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Timing is everything
Rb's choice in islet-cell fate

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Retinoblastoma tumor suppressor (Rb) is best known for its role as a negative regulator of cell cycle entry through inhibition of E2f transcription factors and their target genes. As such, Rb loss promotes tumorigenesis and is a hallmark of human cancer.[1] However, Rb also regulates other cellular processes, including differentiation, apoptosis, and autophagy. The specific consequence of Rb loss is context-specific. In the pancreas, deletion of Rb alone in differentiated β-cells via an insulin promoter-Cre transgene does not cause cell cycle re-entry or any discernible effect.[2] We have reported, in a recent issue of *PNAS*, that deletion of Rb in proliferating pancreatic progenitors via Pdx1-Cre alters α- and β-cell specification.[3] Altered specification is caused by differential effects of Rb on proliferation and survival of α- and β-cells, leading to increased β- to α-cell ratio and resistance to experimentally induced diabetes.

Pancreatic islets control glucose homeostasis by tightly regulated secretion of insulin (β-cells) and glucagon (α-cells). Loss of β-cell mass and function underlies both type 1 and type 2 diabetes. There is therefore a great interest in how to boost β-cell mass as a possible therapeutic strategy for diabetes. Pancreatic insulin-secreting cells are mostly post-mitotic in a quiescent/G_0/G_1 state and regenerate through duplication of pre-existing β-cells.[4] Thus, Rb is a logical target for switching on cell cycle re-entry and regenerating islet mass.

As noted, deletion of Rb in matured β-cells had no discernible effect.[2] However, previous studies showed that Rb is required during the transition of cells from proliferative to differentiated states, but is dispensable once cells exit the cell cycle and differentiate.[5] We therefore used a Pdx1-Cre deleter line to disrupt Rb in proliferating pancreatic progenitors (Pdx1-Cre:$Rb^{fl/fl}$ mice). This has resulted in a dramatic increase in multipotent neurogenin 3 (Ngn3)-expressing cells at embryonic day 16.5, which persisted into adulthood.[3] The expanded Rb-deficient $Ngn3^+$ islet precursors likely differentiated into mature β-cells, leading to the observed increase in adult β-cell mass in Pdx1-Cre:$Rb^{fl/fl}$ mice. In sharp contrast, Ngn3 was undetectable in islets of wild-type littermates by 4 wk of age.

In addition to induction of β-cell expansion, disruption of Rb in pancreatic progenitors led to concomitant reduction in α-cell number. We showed that Rb-E2f1 bound exon 2 of the α-cell differentiation factor, aristaless related homeobox (Arx), and induced its expression. In the absence of Rb, E2f1 suppressed *Arx* gene expression, leading to inhibition of α-cell differentiation during embryogenesis. Rb-deficient α-cells also showed increased apoptosis through upregulation of p53. Consequently, loss of Rb in proliferating islet precursors resulted in increased β- to α-cell ratio and improved glucose homeostasis and resistance to streptozotocin (STZ)-induced β-cell apoptosis and diabetes (**Fig. 1**).

Our results demonstrate a contrasting role for Rb in proliferating pancreatic progenitors vs. differentiated β-cells. Moreover, our results suggest that Rb controls α- vs. β-cell specification through 3 distinct mechanisms: expansion of progenitor stem cells, survival (through differential regulation of p53), and differentiation (through upregulation of Arx in α-cells). Previous work has demonstrated a role for Rb in cell fate specification in other tissues. For example, inactivation of Rb in mesenchymal stem cells results in increased differentiation into brown adipose tissue at the expense of bone differentiation.[6] In principle, the effect of Rb on stem cell expansion and survival suffice to control lineage determination. Whether Rb is also actively required for differentiation is a contentious issue. Indeed, it was shown that various survival factors such as Bcl-2 could compensate for Rb deficiency and promote terminal muscle differentiation in vitro.[7]

Importantly, while Rb loss in most tissues analyzed so far has resulted in deleterious consequences, our work shows that in pancreatic progenitors, Rb deficiency increases β- to α-cell ratio and improves islet function. The challenge ahead will be to translate these findings into novel therapeutic avenues. In this regard, recent demonstrations that transient Rb loss can induce cell cycle re-entry followed by regeneration,[8] and that Rb plays a potential role in stem cell expansion,[1] combined with our results, suggest that one approach may involve therapeutic reprogramming of pre-existing β-cells to pancreatic progenitors followed by spontaneous re-differentiation into mature, insulin-secreting cells.

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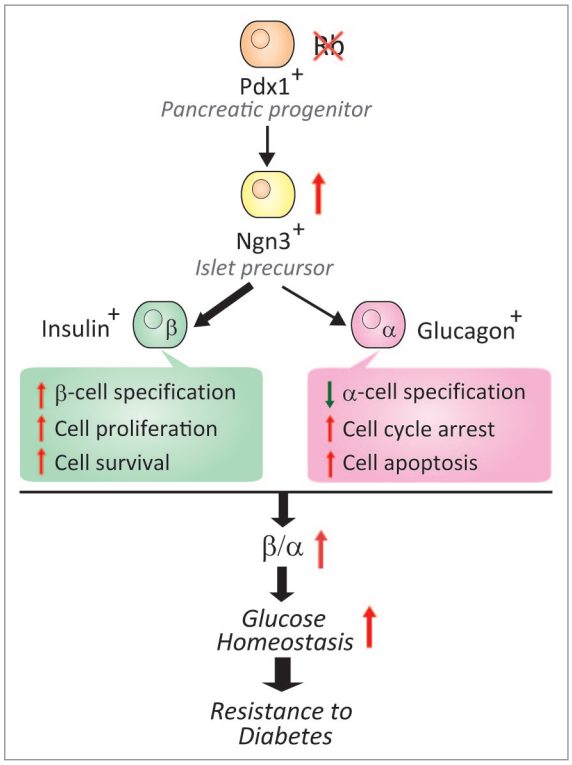

Figure 1. Mechanisms of Rb in regulating α- and β-cell fate. Disruption of Rb in pancreatic progenitors (Pdx1$^+$ cells) led to increased islet precursors (Ngn3$^+$ cells) during embryogenesis with enhanced β-cell specification and postnatal proliferation. In contrast, α-cell differentiation was suppressed along with increased apoptosis, leading to a persistently increased β/α-cell ratio that contributed to improved glucose homeostasis and protection against diabetes.

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