DIRECTONS FOR USE
VitroCol®

PURIFIED HUMAN TYPE I ATELO-COLLAGEN, LYOPHILIZED, STERILIZED
Catalog Number **5008**

Product Description

Advanced BioMatrix offers VitroCol® collagen lyophilized powder which is highly purified atelo-collagen. VitroCol® is about 97% Type I collagen with the remainder being comprised of Type III collagen. The purity of the VitroCol® collagen is ≥99%. SDS-PAGE electrophoresis shows the typical α, β and γ banding pattern for collagen. This product is provided as a lyophilized powder in a 15 mg sterile package size. When reconstituted with 5 ml of 0.01 N HCl, the resulting concentration is 3 mg/ml.

This product is prepared from extracellular matrix secreted by normal human fibroblasts. The human fibroblast cells used to produce this product were intensively tested. Production and purification of the collagen occurs using a multi-step manufacturing process. This process contains built-in, validated steps to insure inactivation of possible viral contaminants.

Note: This product is intended for the coating of surfaces including plasticware but is not intended for use as a 3D gel.

Type I collagen is a major structural component of skin, bone, tendon, and other fibrous connective tissues, and differs from other collagens by its low lysine hydroxylation and low carbohydrate composition. Although a number of types of collagen have been identified, all are composed of molecules containing three polypeptide chains arranged in a triple helical conformation. Slight differences in the primary structure (amino acid sequence) establish differences between the types. The amino acid sequence of the primary structure is mainly a repeating motif with glycine in every third position with proline or 4-hydroxyproline frequently preceding the glycine residue.[1,2] Type I collagen is a heterotrimer composed of two α1(I) chains and one α2(I) chain.

Different collagen subtypes are recognized by a structurally and functionally diverse group of cell surface receptors, which recognize the collagen triple helix. The best-known collagen receptors are the integrins $\alpha_1\beta_1$ and $\alpha_2\beta_1$. $\alpha_1\beta_1$ is the major integrin on smooth muscle cells, while $\alpha_2\beta_1$ is the major form on epithelial cells and platelets. Both forms are expressed on many cell types including fibroblasts, endothelial cells, osteoblasts, chondrocytes, and lymphocytes.[13-15] Some cell types may also express other collagen receptors such as glycoprotein VI (GPVI), which mediates both adhesion and signaling in platelets.[16] Other collagen receptors include discoidin domain receptors, leukocyte-associated IG-like receptor-1, and members of the mannose receptor family.[17,18]

Characterization and Testing

Parameter/Test/Method	Specification
Quantity per Vial	15 mg
Purity - SDS PAGE Electrophoresis – Silver staining	≥ 99%
Electrophoretic Pattern - SDS PAGE Electrophoresis - Coomassie	≥ 85% collagen contained with α, β and γ, < 15% collagen contained within bands traveling faster than alpha
Sterility (USP modified)	No Growth
Endotoxin LAL (EU/ml)	≤ 1.0
Cell Attachment	Pass

Storage/Stability: The product is stored at -10 to -30°C prior to solubilization and ships on frozen gel packs. The product is recommended to be stored at 2 to10 °C after reconstitution. The expiration date is listed on the product label and certificate of analysis for each specific lot. The product shelf life after reconstitution is 3 months. The expiration date is applicable when product is handled and stored as directed.

Precautions and Disclaimer
This product is for R&D use only and is not intended for human or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Coating Procedure

1. Add 5 ml of sterile 0.01 N HCl solution to the VitroCol® serum vial containing 15 mg.

2. Gently mix contents until material is completely solubilized. It may be necessary to agitate at 2 to 10°C overnight.

3. Transfer desired volume of solution from the serum vial to a dilution vessel if required. Further dilute to desired concentration using sterile 0.01 N HCl solution. A typical working concentration may range from ~50 to 100 µg/ml.

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

4. Add appropriate amount of diluted VitroCol® material to the culture surface.

5. Incubate at room temperature, covered, for 1-2 hours. Aspirate any remaining material. Alternatively, incubate at room temperature until surface is dry.

6. After incubation, aspirate any remaining material.

7. Rinse coated surfaces carefully with sterile medium or PBS, avoid scratching surfaces.

8. Coated surfaces are ready for use. They may also be stored at 2-10°C damp or air dried if sterility is maintained.

References

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