Normalization of Snai1-mediated vessel dysfunction increases drug response in cancer

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- Supplemental Data -

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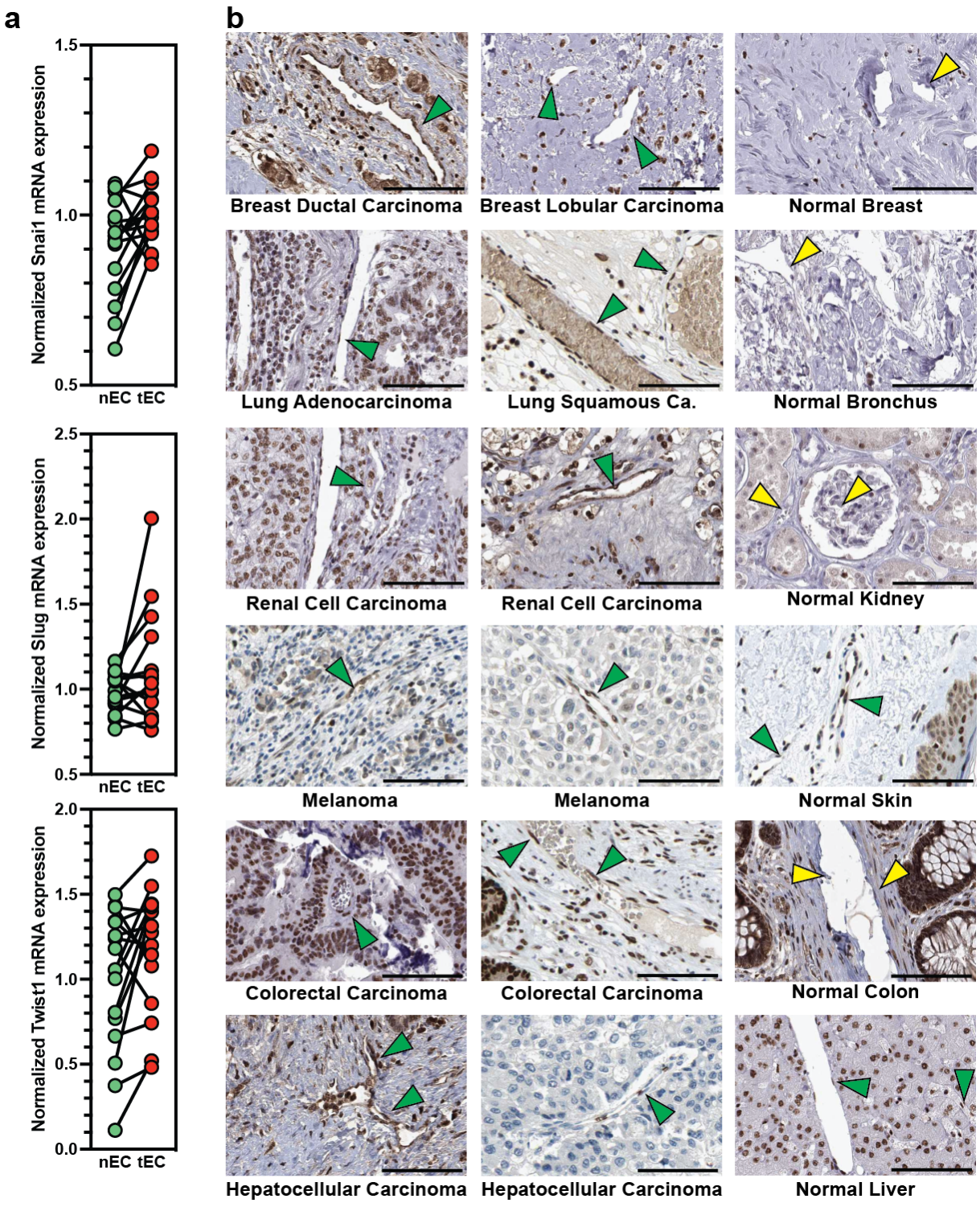
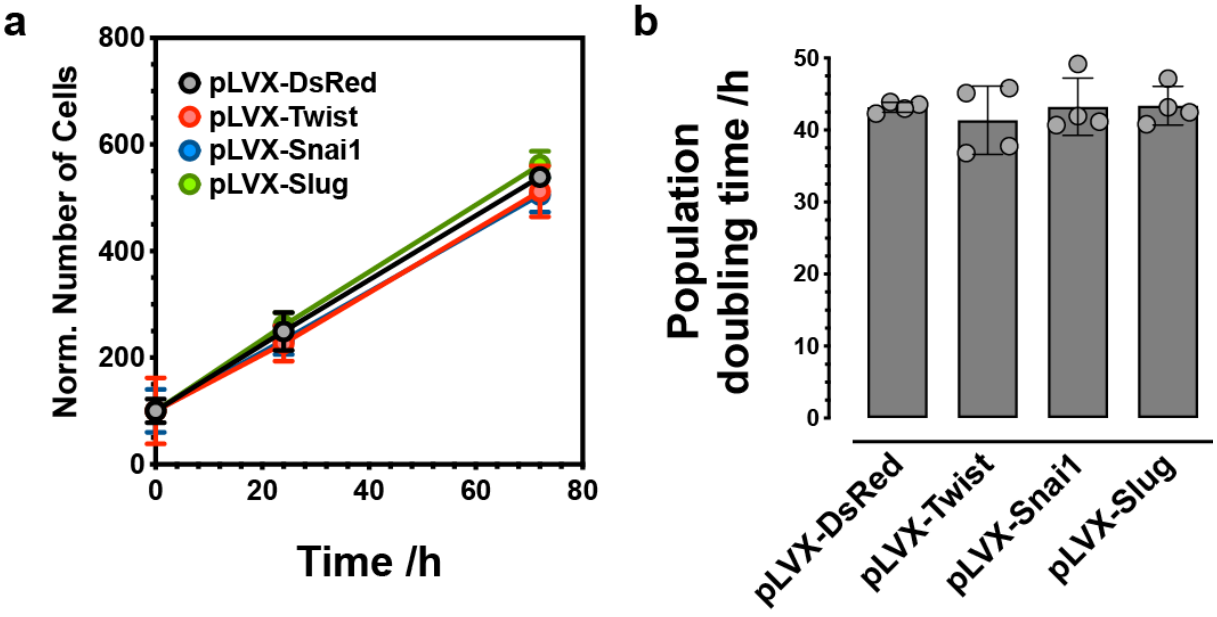
Supplemental Figure 1:



(a) Alignment of paired samples from the GSE 51401 data set for Snai1, Slug, and Twist1 expression (https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE51401).

(b) IHC staining for Snai1 in human tumor samples and in corresponding normal tissue (https://www.proteinatlas.org/ENSG00000124216-SNAI1/pathology). Green arrowheads show Snai1+ ECs and yellow arrowheads show Snai1- ECs (in normal tissue).

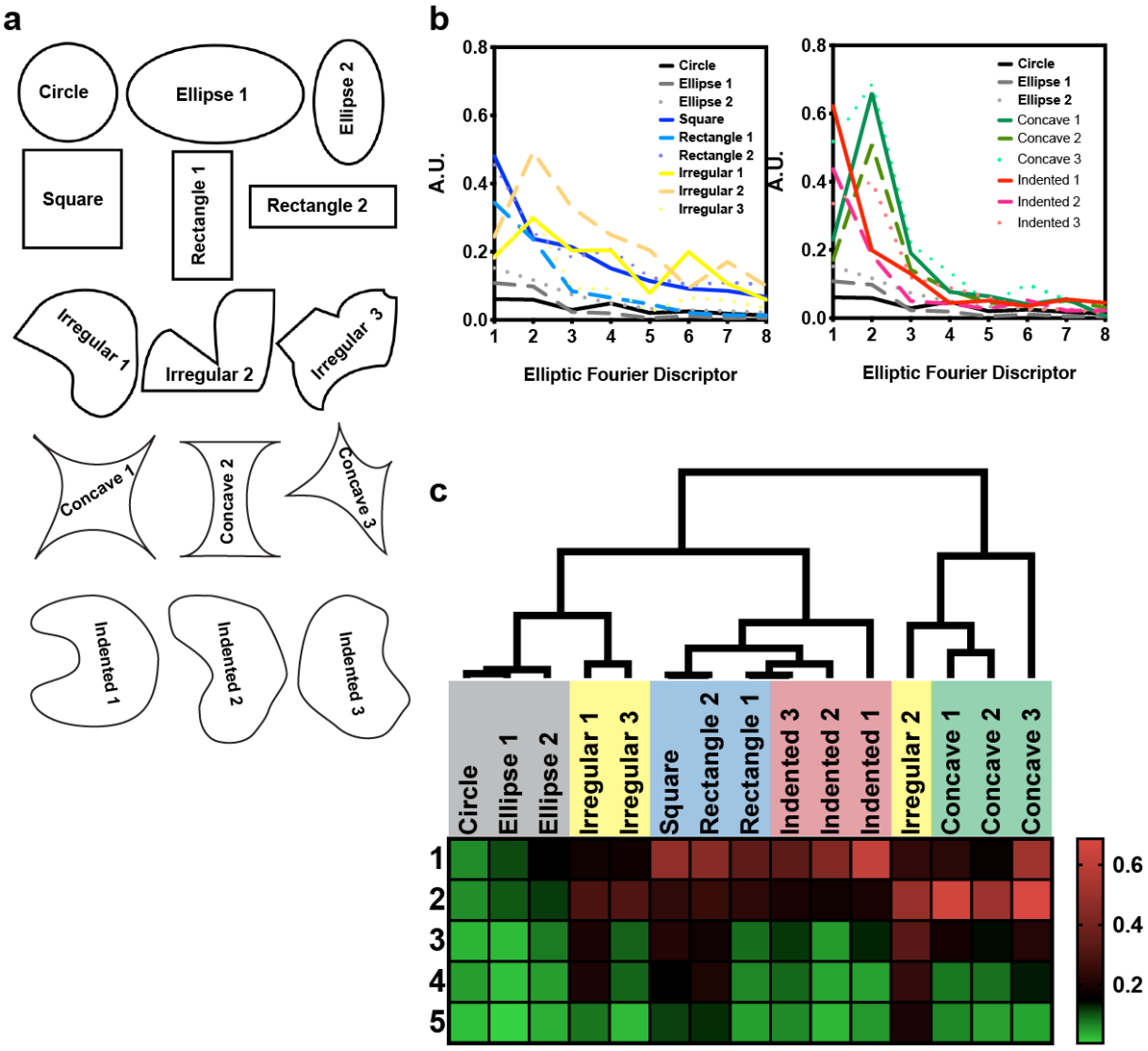
Supplemental Figure 2



(a) Cell proliferation of HUVEC stably overexpressing Twist1, Slug, or Snai1 over 72h in comparison to HUVEC transfected with the control lentiviral vector (pLVX-DsRed).

(b) Cell doubling time of HUVEC stably overexpressing Twist1, Slug, or Snai1 in comparison to HUVEC transfected with the control lentiviral vector (pLVX-DsRed).

Supplemental Figure 3

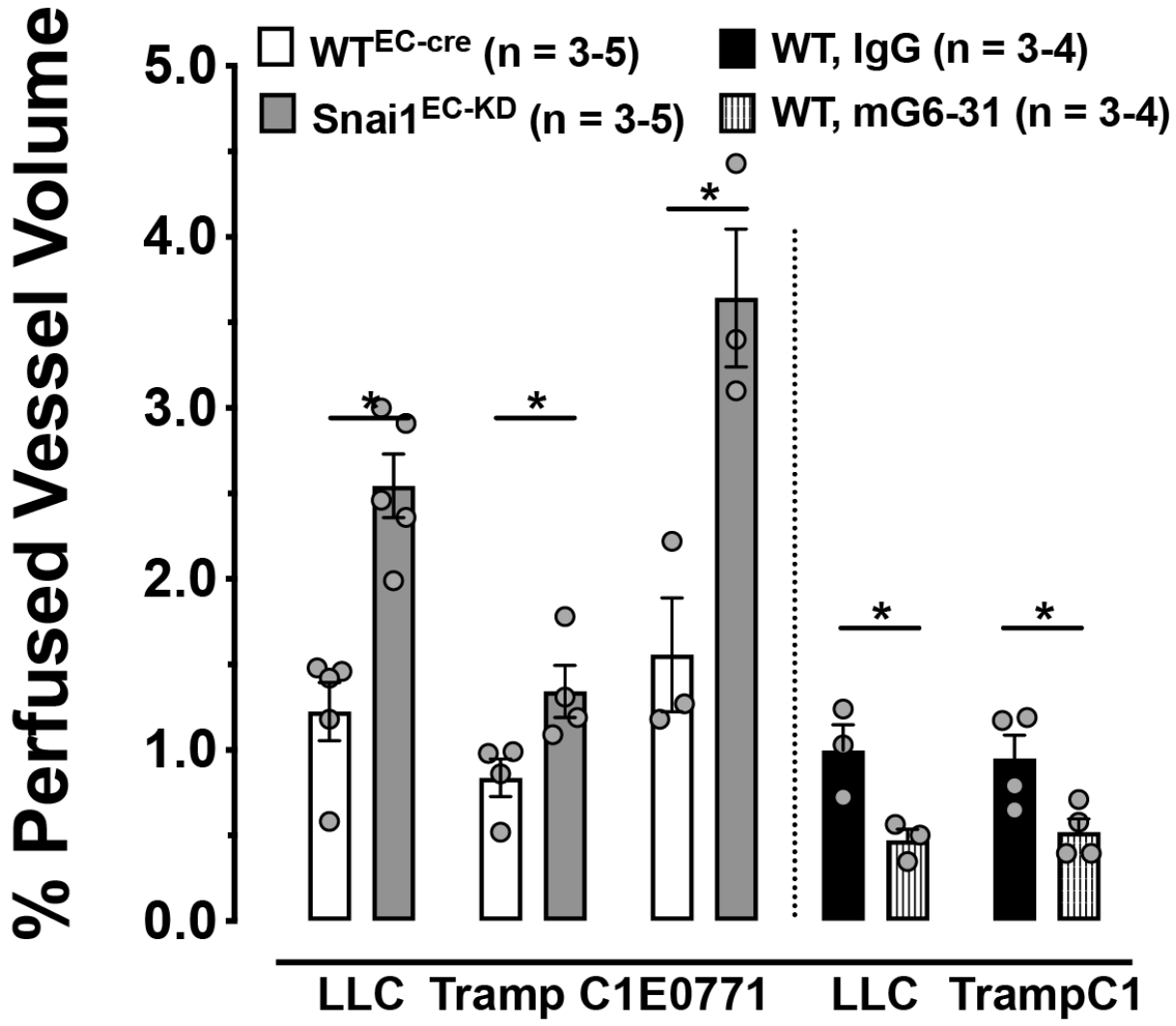


(a) Test shapes to establish Elliptic Fourier Analysis (EFA) as a method to rank vascular irregularities in archival tissue sections.

(b) Results from EFA for the test shapes shown in (a). Displayed are the first eight Fourier descriptors (FD). For clarity the data is divided in two graphs.

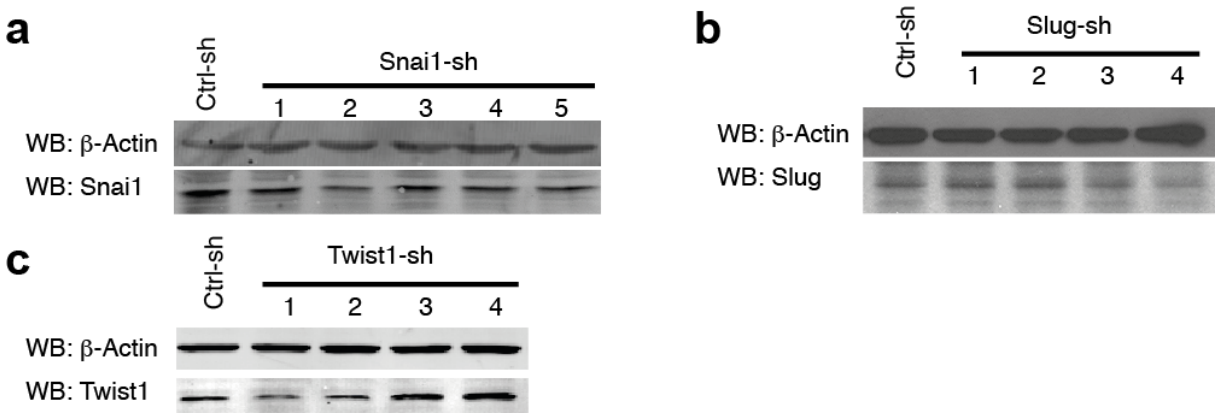
(c) Cluster analysis of the five first FD for the test shapes shown in (a).

Supplemental Figure 4



Evaluation of the relative volume of perfused vessels in various murine tumors. Endothelial-specific knock-down of Snai1 increased vessel volume while VEGF-A sequestration reduced it. (n = 4-5)

Supplemental Figure 5



(a) Western blot analysis of Snai1-expression in HUVEC stably transfected with 5 different shRNA transferring lentiviral particles. HUVEC transfected with Snai1-shRNA-2 and Snai1-shRNA-3 were used for functional assays as they produced the strongest reduction of Snai1 protein levels.

(b) Western blot analysis of Slug-expression in HUVEC stably transfected with 4 different shRNA transferring lentiviral particles. HUVEC transfected with Slug-shRNA-3 and Slug-shRNA-4 were used for functional assays as they produced the strongest reduction of Slug protein levels.

(c) Western blot analysis of Twist1-expression in HUVEC stably transfected with 4 different shRNA transferring lentiviral particles. HUVEC transfected with Twist1-shRNA-1 and Twist1-shRNA-2 were used for functional assays as they produced the strongest reduction of Twist1 protein levels.