

DiscoveryBioMed, Inc. Human ADPKD Primary and Immortal Cell Cultures

Fall 2017



Background: DBM, Inc. acted as a Cell Culture and Engineering Core D for the Baltimore PKD Center in the first 5 years of the NIH NIDDK P30 PKD Research and Translational Core Centers program. These efforts have focused on the generation of mouse PKD vs WT renal epithelial cell lines.

There were as many requests from Research Base investigators of our P30 and beyond about the availability of human cell cultures and cell lines with relevance to PKD. For our own interests and to complement our efforts for the Baltimore PKD Center, we ramped up this parallel effort. We collaborate with this Center and now the Mayo PKD Center and the Kansas PKD Center on the further development of these primary cultures from both human ADPKD donor kidneys and from normal kidney cells and diseased tissues. We have multiple offerings from donors with mutations in either PKD1 or PKD2.

Source Material: DBM, Inc. has multiple commercial vendors/exchanges where it has protocols to obtain fresh human normal or ADPKD kidney tissue. The Baltimore P30 PKD Center and the Mayo P30 PKD Center will also be sources of this tissue material going forward and will collaborate with DBM on the generation of these human cells and on their genotyping for PKD1 and PKD2 gene mutations, respectively. All collaborators are well respected PKD academic investigators.

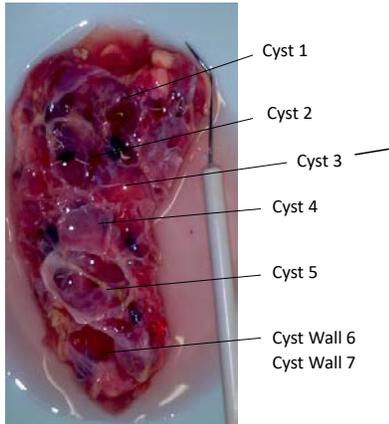
Significant Unmet Need of *in vitro* Models: Primary and immortalized diseased human kidney cells of high quality and relevancy have always been a goal for the research community. And their remains a huge unmet resource need of any novel ADPKD cell line that maintains disease characteristics, *in vivo*-like phenotype, and robust propagation. DBM acknowledges this need and has sought to fill this void.

Primary human ADPKD kidney epithelial cells: Capitalizing on the lessons learned from working with mice tissue and cells, DBM successfully began the culture of these cells in 2015 and has since refined and optimized for 2D and 3D applications, specifically those related to the formation of cysts in 3D culture. Furthermore, over this time DBM has tested and characterized several industry standard controls in this system. See **Figure 1** on next page for 2D and 3D images.

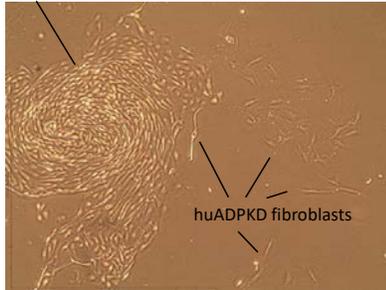
Immortal human ADPKD kidney epithelial cells: DBM is proud to officially offer immortalized versions of its ADPKD primary cells in several varieties. Our offerings for these lines are continually being expanded, so please inquire to learn more about what mutations these ADPKD cells bear and what immortalization constructs were used.

Every immortal cell line has its origins in primary cell cultures established from fresh cellular or tissue material. One then exposes the best primary cell cultures to immortalization constructs (genetic material designed to render primary cells to be more “longer-lived” or immortal).

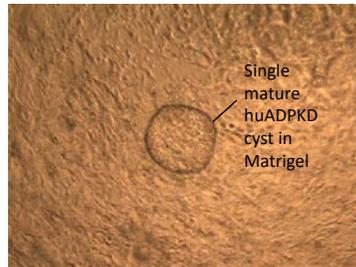
Segment of a Human ADPKD Donor Kidney



“Cyst 3” huADPKD cystic epithelial cells growing out of a cyst wall fragment



Single Cyst-derived Primary Culture



“Cyst 3” huADPKD cysts formed within 3D Matrigel culture

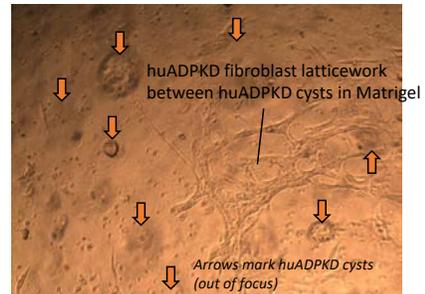


Figure 1: Example of Single Cyst-derived Primary Culture and 2D and 3D Growth Formats in DBM RenaLife Hybrid Media. See text for details and methods.

Supporting Reagents and Materials: DBM has optimized a specialty medium, that it is recommended for use with all human kidney cells and sold through our online store. Additionally, DBM has discovered that to maintain the ability to make cysts, cultures should not be grown on traditional tissue culture plastic and instead it is advised that cells be expanded on permeable filter supports, if cyst forming capacity is important to the investigator.

Final Notes: Our goal is to provide these biologically-relevant and disease-relevant human cell platforms to the renal biology and physiology research community with basic knowledge of characteristics and best growth conditions. However, these efforts remain a work in progress and users can help us with this cause in relaying back what primary cultures and cell lines work for them and why.

Please contact DBM’s Dr. Erik Schwiebert to negotiate a bundled shipment of cells, medium and ‘know how’ to get you started. erik@discoverybiomed.com

* Both the cells and the media are tested and free of pathogens.