ProductInformation

SIGMAScreen™
Collagen I Coated Multiwell Plates

Product Codes:

S 8813	96 well; Clear/Clear Bottom
S 3190	96 well; Black/Clear Bottom
S 3315	96 well; White/Clear Bottom
S 3440	384 well; Clear/Clear Bottom
S 3690	384 well; Black/Clear Bottom
S 3565	384 well; White/Clear Bottom

Product Description

Certain cell lines require an extracellular matrix (ECM) on the growth surface for enhanced cell attachment and proliferation.[1] Collagen I, a fibrous protein and major component of the ECM, is commonly used as an attachment factor.[3] Cells attach firmly to Collagen I coated plates via integrin surface receptors, thereby promoting attachment and spreading.[2,4]

Most often, the presence of serum in culture media provides the necessary growth and attachment factors for optimal culture maintenance. However, the presence of these factors may also influence experimental results and, therefore, need to be reduced or omitted. SIGMAScreen Collagen I Coated Multiwell Plates offer a growth surface that is able to promote cell plating efficiency, improve morphology, and enhance cell proliferation in low-serum or serum-free conditions.

SIGMAScreen Collagen I Multiwell Coated Plates are coated with rat tail collagen (Product Code C 7661) and tested for cell attachment and spreading in serum-free Dulbecco's Modified Eagle's Medium using HT-1080 human fibrosarcoma cells. SIGMAScreen Collagen I Multiwell Coated Plates are manufactured in a highly controlled environment. Each lot is tested for the presence of endotoxins, bacteria, and fungi.

Applications

- Cell culture in low serum or serum-free media
- Promotion of attachment and spreading of many normal and transformed cells including epithelial, endothelial, muscle, and ovarian cells
- Improved survival of primary cultures[5]
- Evaluate the effects of Collagen I on cell genotype and phenotype

The following is a partial list of cells that have been reported to be successfully cultured on Collagen I coated plates:

Epithelial Cells

- Human Nasal Airway epithelial cells[6]

Endothelial Cells

- Primary Human Umbilical Vein Endothelial Cells (HUVEC)[5]
- Bovine aortic endothelial cells[7]

Muscle Cells

- Rat and human smooth muscle cells[8]
- Transfected MM41 skeletal myoblasts[9]

Hepatocytes

- Primary rat hepatocytes[10]
- Primary human hepatocytes[11]

Neuronal Cells

- Rat sympathetic neurons[12]

Other cells:

- Primary spermatogenic cells [13]
- PC 12[14] (rat pheochromocytoma cells)
- COS-7 cells[15]

Precautions and Disclaimer

For research use only, not intended for use in diagnostic procedures.

Storage/Stability

The 5 plate package is in a resealable bag with individual lids and desiccant.
The 100 plate package is 5 sleeves of 20 plates, also with desiccant and individual lids.

For optimal performance, the unopened product should be stored in a dry place at 2-8 °C. The product may be stored at room temperature for up to three months. The product should not be exposed to temperatures above 50 °C.

Refer to the Certificate of Analysis for the expiration date. The Certificate of Analysis can be obtained either from the Sigma-Aldrich website (www.sigma-aldrich.com) or by contacting Sigma Technical Service at 800-325-5832. Once opened, the product should be used promptly.

Procedure

Cell Attachment/Proliferation Assay

1. Using standard protocols, harvest the cells.

2. Centrifuge the harvested cells at 200 to 300 x *g* for five minutes. Discard the supernatant.

3. Resuspend the cell pellet in fresh, complete medium.

4. Count the cells. Dilute in complete medium for a cell stock suspension of 1 to 50 x 10^5 cells/ mL.

5. Open the plate packaging under aseptic conditions. Pipette 100 µL of the cell stock suspension into each well of the 96 well format or 25 µL of the cell stock suspension into each well of the 384 well format. If necessary, add appropriate quantities of test substances to the wells.

6. Cover the plates with lids. Place in a 37 °C, 5% CO_2, humidifying incubator for the desired amount of time.

7. Observe the cell morphology with a microscope.

8. Gently aspirate to remove the medium and non-adherent cells from the wells. Gently wash with 100 µL Dulbecco's PBS (Product Code D 5773) per well for the 96 well format or 50 µL Dulbecco's PBS per well for the 384 well format . Repeat this wash step, using care not to dislodge the cells.

9. Using standard protocols, lyse the cells. A lysis reagent such as CelLytic™-M Mammalian Cell Lysis/ Extraction Reagent (Product Code C 2978) may be used.

10. Add 175 µL of Bradford Reagent (Product Code B 6916) to each well for the 96 well format or 50 µL Bradford Reagent to each well for the 384 well format . Mix thoroughly and incubate at room temperature for 10-20 minutes.

11. Read the absorbance at 595 nm. Calculate the protein content of the attached cells by comparing the sample absorbance with an appropriate standard curve.

References

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Product Profile

Table 1 – Plate Features

Property	96 Well Plate	384 Well Plate
Plate composition	Tissue culture polystyrene	Tissue culture polystyrene
Lid	Yes	Yes
Well configuration	Flat bottom/round	Flat bottom/round
Well width	6.4 mm	2.8 mm
Well depth	11 mm	11 mm
Maximum recommended working volume, per well	250 µL	75 µL
Recommended culture volume, per well	100 µL	25 µL
Growth area, per well	32 mm^2	8 mm^2
Coating coefficient of variability, well-to-well	≤ 10%	≤ 10%

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