

Datasheet for ABIN2344861

CytoSelect™ 24-well Collagen Cell Invasion, Colorimetric



[Go to Product page](#)

3 Publications

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™ Collagen Cell Invasion Assay Kit utilizes Bovine Type I Collagen-coated inserts to assay the invasive properties of tumor cells. It contains sufficient reagents for the evaluation of 12 samples.
Components:	<ol style="list-style-type: none"> 1. Collagen Invasion Chamber Plate : One 24-well plate containing 12 collagen-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:	<ol style="list-style-type: none"> 1. Invasive cell lines 2. Cell culture medium 3. Serum free medium, such as DMEM containing 0.5 % BSA, 2 mM CaCl₂ and 2 mM MgCl₂ 4. Cell culture incubator (37 °C, 5 % CO₂ atmosphere) 5. Light microscope

Product Details

6. 96-well microtiter plate
7. Microtiter plate reader 3

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 Collagen I gel layer

Plate: Pre-coated

Protocol: The CytoSelect™ Collagen 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 Bovine Type I Collagen matrix. This collagen matrix layer serves as a barrier to discriminate invasive cells from non-invasive cells. Invasive cells are able to degrade the collagen matrix 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 collagen invasion chamber plate to warm up at room temperature for 10 minutes.
2. Rehydrate the collagen layer of the cell culture inserts by adding 300 µL of warm, serum-free media to the inner compartment. Incubate at room temperature for 30 minutes.
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 250 µL of rehydration medium (step 2) from the inserts without disturbing the collagen layer (leaving 50 µL inside).
5. Add 250 µL of the cell suspension solution to the inside of each insert.
6. Add 500 µL of media containing 10 % fetal bovine serum or desired chemoattractant(s) to the lower well of the invasion plate.

Application Details

7. Incubate for 12-24 hours in a cell culture incubator.
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 μ L 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 μ L of Extraction Solution per well, then incubating 10 minutes on an orbital shaker.
13. Transfer 100 μ L 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: Loperfido, Jarmin, Dastidar, Di Matteo, Perini, Moore, Nair, Samara-Kuko, Athanasopoulos, Tedesco, Dickson, Sampaolesi, VandenDriessche, Chuah: "piggyBac transposons expressing full-length human dystrophin enable genetic correction of dystrophic mesoangioblasts." in: **Nucleic acids research**, Vol. 44, Issue 2, pp. 744-60, (2016) ([PubMed](#)).

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Noh, Maze, Zhao, Xiang, Wenderski, Lewis, Shen, Li, Allis: "ATRX tolerates activity-dependent histone H3 methyl/phos switching to maintain repetitive element silencing in neurons." in: **Proceedings of the National Academy of Sciences of the United States of America**, Vol. 112, Issue 22, pp. 6820-7, (2015) ([PubMed](#)).