Fibronectin Cell Adhesion Assay

Cat. No. 8018
48 assays

Introduction

Cell adhesion plays an important role in cellular communication and regulation, and is of fundamental importance in the development and maintenance of tissues. Fibronectin is a high-molecular-weight extracellular matrix glycoprotein involved in many cellular processes, including tissue repair, embryogenesis, blood clotting, and cell migration/adhesion. The ScienCell™ Fibronectin Cell Adhesion Assay is designed for the rapid, quantitative and reliable measurement of cell adhesion to fibronectin. The kit is composed of a 48-well plate pre-coated with fibronectin, as well as BSA as negative controls (see plate format in Figure 1). Cells are cultured in the pre-coated wells for a desired period of time, then unbounded cells are washed away, and the adhered cells are fixed and stained, followed by an extraction step which leads to dye elution from stained cells into the supernatant. Thus cell adhesion can be quantified using a colorimetric ELISA plate reader at 595 nm.

Kit Components

Cat. No.	# of vials	Reagent	Volume	Storage
8018a	1	Fn & BSA coated 48-well plate	N/A	4°C
8018b	1	Staining Solution	10 ml	4°C
8018c	1	Extraction Solution	10 ml	4°C

Quality Control

Human Vessel Umbilical Endothelial Cells (Cat. No. 8000, ScienCell™) are cultured in the pre-coated 48-well plate. A linear relationship can be observed between signal produced (OD_{590nm}) and the number of cells, as shown in Figure 2.

Procedures

1. Under sterile conditions, allow the pre-coated 48-well plate to warm to room temperature and rinse once with PBS.
2. Seed cells of interest into the 48-well plate and culture for a desired period of time (at least 30-90 minutes) at 37°C.
3. After the culture is done, remove culture medium and rinse cells with PBS for 3-5 times.
4. Add 200 µl/well of freshly diluted 0.1% glutaraldehyde in PBS, fix for 10 minutes at room temperature. Then discard fixing solution and rinse cells 3 times with PBS.
5. Add 200 µl/well of Staining Solution, incubate for 30 minutes at room temperature on an orbital shaker.
6. After the staining is done, wash plate with DI water for 3-5 times. Pull off remaining wash water, invert plate onto an absorbent diaper pad and let the wells air dry.
7. Add 200 µl/well of Extraction Solution, incubate for 3-5 minutes and read OD_{595nm} using an ELISA plate reader.

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

Figure 1. Layout of the pre-coated plate.

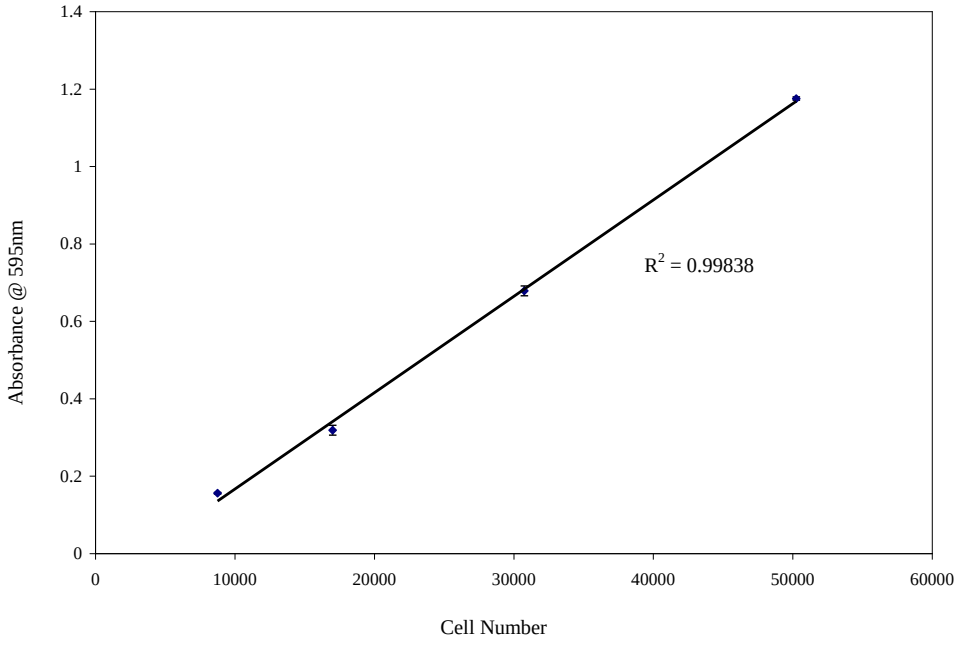

Figure 2. Correlation between cell number and absorbance at 595 nm shows linearity for human vessel umbilical endothelial cells.