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Biozellen® Collagen I Solution, Rat

Catalog No.	B-P-00001	
Specification	10mg、20mg、50mg、100mg	
Storage	Store at 4°C. Keep for two years.	

Specifications

Characterization	Biozellen [®] Collagen
Source	Rat Tail Tendon
Form	Solution (0.01N HCI)
рН	2.0-3.0
Package Size	10mg、20mg、50mg、100 mg
Collagen concentration /Biuret	3.0 - 4.5 mg/mL
Endotoxin (EU/mL) /LAL	≤ 1.0
Sterility (USP)	No growth
Storage Temperature	2 to 10 °C / 35.6 to 50 °F
	Listed on product label and
Expiration Date	Certificate of Analysis
Purity / SDS PAGE /	
electrophoresis / silver stain	≧ 95%
	\ge 90% collagen containing within
	α,βandγbands
Electrophoretic Pattern	\leq 10% collagen containing with
/ SDS PAGE / Silver stain	band traveling faster than α
Cell Attachment Assay	Pass

Applications

Biozellen[®] Collagen I is ideal for the thin coating of surfaces in 2D environments. It promotes cell adhesion for various cell types, such as hepatocytes, fibroblasts, spinal ganglion, muscle cells, Schwann cells, epithelial cells. Coating collagen I can be applied to the study of tumor cell invasion, migration and the chemotaxis of macrophages, monocytes.

Collagen I 3D gel, similar to the animal extracellular matrix. Culturing in 3D *in vitro* configurations, could offer biomimetic microenvironment. The gel stiffness can be manipulated to affect cell migration in 3D than in 2D. It allows you to study the effects of the mechanical properties of the ECM on cell development, chemotaxis, migration and morphology.

Notes

Employ aseptic practices to maintain the sterility of the product throughout the preparation and handling of the collagen product.

It is recommended that the collagen and other working solutions be chilled and kept on ice during the

whole preparation of the collagen.

Coating Procedure

Note: Use these recommendations as guidelines to determine the optimal coating conditions for your culture system.

- 1. Transfer desired volume of collagen solution from the bottle to a dilution vessel if required. Further dilute to desired concentration using a sterile 0.01 N HCl solution.
- 2. Swirl contents gently until material is completely mixed.
- 3. Add diluted collagen material to the culture surface ensuring that the entire surface is coated. (Suggestion: $5 \mu g/cm^2$)
- 4. Incubate at room temperature, covered, for 1 hour.
- 5. After incubation, aspirate any remaining material until the surface is dry.
- 6. Rinse coated surfaces carefully with sterile medium or 1X PBS, avoid scratching surfaces.
- 7. Coated surfaces are ready for use. They may be stored at 2-8 °C for up to one week under sterile conditions.

3-D Gel Preparation Procedure for general

Note: The properties of collagen I gels vary depending on multiple factors during preparation, such as temperature, pH, and collagen concentration.

1. Place the sterile 10X PBS, sterile 1N NaOH, sterile dH₂O, *GEcollTM* Collagen I, and a sterile tube on ice.

2. Determine the final volume and final collagen concentration. (Suggestion concentration: 0.5 - 3mg/ml)

- 3. Calculate the following:
 - $V_{10X PBS} [ml] = \frac{Final Volume [ml]}{10}$
 - $V_{collagen}$ $[ml] = \frac{Final \ volume \ x \ Final \ collagen \ con \ [\frac{mg}{mL}]}{Original \ collagen \ con}$
 - $V_{1 N NaOH}$ [ml] = (volume of collagen) × 0.01 [ml]
 - V_{dH20} [ml] = (Final Volume) (volume10X PBS) (volume 1N NaOH) (volume collagen)

4. Perform the following steps:

- (a) Add 10X PBS into the tube.
- (b) Add 1N NaOH into the 10X PBS to adjust the pH value.
- (c) Add dH₂O into the 10X PBS/1N NaOH to match the final volume, mix the contents and hold in ice.

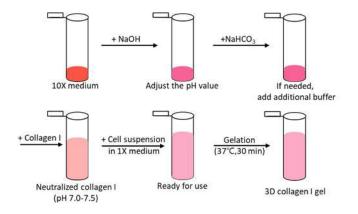
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- (d) Add *GEcollTM* Collagen I to the tube, mix thoroughly without bubbles and place on ice until ready for use.
- 5. For gelation, aseptically transfer the solution into cell culture device, and put into the incubator at 37°C for 30 minutes.

3-D Gel Preparation Procedure for medium

For cell cultured 3D Gel system in medium, the following protocol is recommended.



- 1. Place the sterile 10X medium, sterile 1N NaOH, *GEcollTM* Collagen I, sterile 1X medium (w or w/o cells) and a sterile tube on ice.
- 2. Optionally, if not contained in 10X medium, place the sterile NaHCO3 additional buffers (according

to 10X medium protocol), on ice.

3. Determine the final volume and final collagen concentration. (Suggestion concentration: 0.5 -

3mg/ml)

- 4. Determine the final cell concentration in the 3D gel. (Suggestion: $10^5 \sim 10^7$ cells/ml)
- 5. Calculate the volume of contents using the formula below.
- 6. Perform the following steps:
 - (a) Add 10X medium into the tube.
 - (b) Add 1N NaOH into the same tube.



- (c) If not contained NaHCO3, add the additional buffer in.
- (d) Add GEcollTM Collagen I to the tube, mix thoroughly avoiding bubbles and place on ice. (Note: Use it as soon as possible. The mixture containing collagen will be partial gelation start from 5 minutes.)

1/3

2/3

1X medium w/ cell suspension

> Collagen I 1X medium

NaHCO3

NaOH

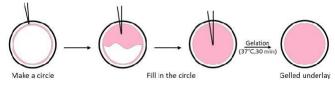
- (e) Add cell suspension in 1x medium into the contents and hold in ice.
- 7. For gelation, aseptically transfer the solution into cell culture device, put into the incubator at 37°C, 5%

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CO2 for 30 minutes.



Calculation formula:

- $V_{10X medium} [ml] = \frac{2}{3} \times \frac{Final Volume [ml]}{10}$
- $V_{collagen}$ $[ml] = \frac{Final \ volume \ x \ Final \ collagen \ con \ [\frac{mg}{mL]}}{Original \ collagen \ con}$
- $V_{1 N NaOH}$ [ml] = (volume of collagen) × 0.01 [ml]
- V_{1X medium (w or w/o cells)} [ml] = (Final Volume) (volume10X medium) (volume 1N NaOH) (volume collagen)