ACCSCIENCE
PUBLISHING

Eurasian Journal of Medicine
and Oncology

ORIGINAL RESEARCH ARTICLE

Marked dysregulation of mature 3p and 5p arms of miR-182 and miR-490 isomiRs lacks prognostic value for 5-year survival in colorectal cancer

Supplementary files

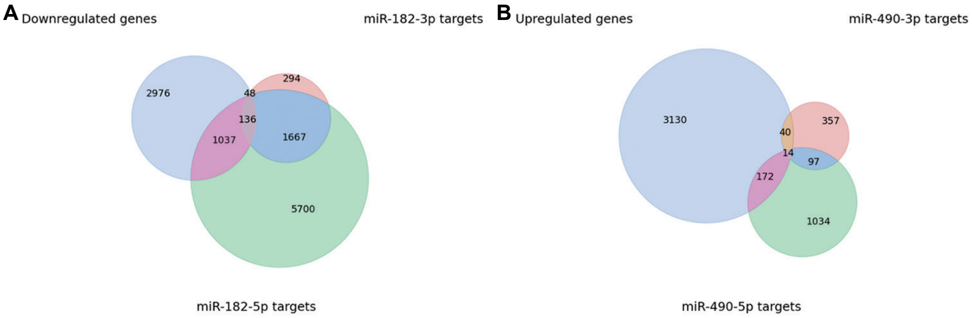

Figure S1. Identification of high-confidence miRNA targets through bioinformatic intersection. Venn diagrams illustrate the strategy used to filter putative miRNA targets. (A) miR-182 Intersection: The blue circle represents genes significantly downregulated in colorectal cancer (CRC) (Gene Expression Profiling Interactive Analysis 2; |log2fold change| > 1, $p<0.01$). The red and green circles represent targets predicted by miRNet 2.0 for miR-182-3p and miR-182-5p, respectively. The intersections (center) reveal the high-confidence targets used for enrichment analysis. (B) miR-490 intersection: The blue circle represents genes significantly upregulated in CRC. These were intersected with predicted targets for miR-490-3p (red) and miR-490-5p (green) to identify genes potentially repressed by this tumor suppressor.

Table S1. Kyoto Encyclopedia of Genes and Genomes pathway enrichment analysis for putative miR-182 targets

Enrichment FDR	nGenes	Pathway genes	Fold enrichment	Pathway	URL	Genes
3.26×10^{-5}	10	200	41.91	Path:hsa04510 Focal adhesion	http://www.genome.jp/kegg-bin/show_pathway?hsa04510	*COL6A2 FLNA FLNC ITGA5 LAMA5 LAMB2 THBS1 TLN1 VCL ACTN1*
7.88×10^{-4}	6	88	22.42	Path:hsa04512 ECM-receptor interaction	http://www.genome.jp/kegg-bin/show_pathway?hsa04512	*COL6A2 HSPG2 ITGA5 LAMA5 LAMB2 THBS1*
7.57×10^{-3}	7	202	135.39	Path:hsa05205 Proteoglycans in cancer	http://www.genome.jp/kegg-bin/show_pathway?hsa05205	*FLNA FLNC GAB1 HSPG2 ITGA5 THBS1 FZD4*
1.15×10^{-2}	9	384	97.78	Path:hsa05010 Alzheimer's disease	http://www.genome.jp/kegg-bin/show_pathway?hsa05010	*CSF1 TUBB2B KLC1 MT-ATP6 MT-CO1 MT-CO3 MT-ND5 FZD4 ULK1*
1.15×10^{-2}	10	476	35.2	Path:hsa05022 Pathways of neurodegeneration-multiple diseases	http://www.genome.jp/kegg-bin/show_pathway?hsa05022	*CSF1 TUBB2B KLC1 MT-ATP6 MT-CO1 MT-CO3 MT-ND5 PRNP FZD4 ULK1*
1.36×10^{-2}	3	29	45.13	Path:hsa04392 Hippo signaling pathway-multiple species	http://www.genome.jp/kegg-bin/show_pathway?hsa04392	*FRMD6 WWTR1 TEAD1*
1.69×10^{-2}	7	266	33.21	Path:hsa05012 Parkinson's disease	http://www.genome.jp/kegg-bin/show_pathway?hsa05012	*DUSP1 TUBB2B KLC1 MT-ATP6 MT-CO1 MT-CO3 MT-ND5*
1.72×10^{-2}	7	273	40	Path:hsa05020 Prion disease	http://www.genome.jp/kegg-bin/show_pathway?hsa05020	*TUBB2B KLC1 MT-ATP6 MT-CO1 MT-CO3 MT-ND5 PRNP*
1.82×10^{-2}	8	364	30.88	Path:hsa05014 Amyotrophic lateral sclerosis	http://www.genome.jp/kegg-bin/show_pathway?hsa05014	*NXF1 TUBB2B KLC1 MT-ATP6 MT-CO1 MT-CO3 MT-ND5 ULK1*
2.40×10^{-2}	7	306	48.89	Path:hsa05016 Huntington disease	http://www.genome.jp/kegg-bin/show_pathway?hsa05016	*TUBB2B KLC1 MT-ATP6 MT-CO1 MT-CO3 MT-ND5 ULK1*
2.40×10^{-2}	6	223	29.83	Path:hsa05208 Chemical carcinogenesis-reactive oxygen species	http://www.genome.jp/kegg-bin/show_pathway?hsa05208	*SLC26A2 ABL1 MT-ATP6 MT-CO1 MT-CO3 MT-ND5*
2.68×10^{-2}	5	161	60.69	Path:hsa05206 MicroRNAs in cancer	http://www.genome.jp/kegg-bin/show_pathway?hsa05206	*ABL1 ITGA5 BCL2L2 THBS1 VIM*
2.86×10^{-2}	6	249	3.43	Path:hsa05132 *Salmonella* infection	http://www.genome.jp/kegg-bin/show_pathway?hsa05132	*AHNAK2 FLNA FLNC TUBB2B KLC1 AHNAK*
2.86×10^{-2}	4	102	18.53	Path:hsa05146 Amoebiasis	http://www.genome.jp/kegg-bin/show_pathway?hsa05146	*LAMA5 LAMB2 VCL ACTN1*
2.86×10^{-2}	7	331	18.15	Path:hsa05165 Human papillomavirus infection	http://www.genome.jp/kegg-bin/show_pathway?hsa05165	*COL6A2 TUBG2 ITGA5 LAMA5 LAMB2 THBS1 FZD4*
3.69×10^{-2}	7	354	22.28	Path:hsa04151 PI3K-Akt signaling pathway	http://www.genome.jp/kegg-bin/show_pathway?hsa04151	*COL6A2 CSF1 ITGA5 LAMA5 LAMB2 THBS1 OSMR*
5.95×10^{-2}	4	134	19.13	Path:hsa00190 Oxidative phosphorylation	http://www.genome.jp/kegg-bin/show_pathway?hsa00190	*MT-ATP6 MT-CO1 MT-CO3 MT-ND5*
7.45×10^{-2}	3	77	13.44	Path:hsa05100 Bacterial invasion of epithelial cells	http://www.genome.jp/kegg-bin/show_pathway?hsa05100	*GAB1 ITGA5 VCL*
8.01×10^{-2}	4	151	14.79	Path:hsa04145 Phagosome	http://www.genome.jp/kegg-bin/show_pathway?hsa04145	*TUBB2B ITGA5 THBS1 COLEC12*
8.67×10^{-2}	4	157	13.44	Path:hsa04390 Hippo signaling pathway	http://www.genome.jp/kegg-bin/show_pathway?hsa04390	*FRMD6 WWTR1 TEAD1 FZD4*
1.00×10^{-1}	3	91	10.41	Path:hsa04970 Salivary secretion	http://www.genome.jp/kegg-bin/show_pathway?hsa04970	*GUCY1A1 ATP2B4 VAMP2*
1.82×10^{-1}	4	203	14.55	Path:hsa05415 Diabetic cardiomyopathy	http://www.genome.jp/kegg-bin/show_pathway?hsa05415	*MT-ATP6 MT-CO1 MT-CO3 MT-ND5*

Notes: Input genes were predicted targets of miR-182-3p and miR-182-5p that were significantly downregulated in colorectal cancer tissues (intersection of miRNet 2.0 and Gene Expression Profiling Interactive Analysis 2 data). Enrichment was performed using ShinyGO v0.77 (hypergeometric test; background: all human protein-coding genes). FDR-adjusted *P*-values are reported; pathways with FDR < 0.05 were considered significant.

Abbreviations: AKT: Protein kinase B; ECM: Extracellular matrix; FDR: False discovery rate; nGenes: Number of genes enriched in the pathway; PI3K: Phosphoinositide 3-kinase.

Table S2. Kyoto Encyclopedia of Genes and Genomes pathway enrichment analysis for putative miR-490 targets

Enrichment FDR	nGenes	Pathway genes	Fold enrichment	Pathway	URL	Genes
1.39×10^{-3}	3	126	41.90659	Path:hsa04110 Cell cycle	http://www.genome.jp/kegg-bin/show_pathway?hsa04110	*MCM3 PRKDC YWHAB*
5.38×10^{-2}	2	157	22.42136	Path:hsa04390 Hippo signaling pathway	http://www.genome.jp/kegg-bin/show_pathway?hsa04390	*WWC1 YWHAB*
7.61×10^{-2}	1	13	135.3905	Path:hsa03450 Non-homologous end-joining	http://www.genome.jp/kegg-bin/show_pathway?hsa03450	*PRKDC*
7.89×10^{-2}	1	18	97.78205	Path:hsa00061 Fatty acid biosynthesis	http://www.genome.jp/kegg-bin/show_pathway?hsa00061	*FASN*
8.53×10^{-2}	1	50	35.20154	Path:hsa00510 N-glycan biosynthesis	http://www.genome.jp/kegg-bin/show_pathway?hsa00510	*STT3B*
8.53×10^{-2}	1	39	45.13018	Path:hsa00513 Various types of N-glycan biosynthesis	http://www.genome.jp/kegg-bin/show_pathway?hsa00513	*STT3B*
8.53×10^{-2}	1	53	33.209	Path:hsa00600 Sphingolipid metabolism	http://www.genome.jp/kegg-bin/show_pathway?hsa00600	*CERS6*
8.53×10^{-2}	1	44	40.00175	Path:hsa00970 Aminoacyl-tRNA biosynthesis	http://www.genome.jp/kegg-bin/show_pathway?hsa00970	*IARS1*
8.53×10^{-2}	1	57	30.87854	Path:hsa01212 Fatty acid metabolism	http://www.genome.jp/kegg-bin/show_pathway?hsa01212	*FASN*
8.53×10^{-2}	1	36	48.89103	Path:hsa03030 DNA replication	http://www.genome.jp/kegg-bin/show_pathway?hsa03030	*MCM3*
8.53×10^{-2}	1	59	29.83181	Path:hsa04330 Notch signaling pathway	http://www.genome.jp/kegg-bin/show_pathway?hsa04330	*JAG2*
8.53×10^{-2}	1	29	60.69231	Path:hsa04392 Hippo signaling pathway-multiple species	http://www.genome.jp/kegg-bin/show_pathway?hsa04392	*WWC1*
9.80×10^{-2}	3	1538	3.43318	Path:hsa01100 Metabolic pathways	http://www.genome.jp/kegg-bin/show_pathway?hsa01100	*STT3B FASN CERS6*
9.80×10^{-2}	1	95	18.52713	Path:hsa01522 Endocrine resistance	http://www.genome.jp/kegg-bin/show_pathway?hsa01522	*JAG2*
9.80×10^{-2}	1	97	18.14512	Path:hsa03015 mRNA surveillance pathway	http://www.genome.jp/kegg-bin/show_pathway?hsa03015	*PABPC1*
9.80×10^{-2}	1	79	22.27945	Path:hsa03018 RNA degradation	http://www.genome.jp/kegg-bin/show_pathway?hsa03018	*PABPC1*
9.80×10^{-2}	1	92	19.13127	Path:hsa04658 Th1 and Th2 cell differentiation	http://www.genome.jp/kegg-bin/show_pathway?hsa04658	*JAG2*
1.02×10^{-1}	1	131	13.4357	Path:hsa04068 FoxO signaling pathway	http://www.genome.jp/kegg-bin/show_pathway?hsa04068	*FOXG1*
1.02×10^{-1}	1	119	14.79056	Path:hsa04071 Sphingolipid signaling pathway	http://www.genome.jp/kegg-bin/show_pathway?hsa04071	*CERS6*
1.02×10^{-1}	1	131	13.4357	Path:hsa04114 Oocyte meiosis	http://www.genome.jp/kegg-bin/show_pathway?hsa04114	*YWHAB*
1.02×10^{-1}	1	169	10.41466	Path:hsa04141 Protein processing in endoplasmic reticulum	http://www.genome.jp/kegg-bin/show_pathway?hsa04141	*STT3B*
1.02×10^{-1}	1	121	14.54609	Path:hsa04152 AMPK signaling pathway	http://www.genome.jp/kegg-bin/show_pathway?hsa04152	*FASN*
1.39×10^{-3}	3	126	10.35339	Path:hsa04310 Wnt signaling pathway	http://www.genome.jp/kegg-bin/show_pathway?hsa04310	*CACYBP*
5.38×10^{-2}	2	157	12.84728	Path:hsa04910 Insulin signaling pathway	http://www.genome.jp/kegg-bin/show_pathway?hsa04910	*FASN*
7.61×10^{-2}	1	13	12.48282	Path:hsa04936 Alcoholic liver disease	http://www.genome.jp/kegg-bin/show_pathway?hsa04936	*FASN*
7.89×10^{-2}	1	18	11.21068	Path:hsa05160 Hepatitis C	http://www.genome.jp/kegg-bin/show_pathway?hsa05160	*YWHAB*
8.53×10^{-2}	1	50	10.86467	Path:hsa05161 Hepatitis B	http://www.genome.jp/kegg-bin/show_pathway?hsa05161	*YWHAB*
8.53×10^{-2}	1	39	11.97331	Path:hsa05224 Breast cancer	http://www.genome.jp/kegg-bin/show_pathway?hsa05224	*JAG2*
8.53×10^{-2}	1	53	8.713252	Path:hsa05203 Viral carcinogenesis	http://www.genome.jp/kegg-bin/show_pathway?hsa05203	*YWHAB*
8.53×10^{-2}	1	44	4.971969	Path:hsa04151 PI3K-Akt signaling pathway	http://www.genome.jp/kegg-bin/show_pathway?hsa04151	*YWHAB*

Notes: Input genes were predicted targets miR-490-3p and miR-490-5p that were significantly upregulated in colorectal cancer tissues. Enrichment was performed using ShinyGO v0.77 (hypergeometric test; background: all human protein-coding genes). FDR-adjusted *P* values are reported; pathways with FDR<0.05 were considered significant.

Abbreviations: AKT: Protein kinase B; AMPK: Adenosine monophosphate-activated protein kinase; FDR: False discovery rate; nGenes: Number of genes enriched in the pathway; PI3K: Phosphoinositide 3-kinase.