PeptiGel®

Redefining Cell Culture for Life Science

APPLETON WOODS

2D Cell Culture in PeptiGels®

This protocol describes the use of PeptiGels® for 2-dimensional (2D) cell culture.

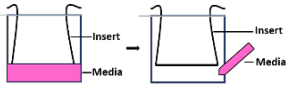
We highly recommend the use a **positive displacement pipette** (such as the Gilson piston pipette) to allow easy pipetting as these are viscous gels.

We recommend the use of cell inserts (such as the Greiner Bio-One cell inserts or equivalent) to increase gel stability and media diffusion.

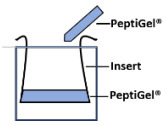
As a guide, this protocol has been written for a total volume of 0.2mL PeptiGel®. Please scale up or down according to culture requirements.

2D Cell Culture Protocol

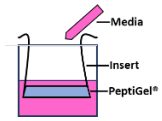
- Pre-wet the inserts in media/PBS for 1 hr to prevent bubbles getting trapped into the membrane pores.



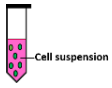
- Remove PeptiGel® from the fridge and pre-warm to room temperature.
  Hint: *If required, centrifuge PeptiGel® for 1 min at 2500 g (3000 rpm) to remove air bubbles.*
- Pipette a volume sufficient enough to cover the surface of the insert. As a guide, 0.2 mL PeptiGel® is enough for a 24 well plate.
  Hint: *Gently tap the plate against a sterile surface approximately 20 times to obtain a flat PeptiGel® surface.*
  Hint*: If necessary, centrifuge the plate containing the insert for 1 min at 2500 g (3000 rpm).*



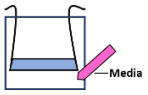
- Add 1 mL of culture media to the well containing the inserts and incubate at 37°C for 30 mins.
- Add 0.2 mL of culture media to the surface of pre-conditioned PeptiGel® in the insert and incubate at 37°C for 30 mins.
  Hint*: Pre-conditioning ensures the pH of PeptiGel® becomes neutral (pH 7).*
  Hint*: Pre-conditioning also ensures a uniform distribution of media nutrients before cell seeding.*



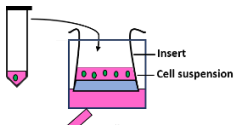
- Resuspend your cells to the required cell density in 0.2 mL of culture media.



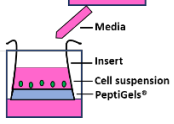
- Remove the media used to pre-condition PeptiGel®.
  Hint*: Leave some media on the surface of pre-conditioned PeptiGel® to prevent the pipette tip disrupting the PeptiGel®.*



- Transfer 0.2 mL of the resuspended cell suspension on top of the PeptiGel® and wait for 5 minutes



- Add 1 mL of fresh culture media to the plate and incubate overnight.



- **Next day**, change the media and repeat as necessary depending on your cell requirements

Manchester BIOGEL

Disclaimer
All standard safety procedures regarding cell culture need to be observed

PeptiGel® sales and technical support

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