

Protocol for preparation of TS medium

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Evercyte Ord. No.:	-
Designation:	TB/SVTERT350, human trophoblasts
Medium components and preparation:	TS medium contains the herein described components and is prepared as described in the following: DMEM/F12 (1:1) (PAN-Biotech, Cat# P04-41154) 0.1 mM 2-Mercaptoethanol (Gibco, Cat# 21985023) 0.2% FBS (PAN-Biotech, Cat# P30-3031) 0.3% BSA (Sigma-Aldrich, Cat# A7906) 1% ITS-X (Gibco, Cat# 51500056) 1.5 µg/ml L-Ascorbic acid (Sigma-Aldrich, Cat# A4544) 50 ng/ml human epidermal growth factor (hEGF) (Sigma-Aldrich, Cat# E9644) 2 µM GSK3 inhibitor/WNT activator CHIR99021 (Stemcell Technologies, Cat# 72052) 0.5 µM ALK5/7/4 inhibitor A83-01 (Stemcell Technologies, Cat# 72022) 1 µM Activin/BMP/TGF-β pathway inhibitor SB431542 (Stemcell Technologies, Cat# 72232) 0.8 mM Valproic acid (VPA) (Stemcell Technologies, Cat# 72292) 5 µM ROCK inhibitor Y-27632 (Stemcell Technologies, Cat# 72302) - take one bottle (500 ml) of DMEM/F12 (1:1) and discard 23 ml - add 909 µl 2-Mercaptoethanol (55 mM, ready-to-use) - add 1 ml FBS (ready-to-use) - add 5 ml BSA (30%, prepared in cell culture grade water) - add 5 ml ITS-X (ready-to-use) - add 107.1 µl L-Ascorbic acid (7 mg/ml, prepared in cell culture grade water) - add 1.25 ml hEGF (20 µg/ml, prepared in cell culture grade water) - add 333 µl CHIR99021 (3 mM, prepared in DMSO) - add 125 µl A83-01 (2 mM, prepared in DMSO) - add 50 µl SB431542 (10 mM, prepared in DMSO) - add 8 ml VPA (50 mM, prepared in PBS) - add 250 µl Y-27632 (10 mM, prepared in PBS) - mix properly and store at 4°C for up to 1 month - temper the medium to room temperature (not 37°C) before use
Related products:	- IMDM-FBS medium (see <i>Protocol preparation of IMDM-FBS medium</i>) - TrophoUp medium (Evercyte, Cat# MHT-070), ready-to-use medium - TB/SVTERT350, human trophoblasts (Evercyte, Cat# CLHT-070-0350)