Supplementary Materials: Dysregulation of miRNA Expression in Cancer Associated Fibroblasts (CAFs) and Its Consequences on the Tumor Microenvironment

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Supplemental Table S1 MiRs identified with specific roles in fibroblasts under pathological conditions

Cell type	miRNA	Direction of deregulation	Type of validation	Functional effect in NFs turning to CAFs	Interaction in CAFs	Ref.
OC	31 214 155	↓	in vivo: paired samples of CAF (metastasis) and NF; in vitro: NF co-cultured with HeyA8 OC cells	proliferation ↑	cytokines ↑ target: CCL5	[5]
EC	31	↓	in vitro: paired samples of EC tissue and tumor free tissue; in vivo: mouse xenograft model	migration, invasion of NFs ↑	target: SATB2 ↑	[11]
	148a	↓	in vitro: paired samples of EC tissue and tumor free tissue		target: WNT10B	[10]
BC	26b	↓	in vitro: paired samples of BC tissue and tumor free tissue culture models	↓	targets: TNKSIBPI, CPSF7, COL12A1	[12]
	31	↑	in vitro: BC tissue compared with tumor free tissue	activation ↑	α-SMA, IL-6 ↑, pathway: TGF-ß ↑	[14]
	221	↑				
	200b	↓				
	200c	↓				
	101	↓				
	141	↓				
	342	↓				
	205, let -7g, 26b	↓				
	127	↓	in vitro: BC cell lines and sets of primary breast tumors and adjacent normal tissues from the same patients	proliferation ↑	p53/p21 ↑ target: BCL6 oncogene ↑	[46]
	22	↑	in vitro: epithelial and BC cell lines, in vivo: by inducing cellular senescence in a mouse model of breast carcinoma	proliferation ↓	targets: CDK6, SIRT1, and Sp1 genes	[71]
	92	↓	in vitro: tissue samples and in breast epithelium and stroma during BC progression	expression of miR in FIBs ↓		[13]
	146b	↓	in vitro and in vivo: tissue samples with and without BC	activation ↑	IL-6 ↑ (p 16 related)	[41]
	200	↓	in vitro	activation ↑	α-SMA ↑ targets: Fli-1 is directly regulated by miR-200c, TCF12 is directly	[3]

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					targeted by miR-141 -> ECM remodeling ↑	
	365	↓	in vitro, in vivo	↓	IL6 ↑ pathway: p38 MAPK ↓ target: NF-kBp65 ↑	[63]
GC	**106b**	↑	in vitro: paired samples of GC tissue and tumor free tissue	proliferation ↑, motility ↑	α-SMA ↑, TGF-ß ↑ target: PTEN	[23]
	200b	↓	in vitro: gastric cancer cell lines compared with CAF; in vivo: mouse xenograft model; clinicopathological: comparing miR-200b expression in 173 patients	activation ↑	α-SMA ↑ targets: ZEB1, ZEB2 ↑ ; repressed E-cadherin expression	[15]
	149	↓	in vitro: GC cell lines, fibroblasts from tumor tissues and non-tumor tissues, in vivo: mouse xenograft model	activation ↑	IL6 ↑ -> EMT (epithelial-to-mesenchymal transition) ↑ target: PGE2 induces the epigenetic silencing of miR-149 in fibroblasts	[79]
	145	↑	in vitro: tissue samples and cell lines	activation ↑	α-SMA ↑ pathway: TGF-ß ↑	[25]
	143	↑	in vitro: tissue samples and cell lines, in situ hybridization	activation ↑	Collagen type III ↑ pathway: TGF-β/SMAD signaling ↑	[24]
EOC	**27a/b**	↑	in vitro: serum expression of miR, from tissue with and without cancer	activation ↑	α-SMA ↑ pathway: TGF-ß ↑	[26]
	21	↑	in vitro: samples from patients with esophageal squamous-cell carcinoma, and a co-culture system of normal fibroblasts and esophageal cancer cells	activation ↑		[40]
PaC	21	↑	in vitro: tumor and tumor free tissue	activation ↑	α-SMA ↑	[17]
	21	↑	in vitro: tumor and tumor free tissue	activation ↑		[17]
	222	↑	in vitro co-cultured experiments with: human normal and cancer associated stellate cells, human cancer associated fibroblast cells, human PaC cell lines	activation ↑	α-SMA ↑	[4]
	221				α-SMA ↑, targets: NF-κB, K-Ras ↑	
	155	↑	in vitro: isolated primary pancreatic fibroblasts from mice co-cultured with pancreatic cancer cell lines	activation ↑ (by microvesicles)	alpha SMA ↑ target: TP53INP1 ↓ by direct targeting of miR-155 and microvesicles	[65]
	200a, 200b	↑	in vitro: cell lines and tissue samples with and without cancer	activation ↑	target: SIP1 by promoter methylation and retain expression of E-cadherin, regulating EMT	[16]
CRC	21	↑	in vitro: human cell lines and tissue samples with and without cancer	activation ↑	cytokines ↑, PDCD4 ↓(inversely correlated with miR expression)	[27]
PC	205	↓	in vitro: cell culture	activation ↑	inflammatory cytokine secretion ↓	[45]

Cell type	miRNA	Direction of deregulation	Type of validation	Functional effect in NFs turning to CAFs	Interaction in CAFs	Ref.
					target: HIF-1 ↑, EMT ↑	
	409	↑	in vitro: prostate and bone and in patient tissue samples and cells , in vivo: mouse xenograft	expression ↑	α-SMA, EMT ↑, extracellular vesicle(EV) release ↑	[19]
	15 **16**	↓	in vitro: 23 non-neoplastic and tumor tissues samples, in situ hybridization, in vivo: mouse model	↓	α-SMA ↑ targets: Fgf-2 and its receptor Fgfr1	[20]
	133b	↑	in vitro: human PC cells and human prostate tissue with or without cancer	activation ↑	α-SMA, IL-6 ↑ pathway: TGF-ß ↑	[22]
LC	**101**	↑	in vitro: human lung cell lines, and human lung tissue with and without cancer	proliferation ↑	pathway: PI3K-AKT ↓ targets: CXCL 12 ↓	[21]
	200b	↑	in vitro: cell lines	activation ↑	α-SMA ↑ target: Flt 1 ↓	[42]
PSC	**143**	↑	Rat PSCs were isolated from the pancreas tissue of male Wistar rats. PSCs were activated in vitro by culture in serum-containing medium	activation ↑	interaction with p39 mitogen-activated protein kinase, extracellular-signal–regulated kinase, and SMAD 2/4 pathway	[74]
	221	↑			interaction with p40 mitogen-activated protein kinase, extracellular-signal–regulated kinase, and SMAD 2/5 pathway	
	31	↑			interaction with p38 mitogen-activated protein kinase, extracellular-signal–regulated kinase, SMAD 2/3 pathway	
	126	↓			interaction with p41 mitogen-activated protein kinase, extracellular-signal–regulated kinase, SMAD 2/6 pathway	
	146a	↓			interaction with p42 mitogen-activated protein kinase, extracellular-signal–regulated kinase, SMAD 2/7 pathway	
	150	↓			interaction with p43 mitogen-activated protein kinase, extracellular-signal–regulated kinase, SMAD 2/8 pathway	
	210	↑	in vitro: Panceatic cancer cells mono-cultured or indirectly co-cultured with PSCs	activation ↑	EMT ↑ pathway: ERK and Akt ↑	[61]

Cell type	miRNA	Direction of deregulation	Type of validation	Functional effect in NFs turning to CAFs	Interaction in CAFs	Ref.
	15b **16**	↓	in vitro: rat PSC	activation ↑	target: BCL-2 ↑	[66]
	29	↓	in vitro: Immortalized mouse pancreatic stellate (mPSC) cell lines and tissue samples, in vivo: mouse model	activation ↑	ECM ↑ pathway: TGF-β1 (SMAD3 dependent) ↑	[57]
HSC	335	↓	in vitro: cultured HSC from rats	activation ↑	α-SMA and Collagen Type 1 ↑ target: TNC (tenascin-C) expression ↓	[51]
	150	↑	in vitro: HSC isolated from sham-operated and bile duct-ligated rats compared with HSC isolated from fibrotic rats	activation ↓	α-SMA and Collagen Type 1 ↓ target: C-myb ↓	[56]
	194	↑			target: Rac 1 ↓	
	146a	↓	in vitro	activation ↑	α-SMA and Col-1 ↑ targets: Wnt1 and Wnt5a ↑	[59]
	146a	↓	in vitro: cell culture, in vivo: rat model	activation ↑	α-SMA ↑, TGF-ß ↑ by direct targeting of SMAD4	[68]
	200a	↓	in vitro: cell culture, in vivo: rat model	activation ↑	α-SMA ↑ pathway: Wnt/β-catenin and TGFβ ↑	[47]
	200a	↑	no information	activation ↓	α-SMA ↓, EMT process ↓, Gli2 (downstream signaling protein of the Hh pathway) ↓	[81]
	9a	↑	in vitro: cell culture, in vivo: rsat model	activation ↑	target: SIRT1	[72]
	17	↑	no information	activation ↑	Collagen Type I and α-SMA ↑, TGF-ß 1 ↑, SMAD7 ↓ (direct target)	[38]
	33a	↑	in vitro: cell culture and human liver and serum samples	activation ↑	α1 Collagen (Col1A1) and α-SMA ↑, TGF-ß1 ↑, PI3K/Akt pathway target: PPAR-α	[30]
	200b	↑	in vitro: human hepatic stellate cell line	proliferation, migration ↑	PI3K/Akt through FOG2 ↓-regulation	[31]
	21	↑	in vitro: human immortalized HSC line	activation ↑	Collagen Type I and α-SMA ↑, PTEN/Akt pathway	[54]
	130a/b	↑	in vitro: immortalized rat HSC line, in vivo: rat model	activation ↑	ECM ↑ target: PPARy ↓	[70]
	30, 193	↓	in vitro: hepatic fibrosis and human tissue samples, in vivo: mouse model	activation ↑	TGF-ß , TGF-β2 and SNAIL1 as potential tagets	[34]
	144	↓	in vitro: human tissue samples	activation ↑	α-SMA ↑, TGF-ß1 ↑	[36]
	146a	↓	in vivo: cell culture	activation ↑	TGF-ß1 ↑ target: SMDA4	[68]

Cell type	miRNA	Direction of deregulation	Type of validation	Functional effect in NFs turning to CAFs	Interaction in CAFs	Ref.
	126	↑	no information	activation ↑	TGF-ß1, IκBα protein expression ↓ pathway: NF-κB	[44]
	221 222	↑	in vitro: cell culture and liver biopsy specimens, in vivo: mouse model	activation ↑	α1-Collagen and α-SMA ↑	[32]
	19b (-1/-2)	↓	in vitro: rat HSCs, in vivo: rat model	activation ↑	Type I Collagen ↑, expression of α1(I) and α2(I) procollagen in mRNAs ↑ pathway: TGF-ß ↑ targets: TGF-ß 2 receptor and SMAD 3	[33]
	101	↑	in vitro: Primary rat and mouse HSCs, in vivo: mouse model	activation ↓	targets: TβRI and KLF6, inhibiting TGFβ signalling pathway	[49]
	31	↑	in vitro: human tissue samples, in vivo: rat model	activation ↑	TGF-ß ↑	[52]
	19b (19b-1, 19b-2)	↑	in vitro: tissue samples, in vivo: rat model	activation ↓	target: GRB2	[64]
	34a/	↑	in vitro: human HSC line, in vivo: rat model	activation ↑	α-SMA ↑ target: PPARγ ↓	[69]
	21	↑	in vitro: human tissue samples, rat HSC cell line	activation ↑	induces hepatocyte EMT pathway: ERK1 targets: SPRY2, HNF4α	[28]
	101	↑	in vitro: Primary rat and mouse HSCs, in vivo: mouse model	activation ↓	TGFß signaling ↓ by suppressing TβRI/KLF6 (targets)	[49]
	181 b	↑	in vitro: human cell line, in vivo: rat model	activation ↑	α-SMA and Type I Collagen ↑ pathway: PI3K/Akt ↑ target: PTEN expression ↓	[54]
	200a	↑	in vitro: HSC-T6 cell lines, in vivo: rat model	activation ↓	pathway/ targets: Keap1/Nrf2; Nrf2 ↑ inhibits TGF-β1 induced growth	[73]
	483	↑	in vitro: cell lines, in vivo: mouse model	activation ↓	TGF-ß ↓ targets: TIMP-2 and PDGF-β ↓	[62]
	21	↑	in vivo: mouse model	activation ↑	TGF-ß, α-SMA ↑ targets: PDCD4/AP-1 ↑	[29]
	126	↓	in vitro: primary rat HSC, in vivo: rat model	activation ↑	PI3K, p-AKT ↑	82
	29a	↓	in vitro: mice HSC, in vivo: mouse model	activation ↑	alpha SMA ↑ target: HDAC4 ↑	[58]
	150	↑	in vitro: human cell lines	activation ↓	alpha SMA ↓ targets: Sp1 and Col4A4	[83]
	133a	↓	in vitro: cell lines, in vivo: mouse model	activation ↑	TGF-ß, collagen ↑	[35]
HSC	**122**	↓	in vitro: rat HSC, in vivo: rat model	activation ↑	collagen maturation and ECM production ↑	[50]

Cell type	miRNA	Direction of deregulation	Type of validation	Functional effect in NFs turning to CAFs	Interaction in CAFs	Ref.
					target: P4HA1 ↓, binding activity of C/EBPα to miR-122 promoter	
HSC	181b	↑	in vitro: serum samples and human cell lines	activation ↑	TGF-ß ↑ target: p27	[39]
HSC	26a	↓	in vitro: primary rat HSC, in vivo: rat	activation ↑	Collagen Type I , Cox-2 protein expression, NF-κB ↑	[60]
	29a	↑				
	214	↓				
	146a	↓				
HSC	199a, 199a*	↑	in vitro: human clinical samples, in vivo: mouse model	activation ↑	TGF-ß ↑	[37]
	200a, 200b	↑				
HSC	16	↓	in vitro: rat HSC	activation ↑	CD1, Bcl-2 ↑	[67]
HSC	**27a/b**	↑	in vitro: rat HSC	activation ↑	target: retinoid X receptor alpha	[84]

(CAFs, HSCs, PSCs) OC (Ovarian Cancer), EC (Endometial Cancer), BC (Breast Cancer), GC (Gastric Cancer), EOC (Esophageal Squamous cell carcionoma), PaC (Pancreatic Carcinoma), CRC (Colorectal Cancer), PC (Prostate Cancer), LC (Lung Cancer).

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