



White Paper.

Applications of JCRB cell lines in spheroid- based cancer research using FUJIFILM Wako Cell Cultureware,

PrimeSurface® Series



Overview

3D tumor spheroids are cancer cell aggregates formed by 3D cell culture *in vitro*.¹ The growth of spheroids from tumor cells mimics the growth of naturally occurring human tumors, as their extracellular matrix and network of cell-to-cell and cell-to-matrix interactions are similar to *in vivo* conditions and differ from the corresponding monolayer cultures.² In addition, spheroids are more stable than monolayer cultures and have a longer lifespan, making them useful for predicting the efficacy and toxicity of drugs in vivo

FUJIFILM Wako provides various cell lines from JCRB, which have high performance in 3D cell culture.

JCRB Cell Lines

JCRB (Japanese Collection of Research Bioresources) is one of the most comprehensive cell banks in the world for pure and applied science, spanning many fields of research across the globe.

FUJIFILM Wako has teamed up with the National Institute of Biomedical Innovation to bring you access to over 1,600 cell lines, which can be used not only as a monolayer cell culture, but also as a 3D cell culture. In Europe, we are the exclusive provider of JCRB's comprehensive catalog of cell lines.



JCRB cell line products in spheroid based research

JCRB No.	Cell-line Name	Profile
JCRB0182	KP-4	Human tumor cell line secreting parathyroid hor- mone-related peptide (PTHrP) established from pancreatic tumor
JCRB0198	OSC-19	Human squamous cell carcinoma cell line from tongue tumor with serum free cell culture
JCRB1054	Hep G2	Hepatoblastoma-derived (previously thought to be hepatocellular carcinoma-derived) cell line
JCRB1594	SK-OV-3/CMV-Luc	Luciferase stably expressing cell line (SK-OV-3; Human ovarian cancer cell line)
JCRB9094	DLD-1	Adenocarcinoma

Applications of JCRB cell lines

1. Toxicology and Drug Screening

Tumor spheroid models derived from JCRB0134 (MCF-7) cell lines are used as valuable models for toxicology and drug discovery. They retain their genome over time and can be used to perform drug screening and facilitate patient specific drug development.

2. Benefit of Variety of Ethnic Background in Patient Targeted Cancer Therapies

JCRB offers a highly representative panel of cell lines in terms of ethnic and gender diversity, which could improve the success rate in identifying effective cancer therapeutics in cell culture models and thereby reduce drug failures in diverse human populations.

JCRB offers a wide variety of cell line products. To see more information or full lineup of cell line products, please refer to the link:

https://labchem-wako.fujifilm.com/europe/cell_bank/index.html

JCRB cell lines related cell cultureware

FUJIFILM Wako provides PrimeSurface® for spheroid culture in regenerative medicine research, including drug efficacy testing and drug screening using 3D models.

1. PrimeSurface® Series

3D Cell Culturing for Drug Screening and Regenerative Medicine

PrimeSurface® is a pre-coated cultureware with a unique ultra hydrophilic polymer that enables the spontaneous spheroid formation of uniform size and shape. In addition, PrimeSurface® has high optical clarity, making it suitable for bright field imaging and confocal microscopy. This cultureware is useful in stem cell research, drug discovery & development, and tissue engineering & regenerative medicine.

We offer various types in U and V bottoms or clear and white colors that could give scientists a choice to form tighter spheroids for specific cell types.



Features

1. Easy formation of cell aggregates (spheroids)

Spheroids can be obtained easily just by seeding cells into a 96-well plate and culturing them statically.

2. Obtainable uniform spheroids

Prevention of cell adhesion to the culture surface allows spheroids to be cultured with a high rate of uniformity and formation.

3. No elution of surface treatment agent There is no need to worry when eluting of the treatment agent because the surface treatment is chemically bonded to the substrate surface.

4. Less stimulation to cells

It is ideal for cells sensitive to external factors due to the less stimulation from the culture surface.

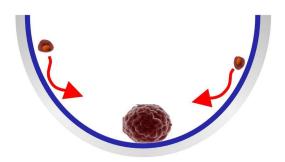


Figure 1 shows single spheroid can be formed in each well.

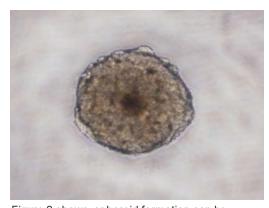
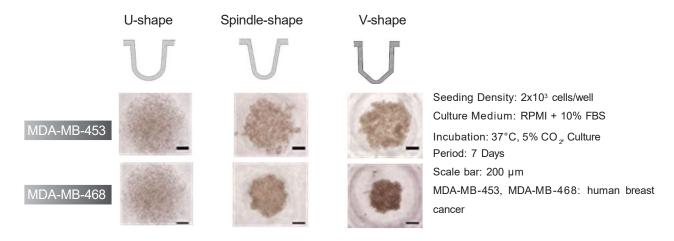


Figure 2 shows spheroid formation can be observed easily.

The inclination of the well bottom allows spheroid formation even in cells with poor cohesion such as MDA-MB-453 and MDA-MB-468.

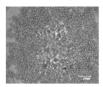
In total Fujiflm Wako offers three well bottom shapes of 96 well microplates.



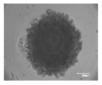
Data are provided by Nishio Lab., Dept. of Genome Bio. Kindai Univ. Faculty of Medicine.

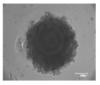
Principle of Spheroid Formation

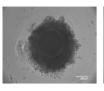
Time-lapse image of Human iPSCs Spheroid (EB: Embryoid Body) Formation













VIABILITY



Experimental Conditions

Culture plate: PrimeSurface® (629-01099)

Kind of cells: hiPSCs (DOI 10.1016/j.cell.2007.11.019)

Seeding density: 9.000 cells/well

Culture medium: DMEM/F12 + 20% (v/v) KSR + 1% (v/v) NEAA + L-Glutamine

 $(2 \text{ mM}) + \beta - \text{Mercaptethanol} (80 \mu\text{M}) + Y - 27632 (30 \mu\text{M})$

Culture environment: 5% CO₂, 37°C

Microscope: BioStudio (Corefront Co.), scale bar 200 µm

Application Example

5-FU NH Mode of action:

Is effective against any cell

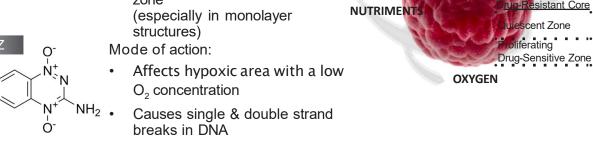
Affects the protein synthesis

Inhibits the DNA replication

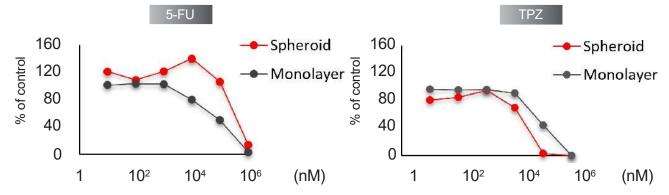
Damage in the proliferative zone (especially in monolayer structures)

Reduces DNA synthesis

Damage in the hypoxic zone (especially in Spheroid structures)



Comparison of cell viability between 5-FU and TPZ



TPZ showed stronger effect in Spheroid than Monolayer. This experiment suggest 3D drug efficacy test environment could be reproduced with

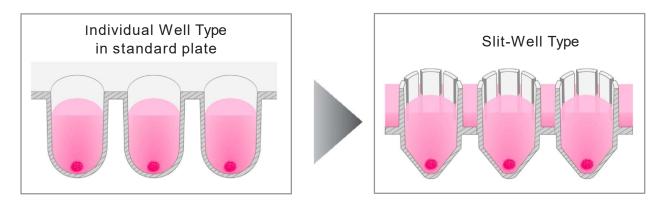
PrimeSurface®.

2. PrimeSurface® 96 Slit-well Plate

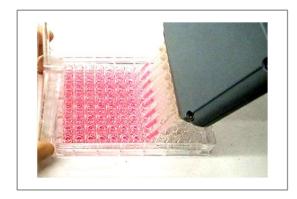
In addition to the standard PrimeSurface® portfolio, FUJIFILM Wako offers a new design of ultra-low attachment 3D plate to facilitate easy handling of media exchange without disturbing spheroid formation.

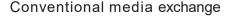
Cell culturing involves frequent media replacement to provide nutrition to growing cells. In a standard 96 well ultra low cell attachment plate, media aspiration or dispensing has to be done extremely carefully to avoid disturbing the unattached spheroid, making this a time consuming operation. With the introduction of PrimeSurface® 96 Slit-well Plate, changing the cell culture media for 96 well plates can be efficiently handled within one step dispensing or aspiration for all 96 wells decreasing the pipetting time by over 80% while minimizing the risk of spheroid damage.

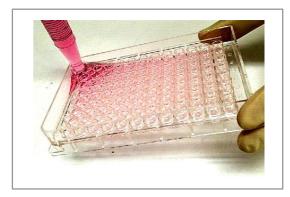
Slit-well structure for simultaneous delivery of cell culture medium to all 96 wells



Minimize media exchange time without disturbing spheroid formation







Easy one step media exchange by tilting the plate and aspirating from the corner.

PrimeSurface® Product Line Up

Wako Code	Product Name	Well Type	Well Bottom	Color	Package Size
627-01419	PrimeSurface® 24F Plate	24	Flat (1.8 cm²)	Clear	10 plates / case
635-21039	PrimeSurface® 96U Plate	96	Round	Clear	20 plates / case
621-01439	PrimeSurface® 96U Plate	96	Round	White	20 plates / case
628-01449	PrimeSurface® 384U Plate	384	Round	Clear	20 plates / case
625-01459	PrimeSurface® 384U Plate	384	Round	White	20 plates / case
622-01109	PrimeSurface® 96M Plate	96	Spindle	Clear	20 plates / case
629-01099	PrimeSurface® 96V Plate	96	V	Clear	20 plates / case
633-47839	PrimeSurface® 96 Slit- well Plate	96	Spindle	Clear	20 plates / case

Citations

- 1) Li W, et al.: Adv Healthc Mater. 2023 Jul; 12(18):e2202609.
- 2) Khaitan D, et al.: J Transl Med. 2006 Mar;4:12.

Online

 $Stem\ cell\ -\ \underline{http://www.sumibe.co.jp/english/product/s-bio/cell-culture/primesurface-96u/spec/list_01.html}$

Cancer research - http://www.sumibe.co.jp/product/s-bio/primesurface-proteo/primesurface-96u/spec/list_02.html

 $Other\ application\ -\ \underline{http://www.sumibe.co.jp/english/product/s-bio/cell-culture/primesurface-96u/spec/list_03.html}$

Distribution & Enquiries

Listed products are intended for laboratory research use only, and not to be used for drug, food or human use. JCRB Cell Bank is testing the cell lines for viruses pathogenic to humans as extensive as possible. However, there is the problem of detection limit and it is practically impossible to examine "all pathogens" as well as unidentified viruses. Therefore, the cell line should be handled as potentially biohazardous materials. Practically, the handling in accordance to biosafety level 2 is recommended. This does not mean that the cell line produces BSL-2 pathogens but is needed to avoid potential risk./ Please visit our online catalog to search for other products from FUJIFILM Wako; https://labchem-wako.fujifilm.com / This leaflet may contain products that can not be exported to your country due to regulations. / Bulk quote requests are welcome. Please contact us.

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