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Senescence controls prostatic neoplasia driven by Pten loss

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ABSTRACT

We report that Pten (phosphatase and tensin homologue) ablation in prostatic epithelial cells of adult mice promotes cell proliferation to generate prostatic intraepithelial neoplasia. Moreover, our results demonstrate that proliferating Pten-deficient cells undergo replication stress and exhibit a DNA damage response, leading to cell senescence, as seen in oncogene-induced senescence.

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Prostate cancer, the most common male visceral cancer, is a multistage disease that progresses over decades. About 40% of men over 50 have prostatic intraepithelial neoplasia (PIN), and even though most of PINs will remain indolent, about 1% of the patients will develop clinically significant tumors. If detected when locally confined, prostate cancer is eradicated in 70 – 80% of cases by radical prostatectomy, radiation therapy or cryotherapy. In contrast, no efficient treatments are available for metastatic prostate cancer, which remains the second leading cause of male cancer-related death.¹

The tumor suppressor gene *PTEN* (phosphatase and tensin homologue) is frequently mutated or deleted in prostate cancer. *PTEN* is a lipid phosphatase that antagonizes the production of Phosphatidylinositol (3,4,5)-trisphosphate by phosphoinositide 3-kinase. Its loss leads to a constitutive hyperactivation of the Protein kinase B (AKT)-mammalian target of rapamycin (mTOR) pathway. Genetic ablation of *Pten* in prostatic epithelial cells of mice induces PINs.^{2,3} Progression toward adenocarcinoma has been shown to be restrained by a cell senescence barrier,² but as no hyperproliferation phase, replication stress or DNA damage response were observed in Pten-deficient cells, in contrast to oncogene-induced senescence (OIS), it was concluded that Pten loss-induced senescence (PICS) is a new type of senescence.⁴ It was proposed that interleukin-1 receptor antagonist (Il-1ra) secreted by infiltrated Gr-1 positive myeloid cells promotes the proliferation of some Pten-deficient prostatic epithelial cells, leading to PIN formation.⁵

To further characterize the impact of Pten loss in prostatic epithelial cells of the mature prostate, we analyzed *PTEN*^{(i)pe-/-} mice, in which Pten is selectively ablated in prostatic luminal epithelial cells of mice after puberty.⁶ Our results show that the proliferation of Pten-null prostatic epithelial cells is stimulated within 1 month, reaches a maximum at 3 months,

and is very low after 6 months. Between 5 and 12 months, most of the prostatic epithelial cells in PINs are senescent, produce a number of chemokines and cytokines, and are infiltrated by Gr-1 positive myeloid cells. As senescence markers and Gr-1 positive cells were however only detected only after 2 months, our data demonstrate that Pten loss in prostatic epithelial cells initiates a phase of enhanced proliferation in the absence of GR-1 positive myeloid cells and before senescence entry. In addition, many Pten-null prostatic luminal epithelial cells exhibited phosphohistone H2AX (γ H2AX) nuclear foci, reflecting an activation of a DNA damage response,⁷ and those proliferating presented characteristics of replication stress [e.g. nuclear foci of Rpa32 (Replication Protein A 32), Rpa32 phosphorylated at Serine 4 and Serine 8 and Atr (Ataxia telangiectasia and Rad3-related)]. Thus, Pten loss-induced senescence resembles OIS, and strategies for cancer prevention or therapies based on PICS induction are at risk, as they will promote replication stress that might lead to additional mutations, allowing senescence escape and cancer progression, and induce the secretion of cytokines and chemokines that might contribute to tumor development.

Interestingly, our results show that transformation related protein 53 (Trp53, best known as p53) is stabilized in Pten-deficient prostatic epithelial cells by replication stress-mediated DNA damage response activation. However, at early time after Pten ablation, p53 is down-regulated by mouse double-minute 2 (Mdm2), most probably through Akt-mediated Mdm2 phosphorylation, which increases its E3 ligase activity towards p53. The progressive p53 stabilization likely involves Casein Kinase I (Cki). Indeed, *Cki* transcript levels are induced 3 and 5 months after Pten ablation, and Cki has been shown to be induced by a DNA damage response and to decrease the interaction between Mdm2 and p53 and/or promote Mdm2 degradation.^{8,9} Ablation of p53 in

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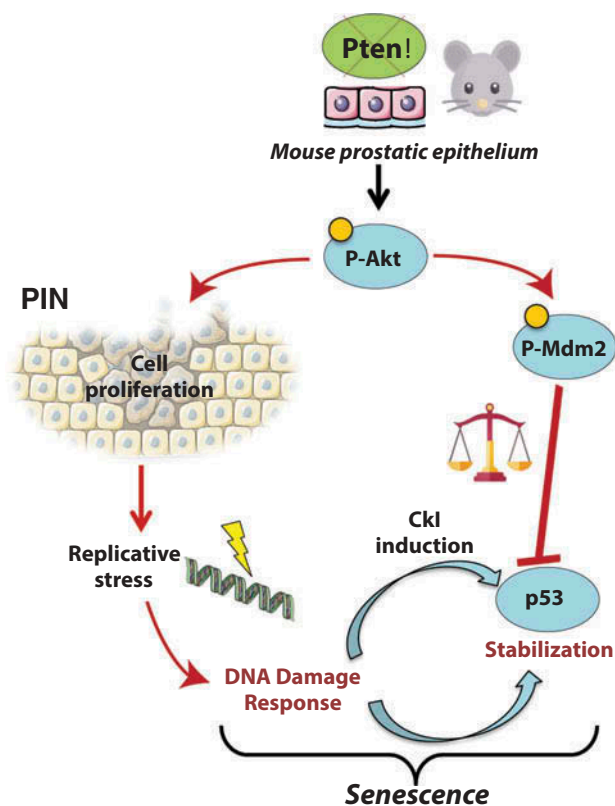


Figure 1. Mechanisms whereby Pten loss drives prostatic intraepithelial neoplasia under the control of cellular senescence.

Ablation of Pten (phosphatase and tensin homologue) in prostatic luminal epithelial cells of adult mice induces the phosphorylation of Protein Kinase B (P-Akt), epithelial cell proliferation and prostatic intraepithelial neoplasia (PIN) formation. Enhanced proliferation promotes replicative stress, a DNA damage response, transformation related protein 53 (p53) stabilisation and epithelial cell senescence. At early time after Pten ablation, Mdm2 (mouse double-minute 2) is phosphorylated (P-Mdm2), most probably by P-Akt, and p53 is downregulated, until DNA damage response-induced Casein Kinase I (Ckl) levels are sufficient to promote Mdm2 degradation and/or impair the interaction between Mdm2 and p53.

Pten-deficient prostatic epithelial cells allows to bypass/avoid the senescence barrier, accelerates the development of adenocarcinoma and promotes the emergence of metastatic sarcomatoid tumors⁶ (and unpublished data).

Our data highlight the importance of analyzing relevant mouse models. Obviously, the possibility to control the time of gene ablation by induction of the CreER^{T2} recombinase activity in luminal epithelial cells of the mature prostate, is a major advantage compared to other models, where target genes are ablated in prostatic epithelial cells before puberty. Moreover, as cell culture conditions imply high levels of growth factors and high partial O₂ pressure, stressors that might facilitate cellular senescence induction,¹⁰ *in vitro* data on senescence should be extrapolated to *in vivo* contexts with care.

Taken together, our study provides important insights into the latency phase of prostate cancer. Indeed, our results show that Pten loss in prostatic epithelial cells induces replication stress-mediated activation of a DNA damage response and progressive cell senescence. Akt activation after Pten ablation is likely to promote Mdm2-mediated p53 degradation and delays

senescence until Ckl levels are sufficient to allow p53 stabilization (Figure 1). Even though selective inhibitors of Akt or of Mdm2-p53 interaction might enhance senescence and thus increase the latency phase, therapeutic approaches allowing to eradicate PTEN-deficient senescent prostatic epithelial cells will probably be required to prevent prostate cancer progression.

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Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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References

- Hodson R. Small organ, big reach. *Nature*. 2015;528:S118–9. doi:10.1038/528S118a.
- Chen Z, Trotman LC, Shaffer D, Lin HK, Dotan ZA, Niki M, Koutcher JA, Scher HI, Ludwig T, Gerald W, et al. Crucial role of p53-dependent cellular senescence in suppression of Pten-deficient tumorigenesis. *Nature*. 2005;436:725–730. doi:10.1038/nature03918.
- Ratnacaram CK, Teletin M, Jiang M, Meng X, Chambon P, Metzger D. Temporally controlled ablation of PTEN in adult mouse prostate epithelium generates a model of invasive prostatic adenocarcinoma. *Proc Natl Acad Sci U S A*. 2008;105:2521–2526. doi:10.1073/pnas.0712021105.
- Alimonti A, Nardella C, Chen Z, Clohessy JG, Carracedo A, Trotman LC, Cheng K, Varmeh S, Kozma SC, Thomas G, et al. A novel type of cellular senescence that can be enhanced in mouse models and human tumor xenografts to suppress prostate tumorigenesis. *J Clin Invest*. 2010;120:681–693. doi:10.1172/JCI40535.

- 155 5. Di Mitri D, Toso A, Chen JJ, Sarti M, Pinton S, Jost TR, D'Antuono R, Montani E, Garcia-Escudero R, Guccini I, et al. Tumour-infiltrating Gr-1+ myeloid cells antagonize senescence in cancer. *Nature*. 2014;515:134–137. doi:10.1038/nature13638.
6. Parisotto M, Grelet E, El Bizri R, Dai Y, Terzic J, Eckert D, Gargowitsch L, Bornert JM, Metzger D. PTEN deletion in luminal cells of mature prostate induces replication stress and senescence in vivo. *J Exp Med*. 2018;215:1749–1763. doi:10.1084/jem.20171207.
- 160 7. Mah LJ, El-Osta A, Karagiannis TC. gammaH2AX: a sensitive molecular marker of DNA damage and repair. *Leukemia*. 2010;24:679–686. doi:10.1038/leu.2010.6.
8. Knippschild U, Kruger M, Richter J, Xu P, Garcia-Reyes B, Peifer C, Halekotte J, Bakulev V, Bischof J. The CK1 family: contribution to cellular stress response and its role in carcinogenesis. *Front Oncol*. 2014;4:96. doi:10.3389/fonc.2014.00096.
9. Inuzuka H, Tseng A, Gao D, Zhai B, Zhang Q, Shaik S, Wan L, Ang XL, Mock C, Yin H, et al. Phosphorylation by casein kinase I promotes the turnover of the Mdm2 oncoprotein via the SCF (beta-TRCP) ubiquitin ligase. *Cancer Cell*. 2010;18:147–159. doi:10.1016/j.ccr.2010.06.015.
- 170 10. Serrano M, Blasco MA. Putting the stress on senescence. *Curr Opin Cell Biol*. 13;2001:748–753.

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