

Supplementary material

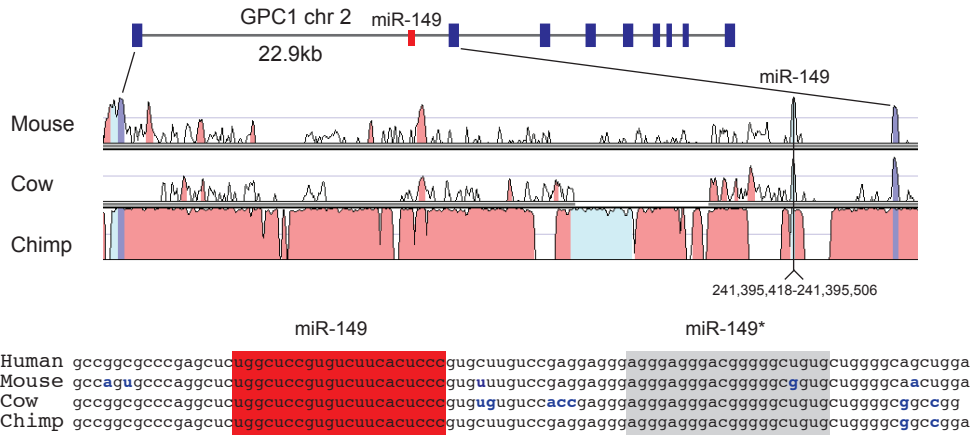
Fig. S1 (A) Schematic representation of the GPC1 gene locus, demonstrating the miR-149 coding sequence within intron 1 and its conservation among species (reference: human genome). (B) Predicted binding sites for miR-149 (in red) or miR-149* (in gray) in human FGFR1 and GPC1 3'UTRs. Point mutations (PM) of the selected predicted binding sites for miR-149 or miR-149* seed sequences in human FGFR1 or GPC1 3'UTRs are indicated in bold. (C) Chromatin State Segmentation track of the potential promoter regions of miR-149 in HUVECs cells (adaptation from ENCODE).

Fig. S2 PANTHER analysis of (A) miR-149 and (B) miR-149* predicted target genes in human based on biological pathways. Data are expressed as number of targets and their respective P values. $P \leq 0.05$. HUVECs transfected for 48 h with (C) CM, miR-149 or miR-149*, and (D) CI, I-miR-149 or I-miR-149*. qRT-PCR analysis of FGFR1 and GPC1. Data are expressed as relative expression to cell transfected with CM or CI and correspond to mean \pm SEM of 3 experiments, $P \leq 0.05$. (E) Cell cycle analysis of HUVECs transfected as indicated in C, then cells were harvested and treated with BrdU for 1 h followed by BrdU detection and propidium iodide staining and analyzed by flow cytometry. Inserts correspond to dot plots that represent DNA content vs incorporation of BrdUrd (active phase S) into DNA. One representative experiment out of 3 with similar results is shown. (F-H) HUVECs were starved for 12 h and then treated with FGF2 (25 ng/mL) or non-treated for the indicated times. (F) qRT-PCR analysis of PAI-2 and Axl expression. Data are expressed as relative expression to non-treated cells and correspond to mean \pm SEM of 3 experiments, $P \leq 0.05$. (G) Flow cytometry analysis of the GPC1 protein expression. Data are expressed as arbitrary fluorescence units to non-treated cells and correspond to one representative experiment out of 3 with similar results. (H) qRT-PCR analysis of FGFR1 expression. Data are expressed as relative expression to non-treated cells and correspond to mean \pm SEM of 3 experiments, $P \leq 0.05$.

Table 1. Predicted targets for miR-149 and miR-149* involved in angiogenesis. Positive regulators of angiogenesis are indicated in red, negative in blue, positive/negative in black, unknown in grey and expressed in endothelial cells in italic.

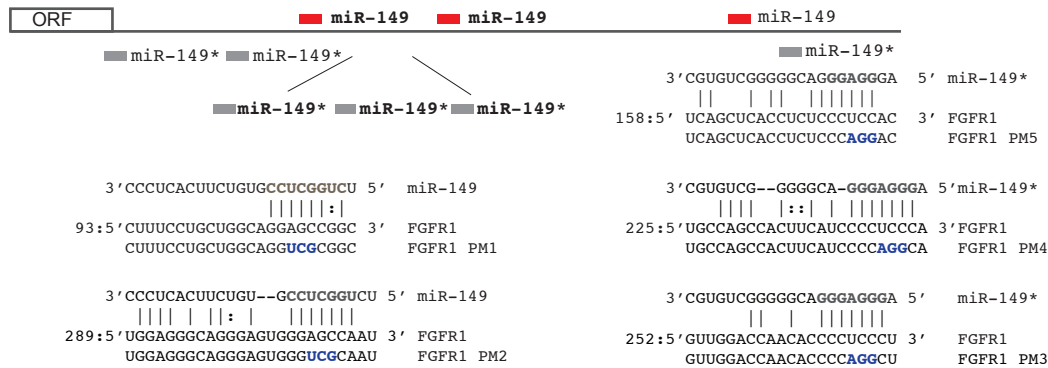
Supplementary material Fig. 1

A

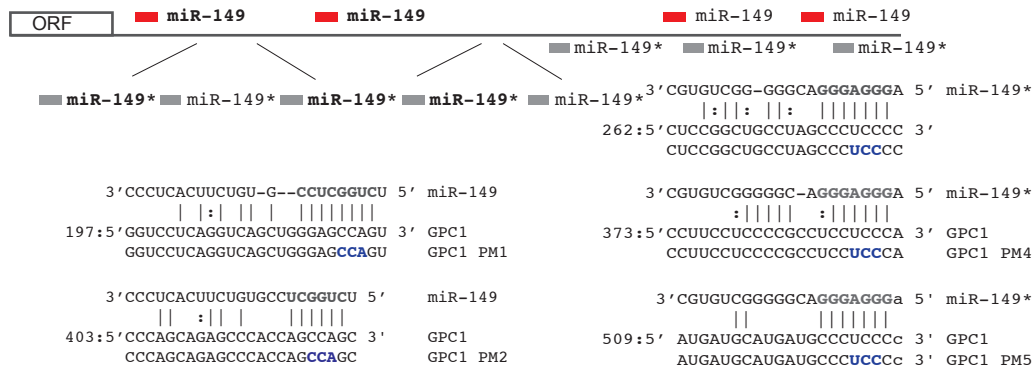


B

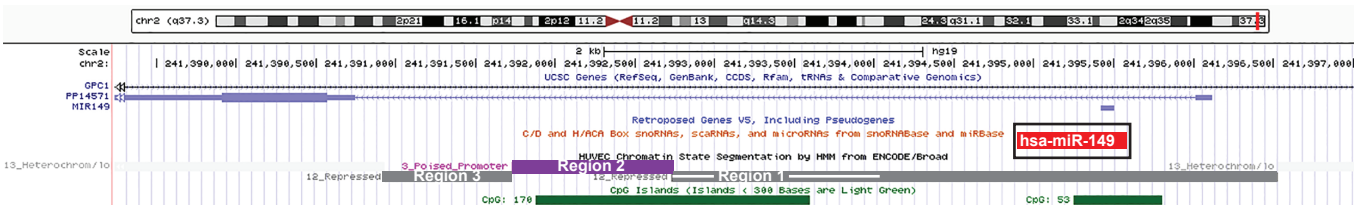
3' UTR FGFR1 (2,490 Kb)



3' UTR GPC1 (1,787 Kb)

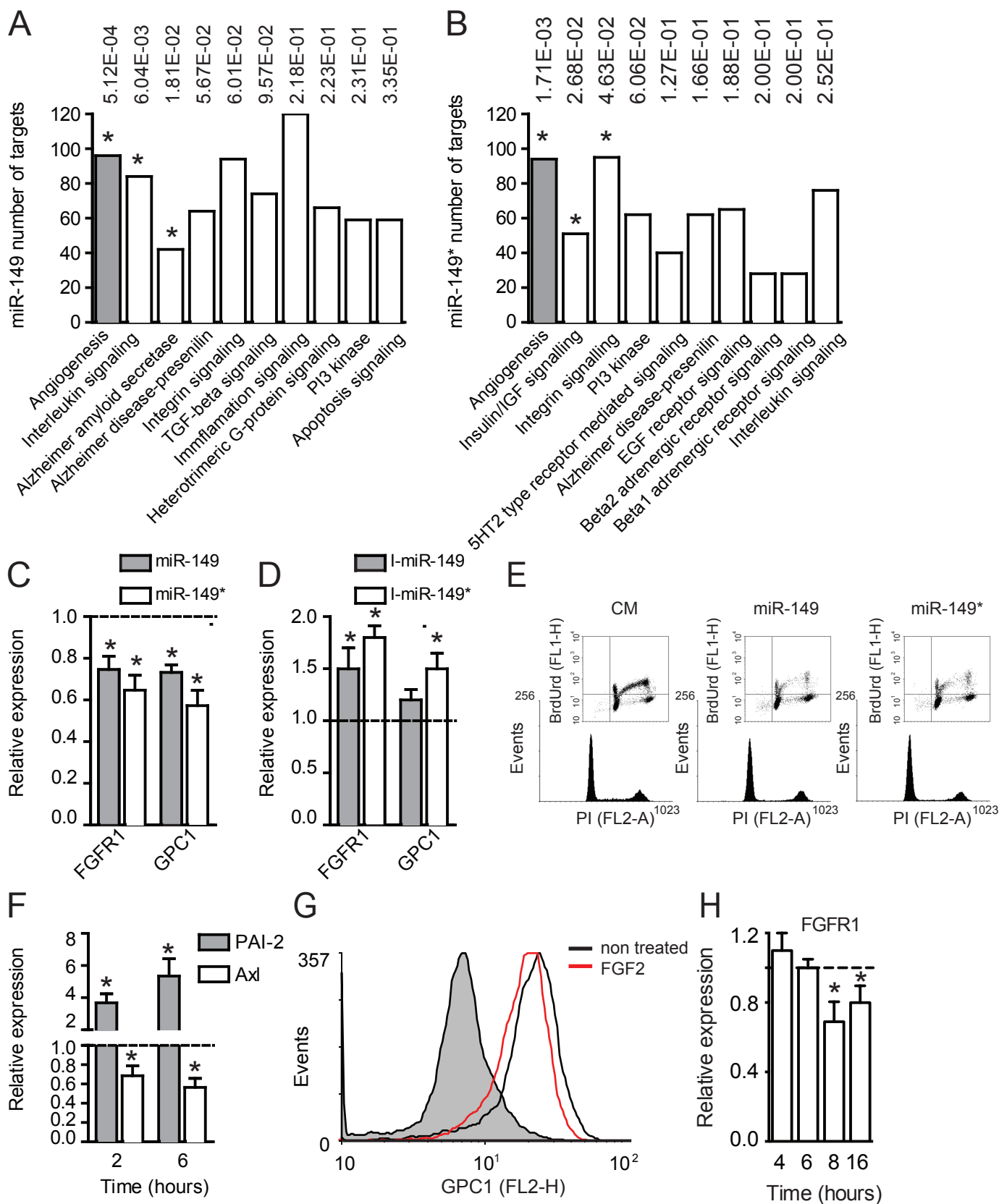


C



Region 1: repressed promoter
Region 2: poised promoter
Region 3: repressed promoter

Supplementary material Fig.2



Supplementary material Table 1

miR-149 targets (36)

FGF1
DOK1
HSH2D
PKD2
FZD3
STAT1
PLCG1
JAG1
PDGFC
MAP3K1
TEK
VEGFR2
CTNNB1
PAK3
AKT3
HN1
PLA2G4C
ANGPT1
PTPN6
ETS1
PLD1
MAPK1
MAPK6
GRB14
PRKCZ
LPXN
NR1I2
FOS
MAPK14
PDGFD
FGF2
DLL1
PIK3R1
BAD
SFRP1
CRK

miR-149-miR-149*targets (61)

PRKCA
EFNB1
 PI4KB
GRB2
PTK2
PKD1
 AXIN2
RAF1
 CRYAA
 PRKD3
WNT5B
PTPRB
FZD5
NOTCH1
 F7
 CRKL
 GRAP
 NOS1
STAT3
 ARHGAP1
 PLA2G4F
RHOA
RBPJL
EPHB3
PXN
EPHB2
 PIK3R3
NOTCH2
GRB7
 SH2D4A
PRKCE
JUN
 PDGFRA
TGFB11

FGFR1
 APC2
MAPKAPK2
 DVL1
WNT7B
 PRKACA
FRS2
EFNB2
 PDGFRB
WNT5A
 MAPKAPK3
 MAP2K4
RHOB
WNT10A
 MAPK4
MAPK3
SRC
GSK3B
AKT1
PAK2
ANGPT2
PIK3C2A
 PIK3CD
 FZD1
 PTPN11
NOTCH3
ITGB3

miR-149* targets (34)

WNT2
PAK1
 SH2D4B
RHOC
AKT2
PIK3CG
PLD2
BIRC5
PRKD2
NOS3
PIK3C2B
 NCK2
 KRAS
 EPHA3
 FRS3
DLL4
PIK3R2
 GSK3A
WNT7A
PRKCG
VEGFA
 AXIN1
NOTCH4
PDGFB
WNT1
WNT10B
PRKCI
 FZD8
FLT4
HRAS
 SHC1
 F3
DVL2
DVL3