Stem Cell Reports
Report
OPEN ACCESS

Basal Progenitors Contribute to Repair of the Prostate Epithelium Following Induced Luminal Anoikis

Roxanne Toivanen,[1,2] Adithi Mohan,[1,2] and Michael M. Shen[1,*]
[1]Departments of Medicine, Genetics & Development, Urology, and Systems Biology, Herbert Irving Comprehensive Cancer Center, Columbia University Medical Center, New York, NY 10032, USA
[2]Co-first author
*Correspondence: mshen@columbia.edu
http://dx.doi.org/10.1016/j.stemcr.2016.03.007

SUMMARY

Contact with the extracellular matrix is essential for maintenance of epithelial cells in many tissues, while in its absence epithelial cells can detach and undergo anoikis. Here, we show that anoikis of luminal cells in the prostate epithelium is followed by a program of tissue repair that is mediated in part by differentiation of basal epithelial cells to luminal cells. We describe a mouse model in which inducible deletion of E-cadherin in prostate luminal cells results in their apoptotic cell death by anoikis, in the absence of phenotypic effects in the surrounding stroma. Quantitative assessments of proliferation and cell death in the luminal and basal compartments indicate that basal cells can rapidly generate luminal cells. Thus, our findings identify a role for basal-to-luminal differentiation in prostate epithelial repair, and provide a normal context to analogous processes that may occur during prostate cancer initiation.

INTRODUCTION

Epithelial integrity and tissue homeostasis are severely challenged by wounding or a range of pathological states. Under such conditions, epithelial cells can detach from the underlying basement membrane and undergo apoptotic cell death through a process known as anoikis (Chiarugi and Giannoni, 2008; Frisch and Francis, 1994; Frisch and Screaton, 2001; Taddei et al., 2012). Subsequently, the missing epithelial cells can be replaced through the activity of endogenous stem/progenitor cells. In the prostate, insults such as bacterial or viral infection can result in inflammation and epithelial cell death, but the process of repair has been poorly studied to date.

The identity of epithelial stem/progenitor cells within the prostate remains a subject of intense study (Shibata and Shen, 2015). The prostate epithelium comprises luminal, basal, and neuroendocrine cells, with both luminal and basal compartments containing stem/progenitor activity (Shen and Abate-Shen, 2010). In addition, rare "intermediate" cells that co-express basal and luminal markers have been proposed to correspond to stem/progenitor cells (De Marzo et al., 1998; Verhagen et al., 1988; Wang et al., 2001; Xue et al., 1998), or a transitional state between basal progenitors and luminal descendants (Bonkhoff and Remberger, 1996; Litvinov et al., 2006; Ousset et al., 2012; van Leenders et al., 2000).

Stem/progenitor activity has been observed in distinct contexts during prostate development and homeostasis in vivo. In the hormonally naive adult prostate epithelium, luminal and basal compartments are maintained by unipotent progenitors (Choi et al., 2012; Lu et al., 2013; Wang et al., 2013), while during prostate organogenesis some basal progenitors are multipotent, giving rise to luminal and neuroendocrine progeny (Ousset et al., 2012; Wang et al., 2014a). In addition, rare bipotential populations exist within both the luminal and basal compartments during androgen-mediated regeneration of the regressed prostate (Lee et al., 2014; Wang et al., 2009, 2013, 2015).

In contrast, both luminal and basal populations display considerable lineage plasticity in specific contexts. Explanted luminal cells can generate basal cells in organoid culture (Chua et al., 2014; Karthaus et al., 2014), whereas basal cells can generate luminal cells in sphere formation assays, and after recombination with embryonic urogenital mesenchyme in renal grafts (Burger et al., 2005; Goldstein et al., 2008, 2010; Hofner et al., 2015; Lawson et al., 2007; Richardson et al., 2004; Wang et al., 2013). Basal-to-luminal differentiation can also occur in pathological contexts, such as during prostate cancer initiation (Choi et al., 2012; Lu et al., 2013; Wang et al., 2013, 2014b), and after acute inflammation in bacterial prostatitis (Kwon et al., 2014b).

Taken together, these findings indicate that basal-to-luminal differentiation can occur in prostate organogenesis, pathogenesis, and ex vivo assays, but rarely during normal tissue homeostasis. Thus, it has been unclear to what extent the plasticity of endogenous adult prostate basal cells in ex vivo models and disease states reflects an in vivo activity. In this study, we introduce a mouse model in which a tamoxifen-inducible Cre driver is used to delete E-cadherin in prostatic luminal cells, which are highly

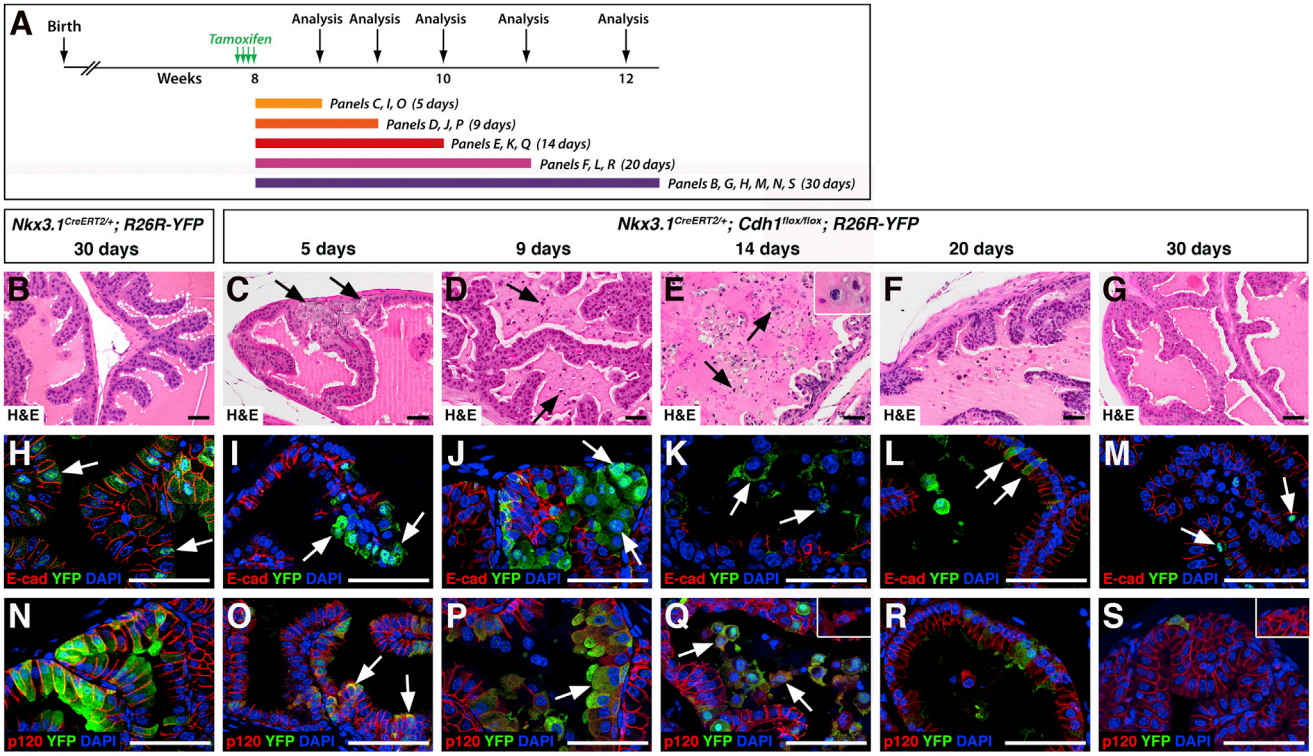

Figure 1. Inducible Deletion of E-Cadherin in the Prostate Epithelium

(A) Schematic timeline of the experiment.

(B–G) H&E staining of histological sections from the anterior prostate. Arrows in (C) show patches of atypical cells, and arrows in (D), (E), and inset in (E) show cells sloughing into the lumen in *Cdh1*^{del} prostates.

(H–M) Immunofluorescence staining for E-cadherin and YFP. Arrows in (H) show intact E-cadherin expression in control mice; arrows in (I), (J), (K), and (M) show E-cadherin loss in YFP^{+} cells of *Cdh1*^{del} prostates; and arrows in (L) indicate rare YFP^{+} cells in which E-cadherin was not deleted.

(N–S) Immunofluorescence staining for p120 catenin and YFP. Arrows in (O), (P), and (Q) show cytoplasmic p120 staining in YFP^{+} cells of *Cdh1*^{del} prostates.

Numbers of mice examined: n = 7 for (B), (E), (H), (K), (N), and (Q); n = 5 for (C), (I), and (O); n = 4 for (D), (F), (G), (J), (L), (M), (P), (R), and (S). Scale bars, 50 μm. See also Figures S1 and S2; Tables S1 and S2.

susceptible to anoikis (Kwon et al., 2014a). This results in rapid sloughing and death of luminal cells, followed by repair of the damaged epithelium. We show that basal-to-luminal differentiation contributes to tissue repair, providing a new approach for studying prostate stem/progenitor activity and epithelial specification.

RESULTS

E-Cadherin Is Essential for Maintenance of Prostate Luminal Cells

To delete *E-cadherin* (*Cdh1*) in the prostate epithelium, we used mice carrying the tamoxifen-inducible *Nkx3.1*^{CreERT2} driver (Wang et al., 2009), the conditional *Cdh1*^{flox} allele (Boussadia et al., 2002), and an *R26R-YFP* reporter (Srinivas et al., 2001). We administered tamoxifen to *Nkx3.1*$^{CreERT2/+\ or\ CreERT2/CreERT2}$; *Cdh1*$^{flox/flox}$; *R26R-YFP* (designated *Cdh1*^{del}) mice at 8 weeks of age, and analyzed prostate tissues at 5, 9, 14, 20, and 30 days thereafter (Figure 1A). In parallel, control *Nkx3.1*$^{CreERT2/+}$; *R26R-YFP* mice were treated with tamoxifen and analyzed at 5, 14, and 30 days later.

Histological analyses of the anterior prostate lobes showed that controls maintained a normal phenotype after tamoxifen treatment (Figures 1B, S1A, and S1B), whereas *Cdh1*^{del} prostates underwent epithelial damage and repair. At 5 days after tamoxifen administration, we observed foci of atypical cells in *Cdh1*^{del} prostates (Figure 1C), and at 9 and 14 days after tamoxifen treatment, many cells had detached from the epithelium and were present in the lumen (Figures 1D and 1E). However, by 20 days after treatment, fewer detached cells were observed, and at 30 days the normal epithelial phenotype was restored