

Protocol for preparation of NHEK/SVTERT3-5 medium

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Evercyte Ord. No.: CLHT-011-0026-5

Designation: NHEK/SVTERT3-5, human epidermal keratinocytes

Growth medium: KGM™ Gold Keratinocyte Growth Medium BulletKit™ (Lonza, Cat# 00192060) supplemented with G418:

Final components:

KBM™ Gold™ Basal Medium (Lonza, Cat# 00192151)

Components of KGM™ Gold™ SingleQuots™ supplements (Lonza, Cat# 00192152: BPE, hEGF, Insulin, Hydrocortisone, Transferrin, Epinephrine, without GA)

50 µg/ml G418 (InvivoGen, Cat# ant-gn5, 100 mg/ml stock solution, ready-to-use)

- take one bottle of KBM™ Gold™ Basal Medium (500 ml)
- add 2 ml of BPE (component of KGM™ Gold™ SingleQuots™)
- add 500 µl of hEGF (component of KGM™ Gold™ SingleQuots™)
- add 500 µl of Insulin (component of KGM™ Gold™ SingleQuots™)
- add 500 µl of Hydrocortisone (component of KGM™ Gold™ SingleQuots™)
- add 500 µl of Transferrin (component of KGM™ Gold™ SingleQuots™)
- add 250 µl of Epinephrine (component of KGM™ Gold™ SingleQuots™)
- add 250 µl of G418 stock solution (100 mg/ml)

- mix properly
- store at 4°C for a maximum of 4 weeks
- temper the medium to room temperature (not 37°C) before use