**Supplemental Materials and Methods**

**shRNA and siRNA**

Oligos for AXL shRNA were synthesized as previously described (Rankin et al., 2014). The sequence pair for AXL targeting and control are as following: 5′-GAGATTCTGTACGTGAATA-3′ (shSCM1), 5′-GAAATCCTCTATGTCAACA-3′ (shAXL1), 5′-GACTTAGAAAATACGCTC-3′ (shSCM2) and 5′-GATTTGGAGAACACACTGA-3′ (shAXL2). Oligos were cloned into the pSiren RetroQ (BD Bioscience) vector. Infected cells were selected in puromycin (Sigma) for 7 days and polyclonal populations were tested for decreased AXL expression levels by western blot analysis.

The ON-TARGET plus smart pool siRNAs for siCON (D-001810-10-20), siAXL(L-003104-00-0010) and siS100A10 (L-011766-00-0005) were purchased from Dharmacon. The knockdown was modified from the manufacture’s instruction. Briefly, 1×105 786-O or M62 cells were plated onto a 6 well plate and cultured overnight. The cells were transfected with DharmaFECT formulation 1 mixed with siCON pool, siAXL pool or siS100A10 pool. Forty-eight hours after the transfection, the cells were treated with 200 ng/ml of GAS6 in serum free medium for 24 hours. The medium was collected and centrifuged at 12000g for 10 min. The supernatant was used for the HUVEC matrigel invasion and in vivo matrigel plug assays.

**S100A10 Expression**

The plasmid containing S100A10 (Catalog number: 107200; YFP-S100A10) was purchased from Addgene. Empty vector pEGFP-C1 was used as control. Lipo3000 (Thermo Fisher Scientific) was used to transfect the cells according to the manufacturer’s instructions. Briefly, 1×105 786-O or M62 cells were plated onto a 6 well plate and cultured overnight. The cells were transfected with Lipo3000 mixed with Empty vector or YFP-S100A10. Forty-eight hours after the transfection, the cells were treated with 200 ng/ml of GAS6 in serum free medium for 24 hours. The medium was collected and centrifuged at 12000g for 10 min. The supernatant was used for the plasminogen, HUVEC matrigel invasion and in vivo matrigel plug assays.

**Real-time PCR**

RNA was isolated using Trizol (Invitrogen) and subsequently treated with DNase I (Thermo). First-strand cDNA synthesis was performed with SuperScript II Reverse Transcriptase and random primers (Applied Biosystems) according to the manufacturer's instructions. Quantitative real-time PCR was carried out using Power SYBR Green Master Mix (Bio-rad), detection and data analysis were executed with the 7900HT Fast Real-Time PCR System (Applied Biosystems) by computing the results relative to a standard curve made with cDNA pooled from all samples, normalized to 18S. For 18S amplification, cDNA was diluted 1:50. Primer sequences used to amplify specific target genes were obtained from the PrimerBank (https://pga.mgh.harvard.edu/primerbank/). The primers used in this paper are as following: AXL-Forward: 5’-GTGGGCAACCCAGGGAATATC-3’; AXL-Reverse: 5’- GTACTGTCCCGTGTCGGAAAG-3’; S100A10-Forward: 5’- GGCTACTTAACAAAGGAGGACC-3’; S100A10-Reverse: 5’- GAGGCCCGCAATTAGGGAAA-3’; 18S-Forward: 5’- GCCCGAAGCGTTTACTTTGA-3’; 18S-Reverse: 5’- TCCATTATTCCTAGCTGCGGTATC-3’; ANXA2-Forward: 5’- GAGCGGGATGCTTTGAACATT-3’; ANXA2-Reverse: 5’- TAGGCGAAGGCAATATCCTGT-3’**.**

**Immunohistochemistry**

For xenograft studies, tumor specimens were fixed in 10% (v/v) neutral buffered formalin, permeabilized with 95% (v/v) ethanol and embedded in paraffin. Tumor sections were subsequently deparaffinized with xylene, rehydrated in ethanol solutions and subjected to antigen retrieval using 10 mM citric acid buffer (pH 6.0) in microwave for 10 min. Slides were probed with primary antibody (Thermo Fisher Scientific) overnight at 4°C, followed by secondary detection using biotinylated anti-rabbit antibody (Vector Laboratories) and streptavidin-HRP conjugated antibody (EMD Millipore), both for 30 min at 37°C. Negative controls for all samples were tissue sections treated with secondary antibodies alone. Proteins were visualized with DAB Chromogen System (DAKO), counterstained with Hematoxylin (VWR) and mounted on slides with Fluoromont-G (Southern Biotech). The primary antibodies used for IHC staining are AXL (1:100, AF154, R&D), S100A10 (1:200, 4E7E10, Santa Cruz), CD31 (1:200, ab28364, ABcam), Ku70 (1:200, AB18560, ABcam), CAIX (1:500, NB-100-417, Novus), CD117 (1:200, CME296, Biocare Medical), CD10 (1:200, CM129AK, Biocare Medical) and CK7 (1:200, OV-TL 12/30, Dako Agilent). Slides were scanned with NanoZoomer (Hamamatsu). Slides were analyzed with equipped NDP view 2 software (Hamamatsu).

**Genomic VHL Sequencing**

DNA was extracted from tissue slice grafts preserved in Allprotect tissue reagent (Qiagen, Valencia, CA) using an AllPrep DNA/RNA/Protein Mini Kit (Qiagen) according to manufacturer’s directions. The 3 exons of VHL were selected for polymerase chain reaction amplification and direct sequencing. The primer used for exon amplification are as followings: Exon 1—Forward: 5'-CTACGGAGGTCGACTCGGGAG-3'; Exon 1—Reverse: 5'-GGGCTTCAGACCGTGCTATCG-3'; Exon 2—Forward: 5'-CCGTGCCCAGCCACCGGTGTG-3'; Exon 2—Reverse: 5'-GGATAACGTGCCTGACATCAG-3'; Exon 3—Forward: 5'-CGTTCCTTGTACTGAGACCCTA-3' and 'Exon 3—Reverse: 5'-GAACCAGTCCTGTATCTAGATCAAG-3'.

**Proliferation Assay**

786-0 and M62 cells were treated with 200 ng/mL of GAS6 for overnight. 1×104 cells were plated onto 96 well plates. After 24 hours or 48 hours of 21% or 2% oxygen treatment, viable cells were quantified with CellTiter-Glo® Luminescent Cell Viability Assay kit (G7570, Promega) according to the manufacturer’s instructions.

**Clonogenic Survival Assay**

786-0 and M62 cells were treated with 200 ng/mL of GAS6 for overnight. 5×102 cells were plated onto 6 well plates. Cells were cultured at 37 ℃, 5% CO2 for colony formation. After 14 days, cells were fixed with ethanol and stained with 1% crystal violet. Colonies were counted manually.

**Tumor Xenografts**

All procedures involving animals and their care were approved by the Institutional Animal Care and Use Committee of Stanford University in accordance with institutional and National Institutes of Health guidelines. To establish orthotropic ccRCC xenograft model, Rag2-/-IL2rg-/- double knockout mice aged from 6 and 12 weeks were injected with a 100 μL of collagen plug containing 1 × 106 ccRCC cells under the renal capsule as previously described (Thong et al., 2014). Briefly, mice were anesthetized and maintained under a heating lamp. The skin at the incision site was shaved and sterilized with three scrubs of povidone iodine followed 70% ethanol. A short incision was made with a scalpel immediately over the kidney. The kidney was gently exposed through the incision and kept moist with sterile physiological saline. A small hole in the renal capsule was made with forceps and raised to create a small pocket between the capsule and the underlying kidney tissue. The 100 μl collagen plug (3.5 mg/mL containing 1 × 106 cells) was inserted into this pocket with forceps. The renal capsule was released and allowed to cover the inserted tissue slice. The kidney was gently placed back through the incision, and the body wall was sutured. The skin incision was sealed with wound clips. Carprofen (4-5 mg/kg) was injected subcutaneously. Sterile practices were followed throughout the surgical procedure.

For intrahepatic injection, mice were anesthetized and maintained under a heating lamp. The skin at the incision site was shaved and sterilized with three scrubs of povidone iodine followed 70% ethanol. A midline abdominal incision of the skin and abdominal muscle layer (starting ~0.5 cm under the xyphoid process) was made. The median lobe of the liver was exposed using a sterile cotton swab soaked in sterile saline solution. A 27-gauge needle was inserted under the liver capsule containing the tumor cell cells in a PBS/Matrigel solution (1:1, 20 µl). After injecting the tumor cell/Matrigel solution, the needle was slowly retracted. Leakage was prevented by placing a sterile Gelfoam for 5 min on the needle track. The muscle layer of the abdominal wall was closed by a continuous suture using 5-0 or 6-0 absorbable sutures. The skin incision was closed with sterile discontinuous suture. Mice were allowed to recover from anesthesia and were monitored every 15 minutes until ambulatory. As part of the recovery, mice were monitored daily and given Nutrical to provide additional nutrition in the first few postoperative days, in addition to standard rodent food pellets. All the mice will be given carprofen every 24 hours for the first three days.

**Patient Derived Xenografts**

All patients who participated in this study provided written informed consent for collection and research use of their materials and use of these samples was approved by the Stanford University Institutional Review Board (IRB #34175). Patient derived tissue RCC054 was obtained from a 44-year-old male undergoing surgery to remove metastatic RCC tissue from the colon at Stanford under an institutional review board–approved protocol with informed consent. We established two additional RCC PDX models from materials received from the NCI Patient-Derived Models Repository (PDMR), NCI-Frederick, Frederick National Laboratory for Cancer Research, Frederick, MD. URL – <http://www.pdmr.nci.gov>. We established a second PDX line from third-generation RCC PDX tissues obtained from NCI Patient-Derived Models Repository (PDMR) (Patient ID: 597326, https://pdmr.cancer.gov/). This patient, a 49-year-old male, received radical nephrectomy surgery upon diagnosis. Sequencing analysis of PDX tissues from our cohort of mice confirmed a single nucleotide deletion within VHL at position 403, leading to a frame shift. Histological analysis of the patient tumor demonstrated that histological features of clear cell renal cell carcinoma. We established a third PDX line from third-generation RCC PDX tissues obtained from NCI Patient-Derived Models Repository (PDMR) (Patient ID: 961994, https://pdmr.cancer.gov/). This patient, a 60-year-old male, received radical nephrectomy surgery upon diagnosis. Sequencing analysis of PDX tissues from our cohort of mice confirmed a deletion of two nucleotides within VHL at position 450, resulting in a premature stop codon. Histological analysis of the patient tumor demonstrated that histological features of clear cell renal cell carcinoma. For the PDX tumor models, we followed procedures are previously described by our group (Thong et al., 2014)

**RNA-Sequencing and Data Analyses**

RNA samples were extracted from 1×106 cancer cells using Qiagen RNeasy Mini Kit. Two μg /sample were delivered to Novogene Corporation for cDNA library preparation and sequencing. Briefly, cDNA was sonicated and subjected to library preparation using the Illumina TruSeq DNA sample preparation kit. Total RNA from 786-O-shSCM1, 786-O-shAXL1, M62-shSCM1 and M62-shAXL1 cells were used for the preparation of RNA-Seq libraries with Illumina's TruSeq RNA Library Prep Kit v2 according to manufacturer's protocol. Sequencing was performed on Illumina HiSeq 2000.

The RNA-Seq data are deposited at the Gene Expression Omnibus ([www.ncbi.nlm.nih.gov/geo/](http://www.ncbi.nlm.nih.gov/geo/)) under the series number GSE125994. The raw RNA-Seq reads were trimmed for quality control by TrimGalore v0.4.2 (https://www.bioinformatics.babraham.ac.uk/projects/trim\_galore/) and then mapped to hg38 using STAR software v2.5.3 with ENCODE options (Dobin et al., 2013). Gene expression quantifications measured in TPM were estimated by RSEM v1.2.30 based on Gencode hg38 v25 annotation (Harrow et al., 2012; Li and Dewey, 2011). Gene for Venn plot is selected with cutoff expression >2, fold change >2.

**Supplementary Table legends**

**Supplementary Table 1 . Gene expression profile of 786-O cell transfected with**

**shSCM1 and shAXL1**

**Supplementary Table 2. Gene expression profile of M62 cell transfected with shSCM1 and shAXL1**

**Supplementary Table 3. List of genes with >2-fold change in 786-O and M62 cells with AXL knockdown**

**Supplementary Table 4. List of genes with >2-fold downregulation in 786-O and M62 cells with AXL knockdown**