Supplemental Data section

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Supplemental Materials and Methods

Genetic crosses used in this study

To generate mice inactivated for both pRb and p130, *K14CreRb$^{f/f}$* (maintained on the inbred FVB/N genetic background) were crossed to *Rb$^{f/f}$p130$^{-/-}$* mice (on the inbred 129/C57 genetic background) and *K14Cre$^{+/-}$Rb$^{f/+}$p130$^{+/-}$* offspring then crossed to *Rb$^{f/+}$p130$^{+/-}$* mice to generate *Rb$^{f/f}$p130$^{-/-}$*, *K14CreRb$^{f/f}$*, and *K14CreRb$^{f/f}$p130$^{-/-}$* mice on a FVB/129/C57 mixed genetic background. Control nontransgenic and *K14E7* transgenic mice were generated on the same mixed genetic background for all comparison made in this study.

To generate mice inactivated for both pRb and p107, *KRT14-cre/Esr1* (i.e., *K14CreERtm$^{+/+}$* on inbred CD1 genetic background, The Jackson Laboratory) were crossed to *Rb$^{f/f}$p107$^{-/-}$* mice (on a mixed 129/C57 genetic background) and *K14CreERtm$^{+/-}$Rb$^{f/+}$p107$^{+/-}$* offspring then crossed to *Rb$^{f/f}$p107$^{-/-}$* mice to generate *K14CreERtmRb$^{f/f}$p107$^{-/-}$*and *Rb$^{f/f}$p107$^{-/-}$* mice on a CD1/129/C57 mixed genetic background. Control nontransgenic and *K14E7* transgenic mice were generated on the same mixed genetic background for all comparisons made in this study.

To generate mice inactivated for pRb, p107 and p130, *K14CreERtm$^{+/+}$* were crossed to *Rb$^{f/f}$p130$^{f/f}$p107$^{-/-}$* mice (on a mixed 129/C57 genetic background) and *K14CreERtm$^{+/-}$ Rb$^{f/+}$p130$^{f/+}$p107$^{+/-}$* offspring then crossed to *Rb$^{f/f}$p130$^{f/f}$p107$^{-/-}$* mice to generate *K14CreERtmRb$^{f/f}$p130$^{f/f}$p107$^{-/-}$*mice on a CD1/129/C57 mixed genetic background.

Supplemental Table 1

Histopathology summary in cervix for the both Rb and p107 conditionally deficient mice treated with estrogen for 6 months. (Without treatment of TAM)

Genotype[1,2]	Grade of Cervical Disease (# of mice)					Cancer Incidence(%)
	H	CIN1	CIN2	CIN3	CC	
NTG(n=11)	11					0
$K14E7^{WT}$ (n=16)			1	11	4	25
$Rb^{f/f}p107^{-/-}$ (n=20)	15	4	1			0
$K14CreERRb^{f/f}p107^{-/-}$ (n=20)	6	6	2	6		0

1. All mice were on the same CD1/129/C57 mixed genetic background (see material and method section for details on breeding scheme).
2. All of *NTG*, *K14E7*, $Rb^{f/f}p107^{-/-}$, $K14CreERtmRb^{f/f}p107^{-/-}$ mice were not treated with TAM.

Supplemental Table 2

Histopathology summary in vagina for the both pRb and p130 conditionally deficient mice treated with estrogen for 6 months.

Genotype[1]	Grade of Vaginal Disease (# of mice)					Cancer Incidence(%)
	H	VIN1	VIN2	VIN3	VC	
NTG(n=22)	18	3			1	4.50
$K14E7^{WT}$ (n=32)			1	22	9	28.1
$K14CreRb^{f/f}$ (n=42)	33	9				0
$Rb^{f/f}p130^{-/-}$ (n=33)	27	6				0
$K14CreRb^{f/f}p130^{-/-}$ (n=29)	7	8	12	1	1	3.4

1. All mice were on the same FVB/129/C57 mixed genetic background (see material and method section for details on breeding scheme)

Supplemental Table 3

Histopathology summary in vagina for the pRb/p107, pRb/p107/p130conditionally deficient mice treated with estrogen for 6 months.

Genotype[1,2,3]	Grade of Vaginal Disease (# of mice)					Cancer Incidence(%)
	H	VIN1	VIN2	VIN3	VC	
NTG(n=22)	19	3				0
$K14E7^{WT}$ (n=24)			2	14	8	33.3
$Rb^{f/f}p107^{-/-}$ (n=20)	17	3				0
$K14CreERRb^{f/f}p107^{-/-}$ (n=23)		2	5	15	1	4.35
$K14CreERRb^{f/f}p130^{f/f}_{7}p107^{-/-}$ (n=15)			2	12	1	6.67

1. All mice were on the same CD1/129/C57 mixed genetic background (see material and method section for details on breeding scheme)
2. All of *NTG*, *K14E*7, $Rb^{f/f}p107^{-/-}$, $K14CreERtmRb^{f/f}p107^{-/-}$mice were treated with TAM at starting point of this study
3. All of $K14CreERtmRb^{f/f}p130^{f/f}p107^{-/-}$mice were treated with 4-hydrixy tamoxifen at starting point of this study

Supplemental Figure 1

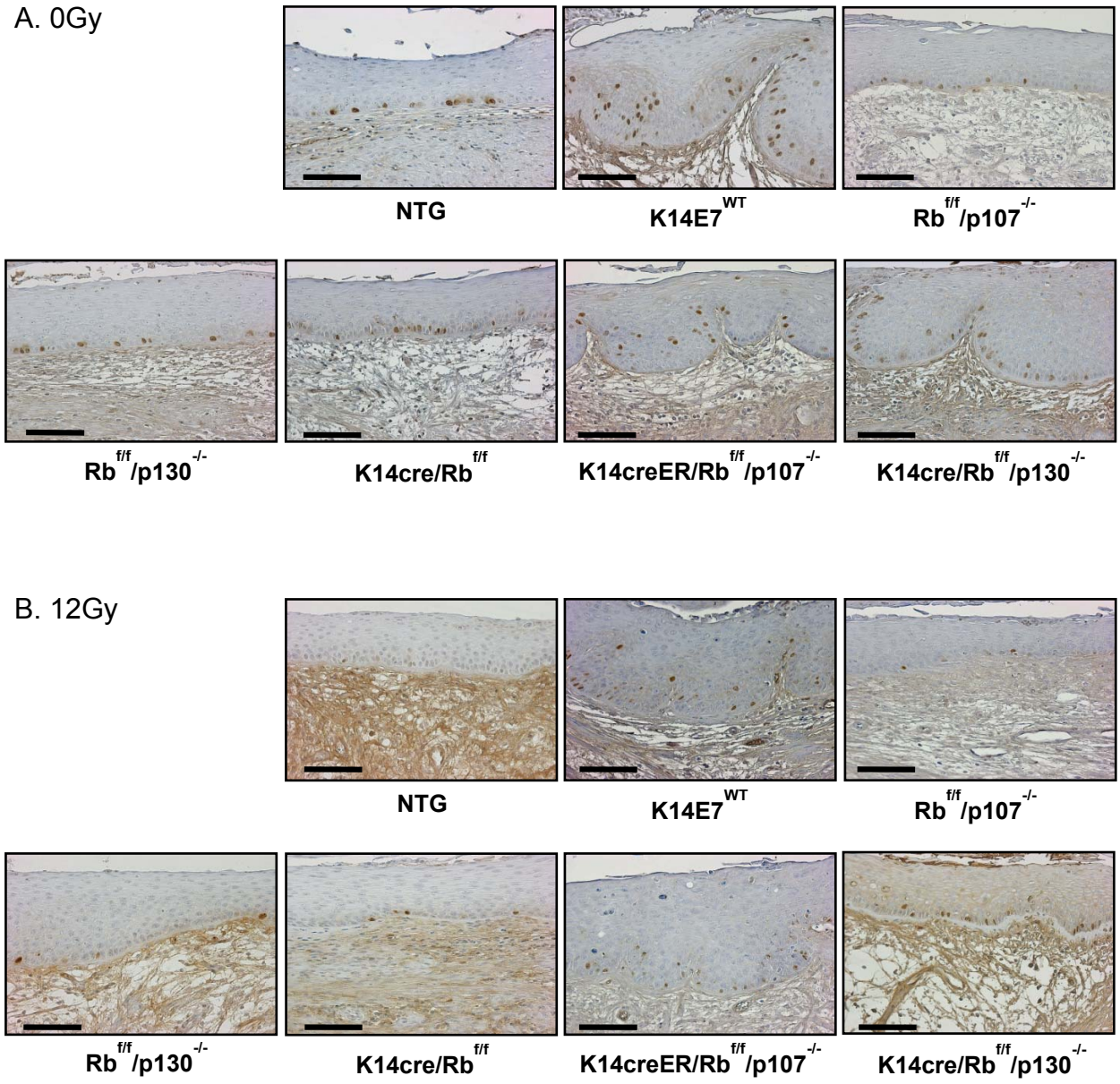

Figure S1. Evaluation of BrdU-incorporation in cervical epithelium of estrogen-treated mice. Representative sections stained with anti-BrdUrd antibody. Brown, positive staining; blue, hematoxylin counterstain. Magnification, x40; scale bar, 200 μm.