Instructions for Use

Huma OsteoGelatin- *Human Gelatin Type A, Lyophilized, High Bloom*

For 2D-3D tissue engineering research, hydrogels and cell culture applications

SKU: BGHL
Tissue Source: Human Bone

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Product Description

Gelatin is a heterogenous mixture of water-soluble proteins with high average molecular masses. Gelatin proteins are derived by denaturing collagen-rich tissue in water [1]. The mechanical properties of gelatin, such as gel stiffness and compressibility, are related to the average molecular weight of the gelatin proteins and characterized by Bloom number [2]. ***Humabiologics offers the first and only human gelatin in the market derived from bone at Bloom numbers between 90-300 g to meet the needs of translational research and regenerative therapies***. Bloom is proportional to the average gelatin molecular mass as described by the table below:

Bloom Number	Average Molecular Mass
50-125 (Low Bloom)	20,000-25,000
175-225 (Medium Bloom)	40,000-50,000
225-325 (High Bloom)	50,000-100,000

Biomedical applications of human gelatin include coating tissue culture surfaces with a thin layer of protein to improve cell attachment for a variety of cell types [3]. Due to excellent biocompatibility of gelatin, it has been used to generate 3D scaffolds and hydrogels for tissue engineering applications [4], and as a delivery material to control the release of bioactive molecules [5]. In the pharmaceutical industry, gelatin is used as an encapsulating agent and binder for tablet production [6]. Gelatin can be combined with other human-derived biomaterials- such as Collagen Type I from human skin (***HumaDerm offered by Humabiologics***) – to make completely human-derived tissue engineered composites [7].

Huma OsteoGelatin Type A, High Bloom is acid treated and ideal for coating tissue culture surfaces with a thin layer of gelatin to support rapid cell attachment and growth or for hydrogel application. The optimal gelatin concentration used may vary depending on cell type or experiment and must be titrated for best results. Type A gelatin is obtained from acid-treated tissues while Type B is obtained from lime-treated tissues. Type A gelatin has an isoelectric point, the charge on the gelatin molecule due to the carboxyl, amino and guanidino groups, is around 7.0-9.0 which corresponds to 80 millimoles of free carboxyl groups in a 100 g of protein [2].

Source Tissue

Huma OsteoGelatin is extracted and purified from human bone sourced strictly from American Association of Tissue Banks (AATB) accredited and FDA registered tissue banks and organ procurement organizations (OPOs). Humabiologics strives to meet research needs by providing high quality biomaterials obtained from tissue partners who comply with requirements for transplantable human tissues under 21 CFR 1271 of the Food and Drug Administration (FDA).

Precautions and Disclaimer

Huma OsteoGelatin is obtained from human tissue that has been tested and found negative for HIV-1 and -2, hepatitis B, and hepatitis C, as well as other infectious agents. Please review the Safety Data Sheet (SDS) for information regarding hazards and safe handling practices. ***Huma OsteoGelatin is for research use only and is not intended for human use, diagnosis, screening, household, food or other uses.***

Storage/Reconstitution

This product can be stored in airtight containers at ambient temperature (15-30 °C) upon receipt and prior to solubilization. The expiration date is 1 year from receipt of the bottle. Dry gelatin, when stored in airtight containers at ambient temperature, can remain unchanged for extended periods of time.

Huma OsteoGelatin can be reconstituted in 37 °C water to the desired concentration with agitation/vortexing for several minutes. Dissolving time depends on the gelatin concentration but usually takes a few minutes for low concentration. Centrifugation might be used to remove undissolved particulate. After reconstitution, solution should be used immediately or stored at 2-10 °C. Solubilized gelatin can be sterilized by filtration through a 0.22 µm membrane.

Preparing Gelatin-Coated Tissue Cultureware

Note: The following are general recommendations. Researcher should optimize parameters based on their specific applications

Optimal gelatin concentrations for tissue culture surface coatings may depend on application and cell type and must be determined for each application. Typically, 100-200 µg/cm^2 of gelatin is used. We recommend making a 1 mg/ml stock solution, but concentration can be increased or lowered based on cell type and application.

1. Dissolve **Huma OsteoGelatin** in DI water to the desired concentration (i.e. 1 mg/ml). Gelatin should readily dissolve with agitation. Higher concentration may require heating to 37 °C with gentle agitation for 30 minutes to ensure complete dissolution.
   NOTE: The optimal concentration of gelatin solution may depend on the experiment and cell type and must be determined for each application.
2. Filter the solubilized gelatin into a sterile container through a sterile 0.22 µm membrane or by autoclaving at 121 °C for 20 minutes.
   Note: for thin coating, thin membranes may be sterilized under UV in the biosafety cabinet for 30 minutes.
3. Aseptically add gelatin solution to tissue culture surface to evenly cover surface.
4. Tilt the cell culture surface so that the solution covers the entire culture surface.
5. Cover, and incubate gelatin coated surfaces at 37 °C for 2 hours.
   NOTE: The optimal incubation time may depend on the experiment and cell type and must be determined for each application.
6. Aspirate excess solution on coated surface. Avoid scratching the coated surface.
7. Transfer gelatin coated surfaces to cell culture hood and remove lid to allow for air drying. Plates can be left overnight in the biosafety cabinet with UV exposure for 30 minutes to sterilize.
8. Use gelatin coated cultureware immediately. Alternatively, keep sterile and store at 2-10 °C.

References

1. Gelatin Manufacturers Institute of America Inc, *Gelatin Handbook*. 2012: 51 W 52nd St, New York, New York 10019, US.
2. Gelatin Manufacturers Institute of America Inc, *Standard Methods for the Sampling and Testing of Gelatins*. 1986: 51 W 52nd St, New York, New York 10019, US.
3. Davidenko, N., et al., *Evaluation of cell binding to collagen and gelatin: a study of the effect of 2D and 3D architecture and surface chemistry.* J Mater Sci Mater Med, 2016. **27**(10): p. 148.
4. Kang, H.W., Y. Tabata, and Y. Ikada, *Fabrication of porous gelatin scaffolds for tissue engineering.* Biomaterials, 1999. **20**(14): p. 1339-44.
5. Dong, Z., Q. Wang, and Y. Du, *Alginate/gelatin blend films and their properties for drug controlled release.* Journal of Membrane Science, 2006. **280**(1): p. 37-44.
6. Corveleyn, S. and J.P. Remon, *Formulation and production of rapidly disintegrating tablets by lyophilisation using hydrochlorothiazide as a model drug.* International Journal of Pharmaceutics, 1997. **152**(2): p. 215-225.
7. Gautam, S., et al., *Surface modification of nanofibrous polycaprolactone/gelatin composite scaffold by collagen type I grafting for skin tissue engineering.* Materials Science and Engineering: C, 2014. **34**: p. 402-409.