3D tissue models for the study of the effects of low-dose irradiation

Celeste M. Nelson, Jimmie E. Fata, Paraic A. Kenny and Mina J. Bissell
Life Sciences Division, Lawrence Berkeley National Laboratory, Berkeley CA 94720

We have previously developed three-dimensional tissue models to study the effects of irradiation of the ability of mammary epithelial cells to form polarized acinar structures and demonstrated that irradiation induced heritable epigenetic perturbations in cell polarity and multicellular organization (1). More recently we have developed a number of more quantitative models of important developmental processes in the mammary gland which can be applied to the study of the interactions between low-dose irradiation, the tissue microenvironment and tissue structure and function.

We have used a micropatterning technique to control the initial three dimensional structure of mammary epithelial tubules (Fig 1A) which can then be stimulated and monitored for changes in the patterns of invasion and branching morphogenesis caused by the stimulus (Fig 1B). The ability to analyze large numbers of similarly sized structures in parallel (by generating stacks of images from multiple samples, Fig 1C) has allowed us to perform robust statistical analyses.

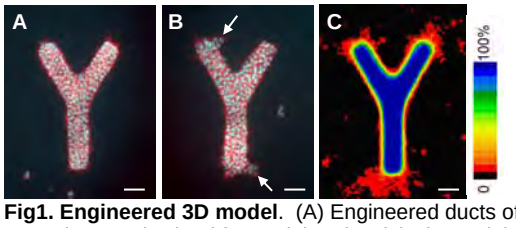

Fig1. Engineered 3D model. (A) Engineered ducts of normal MECs (stained for nuclei and actin), day 0. (B) Cells branch from specific locations (arrows), day 1. (C) Stacking images of nuclei from multiple samples (~50) yields average location of invasion. Bars, 50 μm.

A second model allows us to follow the pattern of branching morphogenesis which takes place in explanted mouse mammary gland organoids cultured on top of matrigel. These structures, composed of all mammary gland cell types, can be induced to reorganize and branch upon stimulation with appropriate growth factors (Fig 2). Many organoids can be analyzed in parallel using time-lapse microscopy. This will facilitate our analysis of the effects of low-dose irradiation heterotypic cell-cell interactions and tissue morphogenesis.

We believe that these models will be exquisitely sensitive to detect the potentially subtle effects of low dose irradiation.

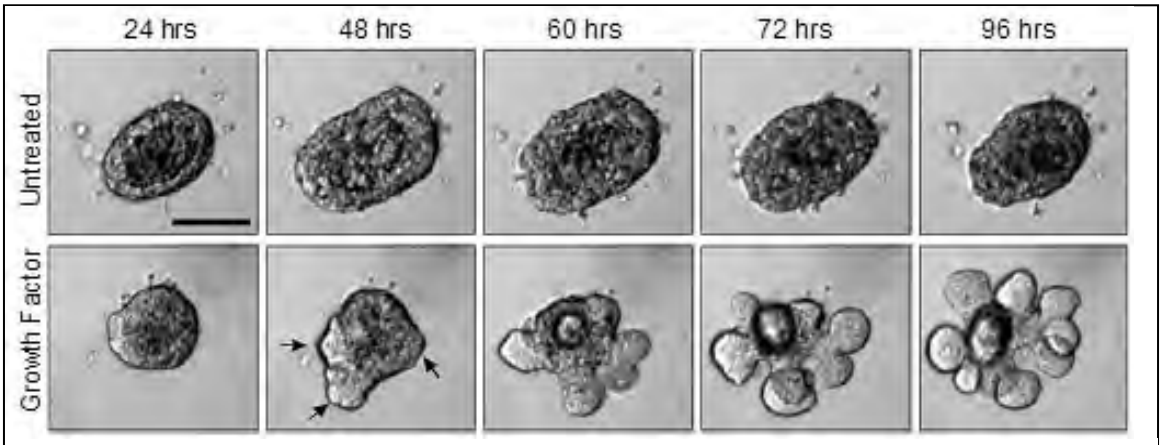

Fig 2. Time lapse of growth factor induced mammary morphogenesis. Untreated organoids fail to undergo and major morphological change, other than a slight lumen expansion and compression from48to60hrs.Incontrast, growth factor stimulation initiates bud formation (arrows) within 48hrs, with the majority of morphogenesis occurring within the 48 to 72 hr window. Images represent frames taken from movies (see supplemental data). Scale bar represents 100 μm.

1. Park, C.C., Henshall-Powell, R.L., Erickson, A.C., Talhouk, R., Parvin, B., Bissell, M.J., and Barcellos-Hoff, M.H. 2003. Ionizing radiation induces heritable disruption of epithelial cell interactions. *Proc Natl Acad Sci U S A* 100:10728-10733.