Supplemental Data:

Supplemental Methods:

Hela array. HeLa cells (ATCC) were transfected with miR-148a-3p mimic or scramble control as described above. DNA Microarray Experiments Biotinylated cRNA was prepared using the Illumina RNA Amplification Kit, Catalog #1L1791 (Ambion, Inc., Austin, TX) according to the manufacturer's directions starting with 250 ng total RNA. For microarray analysis, the Illumina HumanHT-12 v4 Expression BeadChip was used (Illumina, San Diego).

Hybridization of labeled cRNA to the BeadChip, and washing and scanning were performed according to the Illumina BeadStation 500x manual. The arrays were scanned on the Illumina BeadArray Reader, a confocal-type imaging system with 532 (cye3) nm laser illuminations. Image analysis and data extraction was carried out as in accordance with Illumina specifications. Preliminary data analysis and QC was carried out using the GenomeStudio software (Illumina). All array data has been deposited in the EBI ArrayExpress Database. The ArrayExpress accessions is pending (E-MTAB-2411)

Analysis of microarray data

1) Normalization of microarray data

Expression level data from the Illumina Bead Studio software were normalized using a multiple-loess algorithm [35]. Probes whose expression level exceeds a threshold value in at least one sample are called detected. The threshold value was found by inspection from the distribution plots of (log) expression levels.

2) Sorting the probes according to significance

Detected probes were sorted according to their q-value, which is the smallest false discovery rate (FDR) at which the gene is called significant. FDR was the expected fraction of false positive tests among significant tests [36]. We evaluated FDR using Significance Analysis of Microarrays (SAM) and its implementation in the official statistical package samr [37]. In order to not be unduly impressed by accidentally small variances, we set the percentile of standard deviation values used for the exchangeability factor s0 in the test statistic to 75.

3) Statistical analysis of pathways and gene ontology terms.

Each gene ontology term or a pathway was treated simply as a set of genes. The probe list, sorted by q-value in ascending order, was translated into Entrez gene ID's and parsed so that whenever several different probes represent the same gene, only the highest-ranking probe was kept for further analysis. The sorted list of genes was subjected to a non-parametric variant of the Gene Set Enrichment Analysis (GSEA) [38], in which the p-value of a gene set of size n was defined as follows: Let us denote the k-th highest rank in gene set as rk, and define pk as the probability that out of n randomly chosen ranks (without replacement) the k-th highest is not smaller than rk. The p-value of the gene set was defined as mink [Pk] It was designed to detect overrepresented gene sets at the top of the list. Unlike the Kolmogorov-Smirnov statistic used in GSEA, it would not detect underrepresented or other, pathologically distributed, gene sets. Finding the p-value of a gene set of size n

required calculation of n rank-order values pk, however, there was no need to adjust the p-values for multiple testing as the rank-order tests are highly statistically dependent. We performed a Bonferroni adjustment of gene set p-values for the number of gene sets tested, even though there were often several gene sets with overlapping gene content (and therefore are statistically dependent), which was partly due to the design of the gene ontology database and partly because genes tended to be involved in multiple processes. We reported only gene sets with adjusted p-values ≤ 0.01.

Supplemental Table: Top 100 Biologic Process Gene Ontology Terms Modulated in HeLa Cells Transfected with miR-148a-3p Mimic.

GO ID	Biological Process	Bonferroni p-value
6952	defense response	1.06E-34
6955	immune response	4.59E-34
60337	type I interferon-mediated signaling pathway	2.91E-33
71357	cellular response to type I interferon	2.91E-33
34340	response to type I interferon	4.85E-33
45087	innate immune response	8.10E-31
9615	response to virus	5.10E-28
19221	cytokine-mediated signaling pathway	1.09E-27
34097	response to cytokine stimulus	3.19E-27
51607	defense response to virus	3.63E-26
71345	cellular response to cytokine stimulus	3.74E-26
51707	response to other organism	4.62E-24
9607	response to biotic stimulus	2.50E-23
2252	immune effector process	2.84E-23
2682	regulation of immune system process	2.36E-14
50776	regulation of immune response	2.11E-13
9719	response to endogenous stimulus	1.21E-12
34341	response to interferon-gamma	1.77E-12
9611	response to wounding	1.34E-11
71346	cellular response to interferon-gamma	1.66E-11
60333	interferon-gamma-mediated signaling pathway	4.12E-11
45069	regulation of viral genome replication	3.35E-09
45071	negative regulation of viral genome replication	4.69E-09
48525	negative regulation of viral reproduction	4.69E-09
9725	response to hormone stimulus	5.94E-09
31347	regulation of defense response	6.18E-09
32879	regulation of localization	7.87E-09
14070	response to organic cyclic compound	2.82E-08
19079	viral genome replication	4.30E-08
45088	regulation of innate immune response	5.49E-08
80134	regulation of response to stress	7.97E-08
9605	response to external stimulus	1.15E-07
1816	cytokine production	1.19E-07
43900	regulation of multi-organism process	1.42E-07
2684	positive regulation of immune system process	1.85E-07
42127	regulation of cell proliferation	2.15E-07
43901	negative regulation of multi-organism process	2.57E-07
33002	muscle cell proliferation	2.96E-07
44703	multi-organism reproductive process	3.69E-07
33993	response to lipid	3.83E-07
48584	positive regulation of response to stimulus	4.87E-07
1901135	carbohydrate derivative metabolic process	6.64E-07
22415	viral reproductive process	9.35E-07
2000242	negative regulation of reproductive process	2.20E-06

51174	regulation of phosphorus metabolic process	2.58E-06
19220	regulation of phosphate metabolic process	3.04E-06
51049	regulation of transport	4.40E-06
39528	cytoplasmic pattern recognition receptor signaling pathway in response to virus	4.42E-06
1901698	response to nitrogen compound	5.01E-06
16477	cell migration	5.24E-06
48659	smooth muscle cell proliferation	5.37E-06
8285	negative regulation of cell proliferation	5.44E-06
42325	regulation of phosphorylation	6.14E-06
32606	type I interferon production	6.85E-06
1817	regulation of cytokine production	7.26E-06
6954	inflammatory response	8.10E-06
42060	wound healing	8.40E-06
10243	response to organic nitrogen	8.48E-06
7155	cell adhesion	9.63E-06
9628	response to abiotic stimulus	1.02E-05
48585	negative regulation of response to stimulus	1.05E-05
22610	biological adhesion	1.06E-05
6928	cellular component movement	1.17E-05
60759	regulation of response to cytokine stimulus	1.22E-05
10941	regulation of cell death	1.45E-05
51247	positive regulation of protein metabolic process	1.50E-05
71495	cellular response to endogenous stimulus	1.80E-05
43067	regulation of programmed cell death	2.36E-05
48545	response to steroid hormone stimulus	2.73E-05
44093	positive regulation of molecular function	2.80E-05
42981	regulation of apoptotic process	2.92E-05
48661	positive regulation of smooth muscle cell proliferation	2.95E-05
40007	growth	2.98E-05
32480	negative regulation of type I interferon production	3.21E-05
2480	antigen processing and presentation of exogenous peptide antigen via MHC class I, TAP-independent	3.29E-05
16032	viral reproduction	3.79E-05
48660	regulation of smooth muscle cell proliferation	3.89E-05
1932	regulation of protein phosphorylation	3.96E-05
44764	multi-organism cellular process	4.07E-05
50792	regulation of viral reproduction	4.11E-05
1901137	carbohydrate derivative biosynthetic process	4.24E-05
31399	regulation of protein modification process	4.29E-05
1819	positive regulation of cytokine production	4.31E-05
35457	cellular response to interferon-alpha	5.26E-05
51241	negative regulation of multicellular organismal process	5.94E-05
48870	cell motility	7.94E-05
51674	localization of cell	7.94E-05
32479	regulation of type I interferon production	1.18E-04
7167	enzyme linked receptor protein signaling pathway	1.22E-04
1959	regulation of cytokine-mediated signaling pathway	1.23E-04
50920	regulation of chemotaxis	1.25E-04
5975	carbohydrate metabolic process	1.29E-04
50817	coagulation	1.40E-04
2753	cytoplasmic pattern recognition receptor signaling pathway	1.43E-04
50921	positive regulation of chemotaxis	1.54E-04
51240	positive regulation of multicellular organismal process	1.76E-04
32270	positive regulation of cellular protein metabolic process	1.76E-04
31349	positive regulation of defense response	1.77E-04
9617	response to bacterium	1.78E-04
1944	vasculature development	2.05E-04