTCTTTGTGGA	CGGGATACGGCTTGGCATA	140
<i>Lrp11</i>	GAGGACAAGCCTCCCGTTAG	TCCGAGCTTTCTCGACCATCT	92
<i>Lrp3</i>	AGCACACTGAACGTCGAGG	TCACCACGGTCTCCTTGAATG	100
<i>Lrrc16b</i>	GTTTGATCCGGCGTGGAATG	ACACACACTGTGAGTGGTAGAT	95
<i>Lrrc24</i>	CCTGCACAACAATACCCTGC	CAGCCAGGTAAAGCACACGAA	152
<i>Madd</i>	GACGAGCTGTCGGCATTATG	TCAATGGCGTCTGCTCAAAAT	112

<i>Man2b2</i>	AAAGCATGAGAGCCTATGCAG	GCCTCCGAGAACAAACTCCA	194
<i>Maneal</i>	AGGTGGCCTTCCATATCCAAC	GGGAGCCATACGTGTCAATGA	90
<i>Map1b</i>	TCGCACCGCTTCCTAGACA	CTGGTCCAAGTTGCACTCAAT	147
<i>Mapk8ip1</i>	CACTCCCATCGAGACCGAATC	TCTGGATCGGAGCTAACTGAC	161
<i>March11</i>	CGAAACACGCTCCGTATGTAG	CACATCGGCAAGGATTTAACAAC	152
<i>Meis3</i>	CATCCAGGTACTCCGGTTCCA	TAGCGGTGACAGAAGTTGTCTG	75
<i>Mgat5b</i>	GAGACCCTTTCGGCTGTTTGT	CCAGCATATCCATGCGCTTC	209
<i>Myo16</i>	AGGGGACAGAATCTAGTGCTATC	CGTTGACATCTCCGCCTGA	147
<i>Ncan</i>	GCTGGGGATCAGGACACAC	CAGTCTGAACCTTAGTCCACTTG	187
<i>Nefh</i>	GTTCCGAGTGAGGTTGGACC	CCGCCGGTACTCAGTTATCTC	94
<i>Nefl</i>	CGCCGCTCTCAAGGACATC	ACAGCGTCGGTGTTCTTGG	129
<i>Nefm</i>	CTCGGCTATGCTCAGCTCG	CGGGACAGTTTGTAGTCGCC	96
<i>Nppa</i>	ATTGACAGGATTGGAGCCCAGAGT	TGACACACCACAAGGGCTTAGGA T	133
<i>Nppb</i>	AGGGATACAGGAGCTGCTGG	CCTCGCCTCCCAGGCTTCTG	118
<i>Nptxr</i>	GGGCAGGCCAATGAGATCG	GTCCAGGAGATGCAGATATGGT	131
<i>Nrxn1</i>	AACGGACTGATGCTTCACACA	GATATTGTCACCTGACGCAGATT	194
<i>Ogdhl</i>	GGATGCAGACCTAGATTTCCTTG	GGCAGCCGGAACCTCCTTAT	165
<i>Pcbp3</i>	CCCTGAGCTACATTTTGGAGG	GCATCAGTAGGCGGATGGT	86
<i>Pex5l</i>	ATGGAAGCTGCAATTCTTCAGG	CTCTGGAGGGCGACAATAGC	110
<i>Pgbd5</i>	GAAGTTCCAGGAGCGGTTTG	GTACCGACTCACAGTGCAG	116
<i>Phactr1</i>	TTTTGAAGCCTCGGAATGAACA	TTCGGTCATAGTCTTGGGCAT	171
<i>Phactr3</i>	AAGGCCGGGAGGAACCTCAT	GGCTCACTCTGCACAGGTT	109
<i>Phf21b</i>	AGAACGGCGACCTCAAGAAG	TCAGAGTCTTTGGCCTAACCT	180
<i>Ppfia3</i>	GCTGCTAAGTGAATCCAACGA	GCTGGTCGATCTCCATCTGC	182
<i>Ppip5k1</i>	AAAGCTGTGGCTTACTCCAAG	CGGTACACCTCCCTCCTATCTT	92
<i>Prmt8</i>	ACGTGGTAGCAATCGAAGACA	GCTCCTTCATGGCAACATCC	108
<i>Prune2</i>	GCTGAACGATGAAGGGAAACT	TCTCCGGGGTTAATGACTCTG	108
<i>Psd</i>	ATGTCCTCCAAGCTATTGACCG	TGAGGTGCCTCTTCGAGACAA	146
<i>Ptprn</i>	TGTTTGACCGCAGACTTTGT	GGAGCACACCTTGTAGGCG	141
<i>Qsox2</i>	AGCAATGGTTCTTATGTTGGACG	TGCTTTGTCCGTATCCAGTGC	93
<i>Rasl10b</i>	GACAGCTTTGAGTACGTCAAGA	TAGCCGCACTTCCAGGTCT	173
<i>Rest</i>	GTGCGAACTCACACAGGAGA	GAGGTTTAGGCCCGTTGTGA	199
<i>Rgs7</i>	GCGGACGAATCACCCAACAT	TTTGACGGTACGGATGGGAAT	99
<i>Ric3</i>	GGAAACGCCACAGGAAGATT	ACCATTAGGTCCAACCTCTGTTGA	114
<i>Rimbp2</i>	TTGGTGACTATATTCCGCTGTCT	GTCCATCTCGGACTCAAATCTG	110

<i>Rims2</i>	CTGCTCAGCTAGTGGGACG	CCCGGATGATTTCTACCTCCAG	105
<i>Rims3</i>	GGAATGTAGTAAGGAGTTCCAGC	CTGGGTCAAGCCGACGATAG	140
<i>Rpl36al</i>	ATGGTCAACGTACCAAAAACCC	TGGGCATACAGGGAATCCTTG	107
<i>Scn8a</i>	AAGAAGCTGTCAGACGTGATG	TCGTTGAAGTTTATGGGCCAC	125
<i>Sez6l2</i>	GGAGGATGAGATGATGCCAGA	GGTGGCTAGTGTGGGGTCT	148
<i>Slc8a2</i>	ATGGCTCCCTTGGCTTTGATG	CAGCGGTAGGAACCTTGGC	146
<i>Smpd3</i>	ACACGACCCCTTTCCTAATA	GGCGCTTCTCATAGGTGGTG	129
<i>Snap25</i>	GCAAGGCGAACAACCTGGAAC	GGCCACTACTCCATCCTGATT	187
<i>Snch</i>	GGAAGGCGTCCTCTATGTCG	CGTGGCTGCTGCAATGTTC	139
<i>Soga3</i>	TGGACGAGATGAGGGATACTTT	GCTCGCCATCAATTTCCCCA	168
<i>Sost</i>	AGCCTTCAGGAATGATGCCAC	CTTTGGCGTCATAGGGATGGT	134
<i>Sox9</i>	CGGAACAGACTCACATCTCTCC	GCTTGACAGTCGGTTTTGG	163
<i>Spon1</i>	GACCAAGTCGGAGGGCTACT	CTGCTTCCTGGCTTATAGAAGTC	120
<i>Spry4</i>	CCCGCTGTGACCAGGATATTA	GCCATGTGATCTAGGAGCCTC	112
<i>Srpk3</i>	CAAGGGTGGTTACTACCCAGT	GCGCTGAATATCCCAGCAGAG	118
<i>Sult4a1</i>	GACGTGTGGATCGTCACCTAC	TGCTCGTCAATGTTTCATCAGG	116
<i>Svop</i>	AGTTGCCGGTGGTGAAATTC	CATGAAAGTGTCATCAGTGGGAT	179
<i>Syn1</i>	AATATGCGGCTATGGATGTTGG	TCCTGAGTGACCGGAGAGTC	102
<i>Syp</i>	CTCGGCTTTGTGAAGGTGCT	CTGAGGTCACTCTCGGTCTTG	122
<i>Sypl2</i>	CGCACCTCGGACAAGTCTC	CCCGAAGGCGAAAATAGCAAA	129
<i>Syt14</i>	TCCAGAGGCGGTTGGATTTTT	TCCGTGTATTGTATCCTGAACCA	140
<i>Syt7</i>	CCTTACCAGCGAGATGCTCAT	TCTTGGAAGTTGTAGCCAACAC	117
<i>Tmem179</i>	GCTCCTTTGGTGGTAACTGTC	CCAGCCACGAAGTAGCAGA	106
<i>Tmem198</i>	CTCACCCAAGGCCCTTTAACC	GCAGTAGATGATTCCGAAGCAG	134
<i>Trim67</i>	CCACTCTCTGCGAGCAATG	GAAGTAGCCGATGCTTAGCGA	96
<i>Tspan17</i>	GCTGCTGCGGGAAATACTTC	ACCACCACAAACAGCCACA	169
<i>Unc13a</i>	GCTGTGCGTGGGAGTCAAA	CAGCTATGGTAGTGCTCTTCAC	104
<i>Unc5a</i>	CTCCTGGGCATAGTCCTCACT	CAGCACGGGCTTGTCTTG	156
<i>Vwf</i>	CTCTTTGGGGACGACTTCATC	TCCCGAGAATGGAGAAGGAAC	115
<i>Xkr4</i>	ACTTCGCGGATGTGGGAAC	CCAGCGGAAGCTGAACACTT	134
<i>Xkr7</i>	GCGCTGCTCGTGTCTTCT	TGAGGCCGAAGTAGGTGCT	91
<i>Zbtb8b</i>	CCTATTCGCCAACAGTGGCTA	TCCGAGGTGACGATGTCCA	99

Table S3: List of primers used in qChIP

Genes	Primer sequence (sense 5'-3')	Primer sequence (anti-sense 5'-3')	Product (bp)
<i>Abcc8</i>	GCACTCGGGATTCCATCTTC	TTCCACGGACTGCCCTCTAT	206
<i>Cadm3</i>	ACAAGAAGTGCCTGGAGTGA	TCCGACGTAAAGTTCTGTGAGC	126
<i>Cacna1a</i>	CACTGCCAGATGTGGGTCTTATT	CTTCCTTCCGTCCTTCCTTGAT	134
<i>Ccdc64</i>	CGCCAGCAACGCCAGCTCCTCCT	CACTGCTGCCGCCCTCGTCTCCTT	82
<i>Cdkn1a</i>	TGTGACGAGGGTCAGGCTATGTC	GGGCACCGTGAGCTTCATTCT	210
<i>Celf3</i>	CCTGTCTGCCCTTCAACCTG	GCCACGCCCTCTTTCTATCTC	354
<i>Ckmt1</i>	ATAGACGGATTGGGTTTCAGATTTA	GAGGACAGCACCACAGGGATT	86
<i>Clic5</i>	GGTGAGCCAGAAAGAAAGG	TAAGCAGCACTCCAGAAGC	113
<i>Crmp1</i>	GGTCGGACTACCAAGGAGGA	TGAGAAAGTGAACCCAGAGGAA	230
<i>Ctgf</i>	AAATTCTTCCCAGGATGTG	TTCCCTCCAATAGATGAC	291
<i>Diras2</i>	GGACCATGAGACTCCTGTGC	GCTCCTGCTTCCTGACCTT	198
<i>Dpp6</i>	ACAGGCAGAGGGCAAAGC	TGTGAGACCCAGACCAGAATG	205
<i>Edn1</i>	ATCTGATGCCGCCAACCA	GTGAAGTGCTCAGCCACAAA	213
<i>Elavl3</i>	ACATTTGCTGGAGCGGGATT	TTAGGCTAGGTCTTGTGATTCTGG	132
<i>Fam155a</i>	CCATTTCAGCACCCACGGA	GATTAGCAAAGGGGAAGTCT	93
<i>Fgf12</i>	CTTCTAGGCAAGGAAAGGC	TGCTTACCATAACTGGGAACT	140
<i>Fhl2</i>	GCAAGTCGGCGAGGAAAG	CAAGCCAGAGGAAGCAATC	149
<i>Fosl2</i>	TACCCATGCAGCACAGC	AGTCAAGGGAGAAACATAAGGA	219
<i>Gabrb3</i>	AGGCTTCTTTGGTGGGTGTC	CTGTAGGGCTACTGATCTCCTCT	218
<i>Gdap1l1</i>	GTTTCCGCTTTGTAGGAGTGA	AACGAGTTCTGGGCTCTGG	141
<i>Gpr19</i>	TTCCCAATTCTCAACCT	GTCCTCCTAAGTGAAGTAAGATAA	270
<i>Gprin1</i>	TTCAGGACCACGGACAGC	TGCCAGAAAGCCAATAATACTC	177
<i>Hcn2</i>	CCCGAAAAGTGTAGCTTGAAG	ACTGGAGAAGGGACAAGCAG	80
<i>Hcn3</i>	GGGGACAGTGAAATGGCTACA	ATCAGGGACGCTGGGAACG	208
<i>Kcnb1</i>	TCCGTCCCGTCGAGTGCTGA	GCAAGCCCAATGCCATCGTTAT	183
<i>Kcnk3</i>	TCCAGGCTGAACATCCCAAGT	CTCCCACAAGCAAGCTGTGAAT	286
<i>L1cam</i>	TCAGAGGGATTGGAGGCA	GAAGGGTCAGGGAGAAAGC	177
<i>Lin28a</i>	CTGAAGGGAAGAAGAAAGACG	CAGCATGTGCCATGAAGC	327
<i>Lpar3</i>	TGCTCGCACTGCTCAACTCC	TGTGAGACAGGTGAGGACGCTTA	292
<i>Lrp11</i>	GGAGACCGACAGACCTGCGGAAGT	CGAGCGGCCTCTACGTGGATCA	113
<i>Lrp3</i>	CAAACCTCCCAGGATTGTGCC	TGCTCTGGTGGCTATTGCTT	169

<i>Lrrc24</i>	ATCCGTGGGACGCAAAGCC	AGAGGTGAGCCCAGGGTGGTTG	123
<i>Maneal</i>	GGTGCTGCCCCGTGGAAGTGT	GCAAGACTCCGGTGCTAATGTCG	111
<i>March11</i>	AGCAGTAGTGGCGGCGGTAG	GAGGGAAGCTCAGTTGTGGG	203
<i>Meis3</i>	TCCGACCCGCAGTCCCAGAT	CCAGGGCTACATTCGCTCTTT	222
<i>Ncan</i>	CTTCAGCCCTCCCTGTTTCTA	TGGCCTGGATTGGCATTAG	206
<i>Nefh</i>	ATACGGCTCGTTCTTGGTTTGG	CGCACAGAGGGACTGCTTGG	177
<i>Nefm</i>	GCTGTGACGCCCAAGGAT	TGGAGGAGATGAGCCAGAAGT	120
<i>Nppa</i>	AGGGCAGAAGGCAGTATTTG	ATAGTGGCTGAGACTAAGGGTGT	249
<i>Nppb</i>	GGAGAGAGCTTGAAAGTTGGT	ACTGCACTACTTCCGGGTAC	85
<i>Nptxr</i>	CCCTGAAAGGACATGGATTGGC	CCTGGGCGCTTAGATGGAGAAT	115
<i>Nrxn1</i>	GGAAGGAAGAGGCGGTGGT	GGTGGGGTGGCGAATGAC	284
<i>Pex5l</i>	GGAGCTGACCCTCAGGAAGTGT	TGTAGGCTAAGGCAGGAAAGAAG	114
<i>Ppip5k1</i>	GTTCGGGAATCCCTGTGGTG	CAAGGAGCCAGCCAGAGCA	242
<i>Ptpn</i>	CCCTCACTCCATTCAGATAGATTC	CCAAGGCGTTGTCATTACCG	232
<i>Rasl10b</i>	TTGAAAGGCAGAGGCAGGAA	GCAGTGAGCACCCAAGAGG	298
<i>Rgs7</i>	CAAGCTAACTGCACAAGCCTGGAACA	TTGGCCGAGGAGGTAACCGGAGGGA	320
<i>Ric3</i>	CTTTACATACACGGCTCATCAA	CTGGGAGAACGGACTGGA	332
<i>Rimbp2</i>	TATCAATTACAGAGCAGGAAGGGTC	CAGAGTTTGGGTAGGCTTGTGG	213
<i>Rims2</i>	CCCAGCATCCTGTCTTCTGTACCTCT	CGGGAGGATCTCAGGTGGCTG	252
<i>Rims3</i>	ACAATGAAGGGTGAGGAGTTTG	CAGCCAGCACAGGAAGGAA	115
<i>Scn8a</i>	GCCTGGCTGATGGAGAAG	GATTAAGAACGTAGAGGGATTTGT	258
<i>Sez6l2</i>	CGGTGCTCTGGAAGTGCAGTC	TTTGTAAAGGAGCCTGATGGGTATGTG	330
<i>Slc8a2</i>	GGAGCCAATATGAGGAGGCG	AGGGACTGGGATACCACAGCA	241
<i>Snap25</i>	AGGTTTGGTGCGGTGGGC	GTCTTTCTTTTAATCTACTATGCGAGCT	224
<i>Sncb</i>	GGTCGGTAATCCTGAGTTCTG	GAATGTGGACCAGGGTTTGT	270
<i>Soga3</i>	TCCATACCCAAAGGGGCTGTTCC	TGCCTCTTCCGGCTGTCCG	117
<i>Spry4</i>	CCAGGAGTGTTGGTGGTGC	GAGGGAGGAAGGAGAAAGGAA	304
<i>Svop</i>	TGGGCATGACCCTTACATGGTGG	TGGCAGCGGCTGGCTTTGTC	144
<i>Syn1</i>	GCACACTCAGAGGGGAACAT	CTCTTTTCTTTGCCCCGACAG	97
<i>Syp</i>	GACATGGACGTGGTGAATCAGGTACG	CCTTCGCTGCGGCAAAGAGC	229
<i>Sypl2</i>	CGGGAATGCTAAGTAAATCA	GGCAGAAGGGTTAAGGGTC	189
<i>Tmem179</i>	AGGCCACCGGACAGGGAC	CAAGCTGACTTAGTAATGCTCACGAC	294
<i>Trim67</i>	CCTGGGACGCTGCTCAATACCG	CCGCTGCCCCGAGTCTTCTTTCT	293
<i>Tspan17</i>	CGACACTGCCTGTAATCCC	CTCGGTTCCCTTCCTGCTT	384
<i>Unc13a</i>	CGTCCACGCAGAACCTACCTCCCA	CGAGGCTGAGCCAGAAAGCAAAGG	294

<i>Unc5a</i>	ACTTTGCCAGGATTGTGAGA	TGCTCTTGTGATAGCGGTTC	106
<i>Xkr4</i>	GCTGTTGGTGGCGGCGATGT	GGTGCTGGGCTCCCTTTCTGTG	229
<i>Mapk8ip1</i>	CGGGGTTCGGACTGAGC	AAGACGGACTTGGGGAGCAGGA	304
<i>Gabrb3</i>	AGGCTTCTTTGGTGGGTGTC	CTGTAGGGCTACTGATCTCCTCT	218

Table S4: List of primers used in bisulfite sequencing

Gene	Primer sequence (sense 5'-3')	Primer sequence (anti-sense 5'-3')	Product (bp)
<i>Fgf12</i>	AAAGAAAGTTTTTTTGTGTTTTTTT	ATACTAAAATCAACCCCACCC	223
<i>Gabrb3</i>	TGTAGGGTTATTGATTTTTTTT	TCAACCTCTATTTAAAAACCC	191
<i>Kcnb1</i>	TGGGAGGAGATGAGAATTTATTTAA	CCCAAAAACTCCCAAAAAAC	192
<i>Mapk8ip1</i>	GGAATGTTTTTGGGATGTGTAG	ACTAAAATCTTATAACCC CCCAACACCTAACTAACTACACT	191
<i>Nppb</i>	TTTTATAAGTTTTTGAGGGGG	A CTCTAATCCCAACCAAACATCTCT	192
<i>Snap25</i>	ATGTGTTGTAAAGTGGGAGAGTTTT AAGTTAGTTTAAGGAATAGGAAGA	A TCAATCAACCTTTAATACTAAAAA	196
<i>Syn1</i>	G	A	259
<i>Unc5a</i>	AAATTTGTTTTAAGTTGGGTA	TCTAAACCTCAATTTTATATCTACA	231

Table S5. qRT-PCR verification of differential expression genes (DEGs) in E12.5 ventricles from RNA-seq between wild type and myocardial *Rest* or *Dnmt3b* knockout (KO) embryos (n=3). * $p<0.05$ by the Student's t test and Mann-Whitney test, as appropriate. Nc, no change.

Gene name	Wild type Mean±SD	<i>Rest</i> KO		<i>Dnmt3b</i> KO	
		Mean±SD	Regulation	Mean±SD	Regulation
<i>Abcc8</i>	1.00±0.14	2.70±0.57*	Up	5.37±0.77*	Up
<i>Ache</i>	1.00±0.16	1.86±0.05*	Up	0.87±0.13	Nc
<i>Acsf6</i>	1.00±0.14	2.69±0.10*	Up	0.94±0.08	Nc
<i>Acta1</i>	1.00±0.24	3.45±0.20*	Up	1.42±0.19	Nc
<i>Aebp1</i>	1.00±0.09	2.52±1.72	Nc	1.72±0.28	Nc
<i>Afp</i>	1.00±0.35	0.44±0.16*	Down	0.50±0.41	Nc
<i>Ankrd1</i>	1.00±0.13	1.67±0.24*	Up	1.12±0.13	Nc
<i>Ap3b2</i>	1.00±0.21	3.05±0.47*	Up	1.89±0.91	Nc
<i>Aplp1</i>	1.00±0.06	1.44±0.18*	Up	0.55±0.33	Nc
<i>Atp2b2</i>	1.00±0.15	0.84±0.40	Nc	2.36±1.05*	Up
<i>Bdh1</i>	1.00±0.09	0.82±0.05	Nc	2.26±0.54*	Up
<i>Bex2</i>	1.00±0.15	1.58±0.13*	Up	1.56±0.38	Nc
<i>Bmp6</i>	1.00±0.19	1.62±0.27*	Up	1.18±0.08	Nc
<i>Bsn</i>	1.00±0.05	1.12±0.96	Nc	3.37±1.07*	Up
<i>Cacna1a</i>	1.00±0.30	3.51±4.61*	Up	3.84±1.02*	Up
<i>Cacna2d2</i>	1.00±0.14	1.49±0.28	Nc	1.20±0.17	Nc
<i>Cadm3</i>	1.00±0.40	3.39±0.35*	Up	2.32±0.83*	Up
<i>Cadps</i>	1.00±0.32	2.44±0.64*	Up	1.65±0.44	Nc
<i>Ccdc64</i>	1.00±0.17	4.38±0.44*	Up	1.89±0.20*	Up
<i>Ccnd2</i>	1.00±0.59	4.83±1.74*	Up	1.06±0.19	Nc
<i>Cdkn1a</i>	1.00±0.25	2.58±0.75*	Up	3.66±0.79*	Up
<i>Celf3</i>	1.00±0.53	3.67±0.78*	Up	3.20±0.75*	Up
<i>Ckmt1</i>	1.00±0.30	3.59±0.63*	Up	1.78±0.15*	Up
<i>Clic5</i>	1.00±0.22	0.52±0.19*	Down	3.23±0.80*	Up
<i>Clu</i>	1.00±0.29	1.45±0.23	Nc	4.14±2.07*	Up
<i>Clvs1</i>	1.00±0.10	2.56±0.29*	Up	0.61±0.38	Nc
<i>Cpeb1</i>	1.00±0.39	1.99±0.24*	Up	0.71±0.32	Nc
<i>Crmp1</i>	1.00±0.06	2.52±0.34*	Up	0.48±0.06*	Down
<i>Ctgf</i>	1.00±0.04	2.44±0.49*	Up	4.50±0.65*	Up
<i>Diras2</i>	1.00±0.22	0.59±0.41	Nc	2.61±1.16*	Up
<i>Dlgap3</i>	1.00±0.14	1.72±0.37*	Up	1.49±0.22	Nc
<i>dnmt3b</i>	1.00±0.17	0.78±0.24	Nc	0.39±0.16*	Down
<i>Dpp6</i>	1.00±0.17	4.85±0.25*	Up	6.35±2.06*	Up
<i>Drd2</i>	1.00±0.20	1.50±0.13	Nc	1.81±2.29	Nc

<i>Edn1</i>	1.00±0.13	2.14±0.37*	Up	2.13±0.31*	Up
<i>Elavl2</i>	1.00±0.17	1.66±0.18*	Up	1.48±0.60	Nc
<i>Elavl3</i>	1.00±0.04	1.59±0.10*	Up	0.31±0.26*	Down
<i>Enox1</i>	1.00±0.11	1.69±0.57*	Up	0.74±0.16	Nc
<i>Fam155a</i>	1.00±0.25	3.94±1.85*	Up	3.49±0.89*	Up
<i>Fbxl16</i>	1.00±0.20	0.99±0.51	Nc	1.99±0.84	Nc
<i>Fgf12</i>	1.00±0.11	3.44±0.20*	Up	3.60±1.47*	Up
<i>Fhl2</i>	1.00±0.20	0.43±0.14*	Up	2.20±0.22*	Up
<i>Fn1</i>	1.00±0.16	0.70±0.17	Nc	2.37±0.16*	Up
<i>Fosl2</i>	1.00±0.05	2.76±0.12*	Up	2.55±0.08*	Up
<i>Gabrb3</i>	1.00±0.23	2.90±0.51*	Up	2.59±0.24*	Up
<i>Gdap111</i>	1.00±0.05	3.59±0.86*	Up	4.67±1.22*	Up
<i>Gnao1</i>	1.00±0.21	1.70±0.21*	Up	0.75±0.42	Nc
<i>Gnptg</i>	1.00±0.35	1.96±0.48*	Up	1.81±2.73	Nc
<i>Golga7b</i>	1.00±0.53	2.76±0.11*	Up	1.66±0.47	Nc
<i>Gpr19</i>	1.00±0.12	2.81±0.19*	Up	1.95±0.30*	Up
<i>Gprin1</i>	1.00±0.16	3.17±0.41*	Up	2.13±0.13*	Up
<i>Hcn2</i>	1.00±0.28	2.73±0.65*	Up	2.37±0.56*	Up
<i>Hcn3</i>	1.00±0.31	3.16±0.47*	Up	1.67±0.12*	Up
<i>Hrc</i>	1.00±0.61	2.36±0.53*	Up	1.16±0.51	Nc
<i>Igsf9b</i>	1.00±0.12	1.48±0.04	Nc	0.38±0.20*	Down
<i>Kcnb1</i>	1.00±0.20	2.82±0.90*	Up	1.83±0.23*	Up
<i>Kcnh7</i>	1.00±0.24	2.49±0.63*	Up	1.12±0.20	Nc
<i>Kcnk3</i>	1.00±0.49	3.67±0.74*	Up	2.36±0.58*	Up
<i>L1cam</i>	1.00±0.27	3.77±0.63*	Up	2.65±0.85*	Up
<i>Lad1</i>	1.00±0.16	3.72±1.13*	Up	0.75±0.06	Nc
<i>Lin28a</i>	1.00±0.14	2.74±0.15*	Up	1.90±0.31*	Up
<i>Lpar3</i>	1.00±0.48	0.62±0.04*	Down	2.89±0.38*	Up
<i>Lppr2</i>	1.00±0.49	1.52±0.81	Nc	1.08±0.39	Nc
<i>Lrp11</i>	1.00±0.13	3.17±0.81*	Up	3.37±0.52*	Up
<i>Lrp3</i>	1.00±0.30	2.68±0.65*	Up	2.30±0.63*	Up
<i>Lrrc16b</i>	1.00±0.20	2.64±0.62*	Up	1.68±0.43	Nc
<i>Lrrc24</i>	1.00±0.10	2.93±0.40*	Up	2.51±0.54*	Up
<i>m11</i>	1.00±0.20	8.57±2.37*	Up	6.65±1.02*	Up
<i>Madd</i>	1.00±0.18	3.55±1.05*	Up	0.74±0.18	Nc
<i>Man2b2</i>	1.00±0.29	6.73±0.48*	Up	1.48±0.24	Nc
<i>Maneal</i>	1.00±0.20	3.00±0.89*	Up	3.27±0.99*	Up
<i>Map1b</i>	1.00±0.16	1.02±0.29	Nc	1.05±0.13	Nc
<i>Mapk8ip1</i>	1.00±0.25	3.10±0.47*	Up	1.92±0.92	Nc
<i>Meis3</i>	1.00±0.22	3.09±0.66*	Up	1.50±0.24	Nc
<i>Mgat5b</i>	1.00±0.11	0.95±0.21	Nc	1.17±0.67	Nc

<i>Myo16</i>	1.00±0.16	1.08±1.30	Nc	1.78±0.41	Nc
<i>Ncan</i>	1.00±0.10	4.80±1.23*	Up	2.72±0.70*	Up
<i>Nefh</i>	1.00±0.35	2.69±0.29*	Up	2.12±0.25*	Up
<i>Nefl</i>	1.00±0.11	2.63±0.63*	Up	1.08±0.24	Nc
<i>Nefm</i>	1.00±0.15	3.22±0.74*	Up	2.27±0.23*	Up
<i>Nppa</i>	1.00±0.30	3.08±0.48*	Up	2.62±0.25*	Up
<i>Nppb</i>	1.00±0.21	3.35±0.96*	Up	2.11±0.14*	Up
<i>Nptxr</i>	1.00±0.34	2.55±0.11*	Up	1.73±0.13*	Up
<i>Nrxn1</i>	1.00±0.54	4.57±0.80*	Up	2.42±0.30*	Up
<i>Ogdhl</i>	1.00±0.19	2.24±0.33*	Up	0.75±0.21	Nc
<i>Pcbp3</i>	1.00±0.19	2.15±0.19*	Up	0.88±0.05	Nc
<i>Pex5l</i>	1.00±0.33	2.31±0.47*	Up	2.41±0.38*	Up
<i>Pgbd5</i>	1.00±0.13	1.77±0.66	Nc	0.65±0.16	Nc
<i>Phactr1</i>	1.00±0.22	1.92±0.53	Nc	2.26±0.43*	Up
<i>Phactr3</i>	1.00±0.14	1.18±0.84	Nc	0.99±0.10	Nc
<i>Phf21b</i>	1.00±0.22	2.78±0.70*	Up	0.64±0.16*	Down
<i>Ppfia3</i>	1.00±0.32	2.67±1.07*	Up	0.75±0.19	Nc
<i>Ppip5k1</i>	1.00±0.17	2.93±0.22*	Up	1.79±0.09*	Up
<i>Prmt8</i>	1.00±0.08	2.86±2.06	Nc	1.69±0.30*	Up
<i>Prune2</i>	1.00±0.28	2.30±0.44*	Up	1.38±0.24	Nc
<i>Psd</i>	1.00±0.25	2.25±0.15*	Up	0.36±0.22	Nc
<i>Ptprn</i>	1.00±0.29	3.38±0.67*	Up	2.34±0.41*	Up
<i>Qsox2</i>	1.00±0.06	1.84±0.22*	Up	0.71±0.19	Nc
<i>Rasl10b</i>	1.00±0.46	2.73±0.56*	Up	2.65±0.88*	Up
<i>Rest</i>	1.00±0.10	0.47±0.15*	Down	1.37±0.49	Nc
<i>Rgs7</i>	1.00±0.06	3.09±0.25*	Up	1.73±0.18*	Up
<i>Ric3</i>	1.00±0.20	2.51±0.65*	Up	1.71±0.29*	Up
<i>Rimbp2</i>	1.00±0.02	2.56±0.63*	Up	1.92±0.29*	Up
<i>Rims2</i>	1.00±0.20	3.17±0.76*	Up	1.89±0.16*	Up
<i>Rims3</i>	1.00±0.16	3.88±0.84*	Up	2.59±0.90*	Up
<i>Rpl36al</i>	1.00±0.09	0.46±0.20*	Down	1.06±0.05	Nc
<i>Scn8a</i>	1.00±0.36	2.78±0.68*	Up	0.37±0.14*	Down
<i>Sez6l2</i>	1.00±0.24	2.85±0.98*	Up	2.18±0.55*	Up
<i>Slc8a2</i>	1.00±0.03	3.38±0.52*	Up	2.26±0.12*	Up
<i>Smpd3</i>	1.00±0.36	1.81±0.22*	Up	0.46±0.25	Nc
<i>Snap25</i>	1.00±0.25	5.99±2.09*	Up	3.94±0.86*	Up
<i>Sncb</i>	1.00±0.16	5.93±0.36*	Up	2.01±0.24*	Up
<i>Soga3</i>	1.00±0.20	2.94±0.89*	Up	0.56±0.18*	Down
<i>Sost</i>	1.00±0.47	1.55±0.15	Nc	0.99±0.16	Nc
<i>Sox9</i>	1.00±0.19	2.69±1.84*	Up	0.65±0.16	Nc
<i>Spon1</i>	1.00±0.34	0.93±0.09	Nc	0.55±0.20	Nc

<i>Spry4</i>	1.00±0.18	3.64±0.67*	Up	1.95±0.27*	Up
<i>Srpk3</i>	1.00±0.86	0.97±0.17	Nc	2.10±1.29	Nc
<i>Sult4a1</i>	1.00±0.25	3.13±0.40*	Up	1.86±0.57	Nc
<i>Svop</i>	1.00±0.31	2.99±0.41*	Up	3.32±0.80*	Up
<i>Syn1</i>	1.00±0.23	2.23±0.40*	Up	1.90±0.07*	Up
<i>Syp</i>	1.00±0.31	3.91±1.45*	Up	3.23±0.72*	Up
<i>Sypl2</i>	1.00±0.19	2.64±0.50*	Up	2.60±0.38*	Up
<i>Syt14</i>	1.00±0.23	3.36±0.18*	Up	1.79±0.35*	Up
<i>Syt7</i>	1.00±0.12	2.30±0.43*	Up	1.83±0.21	Nc
<i>Tmem179</i>	1.00±0.28	2.74±0.41*	Up	1.76±0.11*	Up
<i>Tmem198</i>	1.00±0.29	0.76±0.08	Nc	1.02±0.04	Nc
<i>Trim67</i>	1.00±0.11	3.88±1.00*	Up	2.03±0.46*	Up
<i>Tspan17</i>	1.00±0.30	2.90±0.35*	Up	3.33±0.11*	Up
<i>Unc13a</i>	1.00±0.31	4.00±0.37*	Up	2.36±0.65*	Up
<i>Unc5a</i>	1.00±0.71	7.11±0.65*	Up	4.78±1.84*	Up
<i>Vwf</i>	1.00±0.30	1.94±0.76*	Up	1.21±0.34	Nc
<i>Xkr4</i>	1.00±0.32	2.54±0.48*	Up	2.73±0.14*	Up
<i>Xkr7</i>	1.00±0.63	5.03±0.87*	Up	2.40±0.67	Nc
<i>Zbtb8b</i>	1.00±0.43	1.51±0.06	Nc	0.93±0.10	Nc

Table S6. REST occupancy on RE-1 sites in E12.5 ventricles of wild type, myocardial *Dnmt3a* or *Dnmt3b* KO embryos. Data are mean±SD (n=3). **p*<0.05 compared with wild type by the Student's t test and Mann-Whitney test, as appropriate.

Gene name	Wild type	<i>Dnmt3a</i> -ko	<i>Dnmt3b</i> -ko
<i>Abcc8</i>	2.91±0.33	3.30±0.32	1.14±0.50*
<i>Cadm3</i>	21.76±2.67	17.92±0.04	9.42±1.02*
<i>Cacna1a</i>	0.86±0.21	1.52±0.81	0.31±0.19*
<i>Ccdc64</i>	5.69±2.39	3.83±2.91	2.37±2.23
<i>Cdkn1a</i>	4.49±1.06	2.69±0.91	1.86±0.60*
<i>Celf3</i>	1.53±0.36	1.57±0.21	1.09±0.46
<i>Ckmt1</i>	4.52±0.57	5.26±0.25	1.98±0.16*
<i>Clic5</i>	9.19±2.56	8.02±3.96	4.25±0.79*
<i>Crmp1</i>	19.11±9.47	21.78±13.06	23.41±3.53
<i>Ctgf</i>	0.81±0.30	0.65±0.22	0.19±0.07*
<i>Diras2</i>	1.53±0.07	2.62±0.05	1.57±0.04
<i>Dpp6</i>	7.35±1.68	7.18±0.97	3.46±1.10*
<i>Edn1</i>	14.79±1.91	16.22±2.22	13.45±10.07
<i>Elavl3</i>	4.27±0.97	2.40±0.51	3.96±2.22
<i>Fam155a</i>	12.15±3.85	13.98±4.80	19.50±8.17
<i>Fgf12</i>	4.66±1.41	5.82±0.86	1.41±0.19*
<i>Fhl2</i>	6.38±1.36	8.00±1.56	3.24±1.59*
<i>Fosl2</i>	1.24±0.27	0.69±0.26	0.90±0.07
<i>Gabrb3</i>	6.78±2.96	7.34±1.74	4.91±0.88
<i>Gdap11l</i>	9.77±4.12	6.04±1.89	2.80±0.22*
<i>Gpr19</i>	17.15±6.56	21.46±1.10	6.43±3.05*
<i>Gprin1</i>	15.58±0.22	11.99±6.63	2.93±2.10*
<i>Hcn2</i>	17.78±4.11	17.43±0.01	5.31±1.99*
<i>Hcn3</i>	3.39±0.78	4.62±1.33	1.41±0.09*
<i>Kcnb1</i>	1.95±0.48	1.74±0.53	0.74±0.19*
<i>Kcnk3</i>	1.92±0.08	2.95±0.92	1.69±1.02
<i>L1cam</i>	16.94±2.34	13.20±4.34	9.37±2.86*
<i>Lin28a</i>	3.52±1.62	2.17±1.28	2.80±1.23
<i>Lpar3</i>	15.16±5.97	12.88±5.35	8.62±3.91
<i>Lrp11</i>	5.41±0.78	2.88±1.57	11.85±4.41
<i>Lrp3</i>	6.14±0.63	5.82±0.75	2.88±1.33*
<i>Lrrc24</i>	25.30±2.44	24.12±1.78	8.18±2.76*
<i>Maneal</i>	5.78±1.14	8.37±2.46	4.27±1.28
<i>March11</i>	6.00±0.11	5.15±2.77	6.28±1.59
<i>Meis3</i>	8.50±5.04	11.34±0.53	16.97±13.82
<i>Ncan</i>	8.37±2.83	5.32±2.20	2.96±0.13*
<i>Nefh</i>	8.97±1.36	7.89±0.73	6.79±1.43
<i>Nefm</i>	19.15±0.39	20.03±1.36	17.10±8.26
<i>Nppa</i>	3.97±1.04	3.44±0.55	1.55±0.77*

<i>Nppb</i>	5.05±0.30	3.30±0.67	2.17±1.16*
<i>Nptxr</i>	0.30±0.18	0.34±0.05	0.17±0.11
<i>Nrxn1</i>	2.34±0.62	3.02±1.24	1.09±0.07*
<i>Pex5l</i>	12.67±1.30	10.88±0.85	8.30±3.07
<i>Ppip5k1</i>	12.99±2.97	10.73±1.42	8.79±3.85
<i>Ptprn</i>	4.34±1.57	4.79±1.29	0.83±0.08*
<i>Rasl10b</i>	2.36±0.29	2.52±0.36	0.79±0.01*
<i>Rgs7</i>	2.67±1.29	3.62±0.71	2.31±0.70
<i>Ric3</i>	6.97±0.22	6.83±0.66	6.59±1.79
<i>Rimbp2</i>	4.07±0.73	3.46±0.01	2.81±2.23
<i>Rims2</i>	11.61±1.01	11.71±1.70	6.92±0.20*
<i>Rims3</i>	0.49±0.03	0.50±0.35	0.15±0.01*
<i>Scn8a</i>	3.07±0.24	2.61±0.84	1.11±0.35*
<i>Sez6l2</i>	1.08±0.13	1.38±0.30	0.59±0.07*
<i>Slc8a2</i>	3.27±0.25	4.03±1.21	1.43±0.21*
<i>Snap25</i>	2.23±0.49	2.34±0.30	0.72±0.00*
<i>Sncb</i>	26.92±4.87	21.07±10.26	9.03±3.26*
<i>Soga3</i>	0.32±0.03	0.50±0.18	0.36±0.21
<i>Spry4</i>	3.92±0.61	3.72±1.30	1.83±0.34*
<i>Svop</i>	10.27±1.55	7.96±2.89	3.00±0.30*
<i>Syn1</i>	7.43±0.73	7.62±3.09	3.39±1.34*
<i>Syp</i>	2.16±0.39	2.08±0.50	0.27±0.37*
<i>Sypl2</i>	11.56±1.54	9.27±5.56	6.30±2.97*
<i>Tmem179</i>	5.93±0.40	6.49±2.44	3.24±1.20*
<i>Trim67</i>	11.05±1.97	14.02±1.71	6.05±1.05*
<i>Tspan17</i>	0.19±0.04	0.13±0.06	0.10±0.03*
<i>Unc13a</i>	0.70±0.19	0.48±0.25	0.52±0.25
<i>Unc5a</i>	7.57±2.12	9.34±2.21	3.29±0.33*
<i>Xkr4</i>	7.68±3.49	9.30±1.87	11.63±3.62

Table S7. Non-CpG and CpG methylation (%) of RE1s and RE1 flank regions in ventricles of E12.5 wild type (8 clones/gene) versus *Dnmt3b* KO (7 clones/gene) embryos. Data are mean±SD. **p*<0.05, Student's t test and Mann-Whitney test, as appropriate.

Gene	RE1				RE1 flanking region			
	5m CpG (%)		5m non-CpG (%)		5m CpG (%)		5m non-CpG (%)	
	WT	<i>Dnmt3b</i> -KO	WT	<i>Dnmt3b</i> -KO	WT	<i>Dnmt3b</i> -KO	WT	<i>Dnmt3b</i> -KO
<i>Nppb</i>	31.5±21.8	14.2±17.8	47.5±17.3	14.28±5.0*	22.9±12.4	26.19±16.3	11.1±2.4	4.0±2.6*
<i>Kcnb1</i>	31.25±22.2	21.4±22.5	56.3±11.7	16.6±13.6*	15.4±7.1	8.7±8.2	7.1±1.7	2.0±1.8*
<i>Fgf12</i>	0	0	43.3±12.6	9.5±13.1*	25.0±46.3	14.3±37.8	7.1±4.6	1.93±2.0*
<i>Syn1</i>	25.0±37.8	14.3±24.4	43.3±11.9	11.9±7.9*	15.0±8.5	2.8±3.6	9.4±3.2	4.4±4.0
<i>Snap25</i>	20.0±23.6	12.6±4.8	44.4±12.9	13.4±5.1*	18.2±11.9	8.9±2.9	8.8±2.1	2.9±1.1*
<i>Unc25a</i>	0	0	40.6±17.4	10.7±11.2*	5.7±6.8	3.3±6.1	2.8±2.8	1.1±1.5
<i>Gabrb3</i>	12.5±35.4	14.2±37.8	3.6±6.6	2.0±5.4	0.0	0.0	2.2±2.4	0.8±1.0
<i>Mapk8ip1</i>	6.2±17.7	0.0	6.3±11.6	3.6±9.4	11.4±11.7	9.1±7.5	5.9±5.5	4.1±3.3

Data File S1 (as a separated Excel file). Result of RNA-seq differential expression analysis. In this table, “TPM” is transcripts per millions, converted by Fragments Per Kilobase of exon per million fragments. “logFC” is the log2 fold change upon starvation. “Pvalue” and “Padj” are the p-value from the hypothesis testing for differential expression and the p-value adjusted for multiple testing using the Benjamini-Hochberg method.

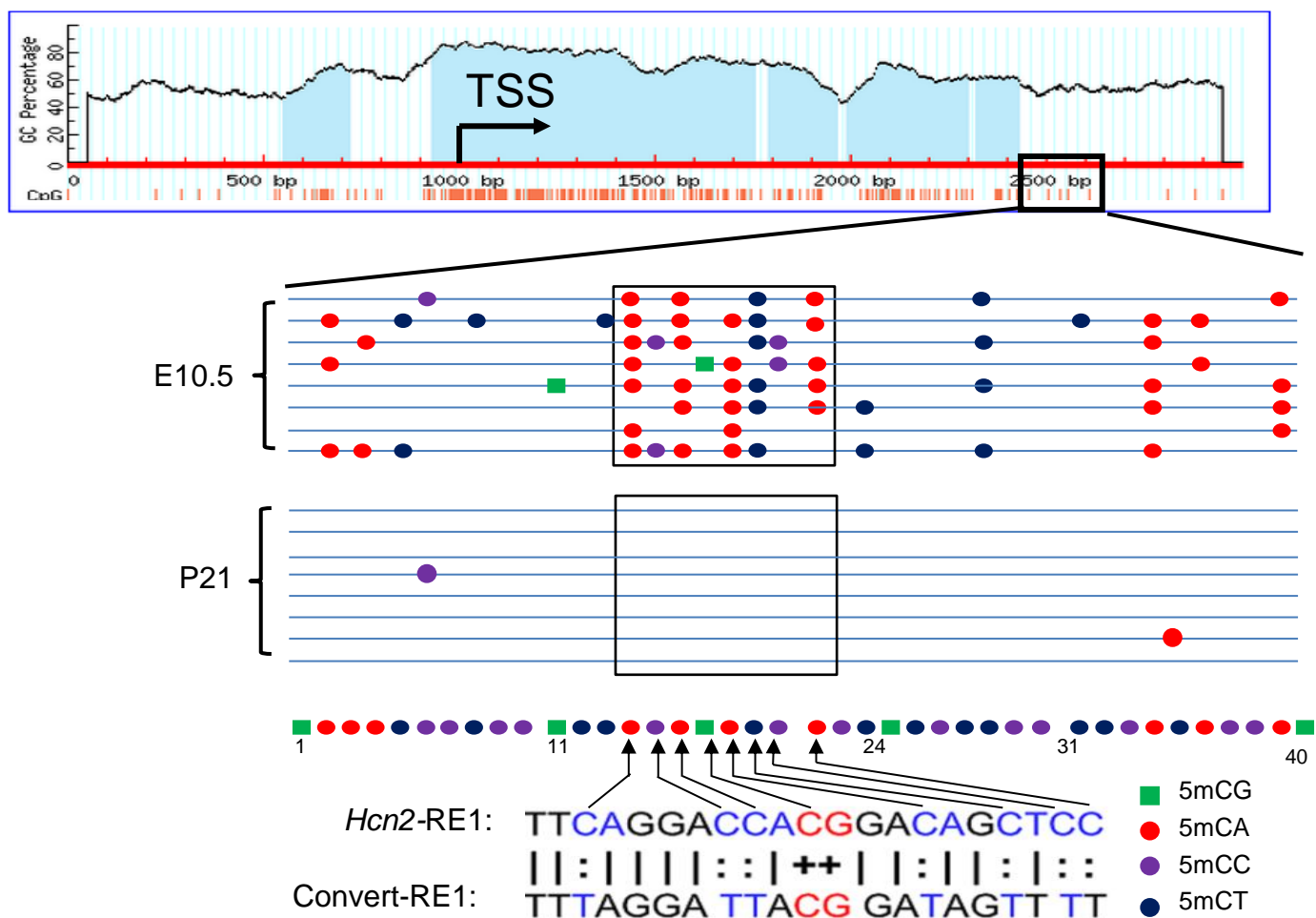


Figure S1. Top panel: the Methprimer histogram of CpG islands (blue) and CpG dinucleotides (red vertical lines) in the regulatory region of Hcn2-RE1 (TSS, transcription start site). Boxes indicate a segment of Hcn2-RE1. Bottom panel: representative bisulfite sequencing of 8 clones at embryonic (E) 10.5 day and postnatal (P) 21 day. Three groups of mouse hearts were analyzed at each stage, with 8 clones selected for bisulfite sequencing for each heart.

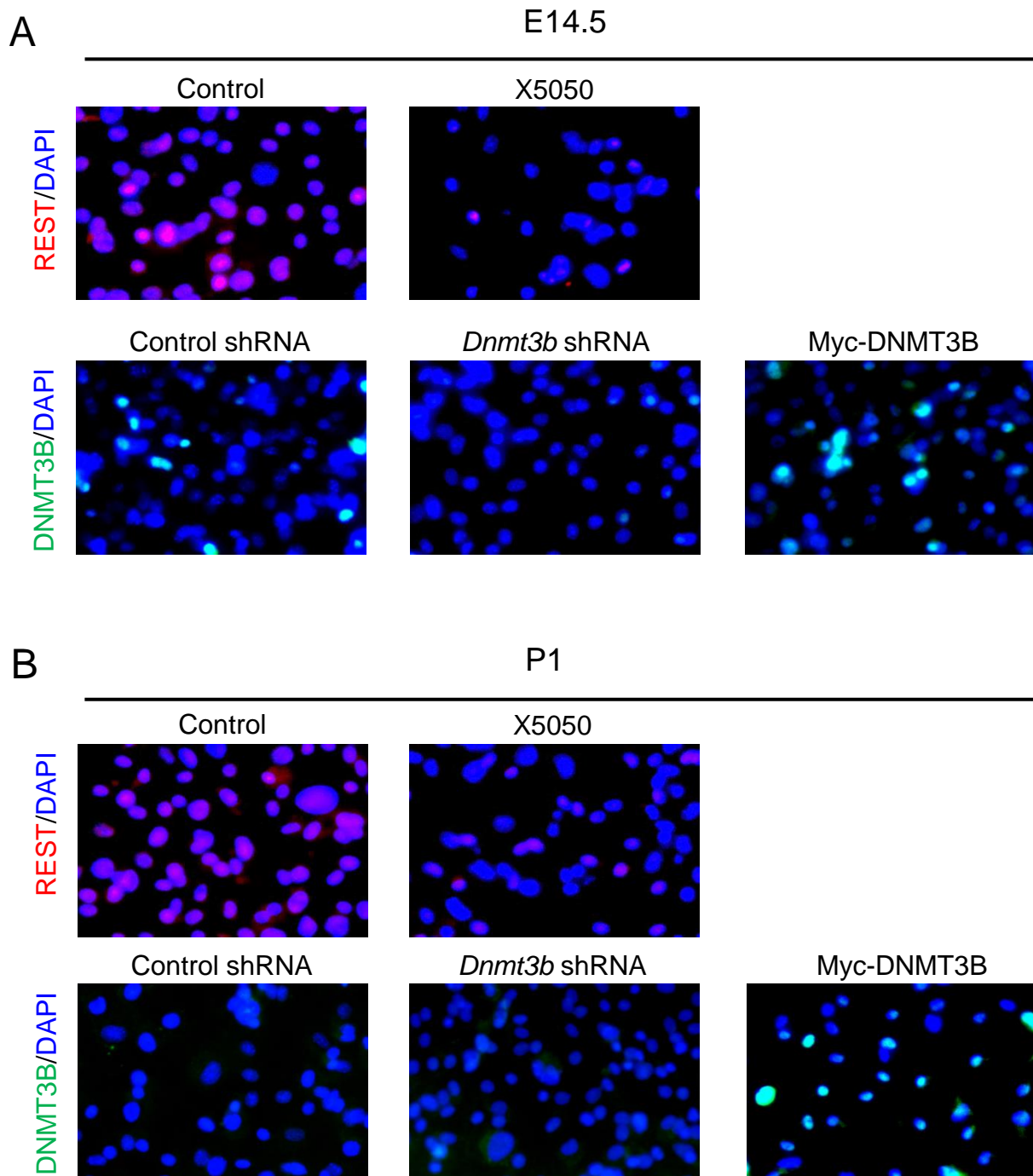


Figure S2. Immunofluorescence of REST (red) and DNMT3B (green) in cultured cardiomyocytes at E14.5 (**A**) and P1 (**B**), with or without the treatment of X5050, or transfected with *Dnmt3b*-shRNA, or Myc-DNMT3B expression plasmid. Nuclei were visualized by DAPI staining (blue). Noted that REST and DNMT3B are present in the nuclei in E14.5 cardiomyocytes, and only REST proteins are detectable at later P1 stage.

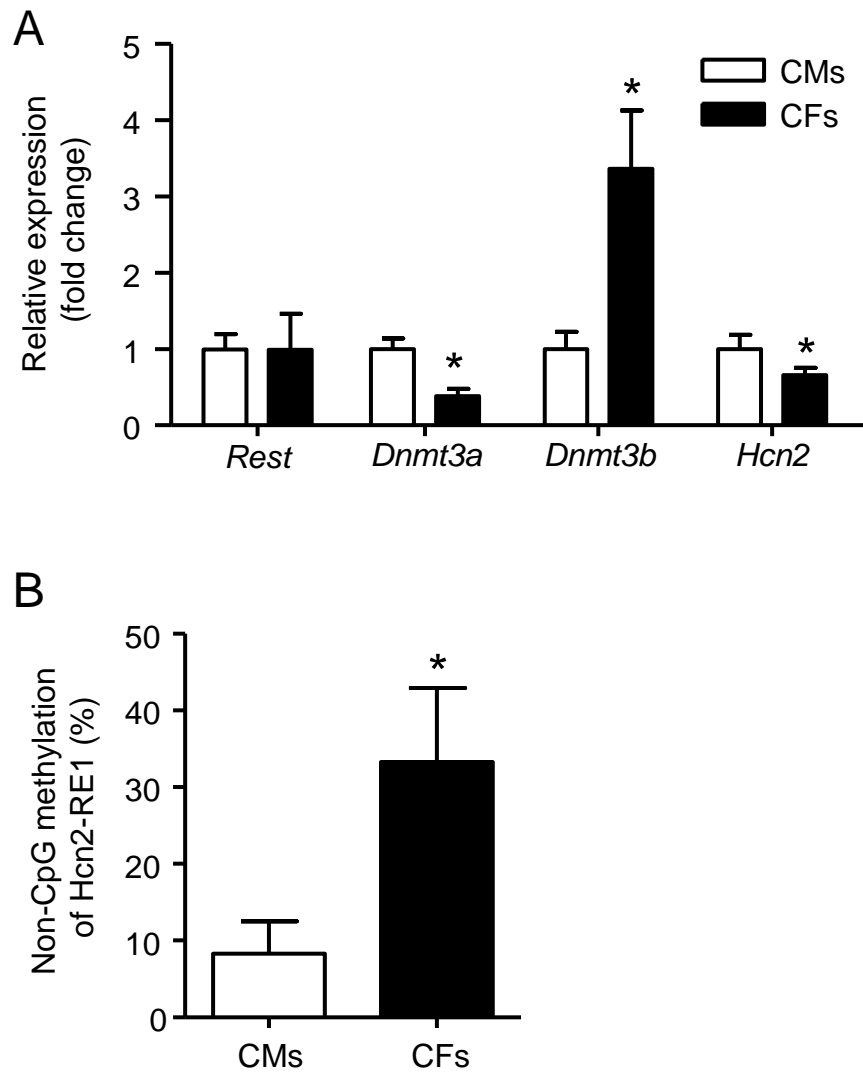


Figure S3. (A) qPCR showing mRNA expression of *Rest*, *Dnmt3a*, *Dnmt3b* and *Hcn2* in the primary cultured neonatal (P1) cardiomyocytes (CMs) and cardiac fibroblast cells (CFs). *Gapdh* was used as an internal control. (B) Bisulfite sequencing showing the level of non-CpG methylation of the *Hcn2-RE1* in CMs versus CFs. Data are presented as mean \pm SD (n=3), * $p < 0.05$ by the Student's *t*-test.

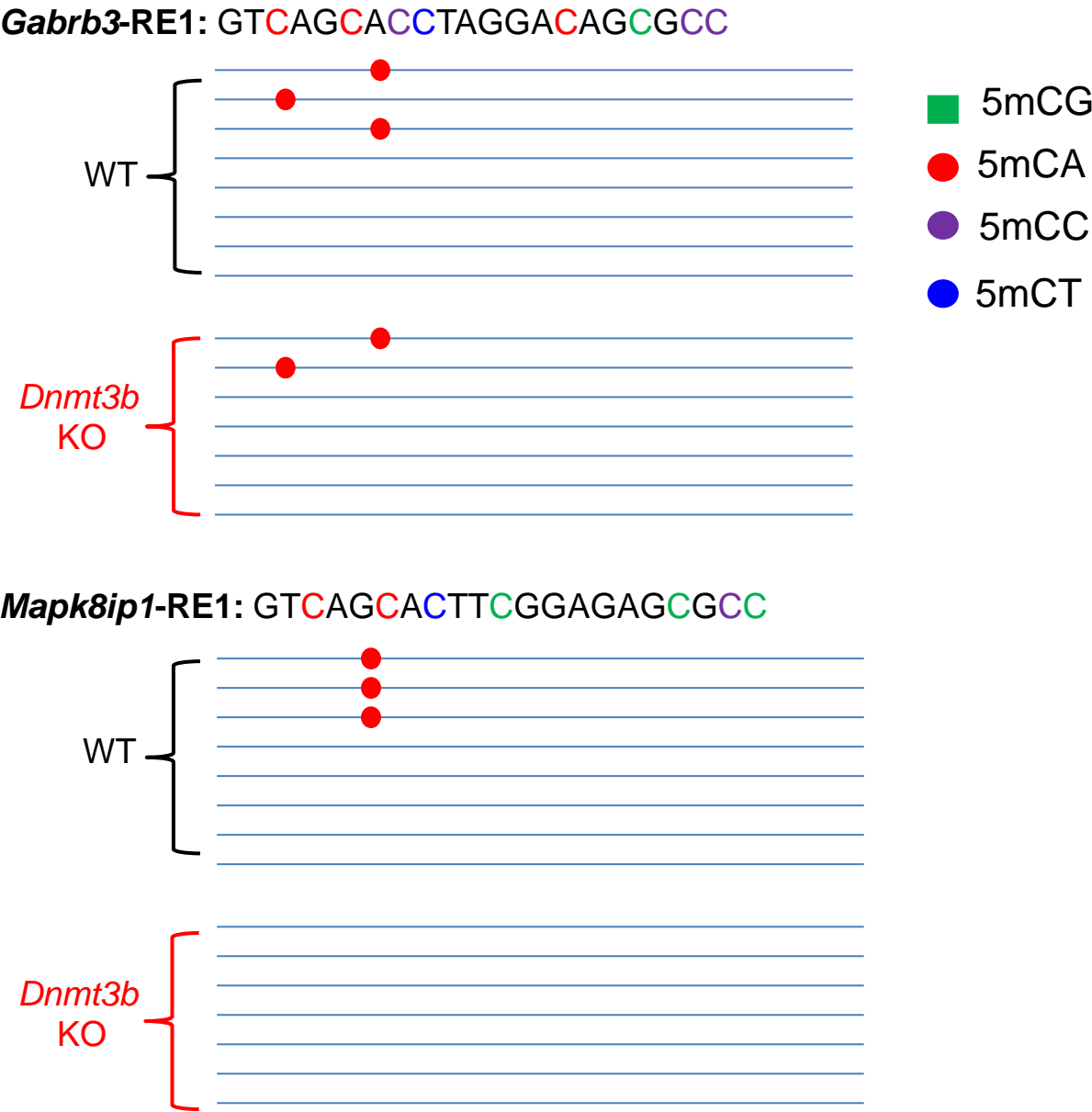


Figure S4. Bisulfite sequencing shows low methylation and no changes in methylation of RE1 sites of *Gabrb3* and *Mapk8ip1* between E12.5 wild type (8 clones/gene) and *Dnmt3b* KO (7 clones/gene) ventricles.

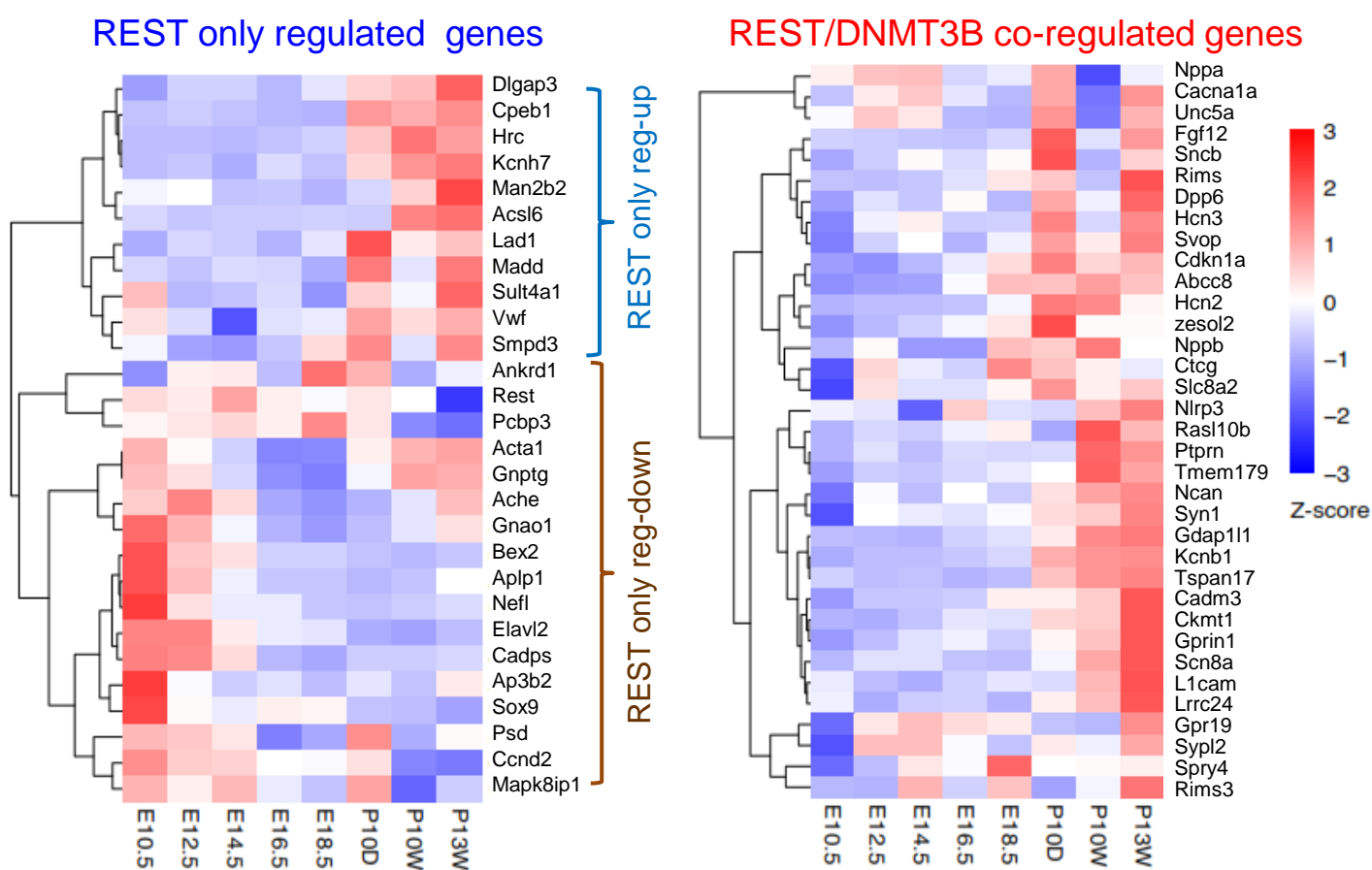


Figure S5. Heatmaps for expression of genes regulated by REST only (left) and by REST/DNMT3B (right) in embryonic and postnatal stages of mouse heart development. Note that expression of REST-only regulated genes has two patterns: upregulation and downregulation, whereas the expression of REST/DNMT3B co-regulated genes are mostly upregulated during development.