Figure S1: Genes differentially regulated and bioinformatically predicted to be directly targeted by miR-200c. Heatmap shows genes that are statistically significantly > 1.5 fold down (blue) or upregulated (yellow) following restoration of miR-200c. These genes are also bioinformatically predicted to be directly targeted by miR-200c as predicted by two or more of the following programs: TargetScan, microRNA.org, PicTar or miRbase.
Figure S2: Validation of microarray results for genes of interest. Breast (MDA-231 and BT549) and endometrial (Hec50 and AN3CA) cancer cells were transfected with miRNA constructs and 72 hrs later harvested for RNA and qRT-PCR was performed for each gene. Results are normalized to actin levels and presented relative to mock. ANOVA, * p < 0.05, ** p < 0.01.
Figure S3. RhoGTPase activating protein 19, ARHGAP19, is directly targeted and down-regulated by miR-200c. Regions of the 3’ UTR where miR-200c is predicted to bind. Hec50 cells treated with transfection reagent only (mock), scrambled negative control (neg), miR-200c mimic (200c), miR-200c antagomiR alone (α200c) or in conjunction with miR-200c (α200c + 200c) and luciferase assay performed. Columns, mean of five replicates, bars, standard deviation of the mean. ANOVA with Tukey-Kramer post-hoc test, ** p < 0.01.
Figure S4. The leptin receptor, LEPR, is directly targeted and down-regulated by miR-200c. Regions of the 3’ UTR where miR-200c is predicted to bind. Hec50 cells treated with transfection reagent only (mock), scrambled negative control (neg), miR-200c mimic (200c), miR-200c antagomiR alone (α200c) or in conjunction with miR-200c (α200c + 200c) and luciferase assay performed. Columns, mean of five replicates, bars, standard deviation of the mean. ANOVA with Tukey-Kramer post-hoc test, ** p < 0.01.
LEPR, a 153 bp section was cloned.
LEPR F 5' – CCACTAGTCAGGCATAGGAACA – 3'
LEPR R 5’ – CTCAGGTTTGCCAAGCGCA – 3'
LEPR mut F 5’ – TATGCACTTTTTAATCCTACATAAG – 3'
LEPR mut R 5’ – TTAAAAAGATGCATAATGACAAATACT – 3'

ARHGAP19, a 321 bp section was cloned.
ARHGAP19 F 5’ – CCACTAGTGGCTGCATTCCT – 3'
ARHGAP19 R 5’ – CTCAGGTTTACCAACTCAGTA – 3'
ARHGAP19 mut F 5’ – ACCTGCACTTTTGAAAAAGGAGAATTCA – 3'
ARHGAP19 mut R 5’ – TTAAAAAGATGCAGTGCAAGGAGAG – 3'

TrkB, a 166 bp fragment was cloned.
TrkB F 5’ – CCACTAGTAGACCGATCCTT – 3'
TrkB R 5’ – CTCAGAGCTTAGTACACACTGC – 3'
TrkB mut F’ – TCTGACAATTAAATGACTCCGA – 3'
TrkB mut R 5’ – TGTTAAAGATGTCAGGGAAGAGAG – 3'

FN1, a 510 bp fragment was cloned.
FN1 F 5’ – CCACTAGTCAGCTTCAGCTCA – 3'
FN1 R 5’ – CTCAGGTTTGCAACTACAGT – 3'
FN1 mut 1 F 5’ – ACCGCTCATTCTTTTAAATGTAAGTTT – 3'
FN1 mut 1 R 5’ – TTAAAAAGATGAGCGGATTGATGAAATCT – 3'
FN1 mut 2 F 5’ – TTCCTATCTTTTTTATACGGAAAAAT – 3'
FN1 mut 2 R 5’ – TTAAAAAGATGGGAAGGGTTGATAAT – 3'

MSN, a 390 bp fragment was cloned
MSN F 5’ – CCACTAGTCAGCTTCAGCTCA – 3'
MSN R 5’ – CTCAGGTTTGCAACTACAGT – 3'
MSN mut 1 F 5’ – TCTACAATTATGTACTCTACTGATA – 3'
MSN mut 1 R 5’ – ACATAAAGATGAGCGGTATTGATGAAATCT – 3'
MSN mut 2 F 5’ – CTTTCACTTTTTTATACGGAAAAAT – 3'
MSN mut 2 R 5’ – TTAAAAAGATGGGAAGGGTTGATAAT – 3'

ARHGAP19 F 5’ – TGTGGCTTGTCACCAATGTT – 3'
ARHGAP19 R 5’ – CACAGGGTGAGAAGGTTGT – 3'

Table S1: Primers used for PCR, mutagenesis and qRT-PCR.