

Table S1. MicroRNAs and reference genes used for real-time qPCR analyses.

miRNA / reference gene name	miRBase ID ^a / NCBI ID ^b	miRBase ^a / NCBI Accession No. ^b	Sequence Primer Forward (5' – 3') Primer Reverse (5' – 3')	Chromosome Location	miRNA Gene Family
miRNAs					
miR-21	hsa-miR-21-5p	0000076	UAGCUUAUCAGACUGAUGUUGA 5' - GCAGTAGCTTATCAGACTGATG - 3' 5' - GGTCCAGTTTTTTTTTTTTTTTCAAC - 3'	chr17: 59841266- 59841337 [+]	miR-21
miR-27b	hsa-miR-27b-5p	0004588	AGAGCUUAGCUGAUUGGUGAAC 5' - GAGAGCTTAGCTGATTGGTG - 3' 5' - GGTCCAGTTTTTTTTTTTTTTTGTTT - 3'	chr9: 95085445- 95085541 [+]	miR-27
miR-93	hsa-miR-93-5p	0000093	CAAAGUGCUGUUCGUGCAGGUAG 5' - GCAAAGTGCTGTTCTGTG - 3' 5' - GTCCAGTTTTTTTTTTTTTTTCTACCT - 3'	chr7: 100093768- 100093847 [-]	miR-17
miR-141	hsa-miR-141-3p	0000432	UAACACUGUCUGGUAAGAUGG 5' - CGCAGTAACACTGTCTGGT - 3' 5' - GTCCAGTTTTTTTTTTTTTTTCCATCT - 3'	chr12: 6964097- 6964191 [+]	miR-8
miR-182	hsa-miR-182-5p	0000259	UUUGGCAAUGGUAGAACUCACACU 5' - GTTTGGAATGGTAGAACTCA - 3' 5' - GGTCCAGTTTTTTTTTTTTTTTAGTGT - 3'	chr7: 129770383- 129770492 [-]	miR-182
miR-205	hsa-miR-205-5p	0000266	UCCUUCAUUCCACCGGAGUCUG 5' - CCTTCATTCACCGGAGT - 3' 5' - GGTCCAGTTTTTTTTTTTTTTTCAGA - 3'	chr1: 209432133- 209432242 [+]	miR-205
miR-221	hsa-miR-221-5p	0004568	ACCUGGCAUACAAUGUAGAUUU 5' - GCAGACCTGGCATAACAATG - 3' 5' - GGTCCAGTTTTTTTTTTTTTTAAATCTAC - 3'	chrX: 45746157- 45746266 [-]	miR-221
miR-375	hsa-miR-375	0000728	GCGACGAGCCCCUCGCACAAACC 5' - AGTTTGTTTCGTTCCGGCTC - 3' 5' - GGTCCAGTTTTTTTTTTTTTTTTCAC - 3'	chr2: 219001645- 219001708 [-]	miR-375
let-7a	hsa-let-7a-1	0000062	UGAGGUAGUAGGUUGUAGUU 5' - GCAGTGAGGTAGTAGTTG - 3' 5' - GGTCCAGTTTTTTTTTTTTTTAACTATAC - 3'	chr9: 94175957- 94176036 [+]	let-7

Reference gene				
			GTGCTCGCTTCGGCAGCACATATACTAAAATTGGAACGAT	
			ACAGAGAAGATTAGCATGGCCCCTGCGCAAGGATGACAC	
RNU6	RNU6 (SNORD6. U6)	004394 ^b	GCAAATTCGTGAAGCGTTCCATATTT	15q23
			5' - CGCTTCGGCAGCACATATAC - 3'	
			5' - AGGGGCCATGCTAATCTTCT - 3'	

^a - Sanger Institute. Manchester. UK; <http://www.mirbase.org>

^b - National Center of Biotechnology Information (NCBI) Reference Sequence Number; <https://www.ncbi.nlm.nih.gov/nucleotide/>

Table S2. Spearman rank correlation coefficients (r_s) between clinicopathological factors and the examined microRNAs in all tissues ($n = 54$) and serum ($n = 102$) samples. The significantly differentially expressed miRNAs and correlations are highlighted in **bold** and indicated as: *** $p < 0.0001$, ** $p < 0.0005$, * $p < 0.005$; the correlation strength is marked with an appropriate color, the darker the color, the greater the relationship between variables.

		Serum-derived miRNAs											
r_s		Age	GS	PSA(ng/mL)	miR-21	miR-27b	miR-93	miR-141	miR-182	miR-205	miR-221	miR-375	let-7a
Tissue-derived miRNAs	Age	-	0.267	0.174	-0.045	-0.247	0.035	0.171	0.202	0.034	0.022	0.221	0.106
	GS	0.101	-	0.421	0.157	0.054	0.511**	0.299	0.460*	0.053	0.344	0.563**	0.276
	PSA (ng/mL)	0.341	0.094	-	0.124	-0.276	0.172	0.146	0.042	0.131	-0.059	-0.022	-0.212
	miR-21	0.276	-0.306	0.172	-	0.161	0.388	0.107	0.354	-0.023	-0.050	0.256	0.069
	miR-27b	0.505	-0.277	0.416	0.602*	-	0.310	0.181	0.322	0.328	0.111	0.442*	0.360
	miR-93	0.342	0.401	0.174	0.271	0.436	-	0.383	0.454*	0.035	0.211	0.678***	0.201
	miR-141	0.156	0.176	0.485	0.333	0.331	0.681**	-	0.229	0.005	0.032	0.445*	0.100
	miR-182	0.081	0.120	0.524	0.217	0.363	0.407	0.643**	-	0.159	-0.015	0.447*	0.275
	miR-205	0.234	-0.344	0.283	0.249	0.586*	0.182	0.255	0.099	-	-0.016	0.091	0.037
	miR-221	0.329	-0.117	0.195	0.238	0.652**	0.644**	0.441	0.451	0.635**	-	0.435	-0.056
	miR-375	0.314	0.560*	0.359	0.249	0.372	0.850***	0.733***	0.465	0.169	0.456	-	0.089
	let-7a	0.134	-0.140	0.140	0.240	0.424	0.569*	0.470	0.466	0.574*	0.798***	0.409	-

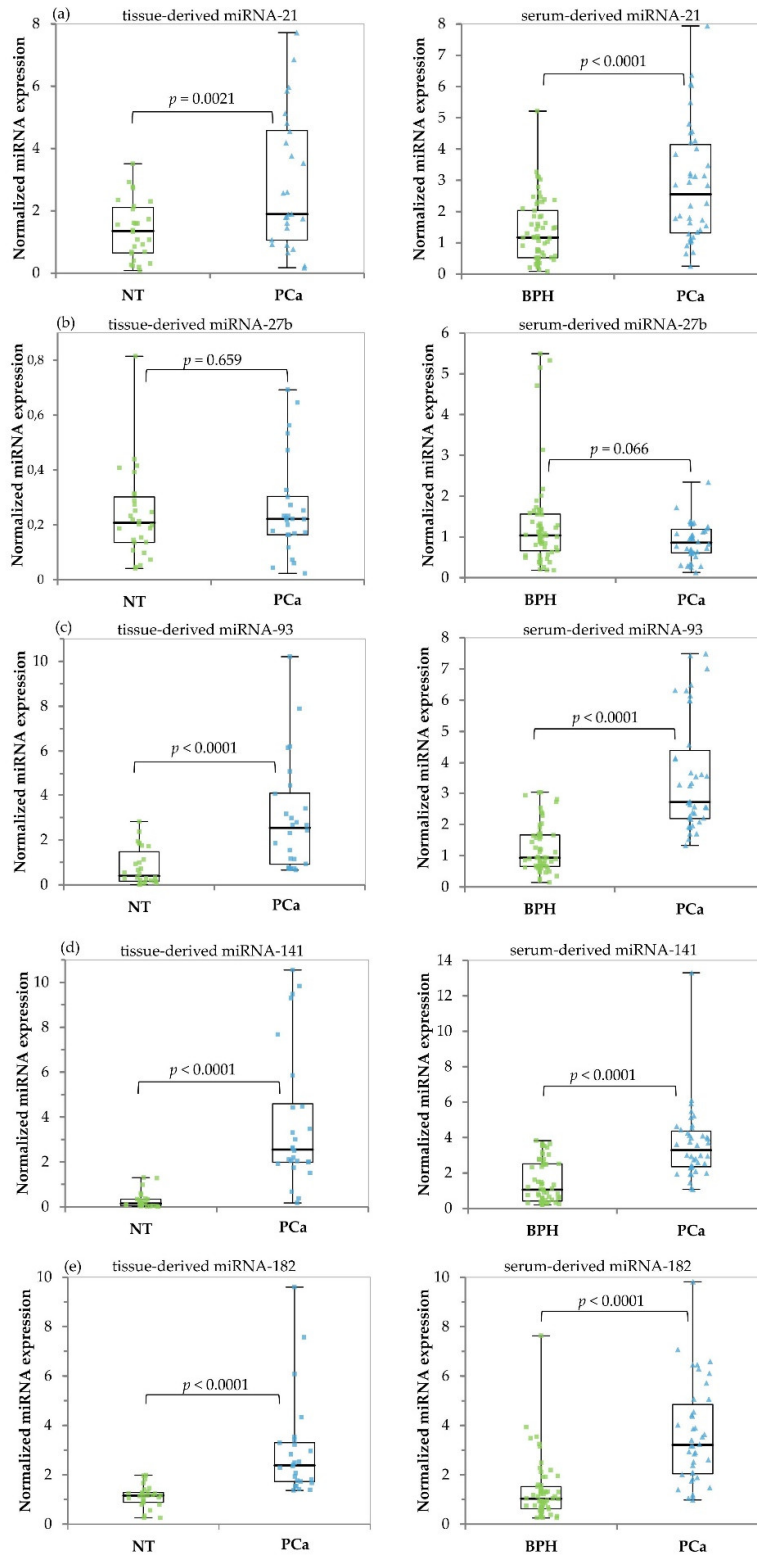


Figure S1. Expression of nine miRNAs (a – i) analyzed in tissue- (NT = 28, PCa = 26) and serum-derived (BPH = 62, PCa = 40) samples. Normalized miRNA expression data are given as box- and whisker plots. Boxes represent the lower and upper quartiles with medians, while whiskers illustrate the ranges of the miRNAs expression levels.

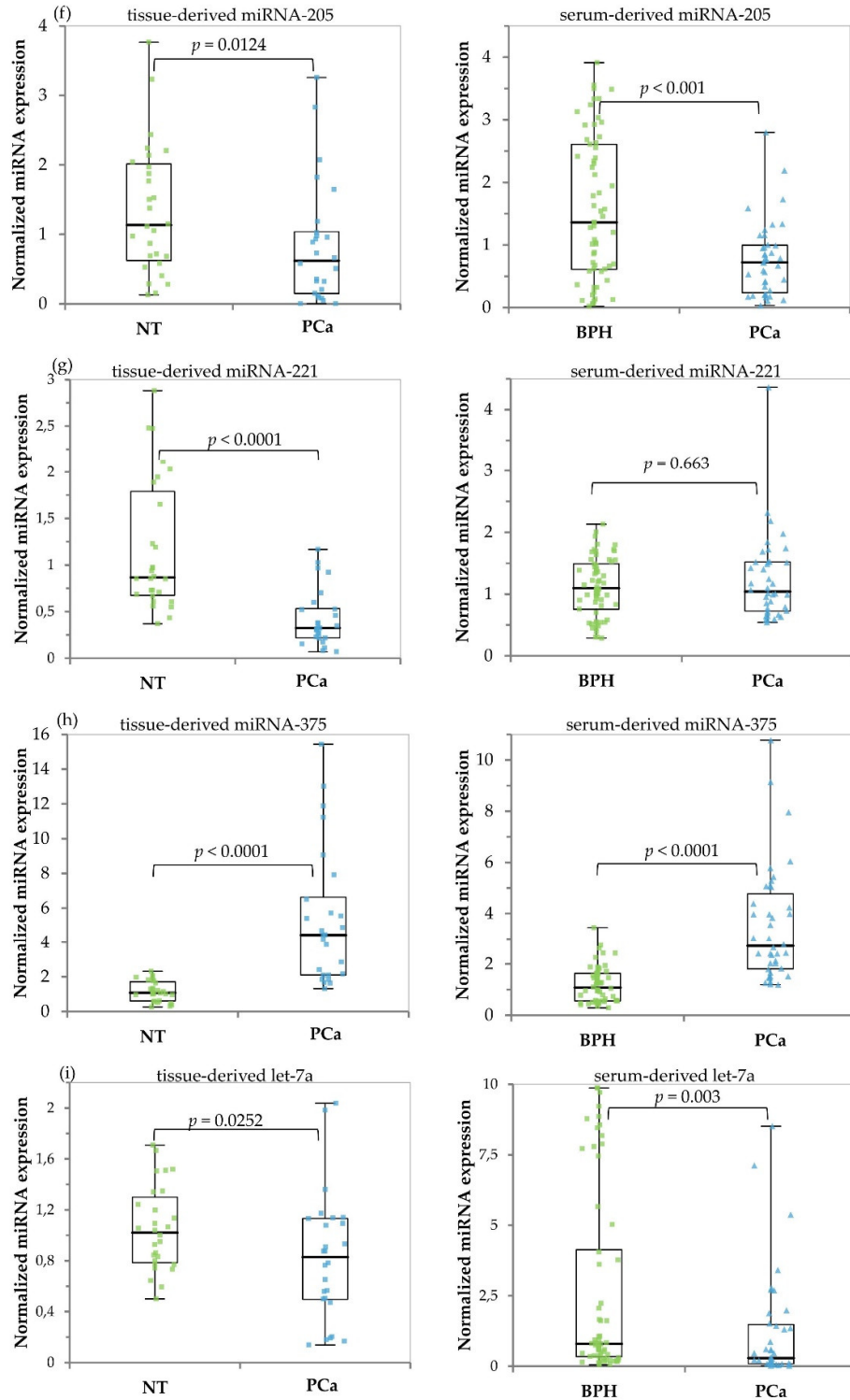


Figure S1 continued. Expression of nine miRNAs (a-i) analyzed in tissue- (NT = 28, PCa = 26) and serum-derived (BPH = 62, PCa = 40) samples. Normalized miRNA expression data are given as box- and whisker plots. Boxes represent the lower and upper quartiles with medians, while whiskers illustrate the ranges of the miRNAs expression levels.

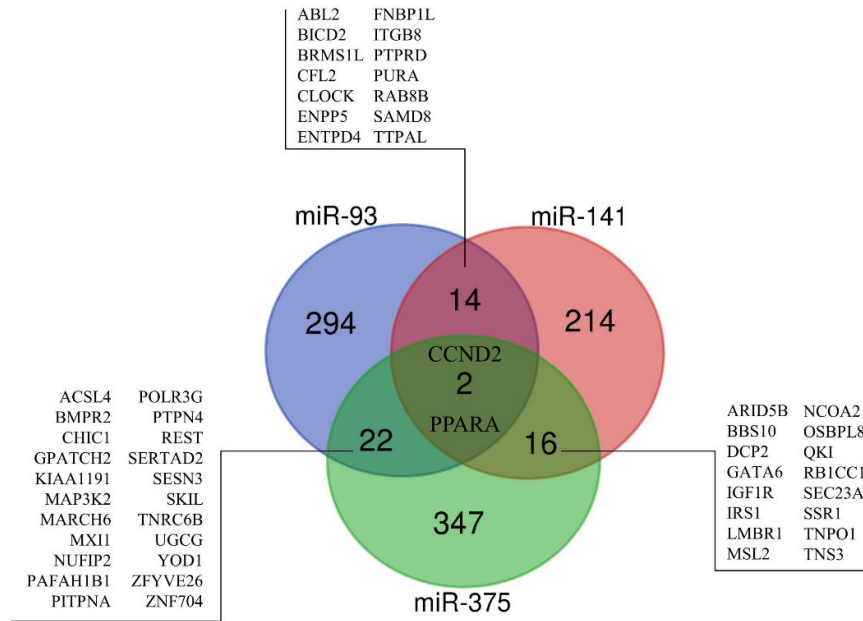


Figure S2. A Venn diagram of the relationships between and among miR-93, miR-141, and miR-375, indicating two common target genes (*CCND2* and *PPARA*).

Table S3. Target genes for miR-93 and/or miR-375 described in The Human Protein Atlas database as PCa prognostic and unprognostic [64].

Ensemble	Gene	Gene description	Chromosome (position)	Pathology prognostics	<i>p</i> value
ENSG00000117020	AKT3	AKT serine/threonine kinase 3	1 (243488233–243851079)	Unprognostic	0.137
ENSG00000110092	CCND1	Cyclin D1	11 (69641087–69654474)	Unprognostic	0.044
ENSG00000118971	CCND2	Cyclin D2	12 (4273772–4305350)	Unprognostic	0.175
ENSG00000140443	IGF1R	Insulin like growth factor 1 receptor	15 (98648971–98964530)	Unprognostic	0.162
ENSG00000169967	MAP3K2	Mitogen-activated protein kinase kinase 2	2 (127298730–127388465)	Unprognostic	0.139
ENSG00000100030	MAPK1	Mitogen-activated protein kinase 1	22 (21754500–21867680)	Unprognostic	0.104
ENSG00000119950	MXI1	MAX interactor 1, dimerization protein	10 (110207605–110287365)	Prognostic unfavourable	<0.0001
ENSG00000007168	PAFAH1B1	Platelet activating factor acetylhydrolase 1b regulatory subunit 1	17 (2593210–2685615)	Unprognostic	0.058

ENSG00000114062	UBE3A	Ubiquitin protein ligase E3A	15 (25333728- 25439056)	Unprognostic	0.048
ENSG00000180667	YOD1	YOD1 deubiquitinase	1 (207043849- 207052980)	Unprognostic	0.010
ENSG00000072121	ZFYVE26	Zinc finger FYVE- type containing 26	14 (67727374- 67816590)	Unprognostic	0.037

Table S4. GO terms and KEGG pathways for miR-93 and miR-375 PCa-related target genes [64] based on in silico through DAVID [65] and DIANA miRpath v.3 database [66].

No.	KEGG pathway Category		Genes	p value
1	hsa04068:FoxO signaling pathway		MAPK1, IGF1R, CCND1, CCND2, AKT3	<0.0001
2	hsa04151:PI3K-Akt signaling pathway			<0.0001
3	hsa04510:Focal adhesion			<0.0001
4	hsa04917:Prolactin signaling pathway		MAPK1, CCND1, CCND2, AKT3	<0.0001
5	hsa05200:Pathways in cancer		MAPK1, IGF1R, CCND1, AKT3	0.008
6	hsa05203:Viral carcinogenesis		MAPK1, CCND1, CCND2, UBE3A	0.001
7	hsa05205:Proteoglycans in cancer		MAPK1, IGF1R, CCND1, AKT3	0.001
8	hsa05214:Glioma			<0.0001
9	hsa05215:Prostate cancer			<0.0001
10	hsa05218:Melanoma			<0.0001
11	hsa04010:MAPK signaling pathway		MAPK1, MAP3K2, AKT3	0.033
12	hsa04014:Ras signaling pathway		MAPK1, IGF1R, AKT3	0.026
13	hsa04015:Rap1 signaling pathway			0.023
14	hsa04066:HIF-1 signaling pathway			0.005
15	hsa04152:AMPK signaling pathway		IGF1R, CCND1, AKT3	0.008
16	hsa04550:Signaling pathways regulating pluripotency of stem cells		MAPK1, IGF1R, AKT3	0.011
17	hsa04630:Jak-STAT signaling pathway		CCND1, CCND2, AKT3	0.011
18	hsa04914:Progesterone-mediated oocyte maturation		MAPK1, IGF1R, AKT3	0.004
19	hsa04919:Thyroid hormone signaling pathway		MAPK1, CCND1, AKT3	0.007
20	hsa05161:Hepatitis B			0.011
21	hsa05162:Measles		CCND1, CCND2, AKT3	0.010
22	hsa05166:HTLV-I infection			0.033
23	hsa05210:Colorectal cancer		MAPK1, CCND1, AKT3	0.002
24	hsa05212:Pancreatic cancer			0.002
25	hsa05213:Endometrial cancer			0.002
26	hsa05220:Chronic myeloid leukemia			0.003
27	hsa05221:Acute myeloid leukemia			0.002
28	hsa05223:Non-small cell lung cancer			0.002
29	hsa05216:Thyroid cancer		MAPK1, CCND1	0.033
30	hsa05219:Bladder cancer			0.047
No.	Category	GO term	Genes	p value
1	BP	GO:0006468~protein phosphorylation	MAPK1, CCND1, MAP3K2, AKT3	0.002
2	BP	GO:0007049~cell cycle	MAPK1, CCND1, CCND2	0.007
3	BP	GO:0008284~positive regulation of cell proliferation	MAPK1, IGF1R, CCND2	0.030
4	BP	GO:0030968~endoplasmic reticulum unfolded protein response	CCND1, YOD1	0.026
5	BP	GO:0033598~mammary gland epithelial cell proliferation	MAPK1, CCND1	0.005
6	BP	GO:0043627~response to estrogen		0.038
7	BP	GO:0045737~positive regulation of cyclin-dependent protein serine/threonine kinase activity	CCND1, CCND2	0.014
8	CC	GO:0005829~cytosol	MAPK1, CCND1, CCND2, UBE3A, MAP3K2, PAFAH1B1, YOD1	0.004
9	CC	GO:0000307~cyclin-dependent protein kinase holoenzyme complex	CCND1, CCND2	0.008

10	MF	GO:0005515~protein binding	MAPK1, IGF1R, CCND1, CCND2, UBE3A, MAP3K2, ZFYVE26, PAFAH1B1, YOD1, MXI1, AKT3	0.001
11	MF	GO:0004672~protein kinase activity	CCND1, MAP3K2, AKT3	0.018
12	MF	GO:0004674~protein serine/threonine kinase activity	MAPK1, MAP3K2, AKT3	0.020
13	MF	GO:0019901~protein kinase binding	CCND1, CCND2, MAP3K2	0.020

Abbreviations: KEGG, Kyoto Encyclopedia of Genes and Genomes; GO, Gene Ontology; BP, biological processes; CC, cell component; MF, molecular function.

Table S5. miRNAs targets among 11 analyzed genes predicted by TargetScan [67].

Gene	miRNA	Position in the UTR	Seed match	CS	P _{ct}
AKT3 ENST00000366539.1 3' UTR length: 5530	miR-93	1142–1148	7mer-m8	-0.16	0.74
CCND1 ENST00000227507.2 3' UTR length: 3192	miR-93	583–589	7mer-1A	-0.03	0.2
		1014–1020	7mer-m8	-0.21	0.74
CCND2 ENST00000261254.3 3' UTR length: 5347	miR-93	968–974	7mer-m8	-0.2	0.74
	miR-375	328–334	7mer-1A	-0.08	0.35
IGF1R ENST00000268035.6 3' UTR length: 7088	miR-93	3998–4004	7mer-1A	-0.01	<0.1
		2276–2282	7mer-1A	-0.01	0.29
		2993–2999	7mer-1A	-0.01	<0.1
MAP3K2 ENST00000409947.1 3' UTR length: 8910	miR-93	65–71	7mer-m8	-0.18	0.74
		265–272	8mer	-0.18	0.96
		6553–6559	7mer-m8	-0.2	0.68
		8869–8875	7mer-m8	-0.14	0.74
		1503–1509	7mer-1A	-0.01	<0.1
		3342–3348	7mer-m8	-0.02	<0.1
		5996–6002	7mer-1A	-0.01	0.17
		2760–2766	7mer-1A	-0.01	0.22
MAPK1 ENST00000215832.6 3' UTR length: 9750	miR-93	1552–1558	7mer-1A	-0.01	<0.1
		445–452	8mer	-0.23	0.96
		414–420	7mer-1A	-0.05	0.13
MXI1 ENST00000332674.5 3' UTR length: 4283	miR-93	7597–7603	7mer-m8	-0.02	<0.1
		1356–1362	7mer-m8	-0.15	0.74
		1691–1697	7mer-1A	-0.01	0.27
		2830–2836	7mer-m8	-0.02	<0.1
		130–136	7mer-1A	-0.13	0.35
PAFAH1B1 ENST00000397195.5 3' UTR length: 3813	miR-375	200–206	7mer-1A	-0.16	0.35
		2870–2877	8mer	-0.16	0.89
PPARα ENST00000396000.2 3' UTR length: 8376	miR-93	1249–1255	7mer-1A	-0.05	<0.1
		416–422	7mer-m8	-0.15	<0.1
		97–103	7mer-1A	-0.01	<0.1
		620–626	7mer-m8	-0.02	<0.1
		1969–1975	7mer-m8	-0.02	<0.1
UBE3A ENST00000232165.3 3' UTR length: 1960	miR-375	4966–4972	7mer-m8	-0.02	<0.1
		3040–3046	7mer-1A	-0.07	<0.1
YOD1 ENST00000315927.4 3' UTR length: 5182	miR-93	1637–1643	7mer-m8	-0.07	<0.1
		218–224	7mer-1A	-0.16	0.35
		1138–1144	7mer-m8	-0.13	0.2
		3495–3502	8mer	-0.24	0.94
ZFYVE26 ENST00000347230.4 3'UTR length: 1916	miR-93	1601–1607	7mer-m8	-0.09	<0.1
		2217–2223	7mer-1A	-0.03	0.17
		85–91	7mer-1A	-0.11	0.31
ZFYVE26 ENST00000347230.4 3'UTR length: 1916	miR-375	353–360	8mer	-0.2	0.91
		1659–1666	8mer	-0.23	0.94
		321–327	7mer-1A	-0.14	0.35

Abbreviations: CS, the context++ score for a specific site is the sum of the contribution of 14 features [100]; P_{ct}, probability of conserved targeting; 3' UTR, untranslated region; 8mer, an exact match to positions 2–8 of the mature miRNA (the seed + position 8) followed by an 'A'; 7mer-m8, an exact match to positions 2–8 of the mature miRNA (the seed + position 8); 7mer-A1, an exact match to positions 2–7 of the mature miRNA (the seed) followed by an 'A' [67].