Recombinant Human Serum Albumin - Cellastim-S

Cat #: orb419911 (manual)

Workflow Summary

Human serum albumin (HSA) has been identified as a powerful tool for the expansion of many cell types *in vitro*. HSA is a multifaceted protein capable of binding many different types of molecules. HSA therefore plays diverse roles in cell biology, acting as an antioxidant to improve health of cells by binding to reactive oxygen species, as an energy-delivery system by chaperoning fatty acids in the bloodstream, and as a mediator for solubilization and stabilization of metal ions required for cellular processes. These functions make HSA supplementation of cell culture media an integral step in facilitating cell growth and expansion *in vitro*, especially when removing serum or blood-derived proteins for use in clinical manufacturing. Using Cellastim S as a recombinant and scalable source of albumin in cell culture is an enabling tool for large-scale manufacture of cell-based and gene therapies. It is the goal of this app note to provide a step-by-step overview for preparing Cellastim S for use as a stock liquid solution for *in vitro* cell culture applications.

Introduction

Human serum albumin (HSA) has been identified as a powerful tool for the expansion of many cell types *in vitro*. HSA is a multifaceted protein capable of binding many different types of molecules. HSA therefore plays diverse roles in cell biology, acting as an antioxidant to improve health of cells by binding to reactive oxygen species, as an energy-delivery system by chaperoning fatty acids in the bloodstream, and as a mediator for solubilization and stabilization of metal ions required for cellular processes. These functions make HSA supplementation of cell culture media an integral step in facilitating cell growth and expansion *in vitro*, especially when removing serum or blood-derived proteins for use in clinical manufacturing.

As cell-based immunotherapies, gene therapy, and stem-cell therapy continue to show promise in the lab and in the clinic, the availability of safe and consistently high- performing HSA grows in importance for the future of these technologies. Currently HSA used in the expansion of mesenchymal stem cells (MSC), hematopoietic stem cells (HSC), T Cells, HEK293, VERO, MDCK, BHK-21, and many other key cell types is sourced directly from human serum. This albumin source has been instrumental to the conception and development of these novel therapeutics. However, supplementation of cell-culture growth media with serum-derived media supplements and other animal-derived components can introduce adventitious pathogenic agents, create variability when cell expansion processes are scaled up for clinical manufacturing, and can also present the risk of a supply-chain bottleneck. Accordingly, chemically defined media, supplemented with recombinant and well-characterized alternatives to animal-derived components, is increasingly being used as a viable strategy for reducing variation, improving consistency, and enabling scalability for cell- based therapies.

Cellastim S is a completely blood-free and animal-free human albumin produced in a scalable, non-mammalian recombinant expression system. It is naturally free of prion agents (bovine spongiform encephalopathy and other transmissible spongiform encephalopathy agents) and carries less potential risk from other mammalian adventitious agents compared to serum-derived components. In addition to improved safety, Cellastim S has shown consistent performance in supporting the culture of T-Lymphocytes, HEK293, MSC, HSC, and VERO, among other cell type. Using Cellastim S as a

Biorbyt Ltd
7 Signet Court, Swann Road, Cambridge, CB5 8LA. United Kingdom
Email: info@biorbyt.com | Phone: +44 (0)1223 859-353 | Fax: +44 (0)1223 280-240

Biorbyt LLC
68 TW Alexander Drive, Durham, NC 27709. United States
Email: info@biorbyt.com | Phone: +1 (415) 906-5211 | Fax: +1 (415) 651-8558

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Materials

Recombinant Human Serum Albumin

√ Cellastim S

Reconstitution Supplies

√ 125ml sterile PETG media bottle

√ Cell culture grade DPBS, PBS, or basal media such as DMEM F/12, DMEM, MEM, RPMI

Vacuum Filtration System

√ 500ml 0.2-micron vacuum filtration system

Protocol: Cellastim S Stock Preparation

1. To prepare a sterile, 10% concentrated liquid stock solution of Cellastim S, first weigh an empty, 125ml Sterile PETG Media Bottle.
2. Under a laminar flow hood, add 10 g Cellastim S to the bottle. Weigh the bottle containing the powder to determine the actual weight of Cellastim S added.
3. Once the exact weight of Cellastim S is known, calculate the volume required to produce a 10% concentration of Cellastim S. Then, gently reconstitute the powder in cell-culture grade DPBS, PBS, or the selected basal media by adding 70% of the calculated final volume directly to the 125ml bottle. Do not add the full final volume of buffer at this stage. An allowance must be made to account for volume displacement caused by the powder.
4. Cap the 125ml bottle and gently turn it on its side to allow the powdered albumin to fall into the buffer. Avoid the formation of bubbles during this step. Return the container upright and allow the albumin to dissolve, undisturbed (no shaking) at 4°C in the dark for a minimum of 4 hours and preferably overnight.
5. Once all the powdered Cellastim S has dissolved, bring the volume up to the calculated target by adding additional reconstitution liquid. To achieve this easily, use a 50ml pipette to move exactly 50ml of the reconstituted albumin powder to a fresh 125ml bottle. Using the same pipette, remove the remaining liquid from the original bottle, noting the volume. Finally, pipet the difference between the target volume and the volume as measured by pipetting and add this volume of reconstitution liquid to the new bottle.
6. Sterilize the stock solution by filtering through an 0.2µm vacuum-driven disposable filtration system.

Optional: For manual filtration, recommends using a 25 mm, 0.8µm filter, followed by a 0.2µm 30mm PES low-protein-binding filter, using a sterile Luer-lock syringe.

Biorbyt Ltd
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Example Calculations

$$\text{Resuspension Volume (ml)} = \frac{\text{Cellastim S weighed (mg)}}{\text{100mg/ml}}$$

$$\text{Resuspension Volume (ml)} = \frac{\text{10828.6 (mg)}}{\text{100mg/ml (10\%)}}$$

Target Resuspension Volume (ml) = 108.3 ml

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