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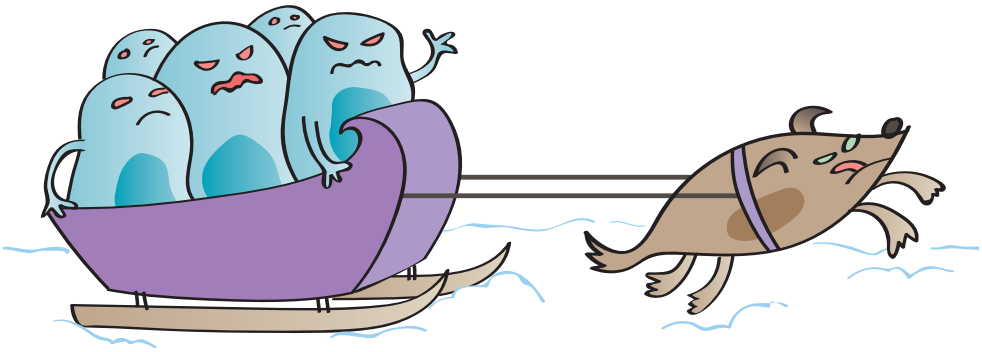

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CELL MIGRATION

Shall we travel together?

> E-cadherin–N-cadherin junctions are mechanically active during CAF-led collective A431 cell migration

Cancer cells have been shown to invade away from the tumour mass either as a single cell or as a collective. The latter of these two invasion strategies is cooperative and can be mediated by stromal cells, such as cancer-associated fibroblasts (CAFs), which remodel the extracellular matrix (ECM) to form tracks through which cancer cells can migrate. This observation has led to the assumption that cancer cell invasion is the outcome of following the path of least resistance. However, Labernadie *et al*. now suggest that the mechanical coupling between CAFs and cancer cells can explain the collective spread of cancer cells.

To explore the CAF-driven collective invasion of cancer cells, the authors used an *in vitro* 3D spheroid assay with a mixture of A431 cancer cells and CAFs, both derived from human vulval squamous cell carcinoma (SCC), embedded within an ECM. Imaging the spheroid over time revealed that by 60 hours, collective strands of cells typically comprising one leading CAF followed by several A431 cells were invading away from the spheroid. To determine whether this pattern of leader CAFs–follower A431 cells was established through signals derived from the surrounding ECM or via an intercellular interaction, a 2D assay was set up in which spheroids composed only of A431 cells were allowed to attach to soft polyacrylamide gels with CAFs subsequently positioned randomly on the substrate. This revealed that independent of a 3D microenvironment, CAFs could contact the spheroid, invert their front to rear polarity and then migrate away followed by A431 cells; a mechanism that confers dispersion away from the tumour mass, leading to movement into the surrounding tissue.

Labernadie *et al*. demonstrated that pulling forces were exerted on cancer cells from the leading edge of CAFs and further analyses showed that there were multiple contacts between the two cell types. These contact zones involved the generation of heterophilic adherens junctions composed of E-cadherin from A431 cells and N-cadherin from CAFs. Knockout of E-cadherin in A431 cells or knockdown of N-cadherin in CAFs was able to reduce the number of leader CAFs in 2D assays and CAF-led invasive A431 strands in 3D assays, indicating that the E-cadherin–N-cadherin junctions are necessary for the guidance of A431 cells by CAFs. Importantly, these heterotypic junctions were also present in patient-derived material including head and neck SCC tissues.

To assess whether heterophilic junctions can both sense and actively respond to physical force, in opposition to previous findings, a magnetocytometry method was used whereby pulling forces are applied to magnetic beads — coated in recombinant cadherin–Fc fusion proteins — attached to cells. Upon contact of N-cadherin-coated beads with A431 cells, E-cadherin was recruited to the interface, and junction protein localization at the bead–cell contact could be increased further by applying a force. In response to a series of force pulses applied through N-cadherin-coated beads, stiffening and mechanical reinforcement of the heterotypic junctions in A431 cells were observed. Moreover, force transmission between CAFs and A431 cells decreased upon knockout of E-cadherin in A431 cells. This suggests that E-cadherin–N-cadherin junctions are mechanically active during CAF-led collective A431 cell migration. Lastly, the authors confirmed the importance of N-cadherin in directing the CAF repolarization by showing that knockdown of N-cadherin in CAFs could inhibit the polarity reversal of CAFs upon contact with A431 spheroids.

This study highlights how the expression of E-cadherin in cancer cells does not necessarily limit their invasive capacity and, together with N-cadherin in heterophilic junctions, can promote cancer cell migration and invasion. Given that heterotypic junctions are absent in normal tissue, it will be interesting to determine whether blocking this interaction between CAFs and cancer cells can prevent metastasis.

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ORIGINAL ARTICLE Labernadie, A. *et al*. A mechanically active heterotypic E-cadherin/N-cadherin adhesion enables fibroblasts to drive cancer cell invasion. *Nat. Cell Biol.* **19**, 224–237 (2017)