CANCER

Oncogenic PI3Kα promotes multipotency in breast epithelial cells

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The phosphoinositide 3-kinase (PI3K) signaling pathway is among the most frequently altered in cancer. Now, two studies show that a mutated oncogenic PI3Kα, commonly found in breast cancer, leads to dedifferentiation or destabilization of luminal and basal epithelial lineages, which in turn leads to increased cancer cell heterogeneity.

Cancer is resilient, often persisting despite extensive immune recognition and our best attempts at curative therapy. A potential explanation for such resilience is development of tumor heterogeneity. Heterogeneity allows cancers to develop functionally specialized cells—for example, those with quiescent properties that are less susceptible to chemotherapy or those with metastatic potential enabling distant spread. Tumor heterogeneity develops despite the single-cell origin of tumors. A gene-centric explanation for this phenomenon is that selection of genetic mutations within tumors causes Darwinian-like clonal evolution, creating subpopulations of tumor cells with distinct phenotypes and mechanisms of evasion from host factors or therapy. Indeed, substantial genetic heterogeneity within individual cancers has been detected through high-resolution genome sequencing approaches and likely drives phenotypic heterogeneity (*1*). However, the observation within tumors of cells with defining characteristics of more than one distinct cellular lineage or state of differentiation has brought nongenetic sources of phenotypic heterogeneity into focus. A dominant framework for understanding nongenetic sources of tumor heterogeneity is the cancer stem cell hypothesis (*2*). According to this hypothesis, cancers establish cellular differentiation hierarchies similar to those found in normal tissues, wherein multipotent stem cells self-renew and produce lineage-restricted progeny.

Although genetic and nongenetic drivers of tumor heterogeneity are not mutually exclusive, few mechanistic links relating genetic alterations and nongenetic drivers of heterogeneity have been proposed. Two papers demonstrate that activating genetic mutations in the *PIK3CA* gene, which encodes the p110α catalytic subunit of phosphoinositide 3-kinase (PI3K), causes loss of lineage restriction and generate tumor heterogeneity from basal and luminal cells of the adult mouse mammary gland (*3*, *4*). PI3Kα is a lipid kinase that generates the second messenger molecule phosphatidylinositol 3,4,5-trisphosphate (PIP_3), which acts as a membrane tether for protein kinases such as phosphoinositide-dependent kinase 1 (PDK1) and AKT. These kinases in turn contribute to activation of the kinase mTOR (mechanistic target of rapamycin) and downstream ribosomal biogenesis and transcriptional pathways, as well as inhibition of transcription factors of the FOXO family. The PI3K pathway has emerged as one of the most frequently activated signaling pathways in cancer (*5*). In breast cancer, *PIK3CA* is mutated in up to 45% of cases, depending on the subtype (*6*). The mutations in *PIK3CA*, which are mostly found at two hotspot locations in the gene, lead to increased kinase activity and are particularly prevalent in the luminal estrogen receptor–positive subtype of breast cancer (*6*).

Classification of breast cancers is based on the assumed cell of origin. These include luminal epithelial cells, which line the milk ducts and are responsive to progesterone or estrogen or both hormones, and basal epithelial cells, which face away from the lumen and tend to be nonresponsive sex hormones. In situ lineage tracking in mice shows that cells of one lineage rarely, if ever, switch to the other lineage, indicating lineage restriction of the two cell types in the postnatal mammary gland (Fig. 1) (*7*). Koren *et al*. and Van Keymeulen *et al*. used inducible Cre recombinase transgenes driven by the *Lgr* or *K5* promoters (expressed by basal epithelial cells) or the *K8* promoter (expressed by luminal epithelial cells) to drive expression of fluorescent fate-tracking reporter alleles (*3*, *4*). In the absence of any further manipulation, the luminal cells did not give rise to basal cells, or vice versa. However, when the activating mutant $p110\alpha^{H1047R}$ was induced in luminal epithelial cells, progeny cells with the transcriptional features and marker expression of basal epithelial cells were detected. Similarly, introduction of $p110\alpha^{H1047R}$ in basal epithelial cells gave rise to luminal epithelial progeny (Fig. 1). Hence, a genetic mutation causing hyperactivation of the PI3K pathway drives mature breast epithelial cells to lose lineage restriction, a process that results in tumor heterogeneity. Although the spectra of tumors induced from luminal and basal precursors were different, both sets were characterized by considerable heterogeneity and overlapping characteristics.

One interpretation of these results is that activated PI3Kα stimulates dedifferentiation of luminal or basal epithelial cells to a common progenitor, which then gives rise to either lineage. If so, a major question is whether such progenitor cells are induced transiently, or whether they establish themselves as self-replicating stem cells that would be capable of seeding tumors with differentiated epithelial cells in the long term. Another possibility is that activated PI3Kα causes direct transdifferentiation of basal to luminal epithelial cells and vice versa. If the former is the case, it is intriguing that activation of the PI3K pathway, which is a potent driver of cellular metabolism, results in acquisition of stem cell–like characteristics, given that stem cells are generally considered to be metabolically quiescent. Moreover, the results evoke questions about the genetic and epigenetic factors that maintain stable lineage identity under physiological conditions and how these are affected by the PI3K signaling pathway.

Not only are the phenotypic characteristics of the primary tumor important for treating the primary tumor, but they also factor into the prognosis and potential for metastasis. Lawson *et al*. demonstrated that early metastases were generally formed by epithelial cells with stem cell–like characteristics, which subsequently acquired more differentiated characteristics (*8*). It will therefore be of interest to determine whether oncogenic PI3Kα contributes to the produc-

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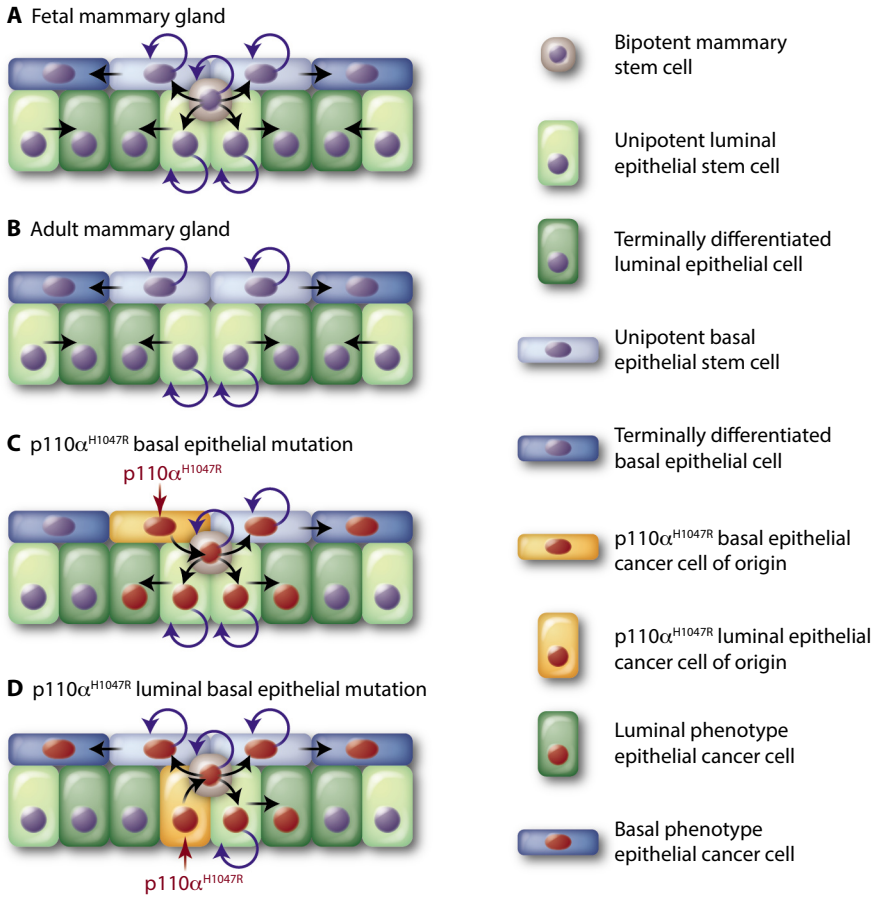

Fig. 1. Lineage relationship of epithelial cells in the fetal, adult, and transformed breast. (**A**) In the fetal gland, mammary stem cells give rise to unipotent self-renewing basal and luminal epithelial stem cells, which in turn generate terminally differentiated basal and luminal epithelial cells, respectively. (**B**) In the adult gland, unipotent self-renewing basal and luminal epithelial stem cells generate terminally differentiated basal and luminal epithelial cells in a lineage-restricted fashion. (**C**) An oncogenic p110α^{H1047R} mutation arising in a basal epithelial cell drives loss of lineage restriction, resulting in a putative multipotent precursor cell, which gives rise to both cancer cells with basal or epithelial characteristics. (**D**) An oncogenic p110α^{H1047R} mutation arising in a luminal epithelial cell drives loss of lineage restriction, resulting in a putative multipotent precursor cell, which gives rise to both cancer cells with basal or epithelial characteristics.

tion of metastatic clones by inducing the dedifferentiation of committed epithelial cells to those with stem cell–like properties. If so, this may suggest that early intervention with PI3Kα inhibitors is warranted to help prevent metastatic disease.

Collectively, the papers by Koren *et al*. and Van Keymeulen *et al*. provide a link between activating mutations in the PI3K pathway and induction of multipotency and tumor heterogeneity in breast cancer. The data also indicate a powerful effect of the lineage of the cancer cell of origin in determining severity of disease and the inadequacy of basal and luminal markers in identifying this cell of origin.

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