

# Corning® PureCoat™ ECM Mimetic Cultureware

## Frequently Asked Questions

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### How are the Corning PureCoat ECM Mimetic surfaces made?

Corning PureCoat ECM Mimetic Cultureware surfaces are prepared by covalently linking biologically active synthetic peptide to a Corning proprietary surface.

### How do the Corning PureCoat ECM Mimetic surfaces work (peptides versus proteins)?

Peptides are rationally designed to mimic the cell attachment regions of the native extracellular matrix (ECM) protein. Surfaces have been tested with a number of cell types (refer to Corning PureCoat ECM Mimetic application notes and brochure for more information).

### Is Corning PureCoat ECM Mimetic a two-dimensional or three-dimensional surface?

Two-dimensional surface.

### How were the peptide sequences designed?

The peptide is rationally designed to mimic the cell binding adhesion regions of the native protein. The Fibronectin peptide contains the RGD amino acid sequence, which supports the attachment of Fibronectin-dependent cell types including alpha-5-integrin positive cells. The Collagen I peptide contains the GFOGER sequence, which supports the attachment of Collagen I-dependent cell types including alpha-2 integrin positive cells.

### Are the peptide sequences available for manual coating?

No, the peptides, as designed, are covalently attached to a proprietary surface for optimal performance.

### Will Corning PureCoat ECM Mimetic Cultureware work for all cells that are dependent on that particular ECM for culture?

Several cell types have been cultured on the Corning PureCoat ECM mimetic surfaces (refer to brochure for more details). Cell culture is also dependent on the culture medium. Researchers need to optimize the protocol for their cells and medium.

### Will anything in the surface coating leach off?

Corning PureCoat ECM Mimetic Cultureware surfaces are prepared by covalently linking biologically active synthetic peptide to a Corning proprietary surface. Residual unbound peptide on the vessel surface is ~0.01 pmoles/cm<sup>2</sup>, based on dilution factor. After immobilization on the surface, peptides are stable as determined by Surface Chemistry Analysis.

### Do the Corning PureCoat ECM Mimetic surfaces need to be washed or pre-incubated prior to use?

Cultureware is ready to use and does not require any special handling.  
**Caution:** Keep product sealed in original packaging until ready for use.

**How long does it take for cells to attach to Corning® PureCoat™ ECM surfaces? Is there any difference in attachment time from a naturally-derived surface?**

We have not compared the time required for cell attachment, but when we monitor plates the day after seeding, cell attachment appears to be comparable on Corning PureCoat ECM mimetic and native ECM surfaces.

**Will my cells look the same morphologically when they are cultured on Corning PureCoat ECM Mimetic and naturally derived surfaces?**

Yes, they look similar. We did not see differences in the cell types we tested.

**What cell dissociation reagents work with Corning PureCoat ECM Mimetic surfaces?**

Corning PureCoat ECM Mimetic surfaces have been demonstrated for use with TypLe, Dispase, BD Accutase™, and Trypsin.

**Is there a need to modify existing dissociation reagent protocols?**

We recommend using the manufacturer protocols as provided.

**Can I use mechanical scraping of cells with Corning PureCoat ECM Mimetic surfaces?**

We have not done this, but we do not anticipate any problems with this dissociation technique.

**Can I use standard methods to isolate DNA or RNA directly from the surfaces?**

We have no experience with DNA or RNA isolation, but we do not anticipate any issues.

**What is the “animal-free” claim for Corning PureCoat ECM Mimetic surfaces?**

Corning PureCoat ECM Mimetic surfaces are manufactured using animal component-free ingredients in a facility segregated from animal-derived materials.

**How soon after I open the package do I have to work with the vessels?**

The package should be opened just prior to use.

**What happens if the Fibronectin peptide vessels are exposed to the laboratory environment without the moisture-proof packaging?**

Product is susceptible to damage by moisture. We do not recommend using the product after the vessels are exposed to the laboratory environment without the moisture-proof packaging.

**How should I re-seal the Collagen I packaging after opening a multi-pack?**

Fold the bag and tape it.

**What are the storage requirements?**

Room temperature (15° to 30°C).

**What is the shelf-life of these products?**

1.5 years at room temperature storage.

**What is the shelf-life after I open the package?**

Shelf-life is based on packaged product. Fibronectin product should be opened immediately prior to use.

### **Are the products manufactured in a cGMP environment?**

Corning® PureCoat™ Collagen I and Fibronectin Mimetic Cultureware are produced at an ISO 13485:2003, ISO 9001:2000 and ISO 14001:2004 certified manufacturing facility. The products are manufactured in a GMP-compliant facility under the requirements of FDA part 21CFR820.

### **What is the quality control for these products?**

**Products Labelled Non-Pyrogenic:** Products labelled non-pyrogenic have been validated per FDA guidelines on LAL (Limulus Amebocyte Lysate) testing for medical devices. The acceptance level for product is  $\leq 0.125$  EU/mL (endotoxin units per milliliter).

**Cytotoxicity:** Testing is conducted to qualify all material resins using USP Class VI and/or ISO 10993 standards for cytotoxicity and have been shown to be non-toxic.

**Leachability:** Residual unbound peptide on the vessel surface is  $\sim 0.01$  pmoles/cm<sup>2</sup>, based on the dilution factor. After immobilization on the surface, peptides are stable as determined by Surface Chemistry Analysis.

**Functionality:** Flasks and Multiwell plates have been tested for the ability to promote attachment of human cell lines for Fibronectin mimetic and Collagen mimetic.

**Sterilization:** Corning PureCoat Collagen I and Fibronectin Mimetic Cultureware have been manufactured using an aseptic process and tested according to USP <71> for which each step has been validated to ensure that all products meet the industry standard sterility assurance level of SAL  $10^{-3}$ .

### **Will the Corning PureCoat ECM Fibronectin mimetic surface work with BD Mosaic™ serum-free medium?**

The Corning PureCoat ECM Mimetic protocol (as currently developed) has not been optimized for use with BD Mosaic medium. A protocol is under development.

### **What commercially available media can be recommended for use in MSC expansion?**

We have evaluated STEMCELL Technologies MesenCult™ XF medium.

### **Are additional vessels, such as dishes or T-25, T-225 flasks or Falcon® Multi-Flasks available?**

Falcon Multi-Flasks with Corning PureCoat ECM mimetic surfaces will be commercially available by the end of 2012. Custom packaging and formats may be made available sooner.

### **Can I use the Corning PureCoat ECM Mimetic Collagen I surface to differentiate MSCs into osteocytes and adipocytes?**

Yes, this has been demonstrated.

### **When the Corning PureCoat ECM mimetic surfaces are exposed to fluorescent dyes, is there any background binding to the surface? If so, how much (low, medium, high)?**

Cells can be stained, but we have no information regarding background binding.

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