0.0E+00

NCNC

Breast

NC

Skin

NCNC

Lung

Lymphnode

Liver

NC

Esophagus

SUPPLEMENTARY FIGURE AND TABLE LEGENDS





Supplementary Figure S1: Analysis of Swiprosin-1 expression in tissue microarray of human cancer. A. Tissue microarray containing 30 normal and 29 cancer tissue sections from human patients was stained with Swiprosin-1 antibody or subjected to H&E. Representative results from two independent experiments are displayed. Images in Figure 1B are marked in black boxes with bold lines, and tumor regions are indicated with yellow dotted lines. B. Expression patterns of Swiprosin-1 were compared in 15 matched TMA tissues. Intensity was quantified using the Positive pixel counts v.9.1 algorithm of AperioImageScope, and the algorithm computed the sum of intensity values for all weak positive pixels (IWP), modest positive pixels (IP), and strong positive pixels (ISP) in scanned images. Total intensity was calculated as the sum of IWP, IP and ISP. Stained images of tissues (red) are presented in Figure 1B.

N C N C N C N C

Colon Kidney

Stomach

NC

Prostate Testis

Urinary bladder

NCNC

Uterine cervix

NCNC

Endometrium

Thyroid

 Normal Goat IgG
 Goat anti-Swi1 Ab

Human melanoma pT3

Supplementary Figure S2: Validation of anti-Swi-1 antibody specificity in human melanoma. To validate the specificity of anti-Swi-1 antibody in melanoma, tissues were incubated with normal goat IgG according to the standard immunohistochemistry protocol. $Bar = 300 \ \mu m$.



Supplementary Figure S3: Knockdown of EGFR inhibits Swiprosin-1 expression. B16F10 cells were transfected with EGFR siRNA in a dose-dependent manner, and knockdown of EGFR and Swiprosin-1 expression assessed.



Supplementary Figure S4: Swiprosin-1 does not modulate proliferation and MMP expression in B16F10 melanoma cells. A. Swiprosin-1 overexpression or knockdown cells (0.5×10^4) cultured on 24-well plates for the indicated time-periods were subjected to the MTT assay. B. After 24 h of culture, total RNA was isolated and subjected to RT-PCR with specific primers for MMPs.

| Organ | Diagnosis | ^{a)} Tissue type |
|----------------------------|-----------|---------------------------|
| Skin | Normal | 2 |
| Breast | Normal | 3 |
| Spleen | Normal | 2 |
| Lymph node | Normal | 2 |
| Skeletal muscle | Normal | 2 |
| Lung | Normal | 3 |
| Salivary gland, sublingual | Normal | 2 |
| Liver | Normal | 2 |
| Gallbladder | Normal | 1 |
| Pancreas | Normal | 3 |
| Tonsil | Normal | 1 |
| Esophagus | Normal | 3 |
| Stomach, antrum | Normal | 3 |
| Stomach, fundus | Normal | 3 |
| Small bowel | Normal | 3 |
| Colon | Normal | 3 |
| Rectum | Normal | 3 |
| Kidney, cortex | Normal | 3 |
| Kidney, Medulla | Normal | 3 |
| Urinary bladder | Normal | 3 |

(Continued)

| Organ | Diagnosis | ^{a)} Tissue type |
|-----------------|--|---------------------------|
| Prostate | Normal | 2 |
| Testis | Normal | 2 |
| Uterine cervix | Normal | 3 |
| Endometrium | Normal | 1 |
| Myometrium | Normal | 1 |
| Placenta | Normal | 1 |
| Adrenal gland | Normal | 2 |
| Thyroid | Normal | 3 |
| Cerebrum | Normal | 1 |
| Cerebellum | Normal | 1 |
| Skin | Squamous cell carcinoma | 4 |
| Skin | Malignant melanoma | 4 |
| Subcutis | liposarcoma | 4 |
| Breast | ductal carcinoma in situ | 4 |
| Breast | infiltrating duct carcinoma | 4 |
| Lymph node | Hodgkin lymphoma | 4 |
| Bone | Osteosarcoma | 4 |
| Lung | Adenocarcinoma | 4 |
| Lung | Squamous cell carcinoma | 4 |
| Liver | cholangiocarcinoma | 4 |
| Liver | Hepatocellular carcinoma | 4 |
| Liver | Metastatic adenocarcinoma (from rectum) | 4 |
| Esophagus | Squamous cell carcinoma | 4 |
| Stomach | Adenocarcinoma | 4 |
| Stomach | Malignant lymphoma, diffuse large B cell | 4 |
| Stomach | Signet ring cell carcinoma | 4 |
| Duodenum | Gastrointestinal stromal tumor, malignant | 4 |
| Descending | Colon adenocarcinoma | 4 |
| Rectum | Adenocarcinoma | 4 |
| Kidney | Renal cell carcinoma | 4 |
| Urinary bladder | Transitional cell carcinoma | 4 |
| Prostate | Adenocarcinoma | 4 |
| Testis | Seminoma | 4 |
| Uterine cervix | Squamous cell carcinoma | 4 |

(Continued)

| Organ | Diagnosis | ^{a)} Tissue type |
|------------------|---|---------------------------|
| Endometrium | Adenocarcinoma | 4 |
| Ovary metastatic | Adenocarcinoma (from stomach) | 4 |
| Ovary mucinous | Cystadenocarcinoma | 4 |
| Ovary | Serous cystadenoma of low malignant potential | 4 |
| Thyroid | Papillary carcinoma | 4 |

^{a)}Normal and cancer tissues were obtained from different patients. The "tissue type" column includes the following categories:

- 1. normal tissue from non-cancer patients
- 2. normal tissue from cancer patients, but the cancer involves unrelated organs
- 3. normal tissue adjacent to the cancer
- 4. Cancer

LEGENDS FOR SUPPLEMENTARY MOVIES

(Supplementary movies are separately linked)



Supplementary MOVIE SM1: Motility of GFP-control and GFP-Swiprosin-1-expressing cells. Time-lapse images of B16F10 cells stably expressing GFP or GFP-swiprosin-1 were captured every 10 min for a total of 9 h. Analysis of cell migration velocity via nuclear tracking is shown in Figure 4E.



Supplementary MOVIE SM2: Translocation of GFP-Swiprosin-1 in B16F10 cells. Time-lapse fluorescence images of GFP-Swiprosin-1-expressing cells were captured at 5 sec intervals for 10 min using confocal microscopy. Images obtained at the selected time-points are presented in Figure 5A.



Supplementary MOVIE SM3: 3D animation of vertical section images of cells on a FITC-gelatin-coated transwell membrane. Angle rotation movies corresponding to Figure 5E (B16F1 and B16F10).



Supplementary MOVIE SM4: 3D animation of vertical section images of cells on a FITC-gelatin-coated transwell membrane. Angle rotation movies corresponding to Figure 5F. B16F1 cells transfected with Myc-empty vector (B16F1-Myc-ev) and Myc-Swiprosin-1 (B16F1-Myc-Sw1).