Table W1. List of Oligonucleotides Used for Quantitative PCR Analysis.

Gene (Symbol) or Amplicon	Primer Sequences (Sense and Antisense, 5′ → 3′)
CXCL2	GCGCCCAGACAGAAGTCATAG
	AGCCTTGCCTTTGTTCAGTATC
CXCL5	GAAAGCTAAGCGGAATGCAC
	GGGACAATGGTTTCCCTTTT
CXCL10	GCTCCTGCATCAGCACCAGC
	CTTGAACGACGACGACTTTGG
CXCL15	CCA TGG GTG AAG GCT ACT GT
	TCT CAG GTC TCC CAA ATG AAA
Cytokeratin18	CAGCCAGCGTCTATGCAGG
	CTTTCTCGGTCTGGATTCCAC
ICAM1	TGC GTT TTG GAG CTA GCG GAC CA
	CGA GGA CCA TAC AGC ACG TGC AG
TNF	TAGCCAGGAGGGAGAACAGA
	TTTTCTGGAGGGAGATGTGG
E-Cadherin	AAGTGACCGATGATGATGCC
	CTTCTCTGTCCATCTCAGCG
Collagen1	CAC CCT CAA GAG CCT GAG TC
	GTT CGG GCT GAT GTA CCA GT
Flag-IKK2	GAC TAC AAG GAC GAC GAT GAC AAG
	GGT TCA GCC TTC TCA TGA TCT GG
SFRP1	CAACGTGGGCTACAAGAAGAT
	GGCCAGTAGAAGCCGAAGAAC
SFRP4	AGAAGGTCCATACAGTGGGAAG
	GTTACTGCGACTGGTGCGA
Nkx3-1	ATGCTTAGGGTAGCGGAGC
	TGCGGATTGCCTGAGTGTC
Spink3	ATG AAG GTG GCT GTC ATC TTT C
	TCA GCA AGG CCC ACC TTT TCG
Smooth muscle actin	GTCCCAGACATCAGGGAGTAA
	TCGGATACTTCAGCGTCAGGA
Probasin	AAGGCTCACCATTGAGAACCT
	CAGTTGGCACTTAGTCCCTTTC
MYH11	AAGCTGCGGCTAGAGGTCA
	CCCTCCCTTTGATGGCTGAG
HPRT	CAA ATC AAA AGT CTG GGG ACG C
	GCT TGC TGG TGA AAA GGA CCT C

Table W2. List of Antibodies Used in This Study.

Antibody/Antigen	Source/Catalog No. (or Equivalent)	Use/Dilution
CD3e	eBioscience/13-0031	IF-F/1:200
F4/80	eBioscience/13-4801	IF-F/1:200
CD11b	eBioscience/13-0112	IF-F/1:200
Gr-1	eBioscience/13-5931	IF-F/1:200
Ki67	Neomarkers/RM-9106-S1	IF-P/1:200
Lipocalin2	Santa Cruz/sc-18698	IHC-P/1:200
Nkx3-1	Santa Cruz/sc-15022	IB/1:400
Smooth muscle actin	Sigma/C6198	IF-P/1:400
		IF-F/1:400
P63	Santa Cruz/sc-56188	IF-P/1:100
Cytokeratin14	Prof E.B. Lane (University of Dundee)	IF-cells/1:10
Androgen receptor	Santa Cruz/sc-815	IF-P/1:100
β-Tubulin	Santa Cruz/sc-9104	IB/1:400
IκBα	Santa Cruz/sc-371	IB/1:1000, IF-F/1:100
ICAM1	eBioscience/14-0541	IF-F/1:100
A555 goat antirabbit	Invitrogen/A-21428	IF-P, IF-F/1:2000
Biotin antirabbit	Vector Laboratories/BA-1000	IHC-P/1:400
Biotin antigoat	Vector Laboratories/BA-9500	IHC-P/1:400
Biotin antimouse	Vector Laboratories/BA-9200	IHC-P/1:400
Streptavidin, Alexa Fluor 555 conjugate	Invitrogen/S32355	IF-F/1:500

F indicates frozen sections; IB, immunoblot; IF, immunofluorescence; IHC, immunohistochemistry; P, paraffin sections.

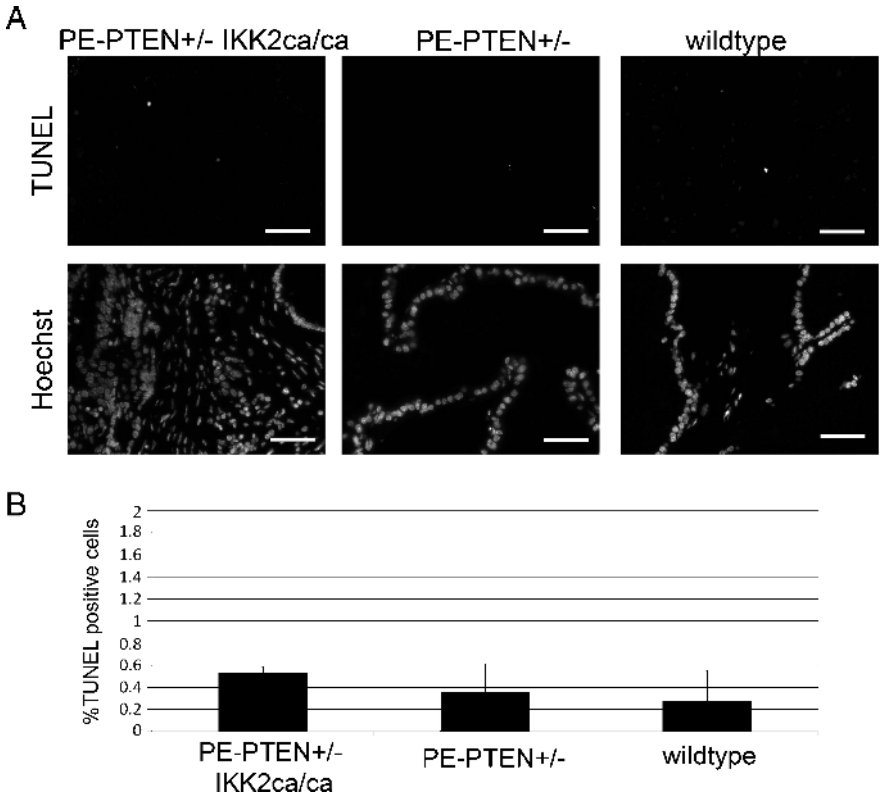

Figure W1. TUNEL stain indicating no change in apoptosis in transgenic *versus* wild-type prostases. (A) TUNEL staining (see Materials and Methods) was performed on paraffin sections from prostates of the indicated genotypes. Hoechst staining is given to indicate tissue architecture and cell density. (B) Quantification of TUNEL-positive cells in lateral cells in the lateral prostates of the indicated genotypes ($n = 4$ per group). Error bars, SEM. No statistical significance was reached in the analysis of variance.

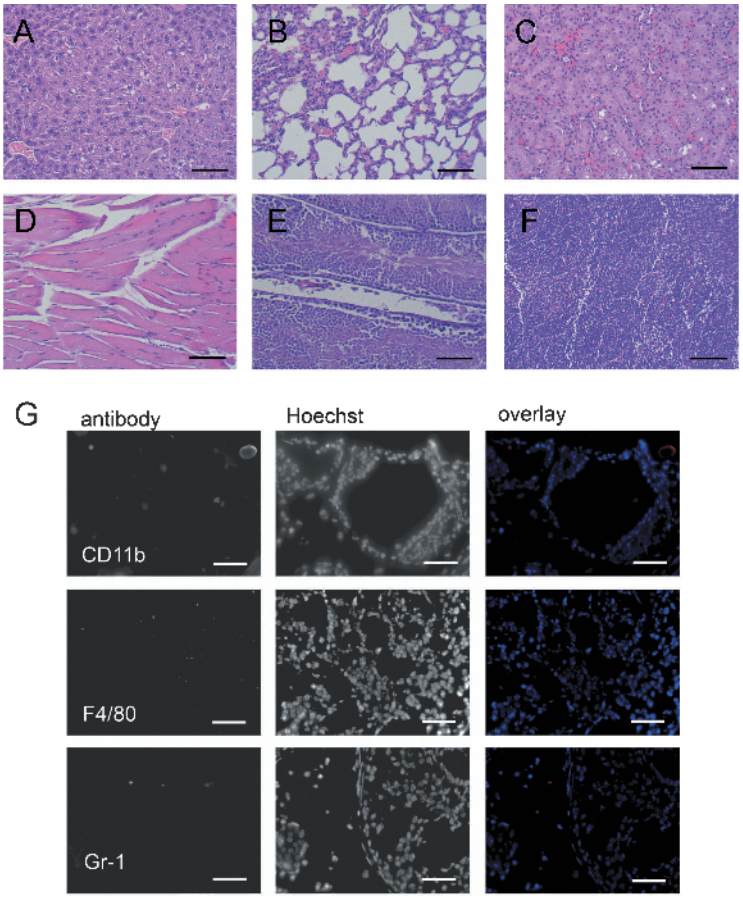

Figure W2. Normal histologic finding of nonprostate tissues from PE−PTEN+/− IKK2ca/ca mice (A-F) and lack of immunoreactivity in PE−PTEN+/− lateral prostates (G). H&E stains are shown for liver (A), lung (B), kidney (C), muscle tissue surrounding the urethra (D), testis (E), and lymph node (F). Scale bars, 100 μm. (G) Antibody staining in PE−PTEN+/− lateral prostates. Scale bars, 50 μm.

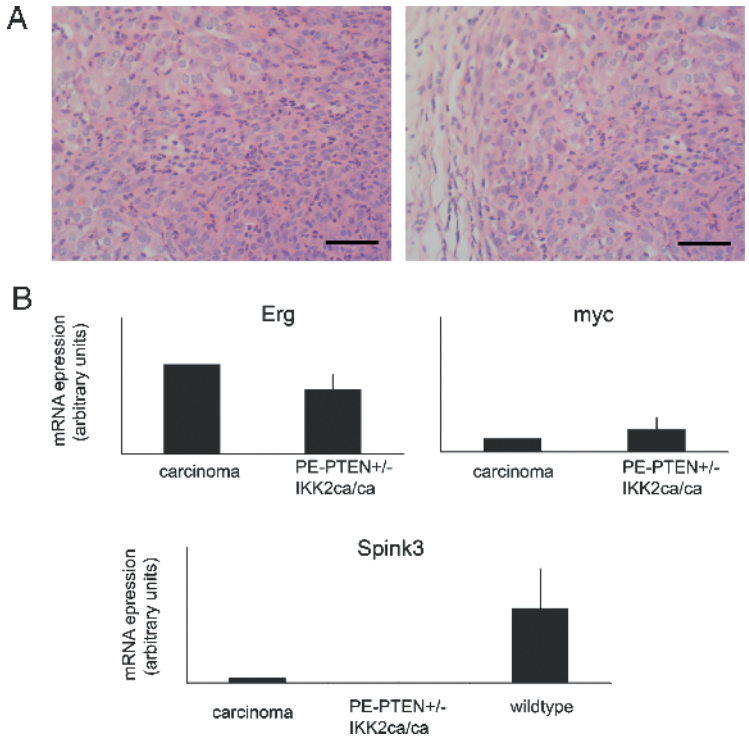

Figure W3. Carcinoma formation in a PTEN+/−IKK2ca/ca lateral prostate (12 months). (A) H&E stain of the tumor itself (left) and adjacent to the stroma (right). (B) Quantitative PCR data of carcinoma ($n = 1$) compared with PE−PTEN+/−IKK2ca/ca prostate ($n = 3$) or wild-type prostate ($n = 3$). Scale bars, 50 μm.

Table W3. The 100 Most Overexpressed Genes in the Microarray Study Comparing PE–PTEN+/–IKK2ca/ca to PE–PTEN+/–.

Gene Symbol	Full Name	Fold Expression
Msln	Mesothelin	33.12
Ly6d	Lymphocyte antigen 6 complex, locus D	13.64
Gabrp	γ-Aminobutyric acid (GABA) A receptor, pi	13.16
Hp	Haptoglobin	11.96
Clca2	Chloride channel calcium activated 2	11.78
Cxcl5	Chemokine (C-X-C motif) ligand 5	11.68
Olfm4	Olfactomedin 4	9.95
Egln3	EGL nine homolog 3 (*C. elegans*)	8.26
Slco1a5	Solute carrier organic anion transporter family, member 1a5	7.98
Sfrp4	Secreted frizzle-related protein 4	7.76
AW112010	Expressed sequence AW112010	7.61
Sftpd	Surfactant associated protein D	7.26
Gdpd3	Glycerophosphodiester phosphodiesterase domain containing 3	7.23
Cxcl15	Chemokine (C-X-C motif) ligand 15	7.22
Ccdc129	Coiled-coil domain containing 129	7.00
Cd14	CD14 antigen	6.97
Icam1	Intercellular adhesion molecule 1	6.94
Tnfsf10	Tumor necrosis factor (ligand) superfamily, member 10	6.90
Car13	Carbonic anhydrase 13	6.90
Muc20	Mucin 20	6.78
Ctla2a	Cytotoxic T-lymphocyte–associated protein 2α	6.66
Atp6v1b1	ATPase, H^+ transporting, lysosomal V1 subunit B1	6.48
Aspa	Aspartoacylase	6.42
C3	Complement component 3	6.07
Ltbp2	Latent transforming growth factor β binding protein 2	5.98
S100a8	S100 calcium binding protein A8 (calgranulin A)	5.74
Car2	Carbonic anhydrase 2	5.57
Cxcl10	Chemokine (C-X-C motif) ligand 10	5.35
Clic6	Chloride intracellular channel 6	5.28
Clip4	CAP-GLY domain containing linker protein family, member 4	5.21
Vsig1	V-set and immunoglobulin domain containing 1	5.20
Wfdc2	WAP four-disulfide core domain 2	5.18
Il8rb	Interleukin 8 receptor, β	5.15
Ccl3	Chemokine (C-C motif) ligand 3	5.11
Ttc9	Tetratricopeptide repeat domain 9	5.05
Cyba	Cytochrome *b*-245, α polypeptide	5.01
Clec4d	C-type lectin domain family 4, member d	4.94
Cp	Ceruloplasmin	4.89
Tnip3	TNFAIP3 interacting protein 3	4.84
Csf3r	Colony stimulating factor 3 receptor (granulocyte)	4.83
Mmp3	Matrix metallopeptidase 3	4.81
Slfn4	Schlafen 4	4.75
Sestd1	SEC14 and spectrin domains 1	4.71
Gm7609	Predicted gene 7609	4.69
Cpxm1	Carboxypeptidase X 1 (M14 family)	4.65
Padi4	Peptidyl arginine deiminase, type IV	4.64
Itgb6	Integrin β6	4.63
Srgn	Serglycin	4.59
Gm7609	Predicted gene 7609	4.58
Cd200r3	CD200 receptor 3	4.56
Clec4n	C-type lectin domain family 4, member n	4.52
H2-Q6	Histocompatibility 2, Q region locus 6	4.49
Csprs	Component of Sp100-rs	4.39
Nfkbie	Nuclear factor of κ light polypeptide gene enhancer in B-cells inhibitor, epsilon	4.38
Mgat3	Mannoside acetylglucosaminyltransferase 3	4.32
Clca1	Chloride channel calcium activated 1	4.29
Ces3	Carboxylesterase 3	4.29
Car8	Carbonic anhydrase 8	4.29
Tnf	Tumor necrosis factor	4.28
Csprs	Component of Sp100-rs	4.26
Wbscr17	Williams-Beuren syndrome chromosome region 17 homolog (human)	4.18
Gpnmb	Glycoprotein (transmembrane) nmb	4.17
Steap4	STEAP family member 4	4.13
Slc39a4	Solute carrier family 39 (zinc transporter), member 4	4.12
Cxcl2	Chemokine (C-X-C motif) ligand 2	4.10
Pglyrp1	Peptidoglycan recognition protein 1	4.04
Dram1	DNA-damage regulated autophagy modulator 1	3.97
Grid2	Glutamate receptor, ionotropic, delta 2	3.94
Atp6v0d2	ATPase, H^+ transporting, lysosomal V0 subunit D_2	3.90
Itga2	Integrin α2	3.85
Il1f9	Interleukin 1 family, member 9	3.85
Capsl	Calcyphosine-like	3.84
Pmepa1	Prostate transmembrane protein, androgen induced 1	3.80
Clec5a	C-type lectin domain family 5, member a	3.78
Dclk1	Doublecortin-like kinase 1	3.73
S100a9	S100 calcium binding protein A9 (calgranulin B)	3.71
Gp49a	Glycoprotein 49 A	3.69
Ly6k	Lymphocyte antigen 6 complex, locus K	3.65
Mpeg1	Macrophage expressed gene 1	3.64
Il1b	Interleukin 1β	3.62
Cxcr4	Chemokine (C-X-C motif) receptor 4	3.60
Eya2	Eyes absent 2 homolog (*Drosophila*)	3.59
Tyrobp	TYRO protein tyrosine kinase binding protein	3.59
Pdgfrl	Platelet-derived growth factor receptor-like	3.58
Sfrp1	Secreted frizzle-related protein 1	3.54
Casp1	Caspase 1	3.54
Gda	Guanine deaminase	3.54
Nfkbia	Nuclear factor of κ light polypeptide gene enhancer in B-cells inhibitor, α	3.52
Gsdmc2	Gasdermin C2	3.49
Nedd9	Neural precursor cell expressed, developmentally downregulated gene 9	3.49
Frzb	Frizzle-related protein	3.49
Arl14	ADP-ribosylation factor-like 14	3.49
Gm7609	Predicted gene 7609	3.47
Krt7	Keratin 7	3.47
Ubd	Ubiquitin D	3.45
Fcrls	Fc receptor-like S, scavenger receptor	3.45
Mmp12	Matrix metallopeptidase 12	3.45
Serpinb11	Serine (or cysteine) peptidase inhibitor, clade B (ovalbumin), member 11	3.44
Tgtp	T-cell–specific GTPase	3.43
Lyz2	Lysozyme 2	3.38

Table W4. The 100 Most Downregulated Genes in the Microarray Study Comparing PE–PTEN+/–IKK2ca/ca to PE–PTEN+/–.

Gene Symbol	Full Name	Fold Expression
Spink3	Serine peptidase inhibitor, Kazal type 3	0.00
Sbp	Spermine binding protein	0.01
Wfdc3	WAP four-disulfide core domain 3	0.02
Pnliprp1	Pancreatic lipase-related protein 1	0.02
Spink5	Serine peptidase inhibitor, Kazal type 5	0.03
4930408F14Rik	RIKEN cDNA 4930408F14 gene	0.04
4930408F14Rik	RIKEN cDNA 4930408F14 gene	0.04
4930408F14Rik	RIKEN cDNA 4930408F14 gene	0.04
Syt10	Synaptotagmin X	0.04
4930408F14Rik	RIKEN cDNA 4930408F14 gene	0.05
Abo	ABO blood group (transferase A, α 1-3-*N*-acetylgalactosaminyltransferase, transferase B, α 1-3-galactosyltransferase)	0.05
Eapa2	Experimental autoimmune prostatitis antigen 2	0.05
Pbsn	Probasin	0.06
G6pc2	Glucose-6-phosphatase, catalytic, 2	0.06
9530002B09Rik	RIKEN cDNA 9530002B09 gene	0.06
Derl3	Der1-like domain family, member 3	0.06
Azgp1	α-2-Glycoprotein 1, zinc	0.07
Atp12a	ATPase, H^+/K^+ transporting, nongastric, α polypeptide	0.07
4930408F14Rik	RIKEN cDNA 4930408F14 gene	0.07
Defb10	Defensin β10	0.07
Tgm4	Transglutaminase 4 (prostate)	0.07
4930408F14Rik	RIKEN cDNA 4930408F14 gene	0.07
D030018L15Rik	Nuclear receptor coactivator 2 pseudogene	0.07
4930408F14Rik	RIKEN cDNA 4930408F14 gene	0.08
Pcp4	Purkinje cell protein 4	0.08
4930408F14Rik	RIKEN cDNA 4930408F14 gene	0.08
Gm5615	Predicted gene 5615	0.08
Defb1	Defensin β1	0.09
Ceacam2	Carcinoembryonic antigen–related cell adhesion molecule 2	0.09
Hmgcs2	3-Hydroxy-3-methylglutaryl-coenzyme A synthase 2	0.09
Htr3a	5-Hydroxytryptamine (serotonin) receptor 3A	0.09
Atp6v1c2	ATPase, H^+ transporting, lysosomal V1 subunit C2	0.09
Cyp3a57	Cytochrome P450, family 3, subfamily a, polypeptide 57	0.10
Agtr1a	Angiotensin II receptor, type 1a	0.10
Mme	Membrane metalloendopeptidase	0.10
Prom2	Prominin 2	0.10
Aass	Aminoadipate-semialdehyde synthase	0.11
Slc9a2	Solute carrier family 9 (sodium/hydrogen exchanger), member 2	0.11
Epha3	Eph receptor A3	0.11
Smgc	Submandibular gland protein C	0.11
Decaf12l1	DDB1 and CUL4 associated factor 12-like 1	0.11
Myh11	Myosin, heavy polypeptide 11, smooth muscle	0.12
Acrbp	Proacrosin binding protein	0.12
Apof	Apolipoprotein F	0.12
Nkx2-6	NK2 transcription factor related, locus 6 (*Drosophila*)	0.12
Hhip	Hedgehog-interacting protein	0.12
Egf	Epidermal growth factor	0.12
Uph1	Ureidopropionase, β	0.13
Prlr	Prolactin receptor	0.13
Spc25	SPC25, NDC80 kinetochore complex component, homolog (*S. cerevisiae*)	0.13
Nkx3-1	NK-3 transcription factor, locus 1 (*Drosophila*)	0.13
Hpgd	Hydroxyprostaglandin dehydrogenase 15 (NAD)	0.13
Prlr	Prolactin receptor	0.14
Gpr165	G protein–coupled receptor 165	0.14
Cbs	Cystathionine β-synthase	0.14
Slc30a10	Solute carrier family 30, member 10	0.14
Bmpr1b	Bone morphogenetic protein receptor, type 1B	0.14
Fcgbp	Fc fragment of IgG binding protein	0.15
Fam38b2	Family with sequence similarity 38, member B2	0.15
Srd5a2	Steroid 5 α-reductase 2	0.15
Fam38b	Family with sequence similarity 38, member B	0.15
Cdh8	Cadherin 8	0.15
Spink11	Serine peptidase inhibitor, Kazal type 11	0.16
Gabra4	γ-Aminobutyric acid (GABA) A receptor, subunit α 4	0.16
Abca5	ATP-binding cassette, subfamily A (ABC1), member 5	0.16
Pdzk1	PDZ domain containing 1	0.16
Slc30a2	Solute carrier family 30 (zinc transporter), member 2	0.16
Acvr1c	Activin A receptor, type IC	0.17
H2-Q10	Histocompatibility 2, Q region locus 10	0.17
Fah	Fumarylacetoacetate hydrolase	0.17
Bhlha15	Basic helix-loop-helix family, member a15	0.17
Slc26a4	Solute carrier family 26, member 4	0.17
Cckar	Cholecystokinin A receptor	0.17
Cnn1	Calponin 1	0.18
Wif1	Wnt inhibitory factor 1	0.18
Ggt1	γ-Glutamyltransferase 1	0.18
Cyp2j13	Cytochrome P450, family 2, subfamily j, polypeptide 13	0.18
Slc5a3	Solute carrier family 5 (inositol transporters), member 3	0.18
Otud7a	OTU domain containing 7A	0.18
Gm1574	Predicted gene 1574	0.19
Defb50	Defensin β50	0.19
Slc18a1	Solute carrier family 18 (vesicular monoamine), member 1	0.20
Chn2	Chimerin (chimaerin) 2	0.20
Tmem45a	Transmembrane protein 45a	0.20
Man1a	Mannosidase 1, α	0.20
Spink8	Serine peptidase inhibitor, Kazal type 8	0.20
Glb1l3	Galactosidase, β1 like 3	0.20
5430419D17Rik	RIKEN cDNA 5430419D17 gene	0.21
9130230L23Rik	RIKEN cDNA 9130230L23 gene	0.21
Fam115c	Family with sequence similarity 115, member C	0.21
Myl9	Myosin, light polypeptide 9, regulatory	0.21
Rab39b	RAB39B, member RAS oncogene family	0.21
Ms4a5	Membrane-spanning 4 domains, subfamily A, member 5	0.22
Gnmt	Glycine *N*-methyltransferase	0.22
P2rx1	Purinergic receptor P2X, ligand-gated ion channel, 1	0.22
Doc2b	Double C2, β	0.22
BC005685	cDNA sequence BC005685	0.22
Bmp7	Bone morphogenetic protein 7	0.22
BC005685	cDNA sequence BC005685	0.22
Upk1a	Uroplakin 1A	0.22
Nxf7	Nuclear RNA export factor 7	0.22
Rnase1	Ribonuclease, RNase A family, 1 (pancreatic)	0.23
BC005685	cDNA sequence BC005685	0.23
Ndrg2	N-*myc* downstream regulated gene 2	0.23
Slco4c1	Solute carrier organic anion transporter family, member 4C1	0.23
Gpr110	G protein–coupled receptor 110	0.23
Slc22a1	Solute carrier family 22 (organic cation transporter), member 1	0.23
Proc	Protein C	0.24
Acrv1	Acrosomal vesicle protein 1	0.24
Aldh6a1	Aldehyde dehydrogenase family 6, subfamily A1	0.25

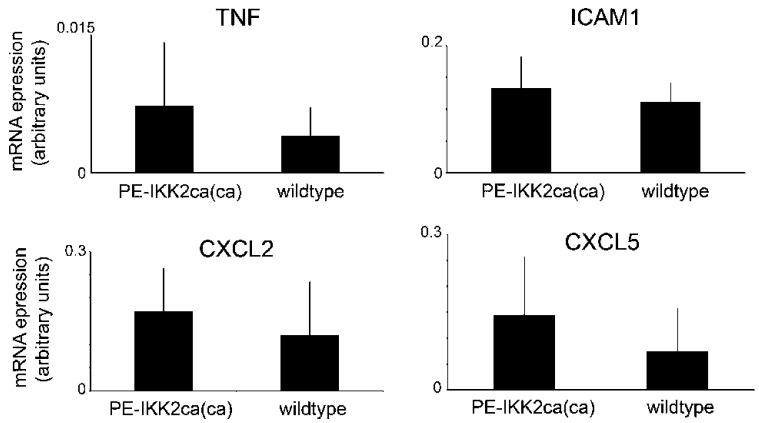

Figure W4. Quantitative PCR analysis of NF-κB–dependent target genes in IKK2ca(ca) lateral prostates compared with wild-type (12 months; $n = 5$ per group). Error bars, SEM.

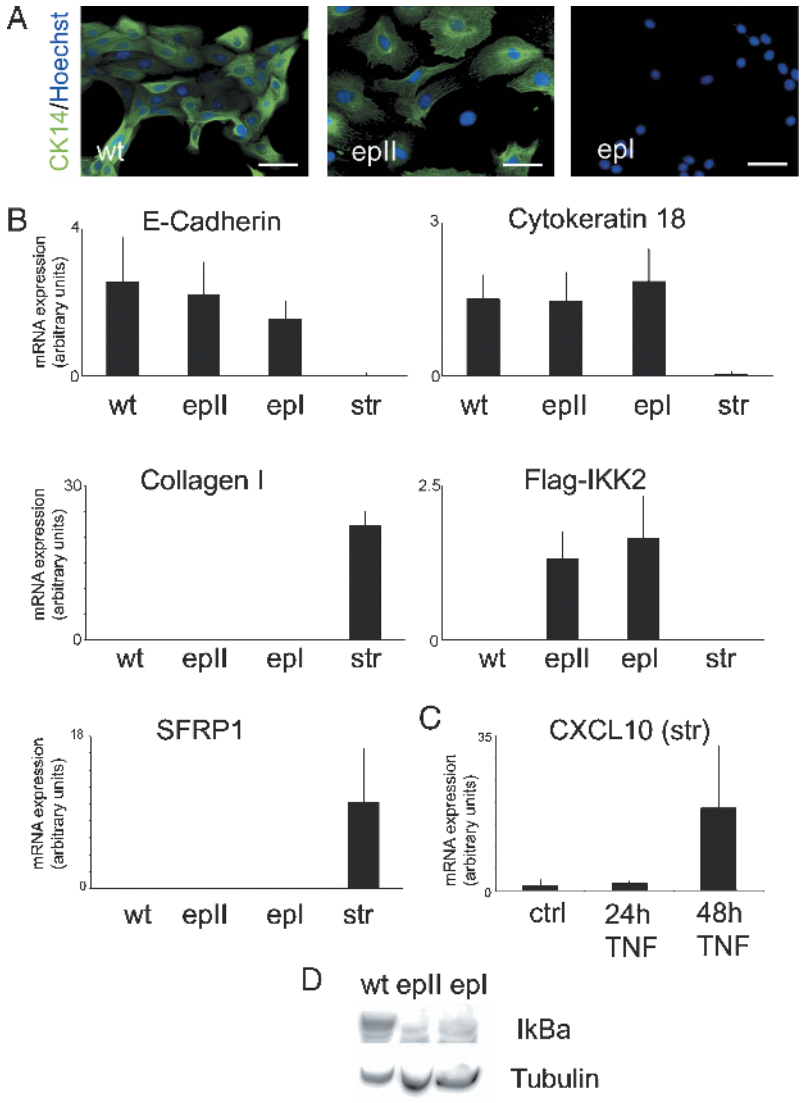

Figure W5. Expression of epithelial and stromal markers in epithelial and stromal cell lines. (A) Colored image of cytokeratin 14 (CK14) staining of indicated epithelial cell lines in culture. Green indicates CK14 signal; blue, Hoechst signal. Scale bars, 50 μm. (B) Expression profile of epithelial and stromal cell lines. The mRNA expression was determined by quantitative RT-PCR. (C) Expression of CXCL10 mRNA in the stromal cell line (str) on treatment with TNF (10 ng/ml). Ctrl indicates control cells (untreated). (D) Western blot showing degradation of IκBα in transgenic cells, indicating IKK2 activity.