

xCELLigence Real-Time Cell Analysis

E-Plate Coating Protocol



Adherent cells attach to and grow on E-Plates in a manner similar to standard tissue culture plates. Pre-coating E-Plates with poly-L-lysine or extracellular matrix proteins is generally not required. For cases where pre-coating is required, this protocol serves as a general guideline. The optimal coating protein and concentration may vary from one cell line to the next. If you have already optimized a coating protocol for your cell line of interest using standard tissue culture plates, it will likely be transferable to E-Plates and we recommend using this as a starting point.

Recommended Coating Volume	
Device	Volume (for each well)
E-Plate L8	150 μ L
E-Plate 16	50 μ L
E-Plate 96	50 μ L
E-Plate 384	25 μ L
E-Plate Cardio 96	50 μ L
E-Plate CardioECR 48	50 μ L
E-Plate Insert	30 μ L

Recommended Final Coating Concentrations	
Extracellular Matrix (ECM) Protein	Concentration
Collagen IV (BD #354233)	10 μ g/mL
Collagen I (BD #354231)	10 μ g/mL
Fibronectin (BD #354008)	10 μ g/mL
Laminin (BD #354232)	>10 μ g/mL
Poly-L-lysine (Sigma #P4707)	10 μ g/mL
Vitronectin (Invitrogen #PHE0011)	10-15 μ g/mL
Bovine Serum Albumin (negative control)	0.5% w/v

Protocol:

1. Dilute protein stock in 1x PBS to the desired final concentration (e.g. 10 μ g/mL).
2. Coat E-Plate well with the desired volume (e.g. 50 μ L).
 - Ensure that the entire bottom surface of the well is covered with the solution.
3. With the plate lid firmly in place, incubate at 37°C for 1-3 hours, or at 4°C overnight.
 - For Vitronectin, incubate overnight.
4. Remove coating solution completely and wash well twice with 1x PBS. (Wash with water for poly-L-lysine coating.)
 - For polyethylene terephthalate (PET) plates, exercise caution when using a pipette to remove solution from the bottom of wells. Apply minimal pressure when the placing pipette tip against the well bottom. Also, be careful when washing the E-Plate Insert device. Do not touch the membrane with the pipet tip as this may compromise the integrity of the membrane.
5. Use plate immediately. In some cases, coated plates may be stored (with the plate lid firmly in place to ensure sterility) at 4°C for up to one week prior to use. The caveat here is that the storage time may affect the status of the protein coat (extent of hydration, oxidation, etc.), which may in turn influence cell adhesion and growth. Careful attention should be paid to the details of plate coating and the timing of plate storage/usage in order to ensure reproducibility.