Supplementary Information

Supplementary Table legends

Supplementary Table 1: miRNA profiling raw data. Expression is presented by the threshold cycle (Ct) values. Ct of 50 indicates a non-detectable expression. C = Control, STD = Sub-Threshold Dose, ED = Effective Dose, H = Hindlimbs, F = Forelimbs.

Supplementary Table 2: Reproductive performance of p53 +/- females, mated with p53 +/- males, treated with 5-aza and tested on GD 18. Statistical analysis was performed on a litter basis using the GT2-method for multiple comparisons. For this, values obtained in each litter of each group were transformed to arsine values by Freeman-Tukey's method as described elsewhere (1) and the means and standard errors of these indices were calculated for each group.

Supplementary Table 3: Reproductive performance of miR-34 +/- females, mated with miR-34 +/- males, treated with 5-aza and tested on GD 18. Statistical analysis was performed on as in Supp. Table 2.

Supplementary Table 4: Percentage of miR-34a +/+ and -/- fetuses suffering from the indicated anomalies following 5-aza treatment on GD 10. Images of such anomalies are presented on figure 6.

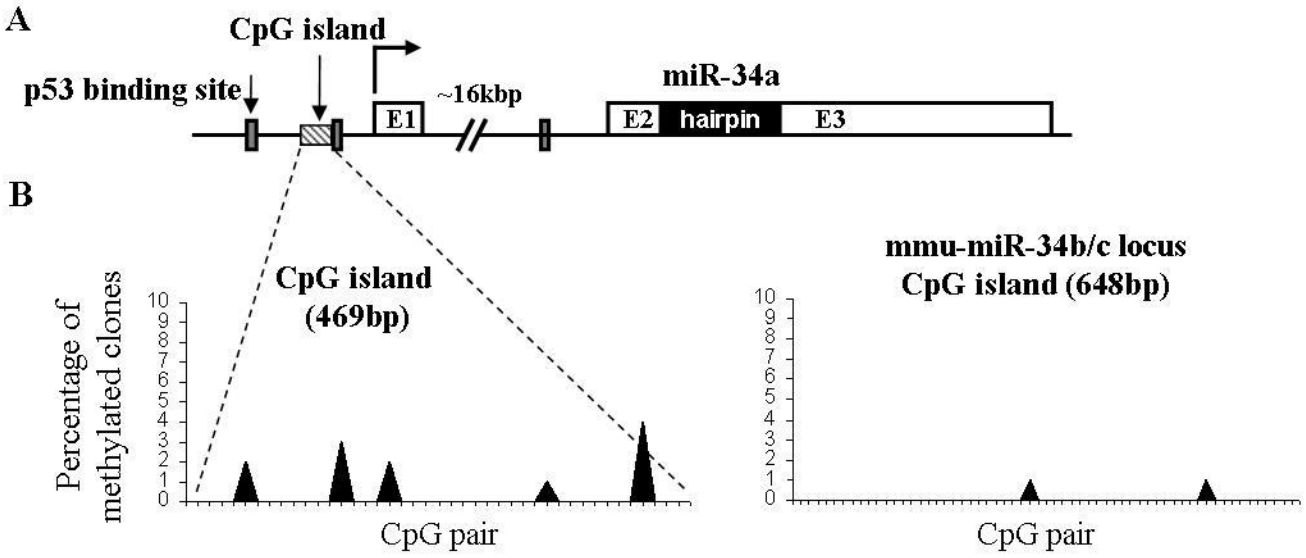
Supplementary Figure legends

Supplementary Figure 1: (A) The genome locus coding for the mouse pri-miR-34a on chromosome 4. Transcription start site is indicated with an arrow. Adapted from (2) with addition of the CpG island. (B) The methylation status of the CpG islands on pri-miR-34a and pri-miR-34bc promoters, in the mouse hindlimb buds on GD 10. The percentage of clones possessing methylated CpG pairs (total of 42, 61 pairs, respectively, indicated on the X-axis) is presented.

References

1. Torchinsky, A., A. Fein, and V. Toder, *Immunoteratology: I. MHC involvement in the embryo response to teratogens in mice.* Am J Reprod Immunol, 1995. 34(5): p. 288-98.
2. Tarasov, V., et al., *Differential regulation of microRNAs by p53 revealed by massively parallel sequencing: miR-34a is a p53 target that induces apoptosis and G1-arrest.* Cell Cycle, 2007. 6(13): p. 1586-93.

Supp. Figure 1



Supp. Table 1 –not presented here

Supp. Table 2

Reproductive performance of $p53^{+/-}$ females treated with 5-aza and tested on GD 18.

	Treatment		
	Control	**0.5 mg/Kg**	**1 mg/Kg**
Number of litters	8	12	12
Implantation sites (total/per litter)	63/7.9	95/7.9	90/7.5
Percent of post implantation loss (arcsine, mean ± S.E.M.)	7.9 (20.4 ± 3.3)	10.5 (22.4 ± 3.8)	62.2 (51.7 ± 2.6)
Number of live fetuses			
Total	58	85	34
(+/+)	17 (29.3%)	27 (31.8%)	11 (32.4%)
(+/--)	30 (51.7%)	42 (49.4%)	17 (50%)
(--/--)	11(19.0%)	16 (18.8%)	6 (17.6%)

Supp. Table 3

Reproductive performance of miR-$34a^{+/-}$ females treated with 5-aza and tested on GD 18.

	Treatment	
	Control	**0.5 mg/Kg**
Number of litters	6	11
Implantation sites (total/per litter)	45/7.5	90/8.2
Percent of post implantation loss (arcsine, mean ± S.E.M.)	11.1 (22.9 ± 2.6)	12.2 (23.6 ±3.1)
Number of live fetuses		
Total	40	79
(+/+)	12 (30.0%)	22 (27,8%)
(+/--)	18 (45.0%)	40 (50.6%)
(--/--)	10 (25.0%)	17 (21.6%)

Supp. Table 4

Proportions of of miR-34a +/+ and -/- embryos suffering from the indicated anomalies following 5-aza treatment on GD 10. Limbs were examined on GD 18.

Anomalies.	**+/+ (22 fetuses)**	**-/- (17 fetuses)**
Incomplete Ossification	20 (90.9%)	13 (76.5%)
No Ossification	7 (31.8%)	5 (29.4%)
Misshaped	10 (45.5%)	3 (17.6%)
Absent	1 (4.6%)	0
Normal	2 (9.0%)	4(23.5 %)