Product Manual

CytoSelect™ 48-Well Cell Adhesion Assay (Fibronectin-Coated, Colorimetric Format)

Catalog Number

CBA-050 **48 assays**

FOR RESEARCH USE ONLY
Not for use in diagnostic procedures

Introduction

Cell adhesion is a complex process involved in embryogenesis, migration/invasion, tissue remodeling, and wound healing. To perform these processes, cells adhere to extracellular matrix components (via adhesion receptors), forming complexes with components of the cytoskeleton that ultimately affect cell motility, differentiation, proliferation, and survival. The Cell Biolabs CytoSelect™ Cell Adhesion Assay Kit provides a rapid, quantitative method for evaluating cell adhesion. The kit contains sufficient reagents for the evaluation of 48 samples (40 Human Fibronectin-coated wells, 8 BSA-coated wells).

Assay Principle

The CytoSelect™ Cell Adhesion Assay Kit utilizes a Fibronectin-coated 48-well plate (see Adhesion Plate Layout below). First, cells are seeded onto the coated substrate, where the adherent cells are captured. Next, unbound cells are washed away, and the adherent cells are fixed/stained. Finally, the stain is extracted and quantified colorimetrically.

Related Products

1. CBA-051: CytoSelect™ 48-Well Cell Adhesion Assay (Fibronectin-Coated, Fluorometric Format)
2. CBA-070: CytoSelect™ 48-Well Cell Adhesion Assay (ECM Array, Colorimetric Format)
3. CBA-071: CytoSelect™ 48-Well Cell Adhesion Assay (ECM Array, Fluorometric Format)
4. CBA-210: CytoSelect™ Leukocyte-endothelium Adhesion Assay

Kit Components

1. Fibronectin Adhesion Plate (Part No. 105001): One 48-well plate containing 40 Human Fibronectin-coated wells and 8 BSA-coated wells (see layout below)
2. Cell Stain Solution (Part No. 11002): One Bottle – 10.0 mL
3. Extraction Solution (Part No. 11003): One Bottle – 10.0 mL

Adhesion Plate Layout

The layout below indicates the location of wells coated with Fibronectin and those coated with BSA.

	1	2	3	4	5	6	7	8
A	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin
B	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin
C	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin
D	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin
E	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin	Fibronectin
F	BSA	BSA	BSA	BSA	BSA	BSA	BSA	BSA

Materials Not Supplied

1. Cell culture medium
2. Serum free medium, such as DMEM containing 0.5% BSA, 2 mM $CaCl_2$ and 2 mM $MgCl_2$
3. Cell culture incubator (37ºC, 5% CO_2 atmosphere)
4. 1X PBS containing 2 mM $CaCl_2$ and 2 mM $MgCl_2$
5. Light microscope
6. 96-well microtiter plate
7. Microtiter plate reader

Storage

Store all kit components at 4ºC.

Assay Protocol

1. Under sterile conditions, allow the Fibronectin Adhesion Plate to warm up at room temperature for 10 minutes.
2. Prepare a cell suspension containing 0.1-1.0 x 10^6 cells/ml in serum free media. Agents that inhibit or stimulate cell adhesion can be added directly to the cell suspension.
3. Add 150 µL of the cell suspension to the inside of each well (BSA-coated wells are provided as a negative control).
4. Incubate for 30-90 min in a cell culture incubator.
5. **Carefully** discard or aspirate the media from each well (**Note: Do not allow wells to dry**). Gently wash each well 4-5 times with 250 µL PBS.

6. Aspirate the PBS from each well and add 200 µL of Cell Stain Solution. Incubate for 10 minutes at room temperature.
7. Discard or aspirate the Cell Stain Solution from the wells. Gently wash each well 4-5 times with 500 µL deionized water.
8. Discard the final wash and let the wells air dry.
9. Add 200 µL of Extraction Solution per well, and then incubate 10 minutes on an orbital shaker.
10. Transfer 150 µL from each extracted sample to a 96-well microtiter plate and measure the OD 560nm in a plate reader.

Example of Results

The following figures demonstrate typical results with the CytoSelect™ 48-Well Cell Adhesion Assay Kit. One should use the data below for reference only. This data should not be used to interpret actual results.

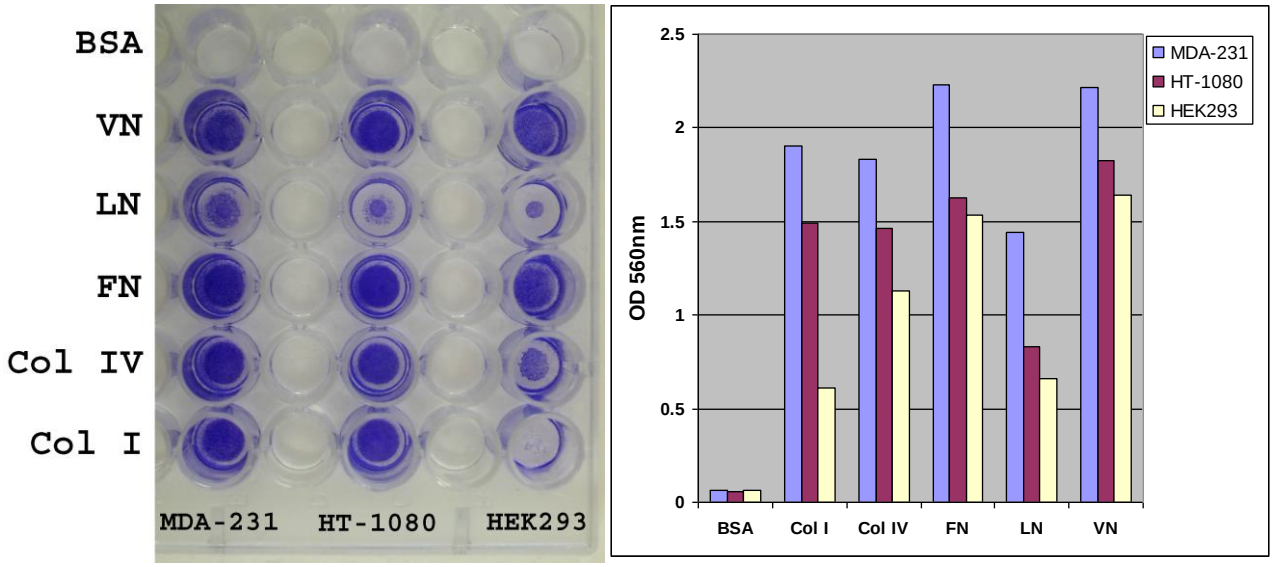

Figure 1. ECM-mediated Cell Adhesion. Serum starved cells were allowed to attach to ECM-coated 48-well plate for 1 hr at 100,000 cells/well. Adherent cells were stained (left panel picture) and quantified at OD 560nm after extraction (right panel figure).

References

1. Hynes, R. O. (1992) *Cell* **69**:11-25.
2. Schwartz, M. A., Schaller, M. D. and Ginsberg, M. H. (1995) *Annu. Rev. Cell Dev. Biol.* **11**:549-599.

Recent Product Citations

1. Lee, Y. Y. et al. (2023). The anti-platelet activity of panaxatriol fraction and panaxatriol fraction of Korean Red Ginseng in vitro and ex vivo. *J Ginseng Res*. doi: 10.1016/j.jgr.2023.03.003.
2. Kim, J.Y. et al. (2022). ARL6IP5 reduces cisplatin-resistance by suppressing DNA repair and promoting apoptosis pathways in ovarian carcinoma. *Cell Death Dis*. **13**(3):239. doi: 10.1038/s41419-022-04568-4.
3. Shin, J.H. et al. (2021). Derrone Inhibits Platelet Aggregation, Granule Secretion, Thromboxane A2 Generation, and Clot Retraction: An In Vitro Study. *Evid Based Complement Alternat Med*. **2021**:8855980. doi: 10.1155/2021/8855980.
4. Shin, J.H. et al. (2021). Antiplatelet effect of cudraxanthone B is related to inhibition of calcium mobilization, αIIbβ3 activation, and clot retraction. *Appl Biol Chem*. doi: 10.1186/s13765-020-00575-1.
5. Schweitzer, K.S. et al. (2020). IGSF3 mutation identified in patient with severe COPD alters cell function and motility. *JCI Insight*. doi: 10.1172/jci.insight.138101.
6. Kwon, H.W. (2018). 20(S)-ginsenoside Rg3 inhibits glycoprotein IIb/IIIa activation in human platelets. *J Appl Biol Chem*. **61**:257−265. doi: 10.3839/jabc.2018.037.
7. Li, Y. et al. (2018). Visualization and Quantification of Sortase Activity at the Single-Molecule Level via Transpeptidation-Directed Intramolecular Förster Resonance Energy Transfer. *Anal Chem*. **90**(21):13007-13012. doi: 10.1021/acs.analchem.8b03716.
8. Irfan, M. et al. (2018). Ginsenoside-Rp3 inhibits platelet activation and thrombus formation by regulating MAPK and cyclic nucleotide signaling. *Vascul Pharmacol*. **109**:45-55. doi: 10.1016/j.vph.2018.06.002.
9. Ma, D. M. et al. (2016). SDF-1/CXCR7 axis regulates the proliferation, invasion, adhesion, and angiogenesis of gastric cancer cells. *World J Surg Oncol.* **14**:256.
10. Li, Y. et al. (2016). High stoichiometry phosphorylation of Talin at T144/T150 or S446 produces contrasting effects on Calpain-mediated Talin cleavage and cell migration. *J Cancer*. **7**:1645-1652.
11. Jiang, F. et al. (2015). CYP3A5 functions as a tumor suppressor in hepatocellular carcinoma by regulating mTORC2/Akt signaling. *Cancer Res*. 75:1470-1481.
12. Chen, L. et al. (2015). Both mTORC1 and mTORC2 are involved in the regulation of cell adhesion. *Oncotarget*. **6**:7136-7150.
13. Cervera, A.M. et al. (2008). Cells silenced for SDHB expression display characteristic features of the tumor phenotype. *Cancer Res.* **68**:4058-4067.

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