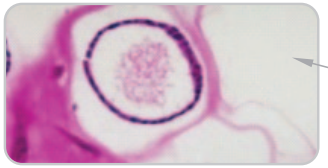
Assays For Human Mammary Stem and Progenitor Cells

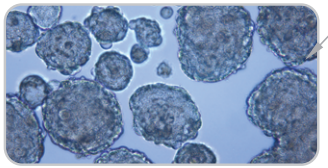
STEMCELL TECHNOLOGIES

EpiCult®-B
The 3D Morphogenesis Assay



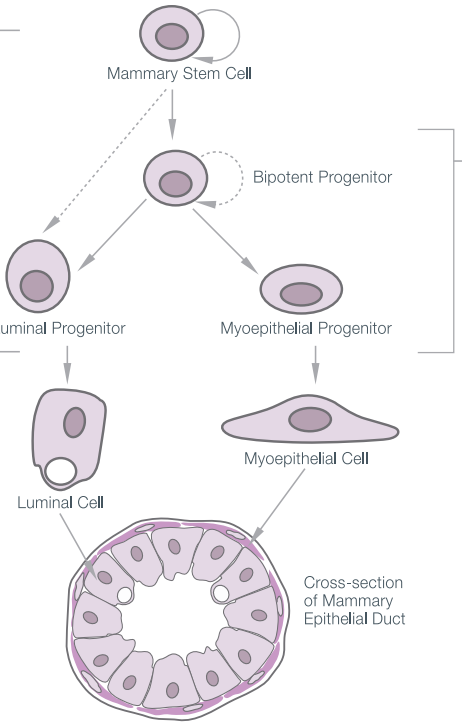
Cells cultured in a semi-solid matrix such as Matrigel™ supplemented with EpiCult®-B and FBS form 3-dimensional mammary structures, analogous to those detected *in vivo*. The structures are comprised of polarized luminal cells surrounded by a layer of myoepithelial cells. Investigators use this assay to characterize mechanism of mammary epithelial cell development.[4]

MammoCult®
Mammosphere Culture

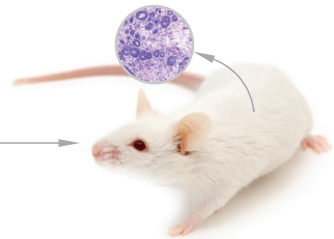


Cells cultured in a liquid medium such as MammoCult® under anchorage-independent conditions form mammospheres. Some of the cells that survive and proliferate in liquid suspension have demonstrated multi-lineage differentiation potential (in the Ma-CFC assay). These cells are thus believed to represent primitive mammary epithelial progenitors. Breast cancer stem cells can also be enriched and detected using mammosphere (or so-called tumorsphere) culture systems.[5]

Mammary Epithelial Cell Differentiation



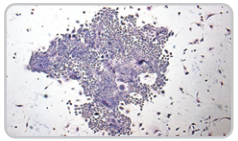
The Mammary Repopulating Unit Assay



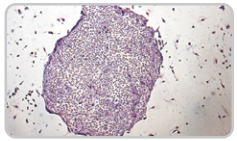
The Mammary repopulating unit (MRU) assay is used to estimate the number of mammary stem cells. MRUs, the operational term for mammary stem cells, can be detected by embedding a single cell suspension of normal breast cells within collagen gels along with irradiated stromal feeder layers, and placing these cells under the renal capsule of a female immune-deficient mouse.[6] The MRUs can generate histologically normal-looking mammary epithelium after 4 weeks *in vivo*. To determine the relative numbers of MRUs seeded within the collagen gels, the gels are then dissociated and seeded into mammary colony-forming cell (Ma-CFC) assay. The number of colonies generated in the Ma-CFC assay is linearly related to the number of MRUs initially seeded within the collagen gel. Human mammary stem cells are perceived to reside within the basal layer of the human mammary epithelium since they have an $EpCAM^{low}CD49f^{high}$ phenotype.[7]

EpiCult®-B - The Mammary Colony Forming Cell (Ma-CFC) Assay

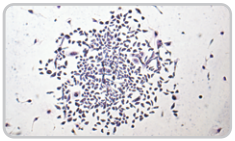
The Mammary Colony-Forming Cell (Ma-CFC) assay detects three distinct types of progenitors based on their ability to form morphologically distinct colonies. Epithelial cells are plated at low seeding density onto a layer of irradiated NIH 3T3 fibroblasts to ensure formation of robust clonally-derived colonies.



Bipotent progenitors generate colonies that contain both luminal and basal cells. The characteristic pattern of these ***mixed colonies*** is that they contain a central core of tightly arranged luminal cells that are surrounded by more elongated teardrop-shaped basal cells. The centrally located cells express the luminal-specific proteins keratins, 8, 18, and 19 as well as high levels of EpCAM and MUC1. The peripherally-located basal cells express keratin 14 and CD10. Bipotent progenitors have an $EpCAM^{low}CD49f^{high}$ MUC1- phenotype.[8,9]



Luminal-restricted progenitors generate colonies that are composed solely of luminal epithelial cells. The *luminal colonies* are characterized by their smooth scalloped borders and closely arranged cells with cell boundaries that are difficult to resolve. Cells of these colonies express luminal-specific proteins such as keratins 8, 18 and 19, as well as high levels of EpCAM and MUC1. Luminal-restricted progenitors have an $EpCAM^{high}CD49f^{-}MUC1^{+}$ phenotype.[8,9]



Myoepithelial-restricted progenitors generate colonies that are composed solely of basal-like epithelial cells. The colonies are characterized by dispersed teardrop-shaped cells that typically do not make cell-cell contact with one another. Cells of ***myoepithelial colonies*** express basal specific proteins such as keratin 14 and CD10 and do not express smooth muscle actin, indicating their inability to undergo terminal differentiation *in vitro*.[8,9]

EpiCult®-C is our new defined medium for the robust culture of human mammary epithelial cells. Contact us for your free sample.

An Optimized Dissociation Protocol is essential for obtaining viable, functionally competent precursor cells. Contact us for detailed protocols.

Need hands-on training? Learn how to process and assay mammary tissue with the Mammary Training Course.

We also offer a complete line of products for Mouse Mammary Epithelial Cell Isolation and Culture!

References

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