



# Instructions for Use

## NativeCoat™ ECM Surface Coating Kit

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For research use only. Not for human or animal therapeutic or diagnostic use.

### Contents and Storage

The components of the NativeCoat™ ECM Surface Coating Kit are shipped on ice. Upon receipt, store all components at 4°C. Avoid freezing. Kit components are listed in the table below.

Component	Quantity
NativeCoat™ ECM component	1 mL × 1
10× Buffer	1 mL × 1

### Materials (required but not provided)

- water (sterile cell culture grade, for diluting 10× Buffer component)
- 1× phosphate-buffered saline (PBS)
- tubes (for mixing components)
- multi-well plate or other cell culture surface
- micropipettes & tips

## Preparation of NativeCoat™ ECM Surface Coating for Cell Culture

**Important:** Prior to proceeding with NativeCoat™ ECM Surface Coating, please review Instructions for Use and see Appendix A, sections A1 – A5 for instructions and example to calculate reagent volumes. We recommend a working concentration between 0.1 mg/mL to 0.2 mg/mL.

**Important:** NativeCoat™ ECM Surface Coating should be prepared right before use and can't be stored.

1. Calculate the volumes of all reagents and dilutions according to the desired NativeCoat™ ECM Surface Coating concentration using the instructions and example provided in Appendix A. We recommend a working concentration between 0.1 mg/mL to 0.2 mg/mL.
2. Add volume of 10× Buffer component (calculated in A4) to volume of sterile cell culture grade water (calculated in A5) to obtain Working Buffer. Mix thoroughly by pipetting up and down. Avoid introducing bubbles.
3. Add volume of NativeCoat™ ECM component (calculated in A3) to Working Buffer to obtain NativeCoat™ ECM Surface Coating. Mix thoroughly by pipetting up and down. Avoid introducing bubbles.
4. Add NativeCoat™ ECM Surface Coating to the cell culture substrate (e.g., multi-well plate, petri dish) according to your experimental setup. Refer to Appendix B for suggested coating volumes for multi-well formats.
5. Gently tap, swirl, or shake multi-well plate or dish for 30 seconds to ensure even coating of cell culture surfaces with NativeCoat™ ECM Surface Coating.
6. Incubate NativeCoat™ ECM Surface Coating at 37°C in a humidified environment for 2 hours.
7. Aspirate NativeCoat™ ECM Surface Coating. Important: Do not allow coated surfaces to dry.
8. Wash cell culture surfaces with 1× phosphate-buffered saline. Aspirate 1× PBS.
9. Add cell suspension to cell culture surfaces coated with NativeCoat™ ECM Surface Coating.
10. Culture cells according to standard cell culture protocols.

For technical support, please visit [xylyxbio.com](http://xylyxbio.com) or email [info@xylyxbio.com](mailto:info@xylyxbio.com).

## References

O'Neill et al. The regulation of growth and metabolism of kidney stem cells with regional specificity using extracellular matrix derived from kidney. *Biomaterials*. 2013.

## Appendix A

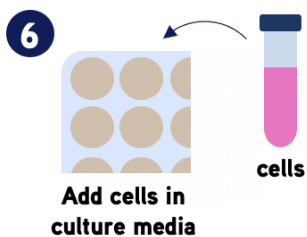
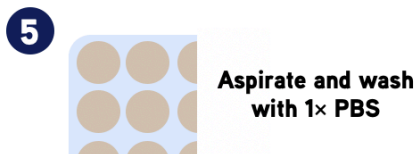
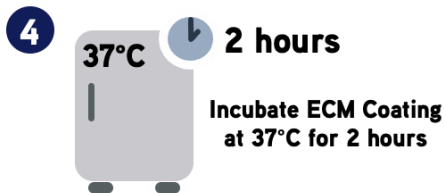
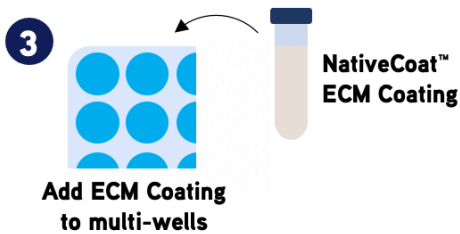
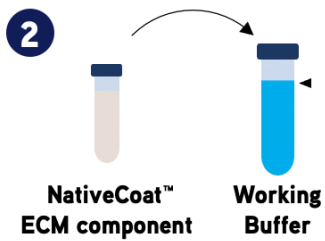
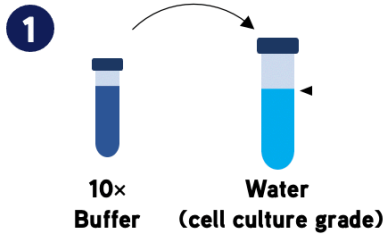
Instructions and example for calculating reagent volumes to prepare NativeCoat™ ECM Surface Coating. We recommend a working concentration between 0.1 mg/mL to 0.2 mg/mL.

**Note:** NativeCoat™ ECM component is provided at a concentration of 1 mg/mL.

Instructions	Example
A1. Determine the desired <b>concentration of NativeCoat™ ECM Surface Coating</b> .	$c = 200 \mu\text{g/mL} = 0.2 \text{ mg/mL}$
A2. Determine the required <b>volume of NativeCoat™ ECM Surface Coating (V<sub>S</sub>)</b> .	$V_S = 4 \text{ mL}$
A3. Calculate the required <b>volume of NativeCoat™ ECM component (V<sub>NC</sub>)</b> .	$V_{NC} = V_S * c = 4 * 0.2 = 0.8 \text{ mL}$
A4. Calculate the required <b>volume of 10× Buffer component (V<sub>B</sub>)</b> .	$V_B = \frac{V_S}{10} = \frac{4 \text{ mL}}{10} = 0.4 \text{ mL}$
A5. Calculate the required <b>volume of sterile cell culture grade water (V<sub>H2O</sub>)</b> .	$V_{H2O} = V_S - V_{NC} - V_B$ $V_{H2O} = 4 \text{ mL} - 0.8 \text{ mL} - 0.4 \text{ mL}$ $V_{H2O} = 2.8 \text{ mL}$

## Appendix B

Multi-well plate	Volume
6	1000 – 1500 μL
12	500 – 700 μL
24	300 – 350 μL
48	100 – 150 μL
96	30 – 50 μL



## NOTES

### Read Instructions for Use

See Appendix A for reagent volume calculations

Mix thoroughly between steps. Avoid introducing bubbles.

See Appendix B for multi-well coating volumes

Gently tap, swirl, or shake coated plate for 30 seconds

**Do not allow coated surfaces to dry**

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