

Supplementary data

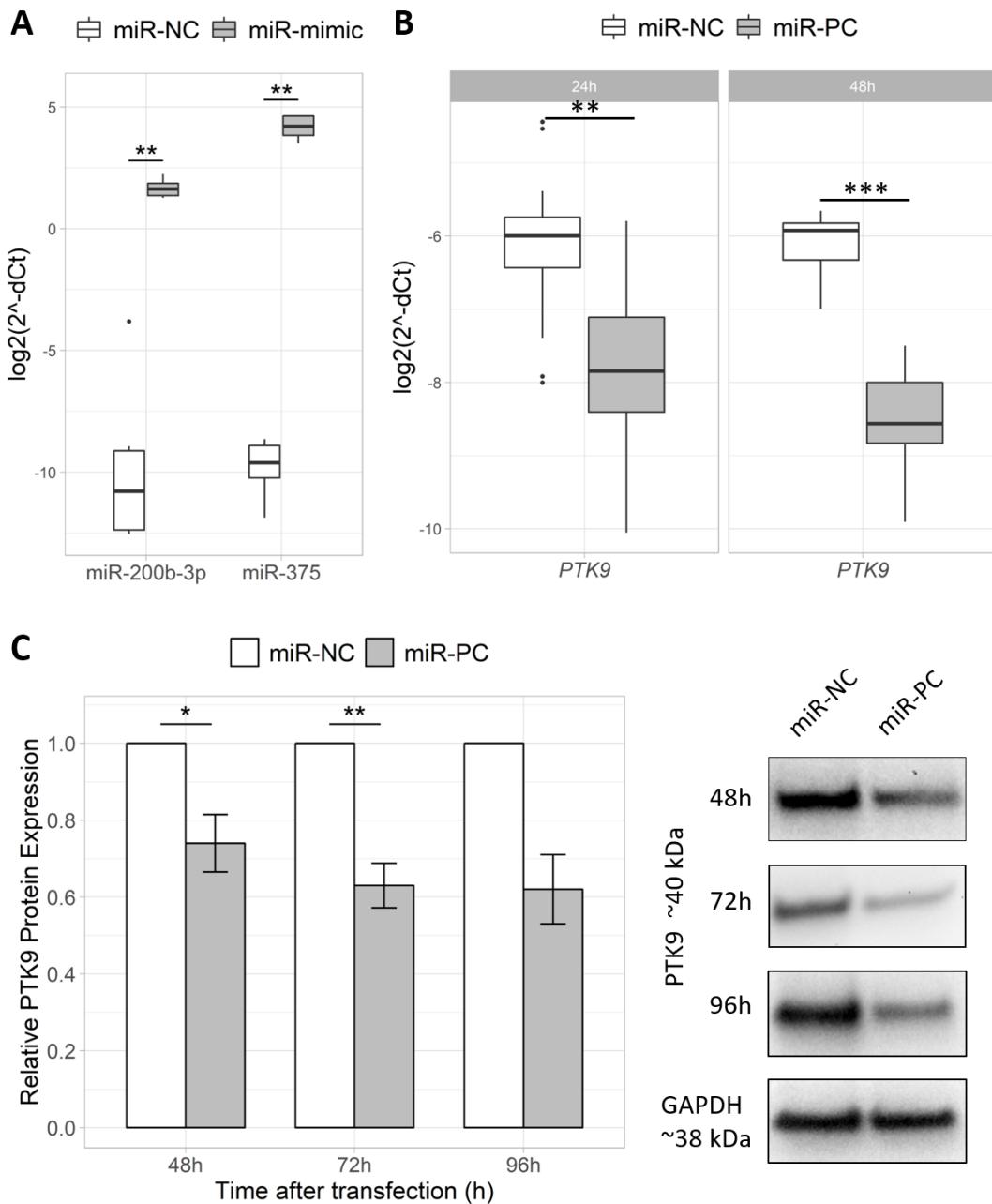


Figure S1: Evaluation of miRNA transfection and silencing efficiency. (A) Upregulation of miRNAs miR-375-3p and miR-200b-3p was observed after transfection with corresponding miRNA mimics; (B) PTK9 gene was downregulated after transfection with miRVana miRNA Mimic miR-1 positive control and (C) PTK9 protein was downregulated after transfection with miRVana miRNA Mimic miR-1 positive control. miR-NC—miRNA mimic negative control, miR-PC—miRNA mimic positive control. * $p < 0.05$, ** $p < 0.005$, *** $p < 0.0005$.

Table S1: Binding positions and insert sequences for the construction of pMIR-REPORT-Luciferase vectors, used for the investigation of direct miRNA-target binding. The sequences coding the predicted microRNA binding sites in the 3'UTR of the target genes are highlighted in grey. Changed nucleotides in the mutant insert sequences are colored in red.

Mature miRNA ID	Mature miRNA Accession Number	Target Symbol	Target Ensembl Number	miRNA Binding Position	Insert type	Insert sequences (5' to 3')	
miR-375-3p	MIMAT0000728	<i>KIT</i>	ENSG00000157404	453-459	Wt	Forward	CTGCGCACTAGTTGGAAAACAGAACATCATTAGAACAAAGGACAGAGTGAACACCTGAAGCTTAATAAA
						Reverse	TTTATTAAAGCTTCAGGTGTTCATACTCTGCCTTGTCTAATGATGTTCTGTTCCAACTAGTGCGCAG
					Mut	Forward	CTGCGCACTAGTTGGAAAACAGAACATCATTAGA TCA AAAGGACAGAGTGAACACCTGAAGCTTAATAAA
						Reverse	TTTATTAAAGCTTCAGGTGTTCATACTCTGCCTTCTAATGATGTTCTGTTCCAACTAGTGCGCAG
				549-555	Wt	Forward	CTGCGCACTAGTAAAAAATGATCCCCAAGTGTGAACAAAAGATGCTCTCTGTGGACCAAAGCTTAATAAA
						Reverse	TTTATTAAAGCTTGGTCCACAGAACAGAGCATCTTGTTCACACTGGGGATCATTTTACTAGTGCGCAG
					Mut	Forward	CTGCGCACTAGTAAAAAATGATCCCCAAGTGTGAAG G AAAGATGCTCTCTGTGGACCAAAGCTTAATAAA
						Reverse	TTTATTAAAGCTTGGTCCACAGAACAGAGCATCTTGTTCACACTGGGGATCATTTTACTAGTGCGCAG
				855-861	Wt	Forward	CTGCGCACTAGTATAACCACAAAGCACAGTTGAACAAAATCTCCTTTAGCTGATGAAGCTTAATAAA
						Reverse	TTTATTAAAGCTTCATCAGCTAAAGAGGAGATTTGTCAAAGTGTGCTTGTGGTTACTAGTGCGCAG
					Mut	Forward	CTGCGCACTAGTATAACCACAAAGCACAGTTGAAG G AAATCTCCTTTAGCTGATGAAGCTTAATAAA
						Reverse	TTTATTAAAGCTTCATCAGCTAAAGAGGAGATTTGTCAAAGTGTGCTTGTGGTTACTAGTGCGCAG
miR-200b-3p	MIMAT0000318	<i>EGFR</i>	ENSG00000146648	5849-5855	Wt	Forward	TGCGCACTAGTAAGTTTGCATGATGAAAGCAGTATTGTACAAATGAAAAGCAGAAAAGCTTAATAAA
						Reverse	TTTATTAAAGCTTTCTGCTTTCATTGTACAAATACTGCTTCATATTGCAAACCTACTAGTGCGCA
					Mut	Forward	CTGCGCACTAGTAAGTTGCATGATGAAAGCAG C TTTGACAAATGAAAAGCAGAAAAGCTTAATAAA
						Reverse	TTTATTAAAGCTTTCTGCTTTCATTGTACAAAGGCTGTTCATATTGCAAACACTACTAGTGCGCAG
				5927-5933	Wt	Forward	TGCGCACTAGTGAATGTTGATTGTGCATTGAGTATTAAGGATGTTGATATTATAAGCTTAATAAA
						Reverse	TTTATTAAAGCTTATAATATACATCTAATTTTAATACTCAATGCAACATCAACATTCACTAGTGCGCA
					Mut	Forward	CTGCGCACTAGTGAATGTTGATTGTGCATTGAGT C CTAAAAAAATTAGATGTATATTATAAGCTTAATAAA
						Reverse	TTTATTAAAGCTTATAATATACATCTAATTTTAGGACTCAATGCAACATCAACATTCACTAGTGCGCAG
		<i>ETV1</i>	ENSG0000006468	4455-4462	Wt	Forward	CTGCGCACTAGTCTTTGCATAGACTCCAGTATTGTTACTTCATTCTAAAGCTTAATAAA
						Reverse	TTTATTAAAGCTTAATTTAGAATGAAAGTAACATAACTGGAGTCTATATGCAAAAGACTAGTGCGCAG
					Mut	Forward	CTGCGCACTAGTCTTTGCATAGACTCCAG G CTTAGTTACTTCATTCTAAAGCTTAATAAA
						Reverse	TTTATTAAAGCTTAATTTAGAATGAAAGTAACATAAGCCTGGAGTCTATATGCAAAAGACTAGTGCGCAG

Table S2. Predicted pairing of target region and miRNA (data obtained from the TargetScanHuman database). The microRNA-target binding sites are highlighted in grey. Changed nucleotides in the mutant insert sequences are colored in red.

		Predicted pairing of target region (top) and miRNA (bottom)	
		Wild type target sequence	Mutant target sequence
Position 453-459 of KIT 3' UTR	hsa-miR-375	5' ...AACAGAACAUCAUUAGAACAAAG... 3' AGUGCGCUCGGCUUGCUGUUU	5' ...AACAGAACAUUUAG T GAAAG... 3' AGUGCGCUCGGCUUGCUGUUU

Position 549-555 of KIT 3' UTR hsa-miR-375	5' ...AUGAUCCCCAAGUGUGAACAAA... 3' AGUGCGCUCGGCUUGCUUGUUU	5' ...AUGAUCCCCAAGUGUGAA GG AAA... 3' AGUGCGCUCGGCUUG GU GUUU
Position 855-861 of KIT 3' UTR hsa-miR-375	5' ...CACAAAGCACAGUUUGAACAAA... 3' AGUGCGCUCGGCUUG-- CUUGUUU	5' ...CACAAAGCACAGUUUG AAG GG AAA... 3' AGUGCGCUCGGCUUG-- CU GUUU
Position 5849-5855 of EGFR 3' UTR hsa-miR-200b-3p	5' ...UUGCAAUGAUGAAAGCAGUAUU... 3' AGUAGUAUAGGUCCGUCAUAAU	5' ...UUGCAAUGAUGAAAGCAG CC UUU... 3' AGUAGUAUAGGUCCGUCAUAAU
Position 5927-5933 of EGFR 3' UTR hsa-miR-200b-3p	5' ...GUUGAUUGUGCAUUGAGUAUUAA... 3' AGUAGUAUAGGUCCGUCAUAAU	5' ...GUUGAUUGUGCAUUGAG CC UUAA... 3' AGUAGUAUAGGUCCGUCAUAAU
Position 4455-4462 of ETV1 3' UTR hsa-miR-200b-3p	5' ...UUUGCAUAUAGACUCCAGUAUUA... 3' AGUAGUAUAGGUCCGUCAUAAU	5' ...UUUGCAUAUAGACUCCAG GC UUA... 3' AGUAGUAUAGGUCCGUCAUAAU