

Supplemental Figure Legends Biddlestone et al.

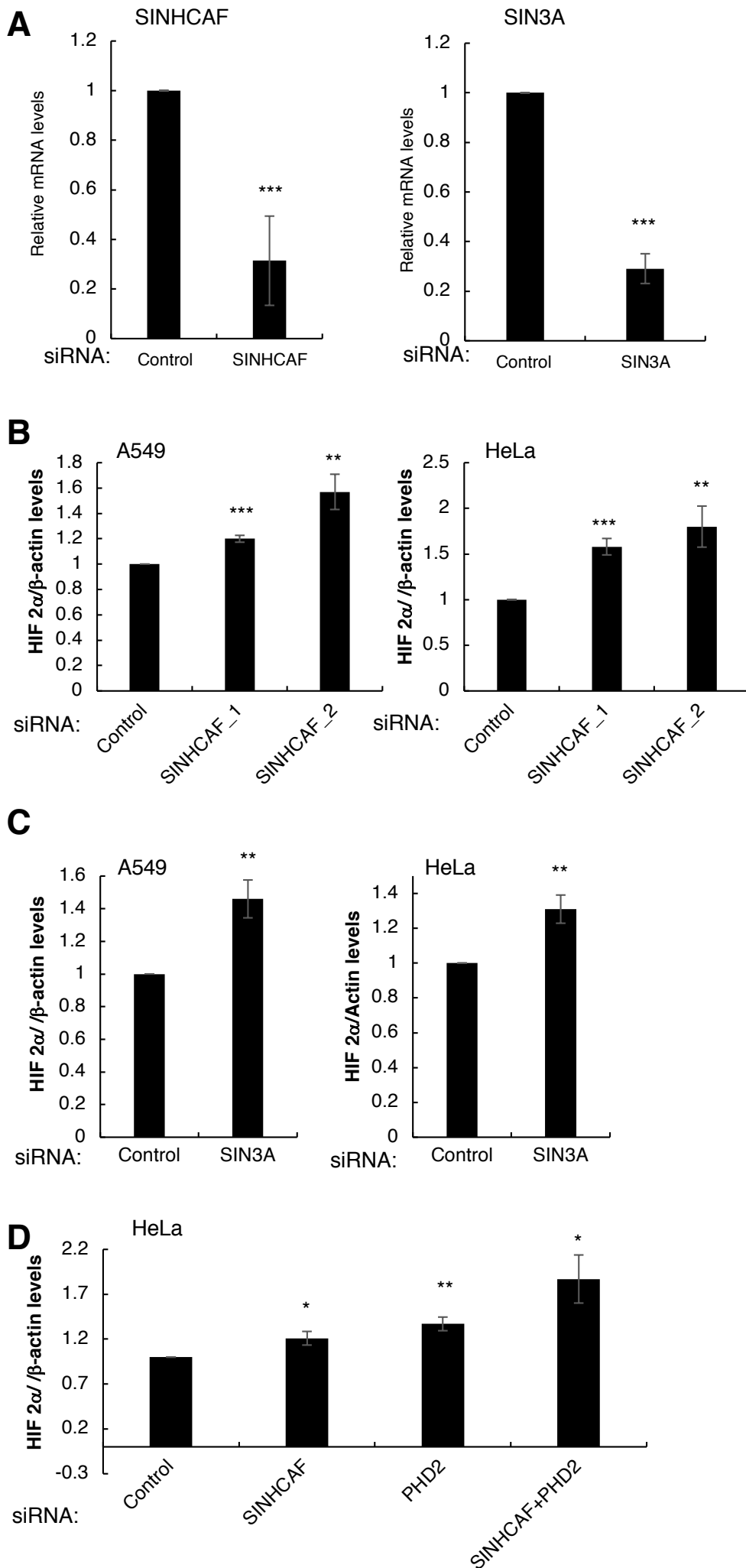
SINHCAF/FAM60A and SIN3A specifically repress HIF 2 α expression

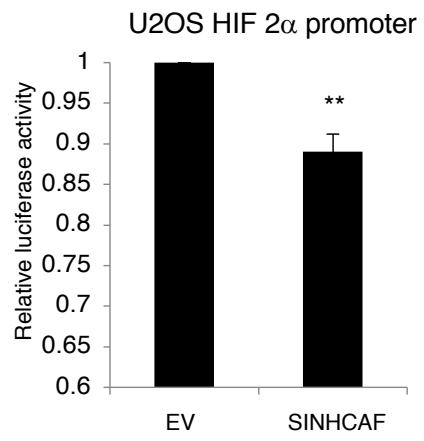
Figure S1: SINHCAF as a HIF 2 α repressor. (A) qPCR validation using HeLa cells, for SINHCAF and SIN3A. Graphs depict mean + S.E.M. * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$. (B) Quantification of HIF 2 α protein levels following SINHCAF depletion compared to control cells exposed to hypoxia. Graph depicts mean and S.E.M. of HIF 2 α protein levels normalised to actin of a minimum of 3 independent experiments. Graphs depict mean + S.E.M. * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$. (C) Quantification of HIF 2 α protein levels following SIN3A depletion compared to control cells exposed to hypoxia. Graph depicts mean and S.E.M. of HIF 2 α protein levels normalised to actin of a minimum of 3 independent experiments. Graphs depict mean + S.E.M. * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$. (D) Quantification of HIF 2 α protein levels following SINHCAF, PHD2 or SINHCAF and PHD2 depletion compared to control cells exposed to hypoxia. Graph depicts mean and S.E.M. of HIF 2 α protein levels normalised to actin of a minimum of 3 independent experiments. Graphs depict mean + S.E.M. * $P \leq 0.05$, ** $P \leq 0.01$.

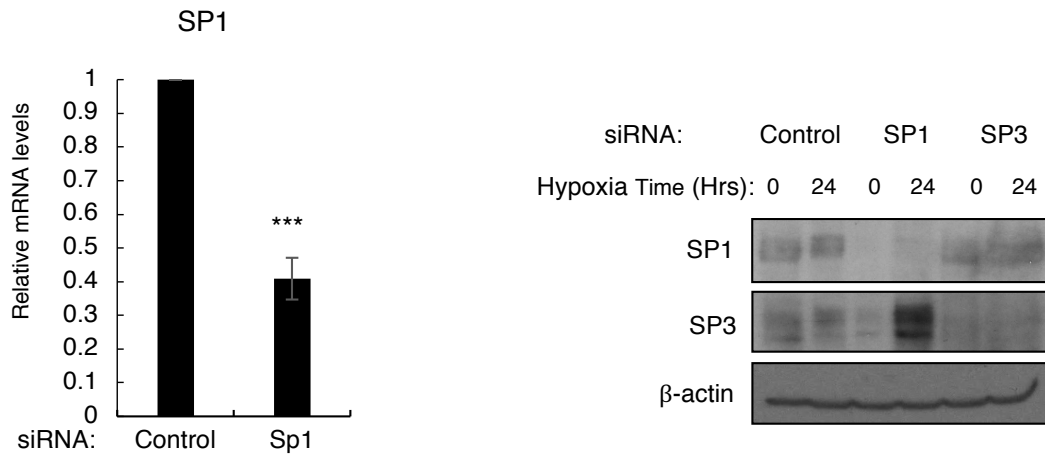
Figure S2: SINHCAF represses the HIF 2 α promoter. (A) U2OS-HIF 2 α promoter cells were transfected with 1 μ g of control or SINHCAF expressing plasmids prior to lysis and luciferase assay being performed. Graphs depict mean + S.E.M. * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$.

Figure S3: Validation of SP1 and SP3 knockdown levels. (A) qPCR and western blot validation for siRNA using HeLa cells. Graphs depict mean + S.E.M. * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$. HeLa cells were transfected with the indicated siRNA oligonucleotides and exposed where indicated to 1% O₂ for 24 hours prior to cell lysis. Whole cell lysates were analysed by immunoblot for the levels of the depicted proteins.

Figure S4: Colony formation assay following SINHCAF and HIF 2 α . (A) Colony Formation: HeLa and DLD-1 cells were seeded for colony formation following siRNA transfection. Representative images are displayed.



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