

The role of microRNAs in gene expression and signaling response of tumor cells to an acidic environment

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Supplementary Material

Table S1: Primers used for quantitative PCR.

Target	forward primer	reverse primer
<i>Acat2</i>	GACTGGAACGGTCACCCCAG	TCGGTCCTACTCCCATGACA
<i>Aox1</i>	GAGCAGACATCTGGACTCGG	GTCAGGACACTTGCGGAGAA
<i>Crem</i>	TCCGAGCTCCTACTACTGCT	TTTCATCAGCCTCAGCTCCC
<i>Gls2</i>	CCTTTTCCTTTAGATGTGGGGC	AAGCAGGTCACCAAGTCGAG
<i>Il6</i>	AGCCAGAGTCATTCAGAGCA	AGCATTGGAAGTTGGGGTAGG
<i>Nos2 (iNos)</i>	GCAGGTTGAGGATTACTTCTTCCA	GCCCTTTTTTGCTCCATAGGAAA
<i>Ccl2 (MCP-1)</i>	CCCAGAAACCAGCCAACT	TGCTGCTGGTGATTCTCTTG
<i>Per3</i>	GAACAGGAAACAACCGCACC	CCACGGGCTTGAATCCTTCT
<i>Pink1</i>	TGTATGAAGCCACCATGCC	GCTTAAGATGGCTTCGCTGG
<i>Spp1 (osteopontin)</i>	CCTCTGAAGAAACGGATGACT	CTGGGCAACTGGGATGACCTT
<i>Tnfa</i>	CCACCACGCTCTTCTGTCTACTGAACT	CCATTGGCCAGGAGGGCGTT
<i>Txnip</i>	CCAGCCTACAGGTGAGAACG	GGCTGGGACGATCGAGAAAA
<i>Ypel3</i>	GCGTGCCTACCTCTTCAACT	TTGCAGTTCTCGCAGTGGAT

Housekeeper		
<i>Hprt1</i>	ACCAGTCAACGGGGGACATA	TTGGGGCTGTACTGCTTGAC
<i>18S</i>	CTGAGAAACGGCTACACATC	CCCAAGATCCAACACTACGAGC

Table S2: List of oligonucleotides used for miRNA-PCR.

miRNA	ID	Accession number (miRBase)	Sequence
<i>control</i>	snoRNA (rat)	U64702 (NCBI)	
<i>control</i>	U6 snRNA	NR_004394	
<i>miR-7a-5p</i>	rno-miR-7a-5p	MIMAT0000606	UGGAAGACUAGUGAUUUUGUUGU
<i>miR-183-5p</i>	rno-miR-183-5p	MIMAT0000860	UAUGGCACUGGUAGAAUUCACU
<i>miR-203a-3p</i>	rno-miR-203a-3p	MIMAT0000876	GUGAAAUGUUUAGGACCACUAG
<i>miR-215</i>	rno-miR-215	MIMAT0003118	AUGACCUAUGAAUUGACAGACA

Table S3: miRNA sequences of mimics and antagomirs used for transient transfection.

target miRNA	function	miRNA strand
<i>rno-miR-7-5p</i>	mimic	5'-UGGAAGACUAGUGAUUUUGUUGU-3'
<i>rno-miR-7-5p</i>	inhibitor (antagomir)	5'-ACAACAAAATCACTAGTCTTCC-3'
<i>rno-miR-183-5p</i>	mimic	5'-UAUGGCACUGGUAGAAUUCACU-3'
<i>rno-miR-183-5p</i>	inhibitor (antagomir)	5'-AGTGAATTCTACCAGTGCCAT-3'
<i>rno-miR-203a</i>	mimic	5'- GUGAAAUGUUUAGGACCACUAG-3'
<i>rno-miR-203a-3p</i>	inhibitor (antagomir)	5'-TAGTGGTCCTAACATTTCA-3'
<i>rno-miR-215</i>	mimic	5'- AUGACCUAUGAAUUGACAGAC-3'
<i>rno-miR-215</i>	inhibitor (antagomir)	5'-GTCTGTCAATTCATAGGTCAT-3'

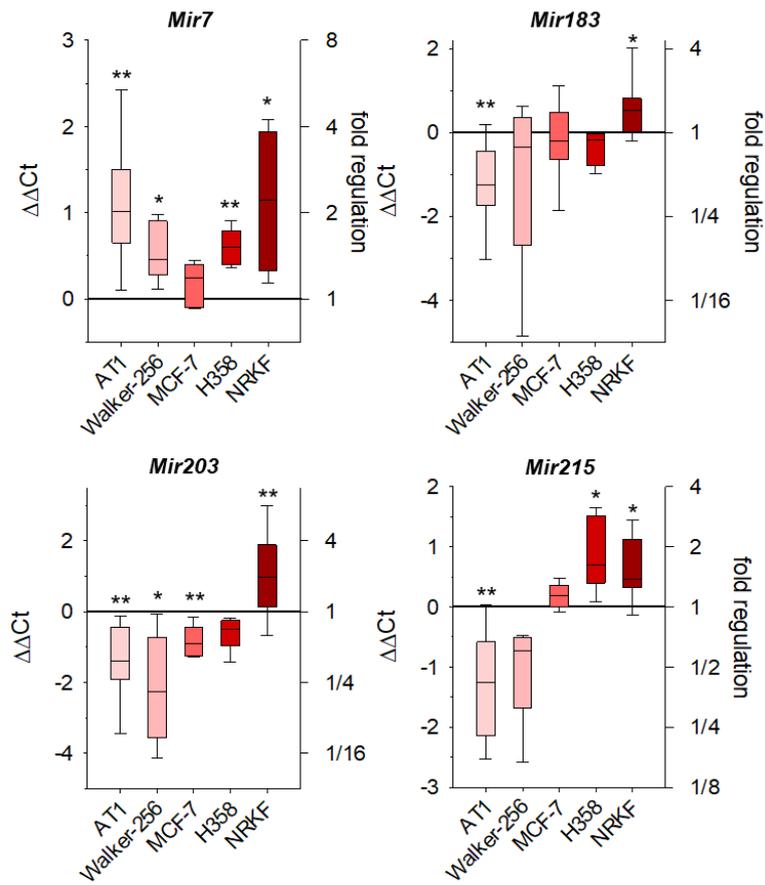


Figure S1: Expression of miRNAs under acidic conditions (pH 6.6, 24 h) in 4 tumor cell lines (AT1: subline of the rat Dunning prostate carcinoma R3327, Walker-256: rat mammary carcinoma, MCF-7: human mammary carcinoma, H358: human non-small cell lung cancer) and one normal cell line (NRKF: normal rat kidney fibroblasts). n=4-16; (*) $p < 0.05$, (**) $p < 0.01$ vs. pH 7.4.