3D Biomatrix™
Three-Dimensional Cell Culture

3D Cell Culture: An Early-Stage Oncology Drug Discovery Tool

White Paper

In this white paper

3D cell culture to enhance physiological relevance of cell-based assays

Introduction

It is often said that the main expense in drug discovery is failure, and oncology drug discovery is no exception. In 2011, nearly 900 anti-cancer medicines and vaccines were in clinical trials or under the Federal Drug Administration (FDA) review [1], yet only 12 oncology drugs were approved in that year [2]. With an average clinical trial length of six years, these numbers represent hundreds of drugs in clinical trials that will not make it to market. Behind those hundreds of unsuccessful drug candidates lie hundreds of millions of dollars spent in research and development (Table 1) and clinical trials costs.

Research Spending Per New Drug

Company	Ticker	Number of drugs approved	R&D Spending Per Drug ($Mil)	Total R&D Spending 1997-2011 ($Mil)
AstraZeneca	AZN	5	11,791	58,955
GlaxoSmithKline	GSK	10	8,171	81,708
Sanofi	SNY	8	7,909	63,274
Roche Holding AG	RHHBY	11	7,804	85,841
Pfizer Inc.	PFE	14	7,727	108,178
Johnson & Johnson	JNJ	15	5,886	88,285
Eli Lilly & Co.	LLY	11	4,577	50,347
Abbott Laboratories	ABT	8	4,496	35,970
Merck & Co Inc	MRK	16	4,210	67,360
Bristol-Myers Squibb Co.	BMY	11	4,152	45,675
Novartis AG	NVS	21	3,983	83,646
Amgen Inc.	AMGN	9	3,692	33,229

Source and adapted from: InnoThink Center For Research In Biomedical Innovation; Thomson Reuters Fundatmentals via FactSet Research Systems

Table 1. The staggering cost of drug development.

Expensive and time-consuming preclinical testing on multiple animal species may not predict how humans will react to drugs

Why Do Drugs Fail?

Currently, standard screening procedures rely on animals. However, the suitability of animal testing to predict the effects of a drug on humans is debated; humans react to drugs differently than animals [3]. Expensive and time-consuming preclinical testing on multiple animal species may not predict how humans will react to the drug, so even if animal trials are promising, the compound may still fail during clinical trials.

These facts, along with strong ethical concerns, are motivating governments and regulatory organizations around the globe to support the implementation of alternative methods for animal testing. Replacement of animal models in drug discovery and toxicity testing is one of the three R's of the Replacement, Reduction, and Refinement (3R) agenda for the humane handling of laboratory animals. 3R-compliance is increasingly required in good laboratory animal practice [4].

In vitro cell-based assays have the potential to replace *in vivo* animal testing and provide more reliable data. However, many current assays are performed using two-dimensional (2D) cell culture, which, due to the unnatural configuration that cells take on a flat surface, do not sufficiently predict how the human body will react to a drug.

3D cell cultures can improve cell-based drug screening and identify toxic and ineffective substances at an early stage of the drug discovery pipeline

Three-dimensional (3D) cell cultures, however, bridge the *in vitro* to *in vivo* gap by growing cells within a micro-environment that mimics real tissues and establishes physiological cell-cell and cell-substrate interactions that regulate proliferation and differentiation. 3D cultures have potential to greatly improve cell-based drug screening and identify toxic and ineffective substances at an earlier stage of the drug discovery pipeline than animal or clinical trials. Moreover, they can reduce ethically-controversial animal testing [5].

Oncology drug development is one therapeutic area that can specifically benefit from the inclusion of 3D cell culture models in the drug-testing process. A growing body of research finds that traditional 2D cell culture may not accurately mimic the 3D environment in which cancer cells reside, as the 2D environment does not allow for areas of hypoxia, heterogeneous populations of cell types (including stromal cells), differing replication behaviors (quiescent vs. replicating), extracellular matrix (ECM) influences, soluble signal gradients, and differential nutrient and metabolic waste transport [6]. As a result, the unnatural 2D environment may provide inaccurate data regarding the predicted response of cancer cells to chemotherapeutics [7]. 3D cell culture models that reproduce the natural tumor environment with all its heterogeneity have great potential in making cell-based assays for anti-cancer drugs much more meaningful, possibly lowering the cost and time of taking a drug to market.

Spheroid Cultures Naturally Mimic Avascular Tumors

Spheroids, self-assembled spherical clusters of cell colonies, are one of the most well-characterized models for 3D culture and screening due to their simplicity, reproducibility, and similarity to physiological tissues compared to other methods involving ECM scaffolds and hydrogel systems [8, 9]. One of the earliest accounts of spheroid culture of mammalian cells was Johannes Holtfreter's work on spherical aggregates of embryonic cells in 1944. Aron Moscona, another pioneer in the 1950s, studied the capacity of embryonic and malignant cells for re-aggregation, proliferation, differentiation, and invasion. This early work sparked a sustained interest in using spheroids as a model system to study the interaction of tumor cells within their micro-environment, as well as their responsiveness to radio- and chemotherapies.

Spheroids mimic tumors with inherent metabolic and proliferative gradients

Spheroids naturally mimic avascular tumors with inherent metabolic (oxygen, carbon dioxide, nutrients, wastes) and proliferative gradients (Figure 1); thus, they serve as excellent physiologic tumor models known to provide more reliable and meaningful therapeutic results compared to 2D tests [8, 9]. Co-culture of cancerous and non-cancerous cells (i.e., endothelial, stromal, and epithelial cells) extends the system's predictive cytotoxicity capabilities. As spheroids are self-organized and produce their own ECM, they encompass the complex cell-matrix and cell-cell interactions that mimic functional properties of the corresponding tissue *in vivo* [8].

One of the most exciting prospects among recent advances in 3D cell culture is standardized mass production of 3D spheroids for high-throughput screening applications. Reproducible mass production of spheroids is a promising path in which 3D culture systems can be incorporated into mainstream drug development processes, including cell-based assays for predictive cytotoxicity for anti-cancer drugs.

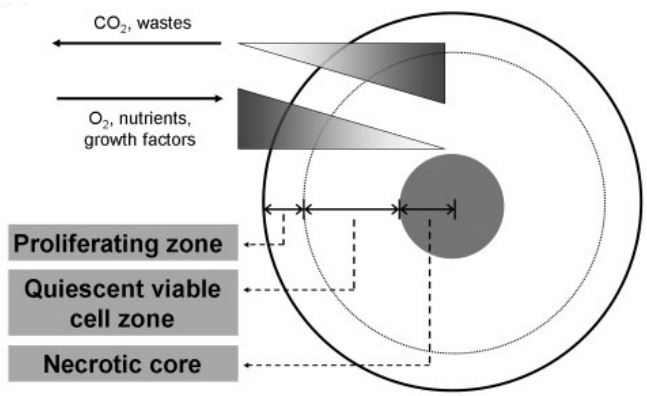


Figure 1. Tumor spheroids exhibit spherical geometry with a concentric organization of proliferating, quiescent and dead cells. The formation of multi-layered arrangements can be attributed to the insufficiency of mass transport through cellular barriers. Figure source [6].

A novel 3D cell culture platform as straightforward to perform as conventional 2D culture plates

Perfecta3D™ Hanging Drop Plates

One such method of mass production of 3D spheroid cultures is the Perfecta3D Hanging Drop Plate. The Perfecta3D Hanging Drop Plate was designed to enable consistent formation of spheroids using conventional liquid handling tools. Access holes in the culture plate allow manipulation of fluids and spheroids from the topside. In addition to the main culture plate in which spheroids are formed, each plate includes a complementary lid and tray, which serve to maintain sterility and reduce evaporation (Figure 2). A water reservoir is constructed around the periphery of the culture plate, additionally contributing to alleviating evaporation. The dimensions of the entire system meet ANSI/SBS standards.

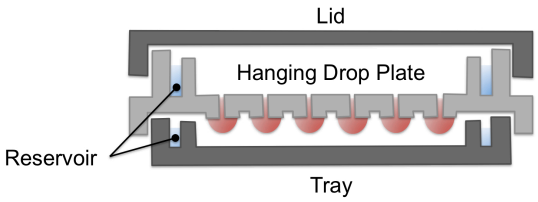


Figure 2. A schematic of the Perfecta3D Hanging Drop Plate.

Hanging drops, of mono- or co-cultures, are created by dispensing small volumes of cell suspension, using standard pipette tips, into the access holes on the top of the plate, just like pipetting into conventional multi-well plates (Figure 3). A plateau structure on the bottom of the plate stabilizes the hanging drops. As the Perfecta3D Hanging Drop Plates do not have a bottom substrate for cells to eventually attach to, cells in suspension aggregate into a spheroid.

Perfecta3D Hanging Drop Plates provide advantages compared to alternative techniques using bioreactors, non-adherent surfaces, magnetic particles, and round- and V-bottom microplates in combination with centrifugation. The culture system inherently offers the convenience of utilizing natural gravity force to aggregate all cell types into a single spheroid. Unlike the use of centrifugation, which generates shear forces that can damage cells, or the use of magnetic particles, which can interfere with cellular behaviors and test compounds, the Perfecta3D platform offers a simple user-interface, free of external forces and substances.

Invented at the University of Michigan

"After struggling to generate reproducible spheroids in spinner cultures and agarose-coated plates, the Perfecta3D Hanging Drop Plates have finally given me consistent cell growth and morphology, and the spheroids' size and shape are remarkably reproducible."

Research scientist at a major Canadian university

Designed to enable consistent formation of spheroids for life sciences research and drug screening

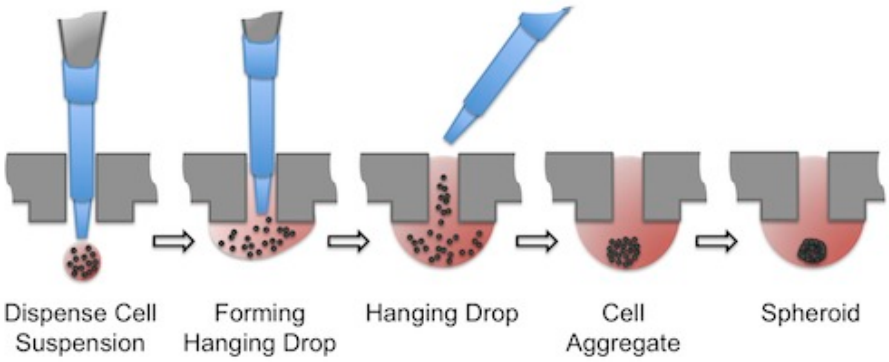


Figure 3. Spheroids are created by dispensing cell suspensions into the access holes of the Perfecta3D Hanging Drop Plate, just like pipetting into conventional multi-well plates.

Reagents, drugs, and cells can be added to or removed from each hanging drop through the access hole in the plate. Importantly, spheroids can be harvested both from the top or the bottom of the plate. The ability to access samples directly from the top of the Perfecta3D Hanging Drop Plates enables more sophisticated patterning of co-culture spheroids that are not possible using other methods. Several configurations of spheroid co-cultures will be discussed in more detail later.

Microscopic imaging of spheroids can be performed directly with the transparent plate, lid and tray assembled. The standard-size 384-well microplate allows performance of colorimetric, fluorescence, and luminescence assays using a plate reader. Furthermore, a recent paper demonstrated that the reliability of Perfecta3D Hanging Drop Plates for fluorescence- and colorimetric-based assays was comparable to that of standard 384-well plates [10].

The platform also offers simplified liquid handling procedures and compatibility with high-throughput screening instruments, such as liquid handling robots like the Biomek® FX and epMotion automated pipetting systems. The Perfecta3D Hanging Drop Plates are positioned to become an integral tool for drug screening and life sciences research.

Advantages

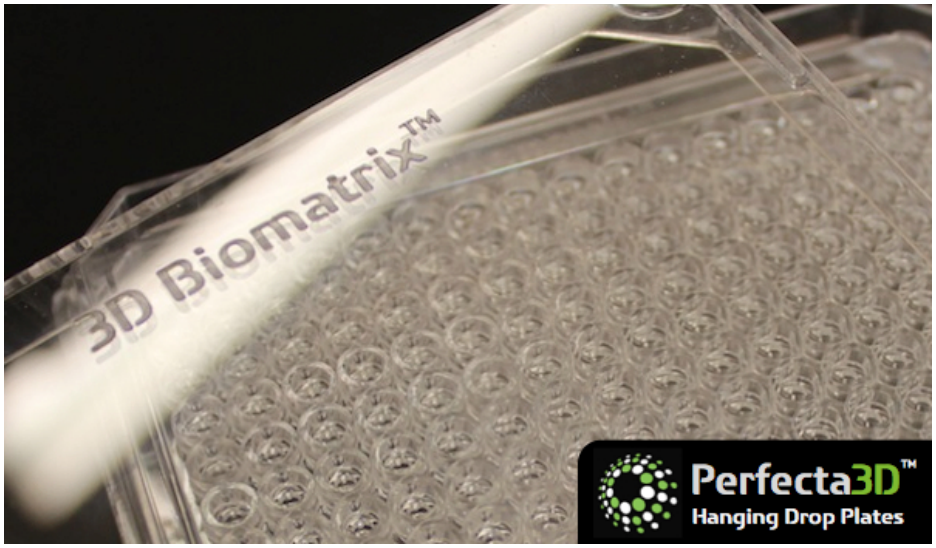
- Physiological and non-expensive spheroid culture system
- Efficient formation of uniform-size spheroids
- Compatible with high-throughput screening (HTS) instruments
- Easily maintained from the top of the plate
- Suitable for long-term culture
- Standardized plate format
- Reduced consumption of media and reagents.

Specifications

- Polystyrene
- Untreated
- Sterile (gamma irradiation)
- Individually packaged
- Standard 384-well format with lid and tray
- Meets ANSI/SBS standards
- Stackable
- Recommended reservoir capacity: Plate 2 mL, Tray 1.5 mL

Applications

- Cancer Research
- Stem Cell Research
- Tissue Engineering
- Drug Discovery and Testing
- High Throughput Screening
- Chemotherapy and Radiotherapy Research



As seen on the cover of *Analyst*

Assessment of Chemosensitivity Using Viability and Proliferation Assays

A 2011 study by Professor Shuichi Takayama's research team at the University of Michigan shows the dramatic difference in results between 2D and 3D cell cultures. The study illustrated the importance and need of using 3D models in drug screening and testing by demonstrating that drugs with different modes of action produce distinctly different responses in the physiological 3D spheroids grown in Perfecta3D Hanging Drop Plates when compared to conventional 2D cell monolayers [11].

AlamarBlue® was used to measure cell viability. In this assay, the active ingredient, resazurin, is reduced to resorufin upon entering the cells. This reaction produces very bright red fluorescence, thereby generating a quantitative measure of viability, or cytotoxicity, after normalization to untreated controls. These viability results were further compared to results obtained by fluorescence microscopy imaging using a LIVE/DEAD® Viability/Cytotoxicity Kit for mammalian cells. This live/dead kit simultaneously stains with green-fluorescent calcein-AM to indicate intracellular esterase activity (live cells) and red-fluorescent ethidium homodimer-1 to indicate loss of plasma membrane integrity (dead cells).

A431.H9 cells, human epithelial carcinoma cells, were treated with two anti-cancer drugs having distinctly different activity profiles. 5-fluorouracil (5-FU) is a conventional compound that inhibits cellular proliferation, and tirapazamine (TPZ) is a hypoxia-trigger cytotoxin that causes DNA damage.

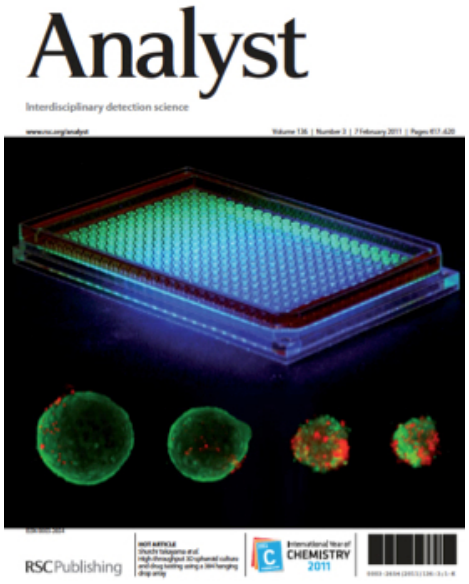


Image adapted from Tung et al. [11].

Data demonstrated that more A431.H9 cells remained viable when treated with 5-FU when grown as a 3D spheroid rather than a 2D monolayer. However, the opposite was true for treatment with TPZ; these data indicated that more A431.H9 cells remained viable when cultured under 2D rather than 3D spheroid culture conditions (Figure 4). 5-FU primarily targets proliferating cells. Therefore, it was more effective against the rapidly proliferating cells in a 2D monolayer culture and would not kill the quiescent cells in the spheroids. In contrast, TPZ is a hypoxia-activated cytotoxin. It was more effective in the spheroids, where active oxygen consumption by cells and limits in diffusive oxygen transport create a hypoxic core similar to that of solid tumors [12].

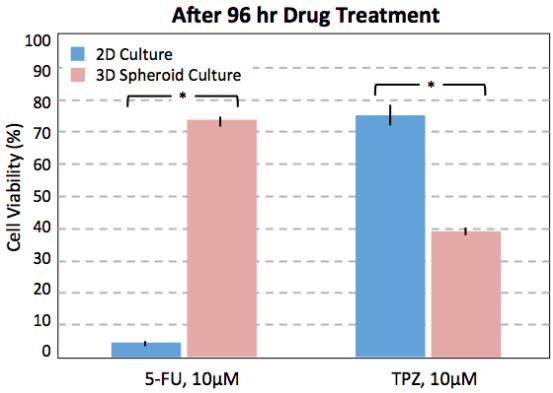
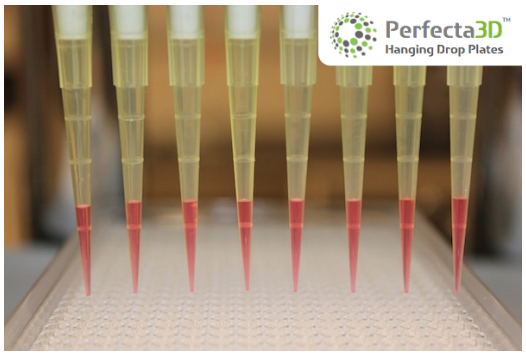


Figure 4. Cells grown in 2D and 3D cultures demonstrated different levels of viability when exposed to anti-cancer drugs. More A431.H9 cells remained viable when treated with 5-FU under 3D spheroid rather than 2D culture conditions. Alternatively more A431.H9 cells remained viable when treated with TPZ under 2D rather than 3D spheroid culture conditions.

The IC_{50} (the concentration of an inhibitor where the maximal response is reduced by half) of A431.H9 cells treated with 5-FU was about 0.1 μM in 2D condition and 1 to 100 μM, depending on cell density, in 3D-spheroid culture. For the TPZ-treated cells, the IC_{50} was about 50 μM when cultured in 2D and 8 μM for all spheroid sizes (data not shown). Such dramatically different viability responses from the same cells to the same drugs tested under different culture conditions illustrated the importance and need of using 3D models in drug screening and testing.

The treatment of 3D spheroids with a combination of the two drugs was shown to be more effective at killing A431.H9 cells than either drug administered alone. When either 5-FU or TPZ was administered at a concentration of 10 μM, cell viability decreased from 75% and 40%, respectively (Figure 4). When spheroids were treated with a combination of the drugs, both at concentrations of 10 μM, viability of cells dropped to 20% (Figure 5). By combining two drugs with distinctly different mechanisms, both the proliferating cells in the peripheral layer and cells in the hypoxic core of the 3D spheroids could be targeted effectively.



Examples of cell types* that the Perfecta3D Hanging Drop Plates have been successfully used with:

Breast Cancer Cell Lines
- BT-20 human breast carcinoma cells
- MCF-7 human breast adenocarcinoma cells
- MCF-10A human mammary epithelial cells
- MDA-MB-231 human breast adenocarcinoma cells
- SUM-159 human breast anaplastic carcinoma cells
- T47D human breast ductal carcinoma cells
- ZR-75-1 human breast ductal carcinoma cells

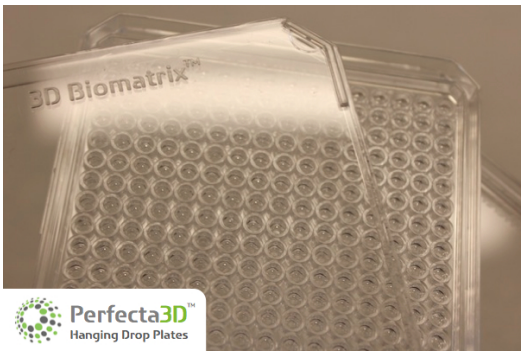
Colon Cancer Cell Lines
- Caco-2 human colorectal adenocarcinoma cells
- CT26 mouse colon adenocarcinoma cells
- HCT-116 human colorectal carcinoma cells
- HRT-18 human colorectal adenocarcinoma cells
- HT-29 human colorectal adenocarcinoma cells

Kidney Cell Lines
- COS7 African green monkey kidney cells
- HEK-293 human embryonic kidney cells

Other Cell Lines
- 3T3-L1 mouse preadipocytes
- A431.H9 human epithelial carcinoma cells
- DU-145 human prostate carcinoma cells
- ES-D3 murine embryonic stem cells
- HBME human bone marrow endothelial cells
- HeLa human epithelial carcinoma cells
- HepG2 human hepatocellular carcinoma cells
- hFOB human fetal osteoblasts
- HUVEC human umbilical endothelial cells
- MT3C3-E1 mouse osteoblastic cells
- PANC-1 human pancreatic carcinoma cells
- SK-OV-3 human ovarian adenocarcinoma cells
- UM-UC6 human bladder carcinoma cells

*complete list available at www.3DBiomatrix.com

Data adapted from Tung et al. [11].



Tumor Histoids: Tissue Engineered Realistic Human Tumor Models for Drug Screening

A poster presented at the 2011 CYTO (Congress of the International Society for Advancement of Cytometry) conference demonstrated that the Perfecta3D Hanging Drop Plates produced highly uniform single-cell-type spheroids suitable for high-throughput drug screening, without further manipulation. To expand the 3D system's utility, the research team demonstrated generation of tumor histoids; stroma (fibroblasts, ECM, endothelium) invaded by cancer cells. In initial experiments, cryo-preserved spheroids of normal fibroblasts were introduced into hanging drops and followed, a few hours later, by introduction of a uniform suspension of BT-20 breast cancer cells into the same 25 µL hanging drops. Although the cultures were maintained for only four days, breast cancer histoids were produced. The research team believes that with improved fluid handling, uniform tumor histoids can be produced in the Perfecta3D Hanging Drop Plates 384-well format, making them suitable for spectrophotometric analysis and high-throughput screening.

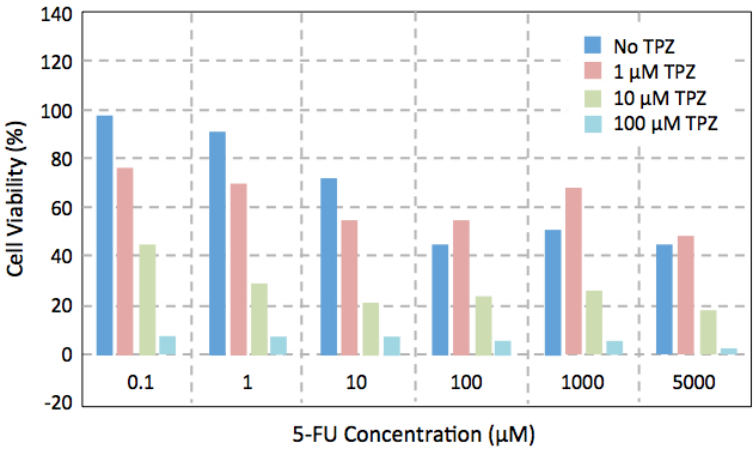


Figure 5. The combined treatment with 5-FU and TPZ was more effective at killing A431.H9 cells grown in 3D spheroids, targeting both the proliferating cells in the peripheral layer and the cells in the hypoxic core.

Formation of Co-Culture Spheroids

The ability to co-culture and pattern multiple cell types in 3D provides new approaches to understand and manipulate heterotypic-cellular interactions, opening up novel ways to study cancer biology, developmental biology, and tissue engineering. Co-culture strategies are fundamental in cell biology and mimic *in vivo* tissue niches *in vitro*. For example, co-culturing different fluorescent-labeled cells allows for real-time tracking of cellular localizations, migration, self-organization, differentiation, cell-matrix interactions and cell-cell communication. Patterned co-cultures have applicability to oncology models, particularly in advancing studies on cellular migration and metastasis, cell-cell signaling, and other cell-cell interactions.

Co-culture spheroids can be easily manipulated using the Perfecta3D Hanging Drop Plates by varying the timing and order of seeding the different cell types into the hanging drops (Table 2, Figure 6). The platform ensures uniform incorporation of all co-culture cell types into the spheroids.

In a December 2011 publication, Dr. Takayama's research team demonstrated the ability to form different types of co-cultures in the Perfecta3D Hanging Drop Plates [10].

Spheroid Type	Cells	Figure
Mixed co-culture	PC-3_DsRed human prostate cancer cells, human umbilical vein endothelial (HUVEC) cells, and MC3T3-E1 mouse pre-osteoblast cells	Figure 7
Concentric layer patterning	PC-3_DsRed human prostate cancer cells and MC3T3-E1 mouse pre-osteoblast cells	Figure 8
Concentric layer patterning	CellTracker Green-labeled and CellTracker Red-labeled COS7 African green monkey kidney fibroblast cells	Figure 9
Janus	CellTracker Green-labeled and CellTracker Red-labeled COS7 African green monkey kidney fibroblast cells	Figure 10

Table 2. Spheroid co-culture examples using Perfecta3D Hanging Drop Plates.

Data adapted from Hsiao et al. [10] and Tung et al. [11].

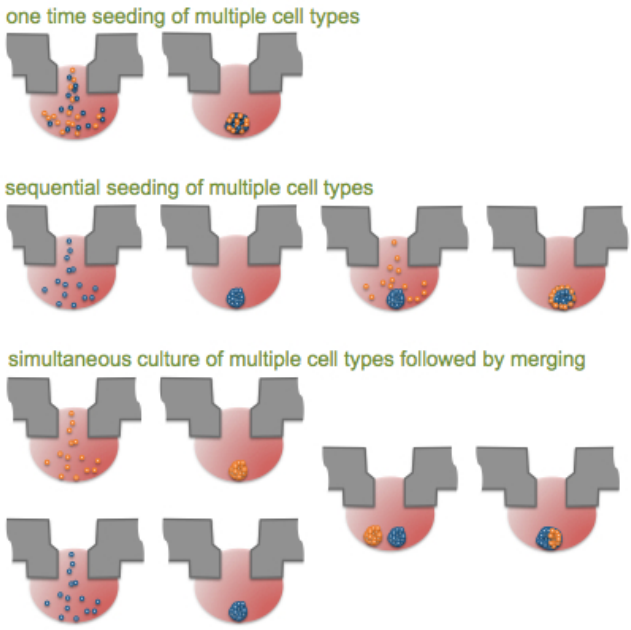


Figure 6. Techniques for forming co-cultures in Perfecta3D Hanging Drop Plates.

Mixed Co-Culture Spheroids. PC-3_DsRed human prostate cancer cells, human umbilical vein endothelial (HUVEC) cells, and MC3T3-E1 mouse pre-osteoblast cells were seeded together at a 1:50:50 ratio to form mixed co-culture spheroids (Figure 7). PC-3_DsRed cells, which were transfected with DsRed lentivirus to express red fluorescent protein, are randomly distributed within the spheroid.

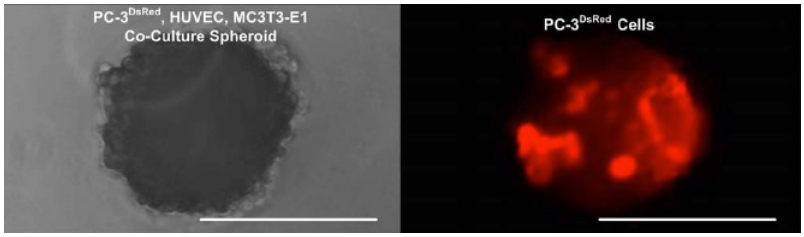
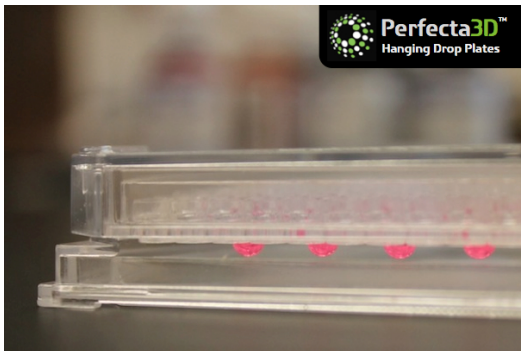


Figure 7. An example of a mixed co-culture with PC-3_DsRed human prostate cancer cells, human umbilical vein endothelial (HUVEC) cells, and MC3T3-E1 mouse pre-osteoblast cells.

Concentric Layer Patterning of Co-Culture Spheroids. Concentric layer patterning was achieved by initially forming a spheroid of one cell type as the inner core. A cell suspension of the second cell type was subsequently added to the existing hanging drop to form an exterior coating around the inner core.

PC-3_DsRed human prostate cancer cells and MC3T3-E1 mouse pre-osteoblast cells were co-cultured at a 1:100 ratio, with PC-3_DsRed cells preferentially patterned in the center core of the spheroid, randomly distributed within the spheroid, or as the exterior coating of the spheroid (Figure 8).



Assays

- Efficacy Testing
- Cell Migration
- Toxicity Testing
- Cell Differentiation
- Embryoid Body Formation
- Angiogenesis
- Colony Formation
- Cell Expansion
- Spheroid Formation
- Cell-to-Cell Interactions
- Genomic Expressions
- Cell-to-ECM Interactions
- Proteomic Expressions

"We are getting excellent results using the Perfecta3D Hanging Drop Plates. This elegant 3D tissue culture system provides more authentic models that can be expected to decrease the high incidence of false positive leads in drug screening."

Senior research scientist at a large US medical research institute

Applications: Co-culture spheroids as new preclinical models for understanding tumor initiation and progression and developing therapies

Upreti et al. have developed a preclinical *in vitro* model that can facilitate the intra/intercellular crosstalk and mimic the *in vivo* tumor and endothelial cell architecture [13]. The researchers hypothesized that tumor cell biology and treatment response would be more informative when done in the presence of stromal components, like endothelial cells, which exist in the tumor microenvironment. To test this hypothesis, they developed a system to grow 3D spheroids of mammary tumor and endothelial cells in hanging drops of cell culture medium *in vitro*.

The research team found that the presence of endothelial cells in 3D co-cultures sensitized the tumor cells to chemotherapy while protecting them from ionizing radiation. When implanted in nude mice, the tumor-endothelial spheroids induced more rapid neovascularization, faster tumor growth rates, and greater metastases than tumor cell-only spheroids. Molecular analysis also revealed pronounced up-regulation of several pro-angiogenic factors in the tumor tissue derived from the co-culture spheroids.

Upreti et al. have demonstrated a new preclinical *in vitro* model system that allows researchers to comprehensively evaluate cancer therapeutics against an environment similar to occult cancer or micro-metastases to generate more predictive preclinical data. Researchers can easily create the same novel spheroid co-culture system using Perfecta3D Hanging Drop Plates, which utilizes gravity to form multicellular scaffold-free spheroids whose size and composition can be easily and precisely controlled.

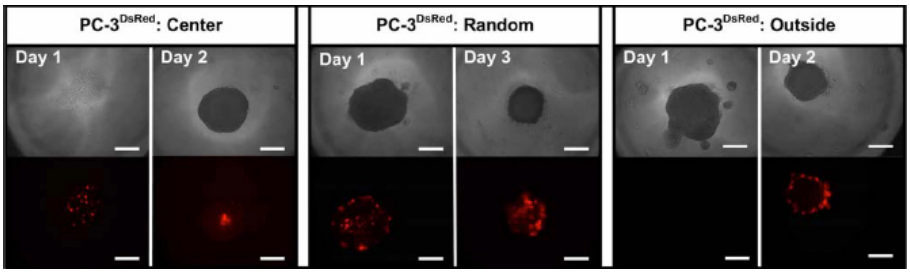


Figure 8. Examples of concentric layer patterning using co-cultures of PC-3_DsRed human prostate cancer cells and MC3T3-E1 mouse pre-osteoblast cells.

CellTracker™ Green-labeled COS7 African green monkey kidney fibroblast cells were cultured to form spheroids. On day 3, CellTracker Red-labeled COS7 cells were added to each hanging drop containing a CellTracker Green-labeled COS7 spheroid. The red cells attached to the periphery of the existing green spheroid and subsequently formed into a single spheroid with the green cells (Figure 9).

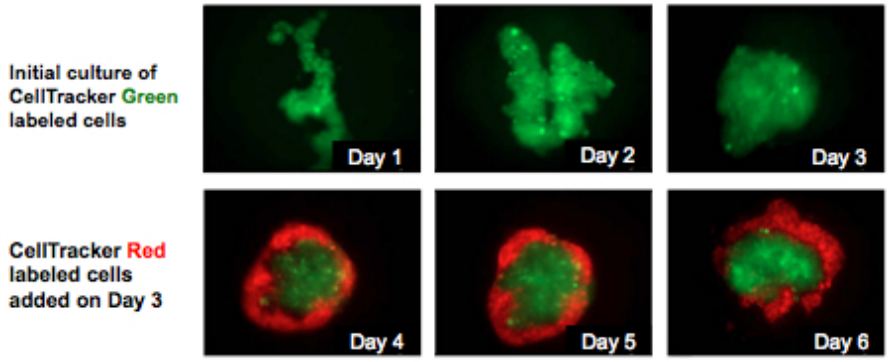


Figure 9. Example of concentric layer patterning using co-cultures of CellTracker Green-labeled and Red-labeled COS7 African green monkey kidney fibroblast cells.

Janus Spheroids. Janus spheroids are spheroids composed of two groups of cells juxtaposed next to each other so that each group essentially forms a hemisphere. This cellular patterning method opens new possibilities for understanding and manipulating interactions between different cell types in 3D cell culture. A Janus spheroid can be formed easily in the Perfecta3D Hanging Drop Plates by retrieving a spheroid from one well and then gently pipetting it into another well containing a spheroid, just like the removal and addition of liquid.

CellTracker Green-labeled and Red-labeled COS7 African green monkey kidney fibroblast cells were cultured separately in different wells to form spheroids. On day 3, a green spheroid was transferred into each hanging drop containing a red spheroid. Over the next few days, the two spheroids slowly aggregated together to form a single Janus spheroid with half green cells and half red cells (Figure 10).

The formation of Janus spheroids is unique to hanging drop plates, such as the Perfecta3D Hanging Drop Plates, which have an access hole large enough to allow the transfer of an additional spheroid with a pipet.

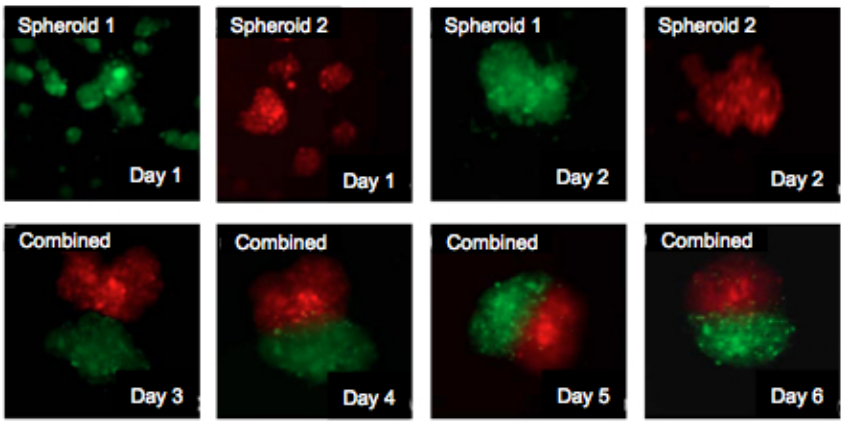
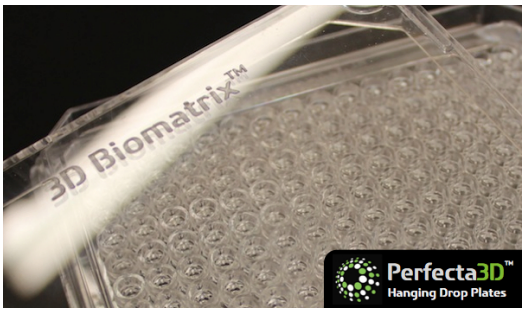


Figure 10. Example of a Janus spheroid using CellTracker Green-labeled and Red-labeled COS7 African green monkey kidney fibroblast cells.

Co-Cultures to Mimic the Tumor Microenvironment. The ability to generate multiple configurations of co-cultures is essential in modeling the tumor microenvironment. The inclusion of fibroblasts, endothelial cells, and other components of the tumor stroma will give the researcher a better and more realistic model of the carcinoma of interest. The three configurations of co-cultures demonstrated in the study above demonstrate that researchers can use spheroids grown in Perfecta3D Hanging Drop Plates to study the interactions between multiple cell types, as well as the migration of cells from a mixed, concentric, or hemispheric configuration.

Summary

New testing methods are imperative at early stages of drug development to reduce the expensive, late-stage failures due to efficacy and safety. Drug testing data, particularly in oncology, from *in vivo*-mimicking 3D cell cultures have the potential to play a pivotal role in early-stage decision-making about the probability of therapeutic candidates' success. The easy-to-use Perfecta3D Hanging Drop Plates are especially designed for spheroid growth and the high-throughput testing of anti-cancer compounds at the preclinical discovery phase. In a standard-size well-plate format, the Perfecta3D Hanging Drop Plates are simple to adopt and include in traditional liquid handling and assay methods.



Perfecta3D Hanging Drop Plates

Case of 8 Plates (HDP1384-8)

384-Well Format with Lid and Tray

Individually Packaged

Untreated

Polystyrene

Sterile

Visit www.3DBiomatrix.com to order.

"The Perfecta3D Hanging Drop Plates work well for formation of embryoid bodies (EBs) from induced pluripotent stem (iPS) cells and embryonic stem cells (ESCs). Overall, the EBs look great using these plates."

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3D Biomatrix
1600 Huron Parkway
Building 520, 2nd Floor
Ann Arbor, MI 48109-2590
USA

www.3DBiomatrix.com
service@3DBiomatrix.com
Phone: 734.272.4688
Fax: 734.818.1999

References

1. 2011 report, Medicines in Development for Cancer. Pharmaceutical Research and Manufacturers of America.

2. CenterWatch, FDA Approved Drugs for Oncology, http://www.centerwatch.com/drug-information/fda-approvals/drug-areas.aspx?AreaID=12.

3. Knight A, Systematic reviews of animal experiments demonstrate poor human clinical and toxicological utility. Altern Lab Anim 2007; 35(6): pp. 641-659.

4. de Boo J and Hendriksen C, Reduction strategies in animal research: A review of scientific approaches at the intra-experimental, supra-experimental and extra-experimental levels. Altern Lab Anim 2005; 33(4): pp. 369-377.

5. Pampaloni F, Stelzer EHK, and Masotti A, Three-dimensional tissue models for drug discovery and toxicology, Recent Patents on Biotechnology, Epub June 2009; 3(2): pp. 103-117.

6. Lin R-Z and Chang H-U, Recent advances in three-dimensional multicellular spheroid culture for biomedical research, Biotechnol. J. 2008, 3, pp. 1172–1184, DOI 10.1002/biot.200700228.

7. Gurski LA, Petrelli NJ, Jia X, and Farach-Carson MC, 3D matrices for anti-cancer drug testing and development, Oncology Issues 2010; 25: pp. 20-25.

8. Friedrich J, Seidel C, Ebner R and Kunz-Schughart LA, Spheroid-based drug screen: considerations and practical approach, Nat. Protoc., 2009; 4(3): pp. 309–324.

9. Kunz-Schughart LA, Freyer JP, Hofstaedter F and Ebner R, The use of 3-D cultures for high-throughput screening: the multicellular spheroid model, J. Biomol. Screening, 2004; 9(4): pp. 273–285.

10. Hsiao AY, Tung YC, Qu X, Patel LR, Pienta KJ and Takayama S, 384 hanging drop arrays give excellent Z-factors and allow versatile formation of co-culture spheroids. Biotechnology and Bioengineering, May 2012: Vol.109, Issue 5: pp. 1293-1304, DOI: 10.1002/bit.24399.

11. Tung YC, Hsiao AY, Allen SG, Torisawa Y, Ho M, Takayama S, High throughput 3D spheroid culture and drug testing using a 384 hanging drop array. Analyst 2011; 136: pp. 473-478, DOI:10.1039/C0AN00609B.

12. Sutherland RM, Cell and environment interactions in tumor microregions: the multicell spheroid model. Science 8 April 1988: Vol. 240, No. 4849: pp. 177-184, DOI: 10.1126/science.2451290.

13. Upreti M, Jamshidi-Parsian A, Koonce NA, Webber JS, Sharma SK, Asea AA, Mader MJ, Griffin RJ, Tumor-endothelial cell three-dimensional spheroids: new aspects to enhance radiation and drug therapeutics. Transl Oncol. 2011 Dec; 4(6): pp. 365-76. Epub 2011 Dec 1.