BioLamina

Coating cell cultureware

with Biolaminin® substrates

INSTRUCTIONS FOR USE 001

BioLamina develops, manufactures, and distributes cell culture reagents that enable the culture of pluripotent stem cells, adult stem cells, and tissue-specific cells in a physiologically relevant, cell-specific environment. In vivo, laminins are key basement membrane proteins that underlie all epithelia and endothelia and surround individual cells. Laminins play essential roles in regulating cellular functions such as adhesion, differentiation, migration, phenotype stability, and survival. BioLamina's Biolaminin® cell culture substrates are full-length, human recombinant laminins that are chemically defined, animal origin-free, and enable easy and standardized cell culture.

IMPORTANT NOTES

- All procedures should be done under sterile conditions using aseptic techniques
- Avoid prolonged exposure of the protein to ambient temperatures
- Store the frozen Biolaminin stock solution at -20°C to -80°C. See the product-specific CoA for expiry date.
- Avoid repeated freeze-thaw cycles. Thawed, undiluted Biolaminin is stable for 3 months at +2°C to +8°C. For long-term storage, aliquot and store at -30°C to -80°C.
- Coated plates can be stored aseptically at +2°C to +8°C for up to 4 weeks.

Transition protocol

1 **Slowly thaw the Biolaminin stock solution at +2°C to +8°C before use.**

Thawed, undiluted Biolaminin stock is stable for at least 3 months when stored at +2°C to +8°C under aseptic conditions. Repeated freeze-thaw cycles should be avoided. For longer storage needs, we recommend dividing the thawed stock solution in smaller working aliquots and to store frozen. Frozen stock can be stored up to three years in -30°C to -80°C.

2 **Calculate the concentration and the amount of coating solution needed for the experiment. The MX and CTG matrix generally should be used in a slightly higher concentration (15–20 µg/mL) compared to the LN matrix (5–10 µg/mL). Once the cells are adapted, a lower coating concentration often can be used but should be optimized empirically for each cell line. Guidelines for surface coating calculations can be found in the table below.**

- *A reduced coating concentration might affect the proliferation rate, extending the culture time with about 1 day. Make sure the coating concentration is high enough to support even cell growth.*
- *When adjusting the coating concentration, it is important to maintain the coating volume recommended in the table below. Make sure that the volume covers the entire culture surface as an uncoated surface will not support cell growth.*
- *When culturing cells on the Biolaminin matrix for the first time, a higher coating concentration (10µg/mL for LN521 and 20 µg/mL for MX521 and CT521) is recommended for the first few cell passages. Some cell lines might need an adaptation period and a higher coating concentration is then recommended for the first few passages. Once the cells are adapted to the Biolaminin matrix, the coating concentration usually can be reduced. The coating should be optimized empirically for each cell line and cell type.*

ID: IN-001-11. Valid from 2024-12-02

3 **Gently invert the vial to mix the Biolaminin stock solution. Do not vortex as this may cause fragmentation.**

4 **Dilute the Biolaminin stock solution with 1×DPBS and add the solution to the cultureware of choice. There is no need to pre-treat the cultureware. Make sure the entire surface is covered by the laminin coating solution. An uncoated surface will not support cell growth. Recommended coating volumes for different cultureware formats can be found in the table below.**

- *The laminin matrices are compatible with most commercial cultureware brands (e.g., Falcon, Sarstedt, Corning).*
- *The laminin matrices can be easily used for coating glass surfaces. For more reliable results, we recommend overnight coating at +2°C to +8°C.*

5 **Seal the plate (e.g. with Parafilm) to prevent evaporation and contamination. Incubate at +2°C to +8°C overnight. If a more rapid coating is required, incubate at +37°C for 2 hours.**

- *Do not allow the coated surface to dehydrate as that will inactivate the Biolaminin coating.*
- *Coated plates can be stored aseptically at +2°C to +8°C for up to 4 weeks. Additional 1× DPBS may need to be added after 1–2 weeks to prevent the plate from drying out.*

Guidelines for surface coating calculations

Cultureware	Coating concentration (µg/Ml)	Coating concentration (µg/cm²)*	Coating solution		Total coating solution volume
			Laminin stock	1×DBPS	
6-well	5	0.45	50 µL/well	950 µL/well	1000 µL/well
12-well	5	0.51	25 µL/well	475 µL/well	500 µL/well
24-well	5	0.55	15 µL/well	285 µL/well	300 µL/well
48-well	5	0.49	7.5 µL/well	142.5 µL/well	150 µL/well
96-well	5	0.46	3.5 µL/well	66.5 µL/well	70 µL/well
T-25cm² flask	5	0.55	150 µL/flask	2850 µL/flask	3000 µL/flask
T-75cm² flask	5	0.51	400 µL/flask	7600 µL/flask	8000 µL/flask

* Calculations based on the entire surface area coated.

BioLamina's human recombinant laminin products

Biolaminin 521 CTG
CT521

Biolaminin 511 LN
LN511

Biolaminin 221 LN
LN221

Biolaminin 521 MX
MX521

Biolaminin 421 LN
LN421

Biolaminin 211 LN
LN211

Biolaminin 521 LN
LN521

Biolaminin 411 LN
LN411

Biolaminin 121 LN
LN121

Biolaminin 332 LN
LN332

Biolaminin 111 LN
LN111

ID: IN-001-11. Valid from 2024-12-02

BioLamina

Keep in touch
E-mail **sales@biolamina.com**

BioLamina AB
Löfströms allé 5
172 66 Sundbyberg, Sweden

BioLamina Inc.,
One Broadway
MA 02142 Cambridge, USA

For more information, visit
www.biolamina.com