Next-generation cancer models from the Human Cancer Models Initiative

James M. Clinton, Ph.D.
Senior Scientist, ATCC Cell Systems

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Outline

- What is Human Cancer Models Initiative?
- What are next-generation cancer models?
- Resources to learn more about the HCMI and the models at ATCC
- HCMI models available
- Summary
The unmet need

- Patients do not respond to treatments as predicted, despite huge advances in genomic analysis of primary tumors and new drug targets.

There is a need for better preclinical models to predict therapeutic outcomes.
Why are new models needed?

- Poor representation of some cancer types/subtypes
- Lack of patient and clinical outcome data, model history
- Lack of comparison to normal reference sample and/or directly compared to primary tumor
- Insufficient to capture the genetic diversity of cancer
- Existing lines may not be biologically/genetically representative of in vivo tumor
HCMI approach and core principles

- Models as a “community resource”
- Awareness of IP issues
- Permissive informed consent language permitting broad use
- Global distribution to ensure wide availability
- Open protocols

Physiological relevance

Number and diversity

Clinical and genomic data

1,000 HCMI models
Diversity of models from the HCMI

- Glioblastoma
- Gastric adenocarcinoma
- Melanoma
- Lung sarcoma
- Wilms Tumor
- Brain metastases
- Chordoma
- Esophageal carcinoma
- Neuroblastoma
- Ewing sarcoma
- Renal medullary carcinoma
- Spindle cell sarcoma
- Osteosarcoma
- Kidney renal clear cell carcinoma
- Rhabdomyosarcoma
- Pancreatic adenocarcinoma
- Mammary triple negative ER+
- Pancreatic adenosquamous carcinoma
- Lung carcinoid
- Primary pancreatic ductal carcinoma
- Colorectal cancer
- Cholangiocarcinoma
- BRCA mutant
- Barrett’s esophagus
- Esophageal adenocarcinoma
- Lung adenocarcinoma
- Desmoid Tumor
- Lung Squamous cell carcinoma

Not a comprehensive list.
Overview of HCMI and ATCC

Founders

- National Cancer Institute
- Cancer Research UK
- Hubrect Organoid Technology Foundation
- Wellcome Sanger Institute

Model Development

- Broad Institute
- Cold Spring Harbor Laboratory
- Wellcome Sanger Institute
- Hubert Organoid Technology Foundation
- University of Verona
- Hubrecht Institute
- Stanford University
- Weill Cornell Medical College

Distribution

ATCC®
Generation and distribution of HCMI models

Cancer model development centers

Primary biopsy sample

In vitro model

Sequencing

ATCC®

Subset will be sequenced

Expand, cryopreserve and perform quality control testing

Global distribution to cancer research community

Anonymized patient/clinical data captured
Characterization of models

**Molecular**
- 15X WGS of model, primary tumor, and normal tissue
- 150X WXS of model, primary tumor, and normal tissue
- RNA-seq of model and primary tumor

**Clinical**
- Disease diagnosis
- Patient demographics
- Treatment and outcomes
Advanced culture technologies

Primary patient-derived organoids

Conditionally Reprogrammed Cells
Shared features of advanced culture methods

- Permits growth and expansion
- Limited starting material required
- Genetically stable
- Maintain *in vivo* phenotype
- Relatively high success rate
Primary tissue derived organoids
Organoid technology

Embedded three-dimensional culture technique that utilizes model-specific growth media formulations in combination with undefined extracellular matrix.

1. Suspend cells or fragments in ECM. Dispense as droplets.
2. Incubate at 37°C to polymerize gel, forming a dome.
3. Overlay gelled dome with media containing niche factors.
4. Organoids form and enlarge.
5. Collect organoids and ECM.
6. Mechanically dissociate and wash to remove ECM.
7. Enzymatically dissociate.

### Primary tissue organoids compared with other models

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Primary tissue-derived organoids</th>
<th>iPSC-derived organoids</th>
<th>Cancer cell line-derived spheroids</th>
</tr>
</thead>
<tbody>
<tr>
<td>Derived from primary patient tissue</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Can be continuously propagated <em>in vitro</em></td>
<td>Yes</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Can be mechanically or enzymatically dissociated to expand culture (i.e., serial passage)</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Self-organize into complex 3D structures</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>Exhibit multiple tissue specific cell types</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>Exhibit tissue or cell type specific functionality</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>Requires multi step differentiation and formation process</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
</tr>
</tbody>
</table>
Organoids expansion, cryopreservation, and recovery

![Graph showing population doublings over days in culture.

- X-axis: Days in culture
- Y-axis: Population Doublings

![Bar graph showing viability over passage number.

- X-axis: Passage Number
- Y-axis: Viability

Legend:
- Pre-freeze
- Post-thaw

![Graph showing viability over passage number.

- X-axis: Passage Number
- Y-axis: Viability
  - Green bars: Pre-freeze
  - Purple bars: Post-thaw

Viability is consistently high for both Pre-freeze and Post-thaw conditions across various passage numbers.
Organoids are amenable to standard lab assays

**Organoids**
- **Esophageal adenocarcinoma**
  - Nucleus
  - Ki67
  - ECAD
  - BCAT
  - MUC

- **Colon adenocarcinoma**
  - CK20
  - Nucleus
  - Ki67
  - ECAD
  - BCAT
  - MUC

- **Lung adenocarcinoma**
  - Nucleus
  - ITGB1
  - ECAD
  - BCAT
  - CD44
HCMI organoid models

Types
- Adenocarcinoma
- Carcinoma
- Primary
- Recurrent
- Metastatic
- Pre-malignant

Tissues
- Lung
- Colon
- Rectum
- Mammary
- Esophagus
- Pancreas
- Prostate
- Liver
- Ovary
- Stomach

Not a comprehensive list

16 unique models from various tissue
Organoid culture guide

Initiation, Expansion, and Cryopreservation of Human Primary Tissue-Derived Normal and Diseased Organoids in Embedded Three-Dimensional Culture

Conditionally reprogrammed cells
Use of Reprogrammed Cells to Identify Therapy for Respiratory Papillomatosis

Hang Yuan, Ph.D., Scott Myers, M.D., Jinggang Wang, Ph.D., Dan Zhou, M.S., Jennifer A. Woo, M.S., Bhaskar Kallakury, M.D., Andrew Ju, M.D., Michael Bazilewicz, M.D., Yvonne M. Carter, M.D., Christopher Albanese, Ph.D., Nazanin Grant, M.D., Aazia Shad, M.D., Anatoly Dritschilo, M.D., Xuefeng Liu, M.D., and Richard Schlegel, M.D., Ph.D.
CRC culture can prevent senescence of primary cells

- Primary Epidermal Keratinocytes
  ATCC PCS-200-011

- Primary Small Airway Epithelial
  ATCC PCS-301-010

- Primary Prostate Epithelial
  ATCC PCS-440-010
HCMI CRC and other non-organoid models

Includes CRC, various other 2D and 3D models types

Tissues
- Adrenal gland
- Soft tissue/bone
- Head and neck
- Brain
- Kidney

Types
- Neuroblastoma
- Glioblastoma
- Ewing Sarcoma
- Rhabdomyosarcoma
- Wilms Tumor

Not a comprehensive list
### Culture conditions for currently released HCMI models

<table>
<thead>
<tr>
<th>Media/Technology</th>
<th>Off-the-shelf growth media</th>
<th>Growth properties</th>
<th>Tissue type(s)</th>
<th>Serum-free/defined</th>
<th>ECM or special surface required?</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Organoid</strong>*</td>
<td>No</td>
<td>3D embedded</td>
<td>Multiple</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td><strong>Conditional Reprogramming</strong></td>
<td>Yes</td>
<td>2D adherent</td>
<td>Multiple</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td><strong>SmGM-2</strong></td>
<td>Yes</td>
<td>2D adherent and suspension</td>
<td>Bone</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td><strong>RETM</strong></td>
<td>Yes</td>
<td>2D adherent</td>
<td>Brain</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td><strong>Neurocult NS-A</strong></td>
<td>Yes</td>
<td>2D adherent and suspension</td>
<td>Brain</td>
<td>Yes</td>
<td>Sometimes</td>
</tr>
</tbody>
</table>

*There are multiple organoid media formulations*
Model resources and data access

- View all models available or grouped by tissue
- Model specific information such as culture images, seeding densities, media change frequencies, etc.
- Individual model product pages include detailed culture protocols
  - Complete media formulation
  - Thawing/subculturing/freezing guides
- Model pages link to other resource pages that host clinical and sequencing data
- Frequently asked questions

www.atcc.org/HCMI
Model resources and data access

- NCI managed website
- Integrates clinical, model, and genomic information.
- Search for models of interest using various filters
  - Primary tumor site/acquisition site
  - Model type
  - Tumor diagnosis/stage/grade/histological type
  - Gender/age/ethnicity
  - Etc.
- Links out to clinical and genomic data, ATCC model product page.

hcmi-searchable-catalog.nci.nih.gov
Model resources and data access

- NCI managed website
- Search and download cancer related datasets for analysis
- Navigate to the “HCMI-CMDC” project for HCMI specific datasets
- Download WGS/WXS/RNAseq data
  - Aligned reads, gene expression, SNVs

portal.gdc.cancer.gov
Model resources and data access

- NCI managed website
- Background information on the HCMI program and organization
- Useful documents
  - Case Report forms for patient enrollment and follow-up
  - HCMI Searchable catalog user guide
  - Informed consent template

https://ocg.cancer.gov/programs/HCMI
28 models currently available from ATCC

Additional models will be made available on a rolling basis, expect releases every 1-2 months.

Pipeline includes mammary carcinoma organoids, primary and metastatic esophageal adenocarcinoma organoids, additional colon, pancreas and glioblastoma models.
ATCC HCMI model distribution material

- Target $\geq 1 \times 10^6$ viable cells per vial
- Product documentation includes
  - Recommended growth medium
  - Recommended culture maintenance parameters

**QC**
- Post-thaw cell count and viability
- Mycoplasma testing
- Sterility testing (bacteria, fungi and yeast)
- Species determination
- STR fingerprinting
- Human pathogenic virus testing (HIV, HBV, EBV, CMV, WNV)
Models currently available

- Bone: 29%
- Brain: 29%
- Bronchus and lung: 3%
- Colon: 29%
- Pancreas: 21%
- Rectum: 7%

Primary: 72%
Metastatic: 10%
Recurrent: 14%
Pre-malignant: 4%
## Models currently available

<table>
<thead>
<tr>
<th>Part No.</th>
<th>Description</th>
<th>Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>PDM-135</td>
<td>Metastatic Adenocarcinoma of stomach</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-1</td>
<td>Primary Adenocarcinoma of colon</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-2</td>
<td>Primary Adenocarcinoma of colon</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-4</td>
<td>Primary Adenocarcinoma of colon</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-5</td>
<td>Primary Adenocarcinoma of colon</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-6</td>
<td>Primary Adenocarcinoma of colon</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-7</td>
<td>Primary Adenocarcinoma of colon</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-8</td>
<td>Primary Adenocarcinoma of colon</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-44</td>
<td>Pre-malignant Adenoma of colon</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-9</td>
<td>Metastatic Adenocarcinoma of rectum</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-43</td>
<td>Primary Adenocarcinoma of rectum</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-106</td>
<td>Metastatic Adenocarcinoma of pancreas</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-36</td>
<td>Primary Adenocarcinoma of pancreas</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-38</td>
<td>Primary Adenocarcinoma of pancreas</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-39</td>
<td>Primary Adenocarcinoma of pancreas</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-40</td>
<td>Primary Adenocarcinoma of pancreas</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-41</td>
<td>Primary Adenocarcinoma of pancreas</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-3</td>
<td>Primary Adenocarcinoma of lung</td>
<td>Organoid</td>
</tr>
<tr>
<td>PDM-127</td>
<td>Metastatic Ewing sarcoma</td>
<td>2D adherent</td>
</tr>
<tr>
<td>PDM-114</td>
<td>Primary Osteosarcoma</td>
<td>2D adherent</td>
</tr>
<tr>
<td>PDM-16</td>
<td>Primary Glioblastoma</td>
<td>2D adherent</td>
</tr>
<tr>
<td>PDM-17</td>
<td>Primary Glioblastoma</td>
<td>2D adherent</td>
</tr>
<tr>
<td>PDM-18</td>
<td>Primary Glioblastoma</td>
<td>Suspension</td>
</tr>
<tr>
<td>PDM-20</td>
<td>Primary Glioblastoma</td>
<td>2D adherent</td>
</tr>
<tr>
<td>PDM-19</td>
<td>Recurrent Glioblastoma</td>
<td>2D adherent</td>
</tr>
<tr>
<td>PDM-21</td>
<td>Recurrent Glioblastoma</td>
<td>2D adherent</td>
</tr>
<tr>
<td>PDM-22</td>
<td>Recurrent Glioblastoma</td>
<td>Suspension</td>
</tr>
<tr>
<td>PDM-23</td>
<td>Recurrent Glioblastoma</td>
<td>2D adherent</td>
</tr>
</tbody>
</table>

www.atcc.org/HCMI
Model availability

- Sign up on the ATCC HCMI website to join the early adopter program and gain access to models prior to release.

- Sign up for the ATCC mailing list to be notified of new HCMI model releases.
Summary

- HCMI models are primary patient-derived models from a variety of tissues and cancer types that are paired with patient clinical and molecular characterization via WGS/WXS/RNA-Seq.

- 28 models from a variety of tissues/cancer types are currently available and new models will be continually released in the coming months.

- ATCC is excited to support the HCMI and their goal of developing and distributing next-generation cancer models.
Thank you for joining today!

- Register for more ATCC webinars at www.atcc.org/webinars

- June 27 | 12:00 PM ET
  iPSC-derived Primary Cells: Expand Your Cell-based Assays With an Unlimited, Biologically Relevant Resource
  Yalin Firinci, M.B.A.
  Product Line Business Specialist, ATCC

Please email additional questions to: tech@atcc.org