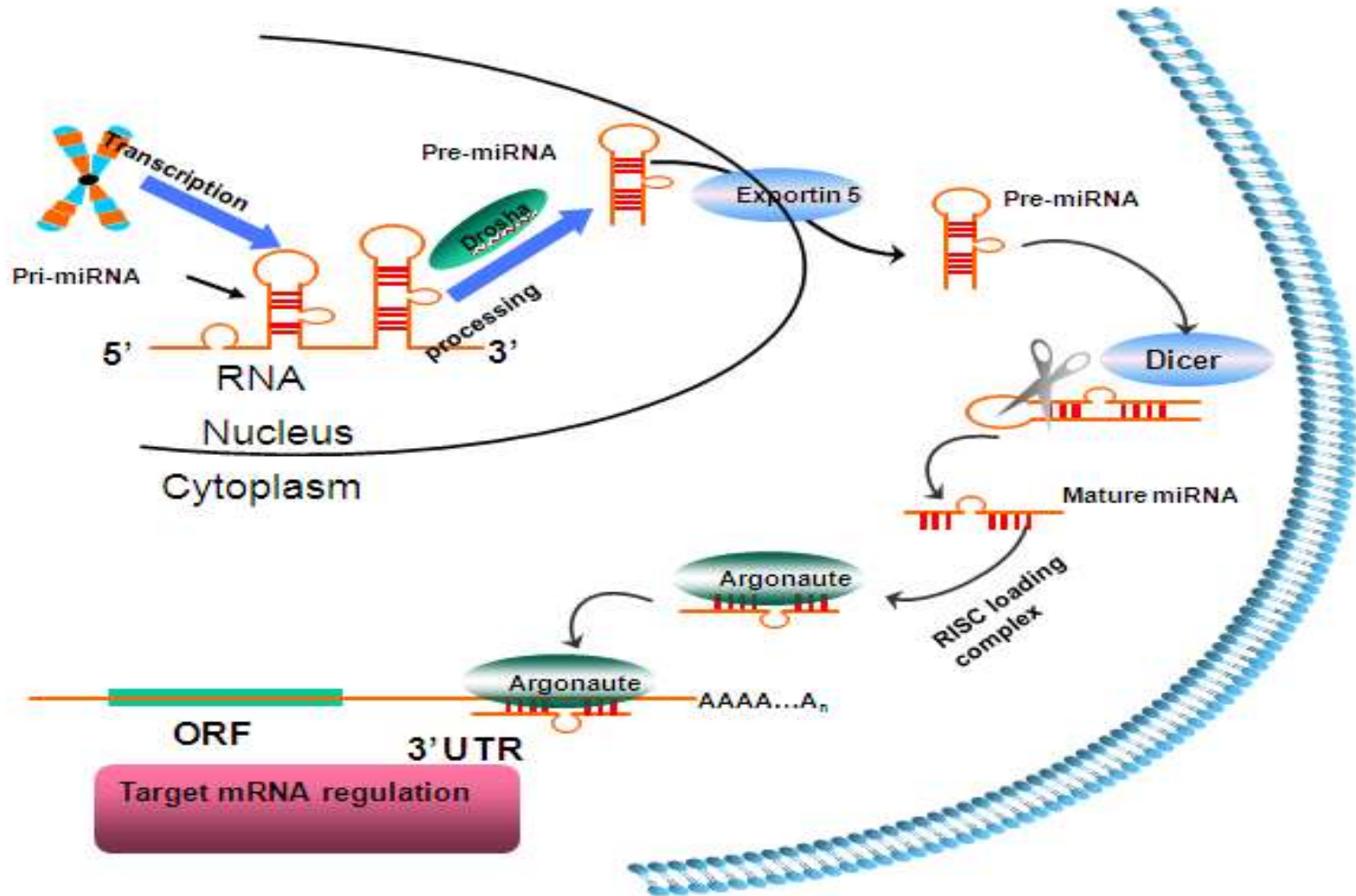


Role of MicroRNA-1 (miR-1) in chordoma and its therapeutic potentials

Zhenfeng Duan

Biological principles of miRNA(miR)



Differential expression of microRNA (miRNA) in chordoma reveals a role for miRNA-1 in Met expression

Zhenfeng Duan^{1,*}, Edwin Choy¹, G. Petur Nielsen², Andrew Rosenberg², John Iafrate², Cao Yang¹, Joe Schwab¹, Henry Mankin¹, Ramnik Xavier³, Francis J. Hornicek¹

Article first published online: 29 DEC 2009

DOI: 10.1002/jor.21055

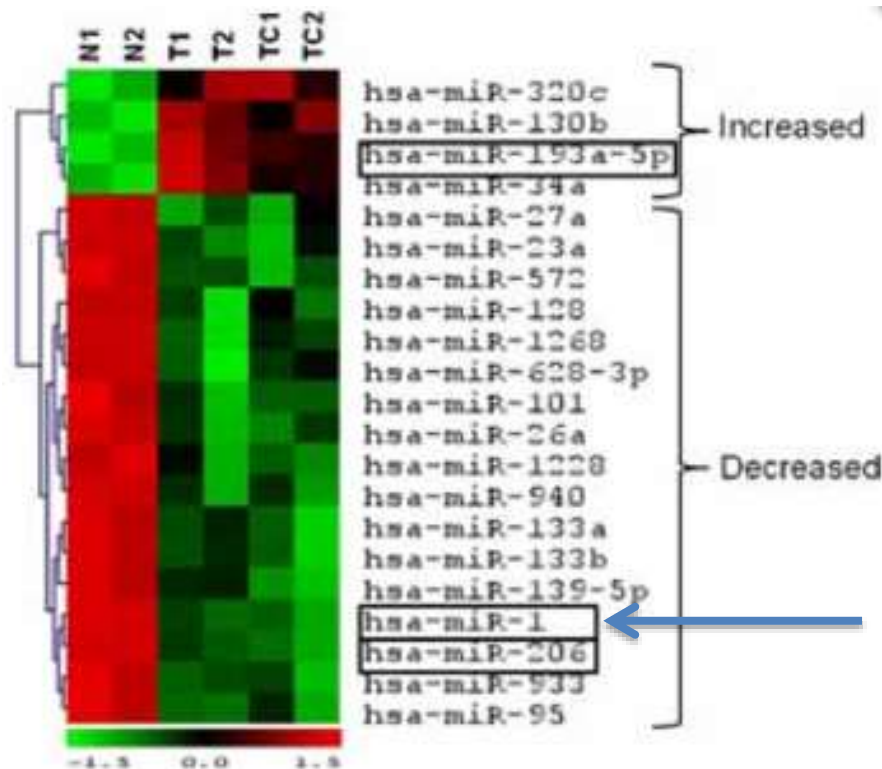
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Issue



Journal of Orthopaedic Research

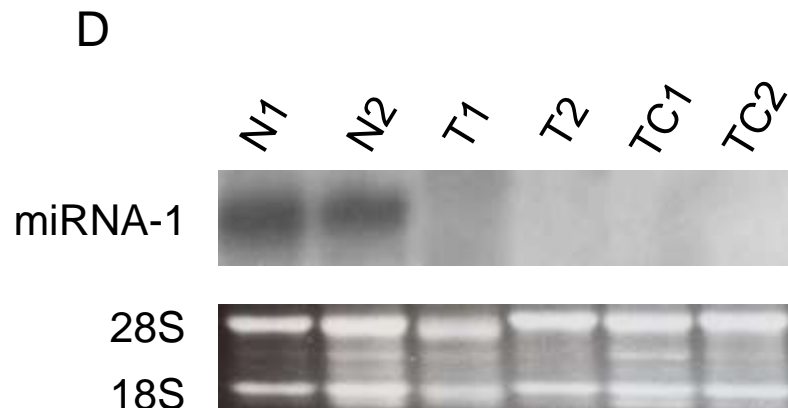
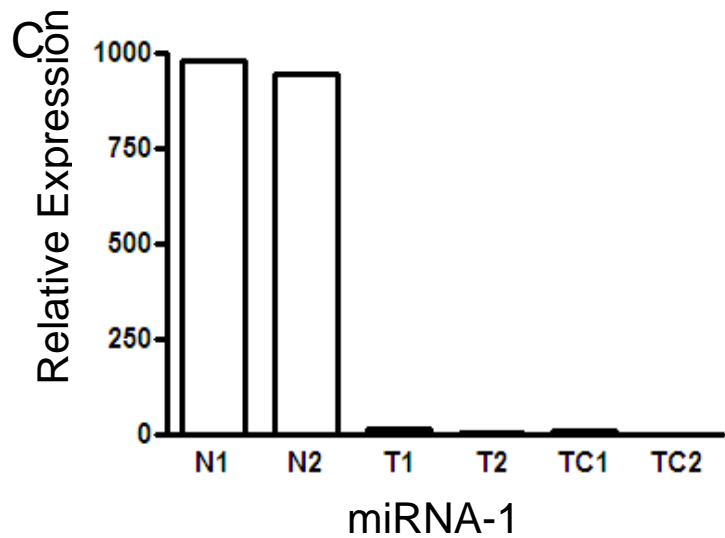
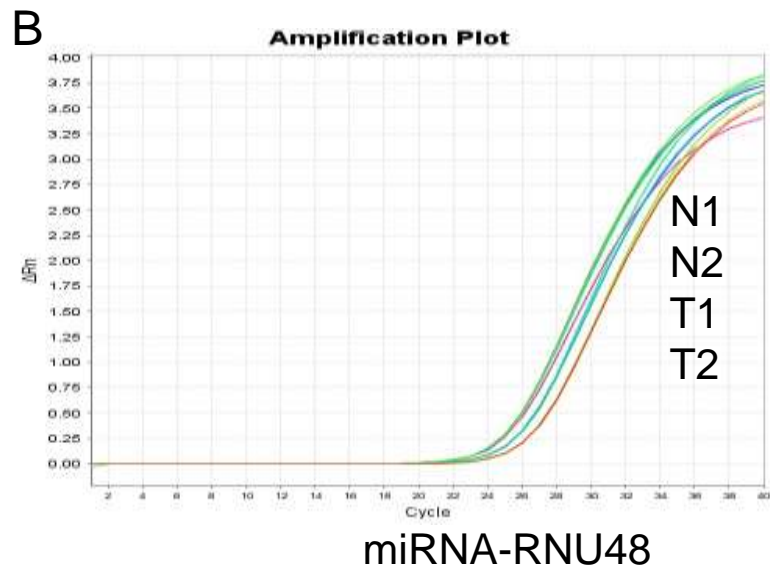
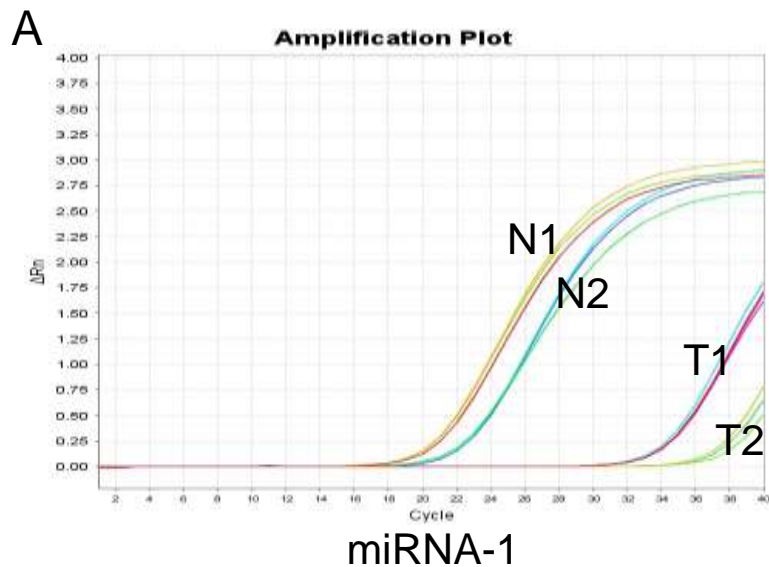
Volume 28, Issue 6, pages 746–752, June 2010



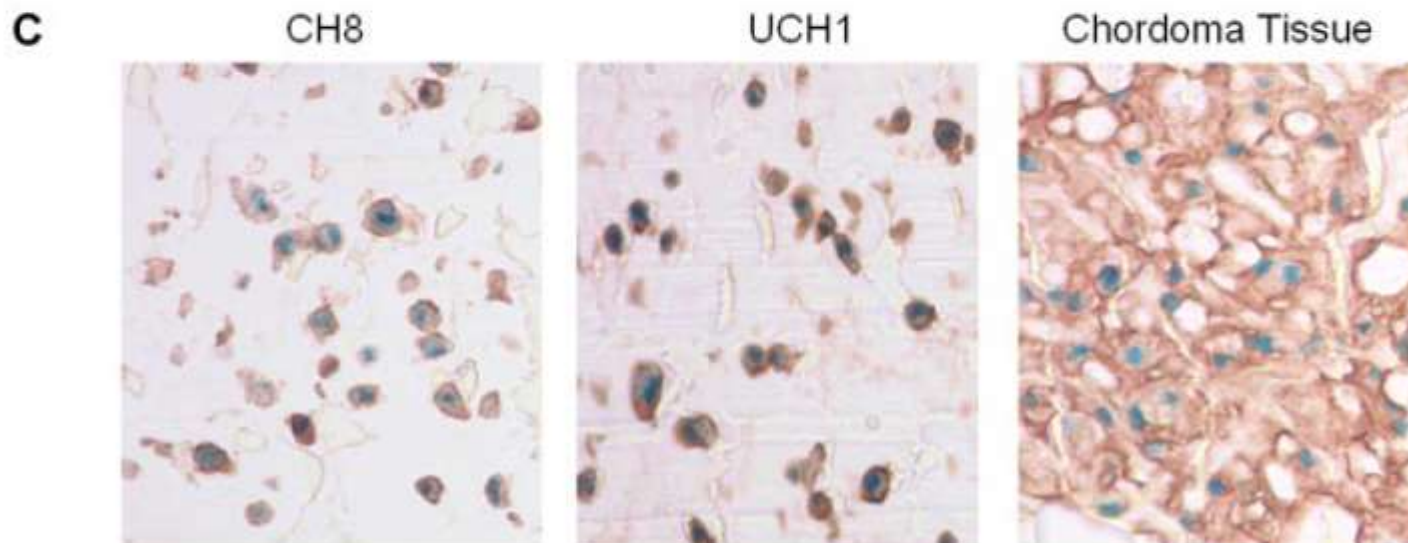
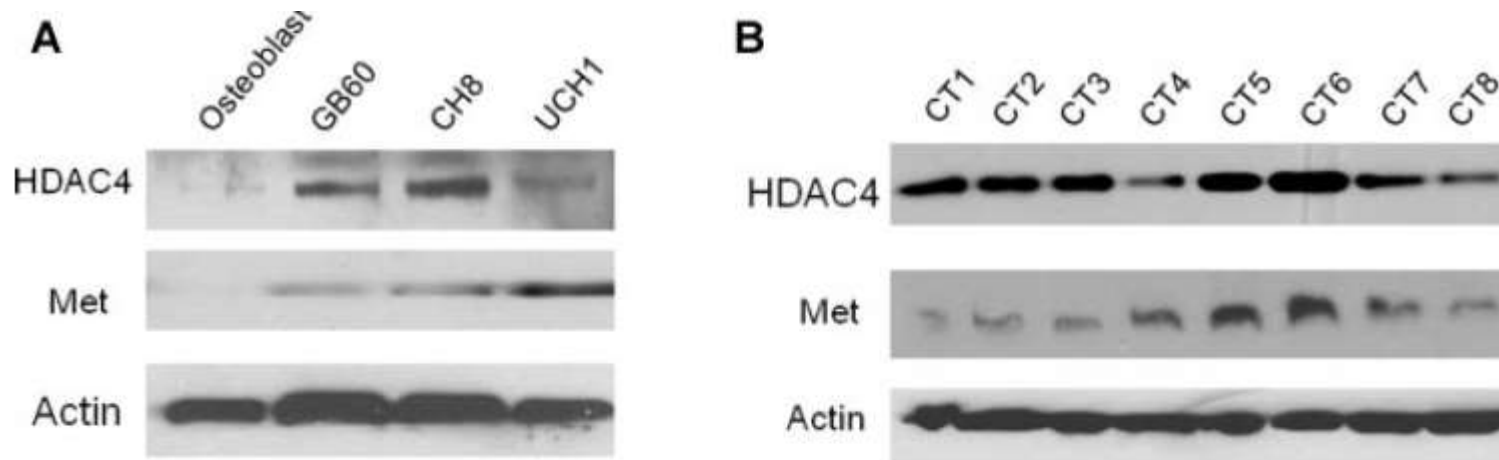
Top decreased expression of miRNAs in both chordoma tissues and chordoma cell lines

Name of miRNA	Normal (mean value)	Tumor tissues and cell lines(mean value)	Fold	p-value (<0.01)
<u>miRNA-1</u>	<u>27,231</u>	<u>31</u>	<u>878</u>	6.75E-04
miRNA-206	16,049	29	553	8.89E-04
miRNA-133b	15,709	68	231	4.41E-03
miRNA-95	4,243	19	223	2.85E-03
miRNA-133a	16,095	75	215	4.49E-03
miRNA-1268	2,043	134	145	4.90E-03
miRNA-101	875	21	42	4.19E-03
miRNA-139-5p	705	17	41	5.90E-03

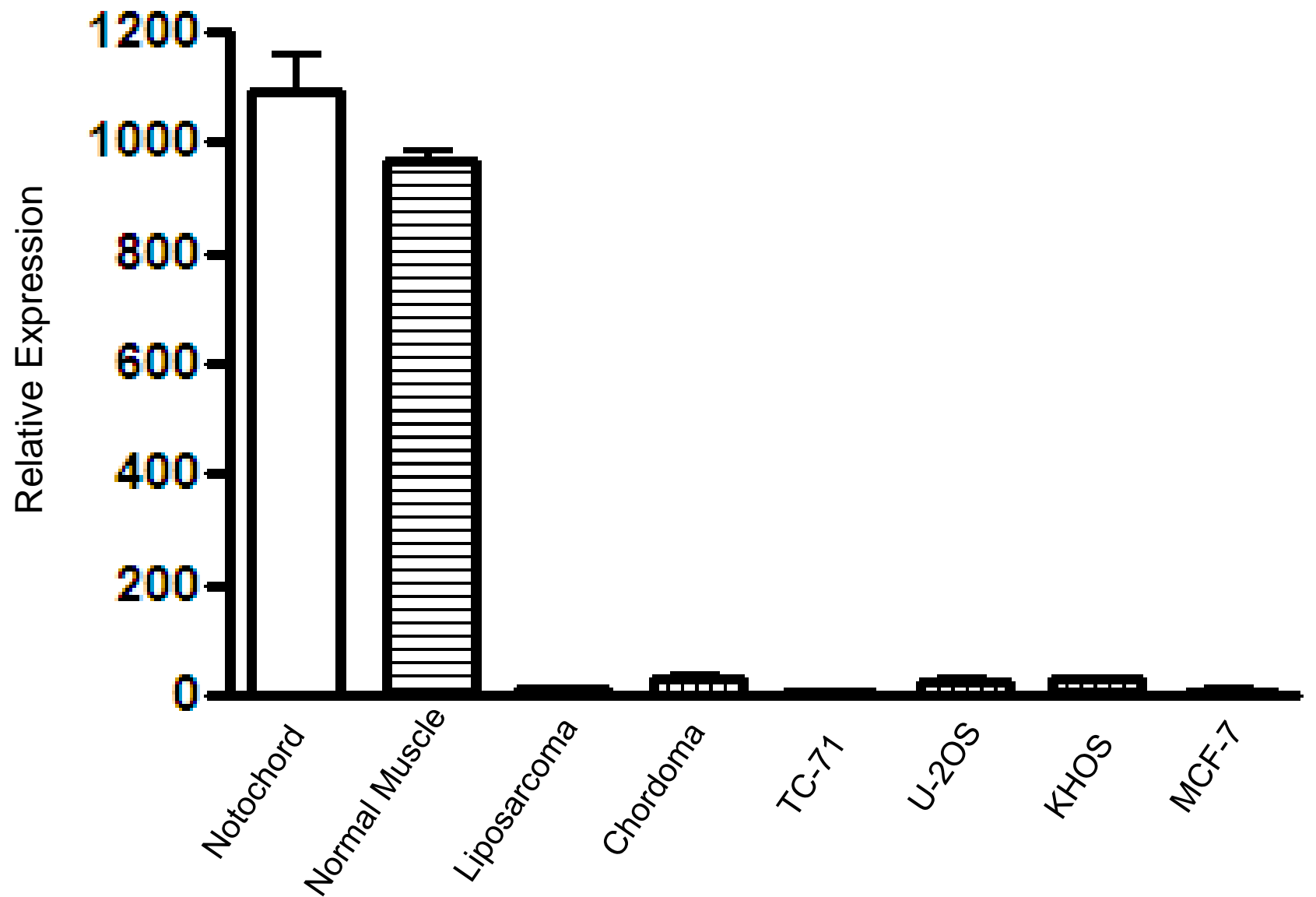
Real-time RT-PCR and Northern blot detection of mature miR-1 in chordoma tissues



Expression of miR-1 target gene HDAC4 and Met proteins in chordoma cell lines and chordoma tissues

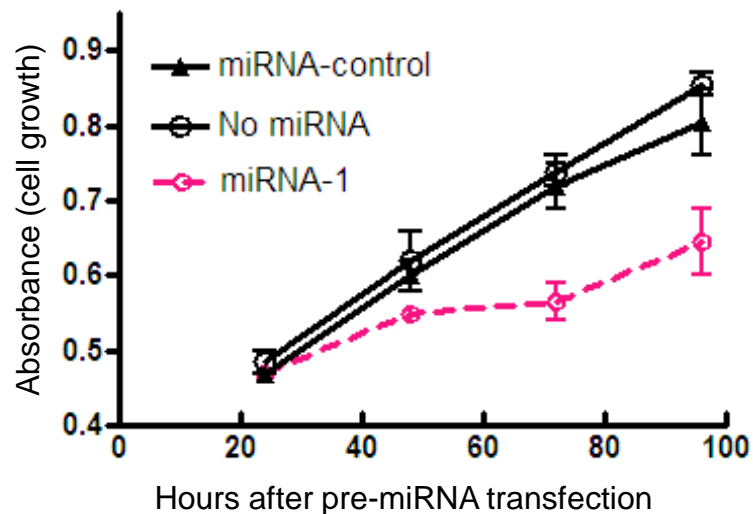


miR-1 expression in other type of tumors

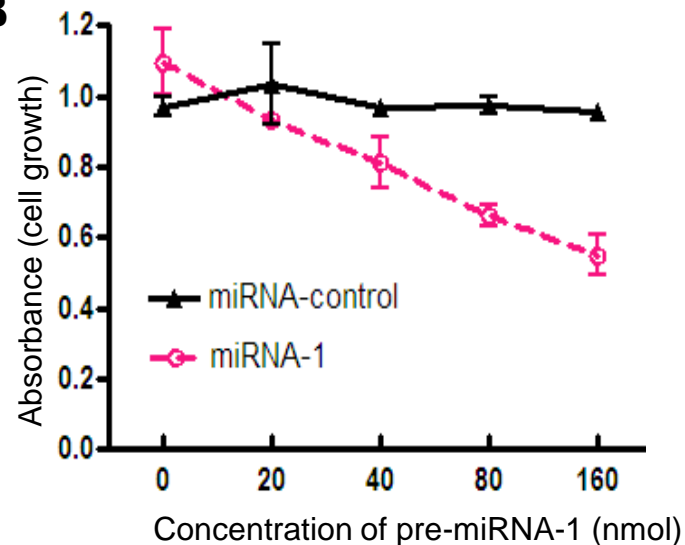


Transfection of miRNA-1 into chordoma cells UCH1 suppresses Met expression and inhibits cell growth

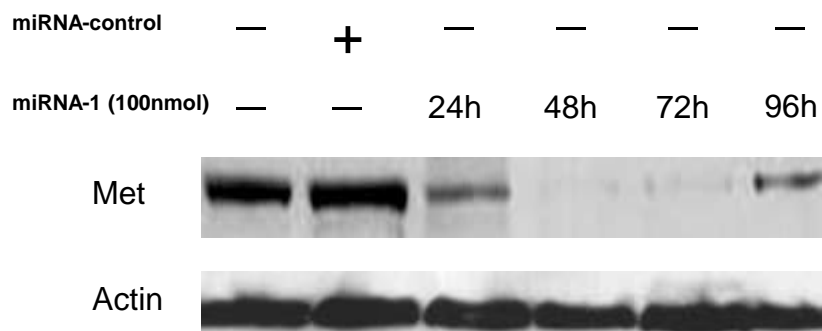
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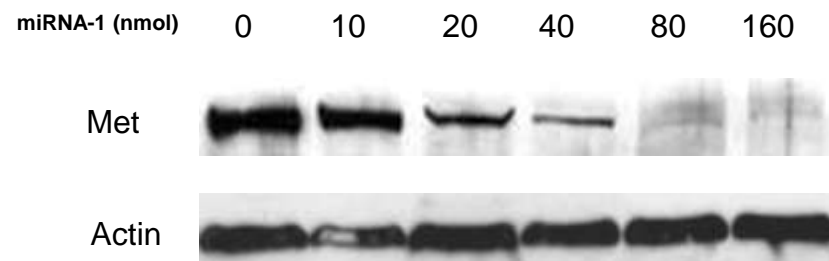
B



C



D



Mod Pathol. 1997 Aug;10(8):832-8.

Expression of c-met proto-oncogene product (c-MET) in benign and malignant bone tumors.

Naka T, Iwamoto Y, Shinohara N, Ushijima M, Chuman H, Tsuneyoshi M.

Department of Pathology II, Faculty of Medicine, Kyushu University, Fukuoka, Japan.

- Met expression was most frequently detected in chordoma (94.4%), followed by chondrosarcoma (54.2%) and osteosarcoma (23.3%).

Spine (Phila Pa 1976). 2008 Dec 1;33(25):2774-80. doi: 10.1097/BRS.0b013e31817e2d1e.

Identifying mechanisms for therapeutic intervention in chordoma: c-Met oncoprotein.

Ostroumov E, Hunter CJ.

From the Centre for Bioengineering Research and Education, University of Calgary, Calgary, Alberta, Canada.

- Met oncoprotein plays a leading role in the metastatic process in chordoma, and that a c-Met-HGF pair is involved in chordoma malignancy.

***MET* overexpressing chordomas frequently exhibit polysomy of chromosome 7 but no *MET* activation through sarcoma-specific gene fusions**

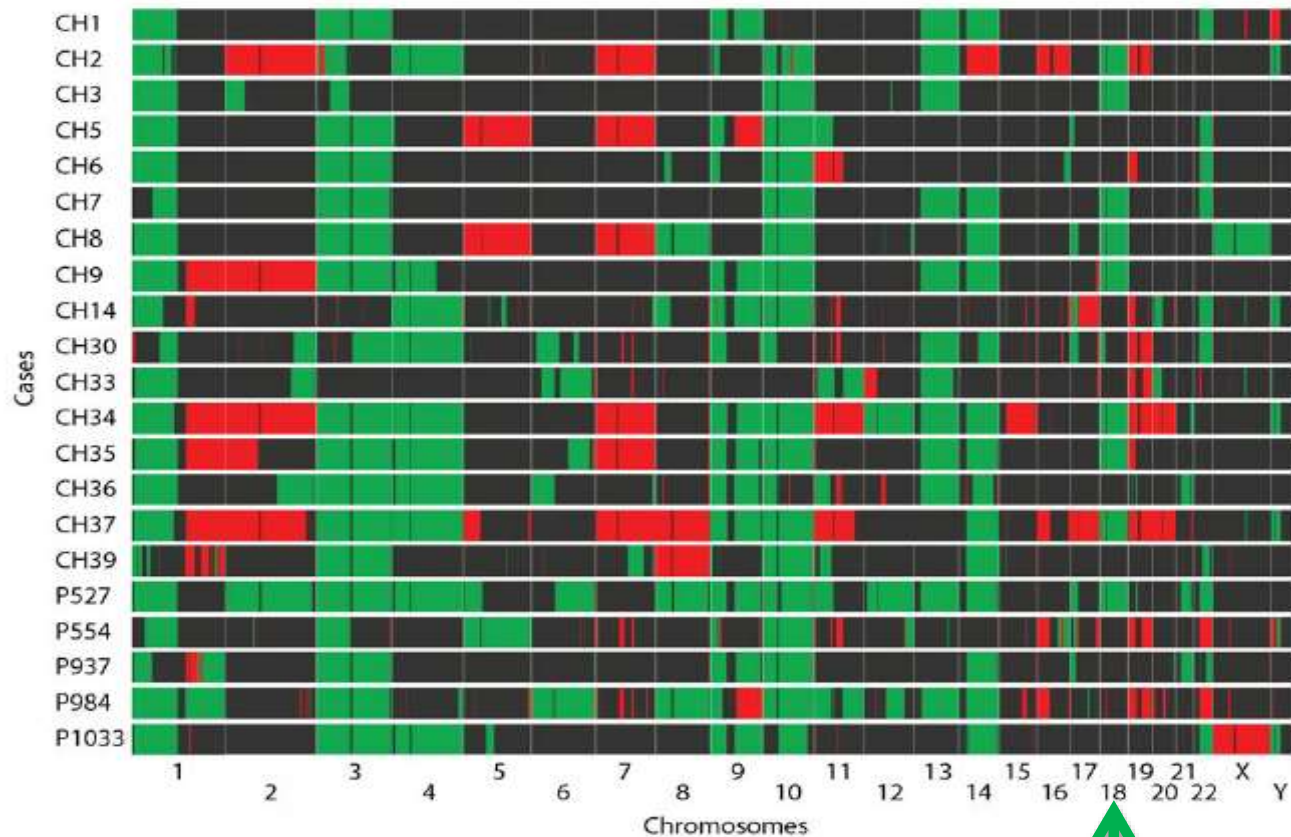
Florian Grabelius · Margarethe J. Konik · Karl Worm · Sien-Yi Sheu · Johannes A. P. van de Nes · Sebastian Bauer · Werner Paulus · Rupert Egensperger · Kurt W. Schmid

	Negative IRS 0	Low IRS 1-3	Moderate IRS 4–8	High IRS 9–12
C-MET	0 (0%)	1 (1.5%)	36 (54.5%)	29 (43.9%)
Pan-CK	0 (0%)	0 (0%)	4 (6.1%)	62 (93.9%)
S100	0 (0%)	3 (4.5%)	29 (43.9%)	34 (51.5%)
EMA	12 (18.2%)	24 (36.4%)	21 (31.8%)	9 (13.6%)

*** FISH analysis demonstrated that *MET* gene amplification is not the cause of *MET* overexpression in chordomas.**

Recurrent Chromosomal Copy Number Alterations in Sporadic Chordomas

Long Phi Le^{1*}, G. Petur Nielsen¹, Andrew Eric Rosenberg¹, Dafydd Thomas², Julie M. Batten¹, Vikram Deshpande¹, Joseph Schwab³, Zhenfeng Duan³, Ramnik J. Xavier^{4,5}, Francis J. Hornicek³, A. John Iafrate¹



Similar to previous findings, large copy number losses (green), involving chromosomes 18 was more common than copy number gains.

Research Article

Laboratory Investigation **92**, 571-583 (April 2012) | doi:10.1038/labinve:

Downregulation of microRNAs miR-1, -206 and -29 stabilizes PAX3 and CCND2 expression in rhabdomyosarcoma

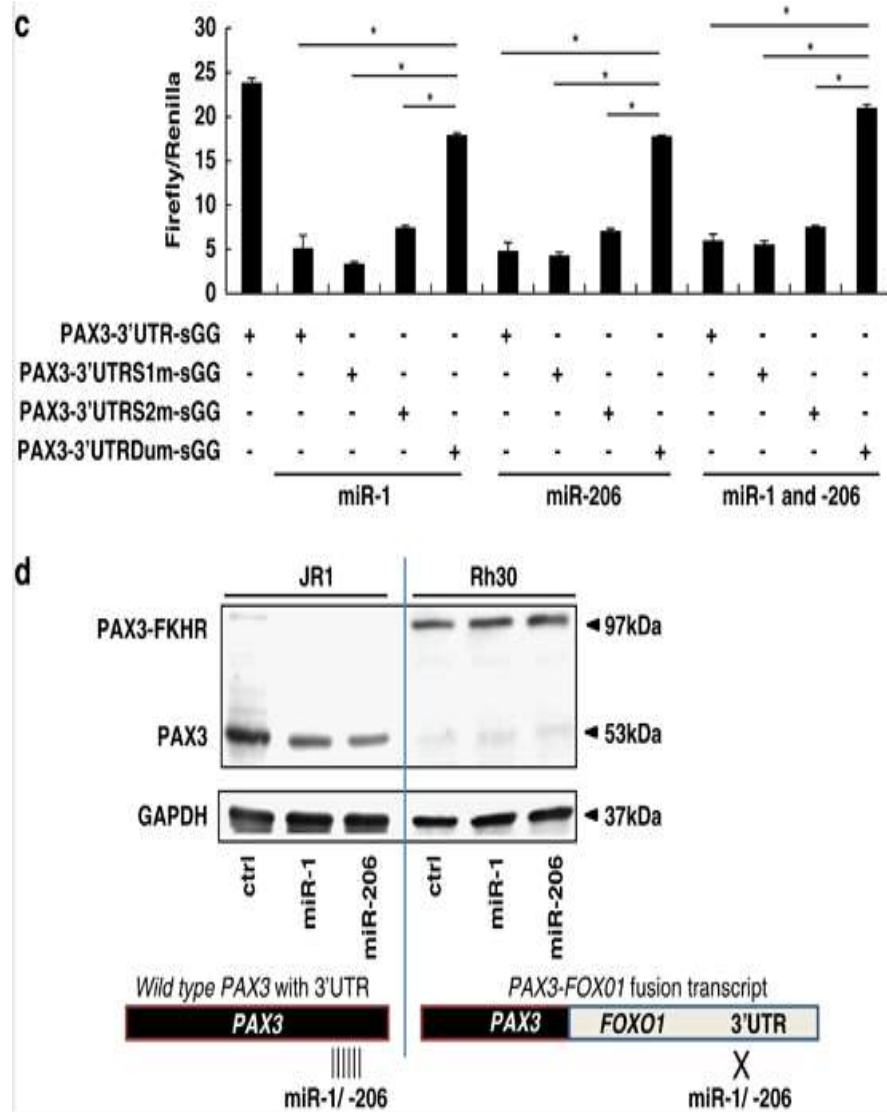
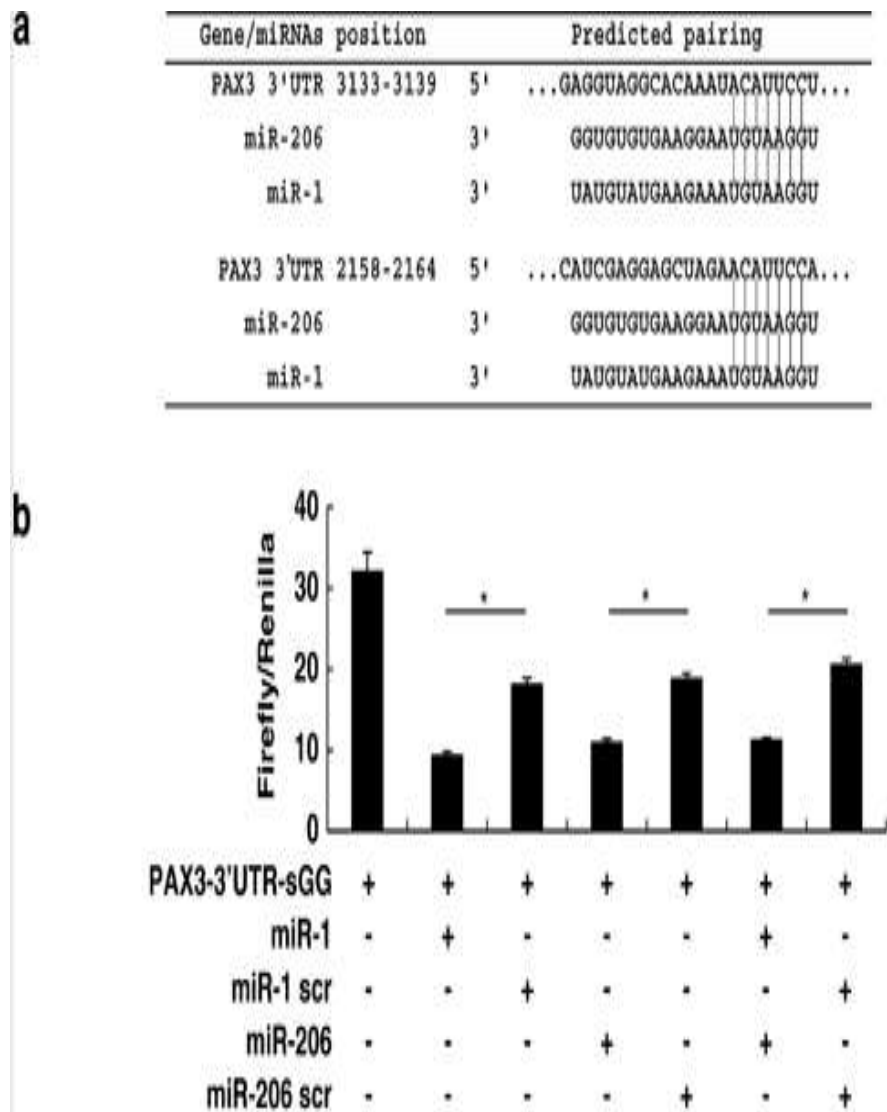
Lihua Li, Aaron L Sarver, Setara Alamgir and Subbaya Subramanian

Proc Natl Acad Sci U S A. 2011 Jul 19;108(29):11936-41. doi: 10.1073/pnas.1105362108. Epub 2011 Jul 5.

MicroRNA regulation of the paired-box transcription factor Pax3 confers robustness to developmental timing of myogenesis.

Goljanek-Whysall K, Sweetman D, Abu-Elmagd M, Chapnik E, Dalmay T, Hornstein E, Münsterberg A.

miR-1 regulates PAX3 expression through sequence-specific binding to its 3' UTR



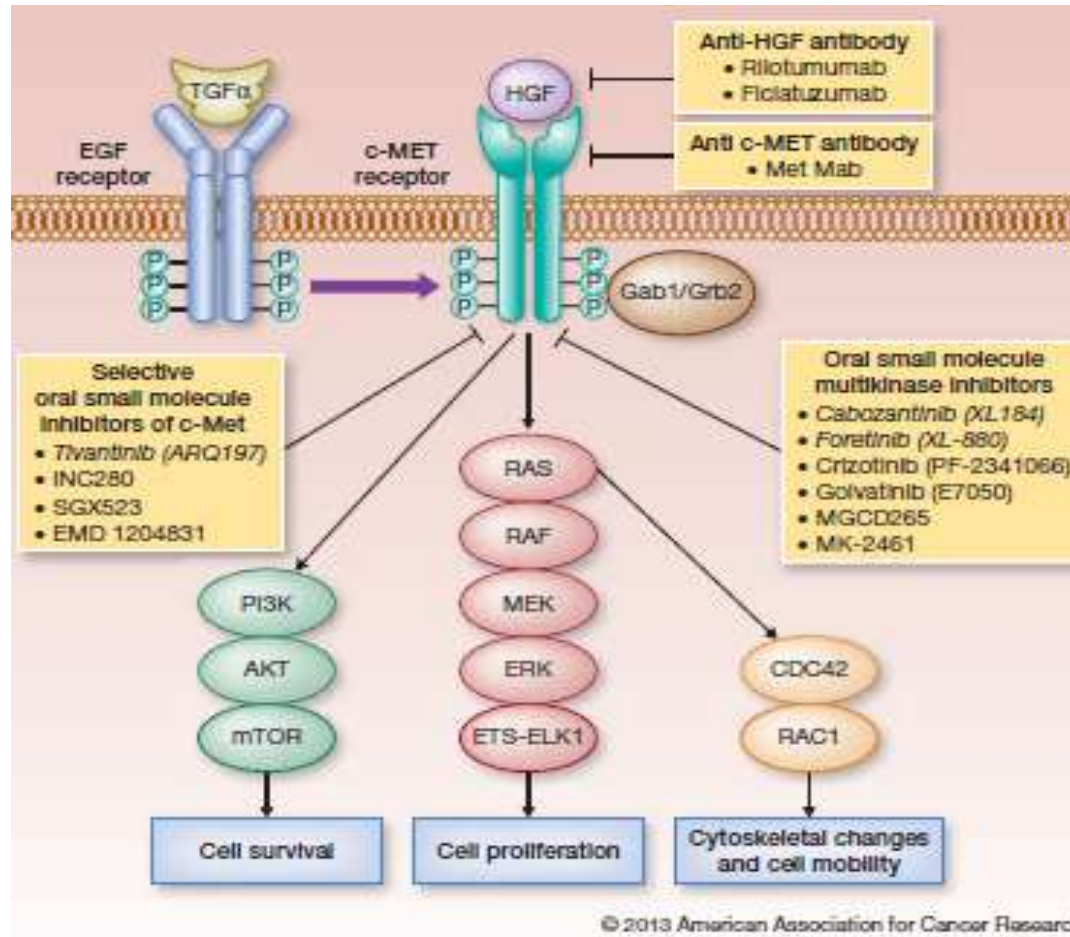
Altered expression of miR-1 in different cancers

Cancer type	miR-1 down regulation	miR-1 target genes
Lung cancer	Primary lung cancer tissue and serum	<u>MET</u> ;FoxP1;HDAC4,Slug
Colon cancer	Different colon cancer tissues	<u>MET</u>
Genitourinary cancer	Cancer cell lines and tissues	LASP1;TAGLN2;SRSF9;PTMA;PNP1
Head & Neck cancer	laryngeal carcinoma and MSSCC	TAGLN2;PTMA;FN1;PNP
Thyroid cancer	Thyroid adenomas and carcinomas	CCND2;CXCR4;CXCL12
Hepatocellular cancer	HCC cell lines and tumor tissues	<u>MET</u> ;FoxP1;HDAC4;ET-1
Sarcoma	Sarcoma cell lines and sarcoma tissues	<u>MET</u> ;CCND2;HDAC4

Oncogenes and oncogenic pathway targets of miR-1 in cancers

Gene name	Genbank Accession #	Chromosome location	Functions
Met	NM_000245.2	7q31	Proto-oncogenic receptor tyrosine kinase
HDAC4	NM_006037.3	2q37.3	Histone deacetylase activity and represses transcription
Pim-1	NM_002648.3	6p21.2	Proto-oncogene
FOXP1	NM_001244810.1	3p14.1	Regulate gene transcription; tumor suppressor
TAGLN2	NM_003564.1	1q21-q25	Earliest marker of differentiated smooth muscle
PNP	NM_000270.3	14q13.1	Purine metabolism
PTMA	NM_002823.4	2q37.1	Enhance cell-mediated immunity
CXCR4	NM_001008540.1	2q21	chemokine receptor
CCND2	NM_001759.3	12p13	Regulator of cyclin-dependent kinase
SRSF9	NM_003769.2	12q24.31	
FN1	NM_212482.1	2q34	Involved in cell adhesion, growth, migration
ETS1	NM_001143820.1	11q23.3	Proto-oncogene
endothelin 1	NM_001955.4	6p24.1	Involve in vascular disorders
Slug	NM_003068.4	8q11	Transcriptional repressor and has antiapoptotic activity
CXCL12	NM_199168.3	10q11.1	Chemotactic for lymphocytes

Therapeutic Inhibitors of the c-MET Signaling Pathway



CCR Reviews

ACR

Clinical Cancer Research

ACR

Targeting the HGF/c-MET Pathway in Hepatocellular Carcinoma

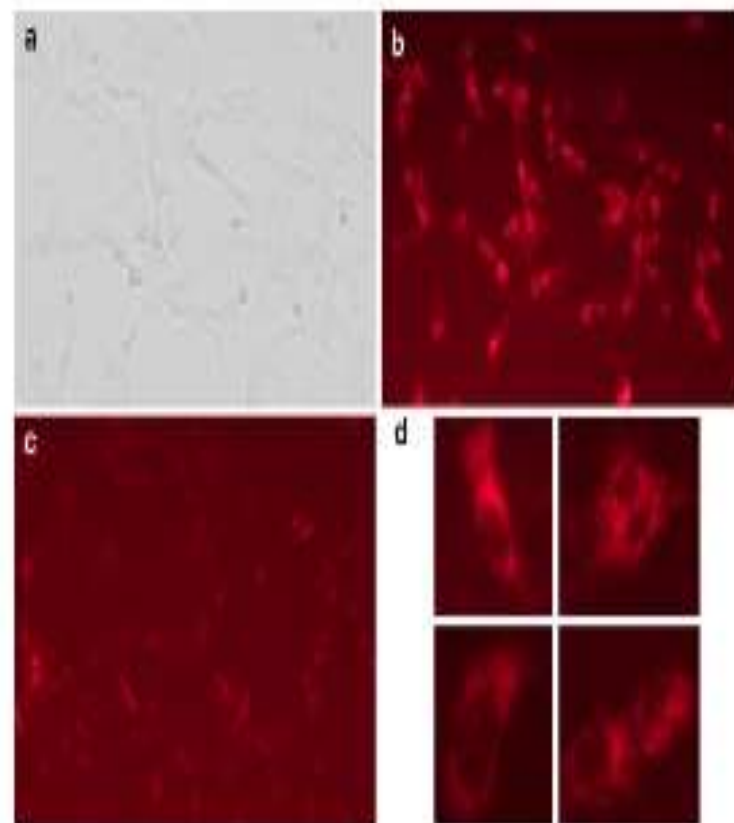
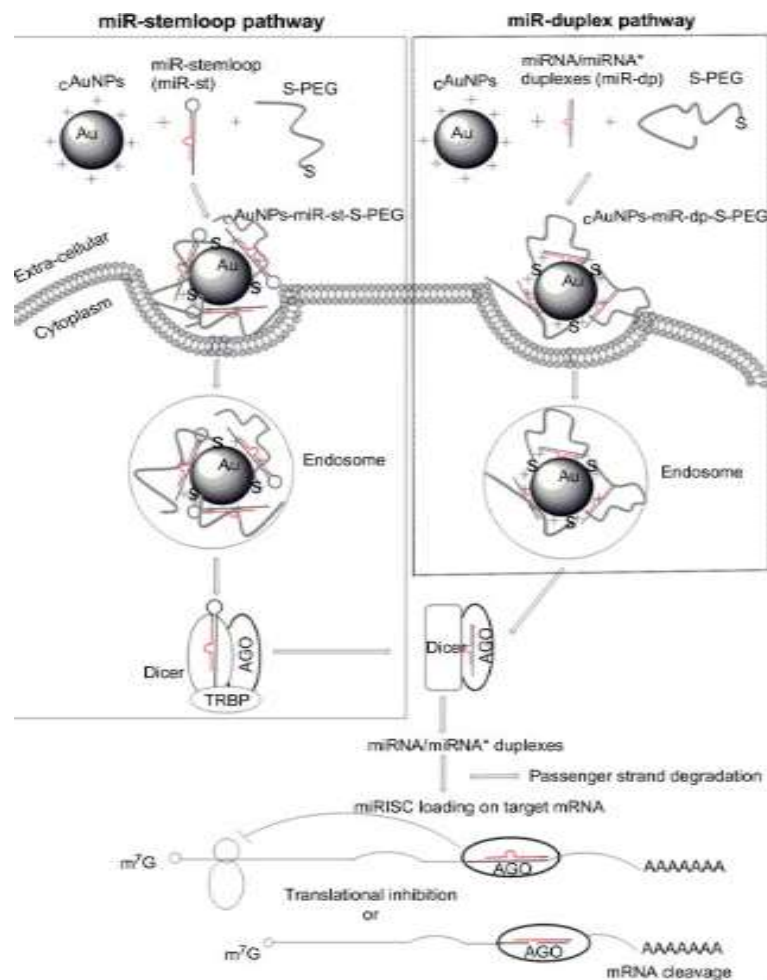
Lipika Goyal, Mandar D. Muzumdar and Andrew X. Zhu
Clin Cancer Res Published OnlineFirst February 6, 2013.

A gold nanoparticle platform for the delivery of functional microRNAs into cancer cells

Rajib Ghosh^{a,1}, Lalithya C. Singh^{a,1}, Jason M. Shoheit^b, Preethi H. Gunaratne^{a,*}

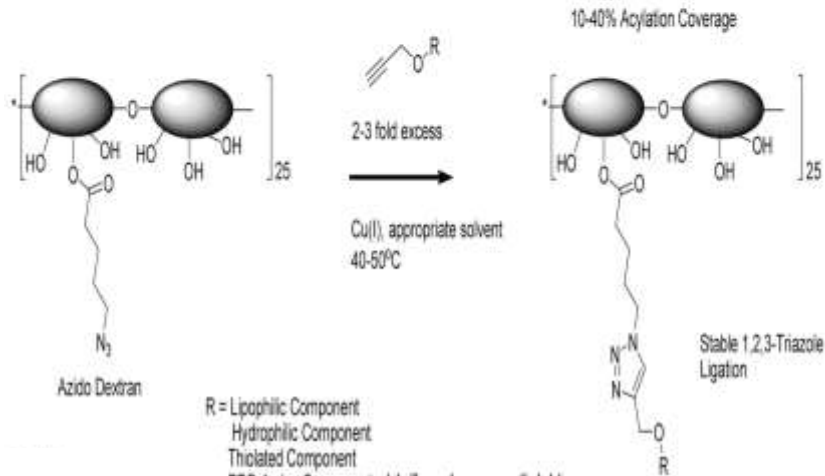
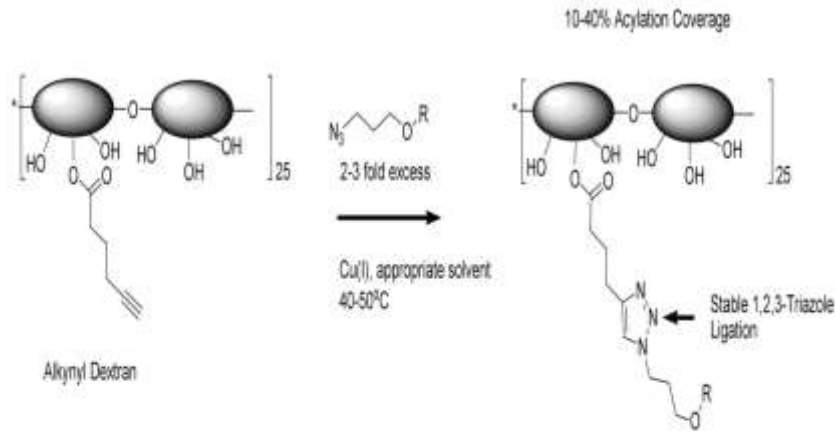
^a Department of Biology & Biochemistry, University of Houston, 4800 Calhoun Road, Houston, TX 77204, USA

^b Texas Children's Cancer Center, Baylor College of Medicine, Houston, TX 77030, USA

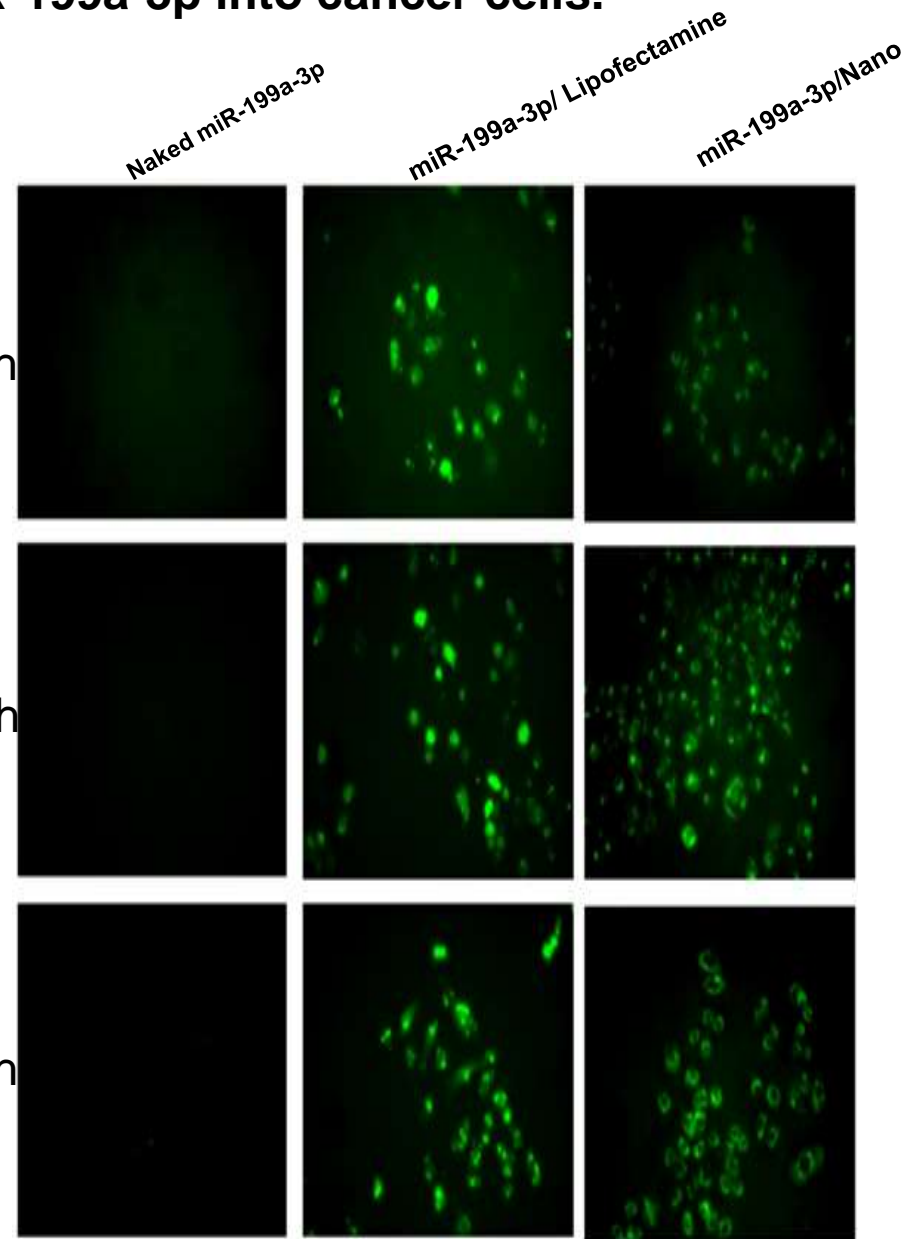


Dextran-nanoparticle delivers miR-199a-3p into cancer cells.

Preparation of Dextran Modules via "Click" Chemical Ligations



R = Lipophilic Component
 Hydrophilic Component
 Thiolated Component
 PEG-Amino Component with fluorophore or radiolabile
 PEG-Maleimido Component



Conclusion

- Chordoma has a distinct miRNA (miR) expression profile.
- miR-1 expression is significantly decreased in both chordoma tissues and cell lines. miR-1 is downregulated in 93.7% of the tumors and its decrease significantly correlated with Met overexpression.
- Downregulation of miR-1 is associated with poor prognosis in chordoma.
- Overexpression of miR-1 in chordoma cells inhibit cell growth and reduced replication potential, miR-1 have a tumor suppressor function in chordoma by directly regulating Met oncogene.
- miR-1 would be a viable small molecule candidate in clinical trials testing Met Inhibitors or directly deliver miR-1 to treat chordoma.

Acknowledgements

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- The Stephan L. Harris Fund
- Gattegno and Wechsler funds
- Sarcoma Foundation of America (SFA)
- Jennifer Hunter Yates Foundation
- Stanton Fund
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- National Cancer Institute (NCI), UO1 grant
- Academic Enrichment Fund of MGH Orthopaedics