# **SPL Life Sciences**

# **Cell Culture Slide**

Catalogue Numbers 30101/30111/30121/30401/30501 30102/30112/30122/30402/30502 30104/30114/30124/30404/30504 30108/30118/30128/30408/30508 33101/33201/33301

# **Instruction for Use**



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# 1. Intended Use

SPL Life Sciences Cell Culture Slides are designed for cell culture based *in vitro* experiments such as cultivation, microscopic observation of mammalian cells.

SPL Life Sciences Cell Culture Slides provide optimized environment for culturing mammalian cells, maintaining their morphology and specific functions. It is recommended that cells cultured in SPL Life Sciences Cell Culture Slides are maintained in appropriate culture medium and culture grade chemicals

Cat. No.	Product	Quantity	Well Dimensions (diameter x width x height; mm)	Volume per Well ( <i>ml</i> )
30101/301 11/30121/ 30401/305 01	Cell Culture Slide 1 well	6 slides/tray Total 12 slides/box	26.50 x 52.40 x 11.30	2.5 ~ 5.5
30102/301 12/30122/ 30402/305 02	Cell Culture Slide 2well	6 slides/tray Total 12 slides/box	26.50 x 22.20 x 11.30.	1.2 ~ 2.5
30104/301 14/30124/ 30404/305 04	Cell Culture Slide 4well	6 slides/tray Total 12 slides/box	23.00 x 11.40 x 11.30	0.5 ~ 1.3
30108/301 18/30128/ 30408/305 08	Cell Culture Slide 8well	6 slides/tray Total 12 slides/box	11.50 x 11.40 x 11.30	0.2 ~ 0.6
33101/332 01/33301	Cell Culture Slide Hybridwell	6 slides/tray Total 12 slides/box	26.50 x 52.40 x 22.00	2.5 ~ 5.5

# 2. Materials Provided

\* Each Cell Culture Dish consists of polystyrene lid and chamber, borosilicate glass, and poly ethylene holder.

# **3. Quality Control**

- Every lot of SPL Life Sciences Cell Culture Slide is tested for strict quality control (according to AQL) to ensure lot-to-lot consistency.
- Functional performance has been validated by culturing commercially available cell lines in appropriate culture media supplemented with 10% fetal bovine serum (FBS) and 1% penicillin-streptomycin. (*Refer to Bench Test Reports*)

## 4. Materials Required But Not Provided

- Cell culture media and necessary components
- Immunofluorescence staining reagents
- Adjustable, automatic micropipettes with disposable tips

### 5. Related SPL Products, Not Supplied

- SPL Serological Pipettes (Cat. No. 91001/91002/91005/91005/91010/91025/91050)
- SPL Conical Tubes (Cat. No. 50015/50050/50115/50150/51015/51115/50040/50250)

#### 6. Precautions

- A. Storage: SPL Life Sciences Cell Culture Slides for cell culture should be stored at room temperature in the original packaging protected from direct sunlight in order to guarantee shelf life. All slides should be used for cell culturing purposes once the package is opened.
- B. All cell culture-related procedures should be performed under aseptic conditions.

### 7. General Guidelines for Use

**7.1. Protocol of general culture of adherent mammalian cells on SPL Cell Culture Slides** The following exemplary procedure is optimized for seeding adherent mammalian cells on cell culture slides, initially grown on conventional cell culture dishes.

Procedure may vary depending upon the cell line used and the specific conditions and health of the cells that are maintained. You should optimize conditions for individual systems.

- (1) Aspirate and discard cell culture media from the culture dishes.
- (2) Gently wash cells using a balanced salt solution without calcium and magnesium (e.g. DBPS).

Note: The wash step removes any traces of serum, calcium, and magnesium that would inhibit the action of the dissociation reagent.

- (3) Aspirate and discard wash solution from the culture dishes.
- (4) Gently add the pre-warmed dissociation reagent such as trypsin to the dish to cover the cell layer and incubate the culture dishes in the humidified 37°C incubator for several minutes (approx. 2-3 minutes).
- (5) Observe the cells under the microscope for detachment. Increase incubation time if needed. You may gently tap the side of the dishes to accelerate cell detachment.
- (6) When cells have detached (approx. 90%), gently add pre-warmed complete growth medium at twice the volume used for the dissociation reagent to end the dissociation reaction.

- (7) Transfer the cells to a conical tube (15 ml) and centrifuge at recommended rpm and minutes.
- (8) Aspirate supernatant and re-suspend the cell pellet in pre-warmed complete growth medium.
- (9) Determine the total number of cells and percent viability using a cell counter and Trypan blue exclusion.
- (10) Dilute cell suspension to appropriate seeding density and insert desired volume into new cell culture dishes.
- (11) Place cells back into the incubator.

Well Type	Grow Area / Well	Working Volume / Well	Recommended Cell Seeding Concentration
1well	<b>9.40</b> cm <sup>2</sup>	2.50 - 5.50 ml	
2well	$4.55 \text{ cm}^2$	1.20 - 2.50 ml	
4well	$2.13 \text{ cm}^2$	0.50 – 1.30 ml	5 x 10 <sup>4</sup> ~ 1 x 10 <sup>5</sup> / ml
8well	<b>0.98</b> cm <sup>2</sup>	0.20 – 0.60 ml	
Hybridwell <sup>TM</sup>	9.00 cm <sup>2</sup>	2.50 – 5.50 ml	

#### General culture conditions of SPL Cell Culture Slides

#### 7.2. Protocol of immuno-fluorescence staining for observation of mammalian cells

The following exemplary procedure is optimized for staining adherent mammalian cells initially grown on cell culture slides.

Procedure may vary depending upon the cell line used and the specific conditions and health of the cells that are maintained. You should optimize conditions for individual systems.

- (12) Aspirate and discard cell culture media from the culture slides.
- (13) Gently wash cells using a balanced salt solution without calcium and magnesium (e.g. DBPS).
- (14) Gently add fixing solution (e.g. paraformaldehye) and incubate for 10 to 15 minutes at room temperature.
- (15) Gently wash cells using a balanced salt solution without calcium and magnesium (e.g. DBPS) several times.
- (16) Add permeabilizing agent (e.g. Trion X-100) and incubate for 10 to 15 minutes at cooled temperature.
- (17) Gently wash cells using a balanced salt solution without calcium and magnesium (e.g. DBPS) several times.
- (18) Add selected antibody solution for observation purposes to cells. Incubate for 30 minutes ~ 12 hours as directed.
- (19) Gently wash cells using a balanced salt solution without calcium and magnesium (e.g. DBPS) several times.

- (20) Repeat step 7-8 for each antibody solution.
- (21) Gently remove holder from the slides and separate chambers from the sample surface.
- (22) Add a small volume of mounting solution to minimize bleaching.
- (23) Cover and seal with optimal grade cover glass.
- (24) Perform fluorescence microscopy.



FLux dissociating method

Using a pair of tweezers, gently push the film outwards from the inside.
Gently peel off the film from the corner.

## 8. Stability

SPL Life Sciences Cell Culture Slides are stable for at least 5 years from date of shipment when stored appropriately.

### 9. Customer and Technical Services

For technical assistance, contact SPL R&D Center at: Tel: +82-31-533-4800; Fax: +82-31-533-1430; e-mail: <u>spl@ispl.co.kr</u>

To place an order, contact your local distributor or Tel: +82-31-533-4800; Fax: +82-31-533-1430; e-mail: <u>business@ispl.co.kr</u>

Visit our website <u>www.spllifesciences.com</u> for additional information on products including:

- Product Literature
- Bibliography
- List of Related Products

# **10. References**

- [1] Jeong et al., "The anticancer effects of Saccharina japonica on 267B1/K-ras human prostate cancer cells", **International Journal of Oncology**, 2012, vol. 41, pp 1789-1797.
- [2]

Symbol	Meaning
(€	European conformity
REF	Catalog number
IVD	In vitro diagnostic medical device
LOT	Batch code
8	Do not reuse
A44	Manufacturer
8	Use by YYYY-MM-DD- or YYY-DD
EC REP	Authorised* Representative in the European Community
-	SPL LIFE SCIENCES SPL LIFE SCIENCES Co., Ltd 26, Geumgang-ro 2047 beon-gil, Naechon-Myeon, Pocheon-si, Gyeonggi-do
	11192, Korea
EC REP	Advena Ltd. Pure Offices, Plato Close, Warwick CV34 6WE UK.

# TABLE OF SYMBOLS