Supplementary Material and Methods

Primers for quantitative RT-PCR.

The sequences for primers are as follows:

Human Cdk1
Forward: 5’- TGGATCTGAAGAAATACTTGATTCTA -3’
Reverse: 5’- CAATCCCCTGTAGGATTTGG -3’

Human Cdk5
Forward: 5’- CGATGACCAGTTGAAGAGGAT -3’
Reverse: 5’- TCTGGCAGCTTGGTCATAGA -3’

Human PCTAIRE1
Forward: 5’- GCAGTGACCCTGGAGAGG -3’
Reverse: 5’- TCAAGTCCCTCGTGCAACAATC -3’

Human p27
Forward: 5’- TTTGACTTTGCATGAAGAGAGC -3’
Reverse: 5’- AGCTGTCTCTGAAAGGGACATT -3’

Human GAPDH
Forward: 5’- GAAGGTGAAGGTCGGAGTTC -3’
Reverse: 5’- ATGGGATTTCCATTGATGAC -3’
All experiments were performed in duplicate and normalized with respect to GAPDH levels.

**In vitro kinase assays.** pcDNA3-HA-tagged PCTAIRE1 (wild type or mutant) were transfected into HEK293T cells in 10 cm dishes. After 24 hours, PCTAIRE1 protein was immunoprecipitated with 2 μg anti-HA antibody. Purified recombinant substrates (1 μg wild type or mutant p27) and immunoprecipitated HA-PCTAIRE1 were incubated in 50 μl kinase buffer (50 mM Tris-HCl, pH 7.5, 150 mM NaCl, 10 mM MgCl₂, 1 mM DTT, 200 μM ATP) for 30 minutes at 30 °C. Reactions were stopped by addition of SDS sample buffer, followed by SDS-PAGE/immunoblotting.

**Time-lapse microscopy.** PPC1 cells containing Tet-inducible shRNA targeting for PCTAIRE1 (shRNA#2) were plated on 35 mm grass bottom culture dishes, and transfected with 1 μg of pEGFP-alpha-tubulin and pmCherry-histone H2B plasmids using Lipofectoamine 2000. After 5 hours, cells were cultured with RPMI media with (Tet-ON) or without (Tet-OFF) 100 ng/ml doxycycline. After 48 hours, dishes were transferred to a heated stage (37°C) on an Inverted IX81Olympus Fluorescence Microscope. Fluorescence images of live cells were collected at 20-minutes intervals for 24 hours.
**Patient specimens.** For prostate cancer analyses, radical prostatectomy specimens were procured retrospectively as part of an Institutional Review Board approved protocol at the University of California, Irvine. 115 patients with locally confined (stage II) and locally advanced (stage III) disease were enrolled during 1992-1996. Prostate cancer tissue microarrays (TMAs) were constructed, as described (1). The TMAs included prostate specimens derived using a cautery-free technique to preserve the neurovascular bundles during robotic laparoscopic radical prostatectomy. All patients were <66 years old and underwent unilateral or bilateral dissections. Each focus of cancer was assigned a Gleason score by two pathologists. For breast cancer analyses, tissue specimens containing normal mammary epithelium (n = 16), in situ breast carcinomas (n = 23), and 121 invasive breast tumors, representing ductal (n = 103), lobular (n = 15), and mucinous (n = 3) histologic subtypes were obtained from St. Vincent’s Hospital in Dublin, Ireland, for TMA generation. The specimens were derived from women who presented in 2001 with symptomatic stage I to III breast cancers, using residual pathologic materials remaining after diagnostic and hormone receptor determination.

**Gene expression.** Individual cancer data sets were downloaded from Oncomine (https://www.oncomine.org), as previously described (2). The prognostic significance of *PCTAIRE1* in overall survival of lung and breast cancers was assessed using Kaplan-Meier
Plotter (http://kmplot.com/analysis/), which employed gene expression microarray data and survival information of patients with breast or lung cancer downloaded from GEO (Affimetrix HGU133A, HGU 133+2, and HGU 133A 2.0) (3).

**Statistical Analysis.** Spearman’s correlation coefficient test was used to identify a significant association of PCTAIRE1 immunoscore with p27 immunoscore.
SUPPLEMENTAL REFERENCES

