

HFSdaFREE™ KIT



A 3-Component, Chemically-Defined Animal Origin-Free (cdAOF) Supplement System, for the Primary Isolation and Serial Propagation of Human Dermal Fibroblasts, Corneal Fibroblasts, and Other Mesenchymal Cells.

DESCRIPTION AND CONTENTS

- AvantBio Products: HFSdaFREE (product no. AVBP-011), HFGE (product no. AVBP-010) and HFGE2 (product no. AVBP-040)
- HFSdaFREE 5 ml Volume, 100X concentrated dAOF supplement. Sterilized by filtration.
- HFGE 0.5 ml Volume, 1000X concentrated dAOF Supplement. Sterilized by filtration.
- HFGE2 1.0 ml Volume, 500X concentrated dAOF Supplement. Sterilized by filtration.
- Shipped and stored frozen (-80° C or -20° C)
- Shelf life: 18 months from date of manufacture when stored at -20° C.
- For laboratory usage only. Not to be used for therapeutic or diagnostic purposes in humans or animals. Use good laboratory practices when handling. Avoid contact and do not ingest. Consult SDS.

DIRECTIONS FOR USE

1. To efficiently support the propagation of human fibroblast primary and post-primary cell cultures in the dAOF environment, AvantBio recommends that HFSdaFREE, HFGE and HFGE2 be used to supplement a **50/50 mixture of EpiLife®** (sold by ThermoFisher Scientific, cat. no. MEPI500CA) and **Human Fibroblast Expansion Basal Medium®** (sold by ThermoFisher Scientific, cat. no. M106500).
2. Thaw supplements at room temperature. Use immediately, or store for up to 6 hours at 4° C before use. Do not leave thawed product at room temperature or higher for longer than 5 minutes. Do not re-freeze. Wipe vial clean with 70% isopropyl alcohol before use. HFSdaFREE, HFGE and HFGE2 quantities (volumes) are intended to supplement 500 mls of a 50/50 mixture of EpiLife® / medium 106®. Use entire contents of each vial (i.e. rinse vial 1 x with basal medium).
3. Pre-coating the cell culture surface (substratum) with animal origin free recombinant human collagen-1 (at 5 ug/cm²), or other animal-sourced collagen-1 (e.g., human / bovine), can enhance cell survival, cell proliferation and cell replicative lifespan. For both primary culture and post-primary serial propagation, pre-coating the cell culture surface (substratum) with collagen-1 **is required**. Purified animal/human-derived collagen-1 can be purchased from a variety of vendor sources. Animal origin free recombinant human collagen-1 can be purchased by request from Collplant Inc. (see: <https://collplant.com/> and <https://collplant.com/contact/>). Additionally, Corning® CellBIND® cell culture-ware (see: www.corning.com/worldwide/en/products/life-sciences/products/surfaces/cell-bind.html) **may replace the requirement for collagen-1 pre-coating**, as CellBIND® has been successfully utilized to propagate post-primary human fibroblasts cultured in the absence of collagen-1 pre-coating. TrypLE™ Select 1X (ThermoFisher Scientific: cat. no. 12563029) is recommended for cell dissociation during post-primary serial propagation.

- Prior to plating fibroblasts into primary culture, make sure that the cells are washed essentially free of residual collagenase activity. Residual collagenase in the primary cell culture can inhibit plating of the primary fibroblasts.



AVANTBIO LIMITED USE AGREEMENT

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