

## Protocol for preparation of PodoUp3 medium

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Evercyte Ord. No.: MHT-033-3

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**Reagents:** The PodoUp3 medium for cultivation of PODO/TERT256 and PODO/SVTERT152 cells can either be ordered from Evercyte as ready-to-use medium (Cat# MHT-033-3) or as basal medium (Cat# MHT-033-3-B) plus supplements (Cat# MHT-033-3-S).  
The medium can also be prepared by mixing the following components:

MCDB131 (Pan Biotech, Cat# P04-80057)  
1.6 mM GlutaMAX-I (Gibco, Cat# 35050-038, ready-to-use)  
9.6 µg/mL bovine brain extract (Lonza, Cat# CC-4098, ready-to-use)  
8 ng/ml hEGF (Sigma-Aldrich, Cat# E9644)  
20 ng/ml Hydrocortisone (Sigma-Aldrich, Cat# H0396)  
20 % FBS (Sigma-Aldrich, Cat# F7524, ready-to-use)  
100 µg/ml G418 (InvivoGen, Cat# ant-gn-5, ready-to-use)

- take one bottle (500 ml) of MCDB131
  - add 5 ml GlutaMAX-I (200 mM stock, ready-to-use), mix properly
  - discard 105 ml from MCDB131 / GlutaMAX-I mixture
  - add 533.6 µl BBE (9 mg/ml stock, ready-to-use)
  - add 200 µl hEGF stock (20 µg/ml, prepared in cell culture grade water)
  - add 200 µl hydrocortisone stock (50 µg/ml, prepared in cell culture grade water)
  - add 100 ml fetal bovine serum (ready-to-use)
  - add 500 µl G418 stock (100 mg/ml, ready-to-use)
  - mix properly and store at 4°C for up to 1 month
  - temper the medium to room temperature (not 37°C) before use
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**Related products:** PodoUp3 ready-to-use medium, 500 ml (Evercyte, Cat# MHT-033-3)  
PodoUp3 basal medium, 500 ml (Evercyte, Cat# MHT-033-3-B)  
PodoUp3 supplements (Evercyte, Cat# MHT-033-3-S)  
PODO/SVTERT152, human urine-derived podocytes (Evercyte, Cat# CLHT-033-0152)  
PODO/TERT256, human kidney tissue-derived podocytes (Evercyte, Cat# CHT-033-0256)

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