COATING PROCEDURE

- 1. Slowly thaw recombinant laminins at $+4^{\circ}\text{C}$ before use. Thawed laminin stock is stable for at least 3 months when stored at $+2^{\circ}\text{C}$ to $+8^{\circ}\text{C}$ under aseptic conditions. For your convenience, the coated plates and diluted coating solution can be kept for up to 4 weeks when stored aseptically at $+2^{\circ}\text{C}$ to $+8^{\circ}\text{C}$.
- 2. Dilute the thawed laminin stock solution with 1xDPBS containingCa2+ and Mg2+.
- 3. Add the diluted laminin solution to tissue culture-treated cultureware for a final coating concentration of 0.5-2 ug/cm2. The optimal coating concentration is cell-dependent. See laminin coating instructions, recommended coating volumes and concentrations.

 4. Seal the plate (e.g. with Parafilm®) to prevent evaporation and incubate at +2°C to +8°C overnight. If a more rapid coating is required, incubate at +37°C for 2 hours. Make sure the laminin solution is spreaded evenly across the surface. Note that the laminin matrix

IMPORTANT NOTES

will be inactivated if let dry.

- The laminin stock solution is stable for 2 years when stored at -20°C. If desired, the laminin stock can be dispensed into working aliquots and stored at -20°C. Repeated freeze thawing should be avoided.
- The protocol can easily be made totally defined and xeno-free with your choice of culture medium and enzyme.
- Before start, all solutions used for cell passaging should be aliquoted in sufficient amounts and prewarmed at +37°C, 5% CO2.