

## Spotlight

Interplay between  
hormonal and  
mechanical signals in  
mammary  
morphodynamicsAlişya A. Anlaş<sup>1</sup> and  
Celeste M. Nelson <sup>2,3,4,\*</sup> 

**Mammographic density is a well-established risk factor for breast cancer. In a recent study, Northey *et al.* reveal that the associated increase in tissue stiffness elevates extracellular signal-regulated kinase (ERK) activity, promoting progesterone receptor-dependent receptor activator of nuclear factor  $\kappa$ B (RANK) signaling. Thus, stiffness alters the context of hormonal signaling and increases mammary stem cells. This mechanism suggests potential treatments for breast cancer.**

The epithelium of the mammary gland has a remarkable capacity for remodeling in response to signals from its microenvironment [1]. Over the lifetime of a mammal, the gland undergoes recursive cycles of pregnancy, lactation, and involution, all of which require significant and repeated tissue remodeling. These unique tissue morphodynamics are dictated by interactions between the cell types that comprise the gland and their surrounding microenvironment [2]. The biochemical signals that regulate morphogenesis and remodeling of the mammary gland are well characterized and occur in the context of a less well-explored mechanical microenvironment. Although recent studies using engineered tissue models have highlighted the impact of mechanical signals on mammary epithelial cells, specifically in the context of breast cancer, those that regulate the

morphodynamics of the intact gland have remained enigmatic.

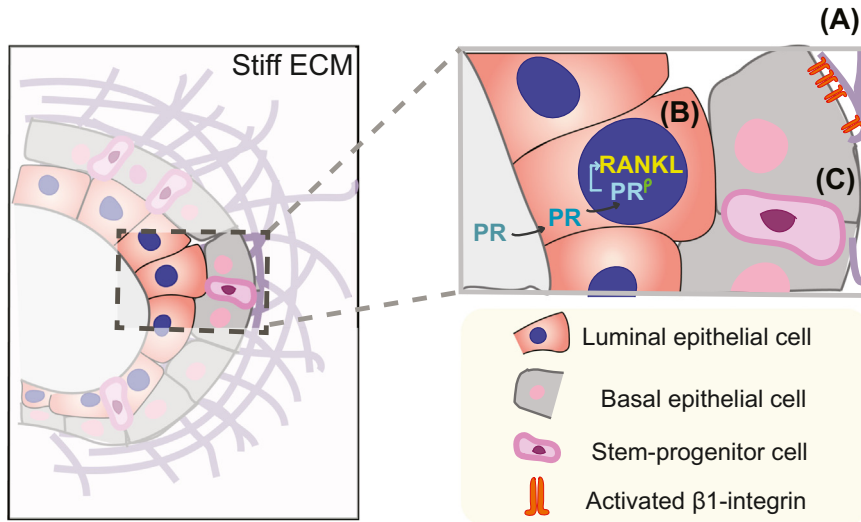
A recent study by Northey *et al.* reveals that tissue stiffness correlates with mammographic density, which has long been associated with an elevated risk of breast cancer [3–5]. The authors combined mouse and organoid models to show that altered tissue mechanics promotes hormonal signaling to expand a specific population of stem cells that aid tumor initiation [4]. These mammary stem and progenitor cells are crucial for morphogenesis of the mammary gland during puberty, as well as expansion of the gland during pregnancy, and have been implicated in breast cancer [6]. Their role is context dependent, because stem and progenitor cells respond to the interplay between tissue stiffness and hormonal signaling. These interactions, some of which were studied by Northey *et al.*, provide valuable insights into the complex biochemical and mechanical factors that promote breast cancer initiation, highlighting the importance of considering both in understanding complex tissue dynamics and disease pathogenesis. This understanding, in turn, paves the way for more holistic breast cancer treatments.

Dynamic reciprocity in the mammary gland involves bidirectional interactions between epithelial and stromal cells and the extracellular matrix (ECM). Cellular behavior influences ECM remodeling and, in turn, ECM composition regulates cell proliferation, differentiation, and migration [7,8]. Increased epithelial content and ECM density, particularly elevated collagen, leads to higher mammographic density [9]. The authors demonstrated that collagen density correlates with tissue stiffness and revealed that increased tissue stiffness correlates with activation of  $\beta$ 1-integrin in tissues with high mammographic density. Increased stiffness also correlates with the expansion of the basal mammary epithelium and enhanced stem progenitor

population. Moreover, the authors used a genetically engineered mouse model, in which the mammary epithelium conditionally expresses a mutant human  $\beta$ 1-integrin, to demonstrate that increased mechanical signaling through  $\beta$ 1-integrin expands the basal epithelial compartment. These findings align with the well-established observation that  $\beta$ 1-integrin regulates the architecture of the mammary epithelium [10].

Hormonal signaling, particularly through progesterone, orchestrates dynamic remodeling of the gland to expand the population of epithelial and stem cells [11]. The authors' findings thus far implicate a role for tissue stiffness in regulating the relative abundance of different cell types within the gland. However, this observation alone does not explain the correlation between high mammographic density and increased stem progenitor cells. Using transgenic mice that express mutant  $\beta$ 1-integrin or a collagenase-resistant form of collagen I that increases collagen density (Col1a1Tm1Jae), the authors showed that mice with enhanced mechanical signaling through  $\beta$ 1-integrin or elevated collagen density in the mammary gland exhibit increased RANK activity. Importantly, Northey *et al.* demonstrated that enhanced mechanical signaling through  $\beta$ 1-integrin drives epidermal growth factor receptor (EGFR)-dependent ERK activity (Figure 1A), increases progesterone-induced RANK signaling (Figure 1B), and expands the stem progenitor population in the mammary gland (Figure 1C). Altogether, these findings align with clinical observations that tissues with high mammographic density harbor a larger population of stem progenitor cells, providing the first substantial link between high mammographic density in clinical samples and the increased stem progenitor population associated with breast cancer.

These findings, that tissue stiffness induces expansion of stem progenitor cells in the mammary gland through progesterone and



**Figure 1. Mechanical regulation of hormonal signaling.** Stiffening of the mammary gland increases (A) mechanical signaling through  $\beta$ 1-integrin. (B) Progesterone (Pr)-mediated receptor activator of nuclear factor  $\kappa$ B (RANK) signaling is mechanosensitive and (C) enhances the stem progenitor cell population. Abbreviation: ECM, extracellular matrix.

RANK signaling, prompt the question of whether increased stiffness in early or premalignant lesions could amplify mechanical signaling through  $\beta$ 1-integrin, thereby enhancing progesterone-induced RANK signaling. To answer this question, the authors embedded cancer cells into soft or stiff hydrogels implanted in the mouse mammary fat pad and found that cancer cells cultured in stiff hydrogels exhibit a higher metastatic potential. This observation highlights bidirectional crosstalk within the mammary gland: elevated tissue stiffness increases signaling through  $\beta$ 1-integrin, which enhances the metastatic potential of cancer cells.

The molecular mechanism identified by Northey *et al.* is an elegant example of biochemical and mechanical factors working in concert during tissue morphogenesis. The study also suggests that alterations in the mechanical microenvironment, such as those that lead to highly fibrotic breast cancer, might exhibit elevated hormonal signaling through RANK, which could be targeted for treatment.

Although the current study focuses on the implications of stiffness in breast cancer, alterations in hormonal signaling can cause a variety of morphological and mechanical changes in the mammary gland. For instance, hormones used in hormone-replacement therapy, such as estrogen and progesterone, can stimulate the growth of glandular and fibrous tissue in the breast, leading to an increase in mammographic density. Progesterone treatment has been causally linked to breast cancer [12]; however, the mechanisms underlying this association remain unclear and could result from a similar effect to that reported by the current study. Likewise, remodeling of the gland during lactation and involution involves the rapid expansion and differentiation of mammary epithelial cells that are responsible for milk production, followed by regression of glandular tissue. Stem and progenitor cell populations within the mammary gland exhibit dynamic changes during these processes [6], and their abundance is likely to be impacted by the corresponding changes in mechanical properties throughout the gland.

In addition to remodeling of the mammary gland, the female body also experiences cyclical, hormone-mediated changes in the endometrium. This tissue undergoes rapid remodeling throughout the menstrual cycle due to fluctuations in the levels of estrogen and progesterone, which alter its thickness, stiffness, and ability to support pregnancy. How an imbalance of biochemical and mechanical signals that maintain the endometrium could lead to malignancy are not yet understood. However, a thorough analysis, similar to that presented in the current study, would shed light on this issue.

#### Declaration of interests

The authors declare no competing interests.

<sup>1</sup>Department of Chemical & Biomolecular Engineering, University of Pennsylvania, Philadelphia, PA 19103, USA

<sup>2</sup>Department of Chemical & Biological Engineering, Princeton University, Princeton, NJ 08544, USA

<sup>3</sup>Department of Molecular Biology, Princeton University, Princeton, NJ 08544, USA

<sup>4</sup>Website: <https://cmngroup.princeton.edu>

\*Correspondence:

[celesten@princeton.edu](mailto:celesten@princeton.edu) (C.M. Nelson).

@Twitter: @celestemnelson (C.M. Nelson).

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