

## BD BioCoat™ Angiogenesis System: Endothelial Cell Migration

### Frequently Asked Questions

#### Can I use VEGF or bFGF as a chemoattractant instead of serum?

Yes. For best results, use serum-free endothelial cell medium supplemented with 0.1% BSA, (*Cat. No. 354331*) without the addition of growth factors, in both the apical and basolateral chambers. The exact concentrations of VEGF and bFGF will need to be determined empirically for your exact experimental conditions. However, a good starting point is 10 ng/ml for VEGF or 50 ng/ml for bFGF.

#### What are the recommended seeding densities for the 96- and 24-Multiwell systems?

Seeding density is dependent upon the migratory ability of a particular cell. To get adequate signal with a less migratory cell you need to seed at a higher density. The reverse is true if the cell is highly migratory. A good starting point is between 50,000-100,000 cells for the 24-Multiwell system and 12,500-25,000 for the 96-Multiwell system. You will need to optimize for your particular cell and experimental conditions.

#### Can I use this system to test both pro- and anti-angiogenic agents?

Yes. This system can be used to screen both pro- and anti-angiogenic compounds. For best results, use endothelial cell media supplemented with 0.1% BSA, (*Cat. No. 354331*) without growth factor supplementation, in the apical and basal compartments. The exact cell seeding densities, assay time, and compound concentrations will have to be determined through experimentation.

#### What template should I use for the 96-Multiwell system when I set up my plate reader?

For more information regarding plate reader set up and template loading, refer to Technical Bulletin #436 located on our website at [www.bdbiosciences.com/discovery\\_labware](http://www.bdbiosciences.com/discovery_labware) or contact Technical Services at 800.343.2035.

#### Do the BD BioCoat™ 24- and 96-Multiwell Angiogenesis Systems: Endothelial Cell Migration come with an extra plate for labeling the cells?

An additional 24- or 96-well plate (*Cat. Nos. 351147 and 353928*, respectively) is needed to label the cells upon the completion of the assay.

#### Can I use a standard BD Falcon™ 96-well plate to label my cells after my cells have migrated?

No. Standard BD Falcon 96-well plates are not compatible. The appropriate plate for use with these inserts is the BD Falcon 96-Square Well, Flat Bottom Plate and Lid (*Cat. No. 353928*).

*Note: The BD Falcon 96-Square Well, Angled Bottom Plate (Cat. No. 353925) is NOT recommended for use with BD BioCoat inserts.*

#### Is there a recommended passage level for HUVEC cells?

The migratory competence of Primary HUVECs varies depending upon the individual from whom they are derived and passage level. As a result, it is recommended that HUVECs used in assays that depend upon their migratory ability be prescreened for this activity and used at a low passage number. Passage 6 or lower is recommended.

#### Can I use a UV or Confocal Microscope to analyze my plates and/or count cell numbers?

Yes. This requires the use of an inverted epifluorescence microscope equipped with the appropriate wavelength filters. This will allow you to directly observe and count the cells. Multiple fields on multiple membranes should be counted.

**BD Biosciences**

Two Oak Park  
Bedford, MA 01730 USA  
tel: 800.343.2035  
fax: 800.743.6200

**BD**

2771 Bristol Circle  
Oakville, Ontario  
Canada L6H 6R5  
tel: 905.855.5550  
fax: 905.829.5405

**Nippon BD**

Akasaka DS Bldg.  
5-26 Akasaka 8-chome  
Minato-ku, Tokyo 107 Japan  
tel: (81) 24 593 5405  
fax: (81) 24 593 5761

**BD Biosciences**

Asia Pacific  
Singapore Branch  
30 Tuas Avenue 2  
Singapore 639461  
tel: (65) 6861 0633  
fax: (65) 6860 1590

**BD Biosciences Europe**

Erembodegem-Dorp 86  
9320 Erembodegem Belgium  
tel: (32) 53 720.211  
fax: (32) 53 720.450  
email: [contact\\_bdb@europe.bd.com](mailto:contact_bdb@europe.bd.com)

**BD Biosciences**

4 Research Park Drive  
Macquarie University Research Park  
North Ryde NSW 2113 Australia  
tel: (612) 8875 5239  
fax: (612) 8875 7200