

HKSdaFREE™ KIT (product no. AVB01HKS)

A 2-COMPONENT, CHEMICALLY-DEFINED ANIMAL ORIGIN-FREE (cdAOF) CELL CULTURE SUPPLEMENT SYSTEM FOR HUMAN KERATINOCYTES, CORNEAL EPITHELIAL CELLS, ORAL EPITHELIAL CELLS AND OTHER EPITHELIAL CELLS

DESCRIPTION AND CONTENTS

- AvantBio Products: HKSdaFREE (product no. AVBP-004) and HKGE (product no. AVBP-005)
- HKSdaFREE 5 ml Volume, 100X concentrated dAOF supplement. Sterilized by filtration.
- HKGE 0.5 ml Volume, 1000X concentrated dAOF Supplement. Sterilized by filtration.
- Shipped and stored frozen at -20° C or lower.
- Shelf life: 18 months from date of manufacture (DOM), when stored at -20° C or lower.
- 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 avoid ingestion. Consult SDS.

DIRECTIONS FOR USE IN EPI LIFE® (THERMOFISHER® - CASCADE BIOLOGICS®) BASAL MEDIUM

1. To efficiently support the propagation of human keratinocytes as well as other epithelial cell primary and post-primary cell cultures in the dAOF environment, AvantBio recommends that both HKSdaFREE and HKGE be used to supplement **EpiLife® basal cell culture medium** (sold by ThermoFisher Scientific, cat. no. MEPI500CA).
2. Thaw supplements at room temperature. Use immediately or store for up to 8 hours at 4 ° C before use. Do not leave thawed product at room temperature or higher for longer than 15 minutes. Do not re-freeze. Wipe outside of vial clean with 70% isopropyl alcohol before use. HKSdaFREE and HKGE quantities (volumes) are intended to supplement 500 mls of EpiLife® basal medium. Mix thawed supplement thoroughly before use, and 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. bovine), can enhance cell survival, cell proliferation and cell replicative lifespan. For primary epithelial cell culture, pre-coating the cell culture surface with collagen-1 **is required** (e.g., 5 ug / cm²). For post-primary epithelial cell culture (serial propagation), pre-coating the cell culture surface with collagen-1 **is optional**. Animal origin free recombinant human collagen-1 can be purchased from CollPlant (<https://collplant.com/>). TrypLE™ Select 1X (ThermoFisher Scientific: cat. no. 12563029) is recommended for animal-free cell dissociation during post-primary serial propagation. For cdAOF cell cryopreservation, it is recommended to use CRYO-free 5 (AvantBio Corp: cat no. AVBP-0102).
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