Datasheet for ABIN2344859

CytoSelect™ 24-well Cell Invasion Assay, Colorimetric

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Overview

Quantity:	12 tests
Reactivity:	Mammalian
Application:	Cellular Assay (CA)

Product Details

Brand:	CytoSelect™
Sample Type:	Serum, Cell Samples
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Characteristics:	CytoSelect™ Cell Invasion Assay Kit utilizes basement membrane-coated inserts to assay the invasive properties of tumor cells. It contains sufficient reagents for the evaluation of 12 samples.
Components:	1. ECM Invasion Chamber Plate : One 24-well plate containing 12 ECM-coated cell culture inserts. 2. Cell Stain Solution : One 10 mL bottle 3. Extraction Solution : One 10 mL bottle 4. Cotton Swabs: 40 each 5. Forceps: One each
Material not included:	1. Invasive cell lines 2. Cell culture medium 3. Serum free medium, such as DMEM containing 0.5 % BSA, 2 mM $CaCl_2$ and 2 mM $MgCl_2$ 4. Cell culture incubator (37 °C, 5 % CO_2 atmosphere) 5. Light microscope

Product Details

6. 96-well microtiter plate
7. Microtiter plate reader

Target Details

Background:	The ability of malignant tumor cells to invade normal surrounding tissue contributes in large part to the significant morbidity and mortality of cancers. Invasiveness requires several distinct cellular functions including adhesion, motility, detachment, and extracellular matrix proteolysis. Metastatic cells produce many proteolytic enzymes (e.g. lysosomal hydrolysates, collagenases, plasminogen activators) while the expression of certain cell surface protease receptors is also increased.

Application Details

Application Notes:	Optimal working dilution should be determined by the investigator.
Comment:	• Fully quantify cell invasion with no manual cell counting • Plate inserts are precoated with ECM basement membrane
Plate:	Pre-coated
Protocol:	The CytoSelect™ Cell Invasion Assay Kit contains polycarbonate membrane inserts (8 µm pore size) in a 24-well plate. The upper surface of the insert membrane is coated with a uniform layer of dried basement membrane matrix solution. This basement membrane layer serves as a barrier to discriminate invasive cells from non-invasive cells. Invasive cells are able to degrade the matrix proteins in the layer, and ultimately pass through the pores of the polycarbonate membrane. Finally, the cells are removed from the top of the membrane and the invaded cells are stained and quantified.
Assay Procedure:	1. Under sterile conditions, allow the invasion chamber plate to warm up at room temperature for 10 minutes. 2. Rehydrate the basement membrane layer of the cell culture inserts by adding 300 µL of warm, serum-free media to the inner compartment. Incubate at room temperature for 1 hour. 3. Prepare a cell suspension containing 0.5-1.0 x 106 cells/mL in serum free media. Agents that inhibit or stimulate cell invasion can be added directly to the cell suspension. Note: Overnight starvation may be performed prior to running the assay 4. Carefully remove the rehydration medium (step 2) from the inserts without disturbing the basement membrane layer. Note: It will not affect the assay performance if a small amount of rehydration medium is left in the compartment 5. Add 500 µL of media containing 10 % fetal bovine serum or desired chemoattractant(s) to the lower well of the invasion plate.

Application Details

6. Add 300 µL of the cell suspension solution to the inside of each insert.
7. Incubate for 24-48 hours at 37 °C in 5 % CO2 atmosphere.
8. Carefully aspirate the media from the inside of the insert. Wet the ends of 2-3 cotton-tipped swabs with water, flatten the ends of the swabs by pressing them against a clean hard surface, and gently swab the interior of the inserts to remove non-invasive cells. Take care not to puncture the polycarbonate membrane. Be sure to remove cells on the inside perimeter of the insert.
9. Transfer the insert to a clean well containing 400 µL of Cell Stain Solution and incubate for 10 minutes at room temperature.
10. Gently wash the stained inserts several times in a beaker of water. Allow the inserts to air dry.
11. (optional) Count invasive cells with a light microscope under high magnification objective, with at least three individual fields per insert.
12. Transfer each insert to an empty well, adding 200 µL of Extraction Solution per well, then incubating 10 minutes on an orbital shaker.
13. Transfer 100 µL from each sample to a 96-well microtiter plate and measure the OD 560nm in a plate reader. 4

Restrictions:	For Research Use only

Handling

Storage:	4 °C
Storage Comment:	Store all components at 4°C.

Publications

Product cited in:

Rodríguez-Mateo, Torres, Gutiérrez, Pintor-Toro: "Downregulation of Lnc-Spry1 mediates TGF-β-induced epithelial-mesenchymal transition by transcriptional and posttranscriptional regulatory mechanisms." in: **Cell death and differentiation**, Vol. 24, Issue 5, pp. 785-797, (2017) (PubMed).

Huang, Luo, Han, Huang, Tang, Wu: "MiR-223/PAX6 Axis Regulates Glioblastoma Stem Cell Proliferation and the Chemo Resistance to TMZ via Regulating PI3K/Akt Pathway." in: **Journal of cellular biochemistry**, (2017) (PubMed).

Ma, Yu: "Over-Expression of TBL1XR1 Indicates Poor Prognosis of Serous Epithelial Ovarian Cancer." in: **The Tohoku journal of experimental medicine**, Vol. 241, Issue 3, pp. 239-247, (2017) (PubMed).

Oba, Nakahara, Hashimoto-Hachiya, Liu, Abe, Hagihara, Yokomizo, Furue: "CD10-Equipped

Melanoma Cells Acquire Highly Potent Tumorigenic Activity: A Plausible Explanation of Their Significance for a Poor Prognosis." in: **PLoS ONE**, Vol. 11, Issue 2, pp. e0149285, (2016) (PubMed).

Slusser-Nore, Larson-Casey, Zhang, Zhou, Somji, Garrett, Sens, Dunlevy: "SPARC Expression Is Selectively Suppressed in Tumor Initiating Urospheres Isolated from As+3- and Cd+2-Transformed Human Urothelial Cells (UROtsa) Stably Transfected with SPARC." in: **PLoS ONE**, Vol. 11, Issue 1, pp. e0147362, (2016) (PubMed).

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