HD Community Biorepository
Cell Line Description and Propagation Instructions

CHDI#	CHDI-90000417
Coriell Ref Number	**CH00285**
Cell Line Name	PC12, Htt-SW2-Q23-TAGRFP, mix, 1-3144, human, clone 34 Alias: PC12-SW2-FLHttQ23-TAGRFP-CL34
Description	PC12 cells inducibly expressing a full length human Huntingtin (Htt) containing 23 polyglutamine repeats with a TAGRFP tag at the C-term.
Host Cell line name, species and tissue source	PC12, rat, pheochromocytoma of adrenal gland
Engineered DNA construct, include reference	Htt-SW2-Q23-TagRFP, mix, 1-3144, human (CHDI-90000427) Alias: SW2-FLHttQ23-TagRFP
Induction system utilized	RheoSwitch
Immortalization method used if any	Not Applicable
Complete growth medium with recommended manufacturer	Kaighn's Modification of Ham's F-12 (ATCC # 30-2004) 15% Horse Serum (Gibco # 16050-122) 2.5% FBS (Hyclone # SH30071) 1% Pen/Strep: Hyclone cat # SV30010 0.2 mg/ml active G418 (Calbiochem cat # 345810) 0.2 mg/ml Zeocin (Invitrogen cat # 46-0072) ***Requires collagen IV substrate - see attachment**
Is it being cultured in the presence of antibiotics?	Yes-see above
Temperature	37ºC
Atmosphere	5% CO2, humidified
Subcultivation ratio	1:3
Max tolerable cell density or confluency	90%
Medium renewal	3-4 days
Appearance/Morphology, etc	Small round and clumpy
Growth Properties (adherent, etc)	Adherent but require collagen IV substrate
Freeze medium	50% Growth medium + 50% Cryoprotective medium (Lonza 12-132A)
Storage temperature	Liquid Nitrogen vapor
Species and tissue of origin, geographical source of isolation, and any known associated hazards (HIV, EBV etc)	rat, pheochromocytoma of adrenal gland (ATCC CRL-1721)
Recommended biosafety level for working with this strain	1
Miscellaneous Background Information, specific notes and supporting data	Sigma Col IV Cat#C5533 Lot#087K3780 or Fluka Biochemika cat # 27663, lot 1314833 31007242

*Coating Cell Culture Surfaces with Collagen IV
Cellumen, Inc.
June 27, 2009

A. Preparation of collagen IV suspension

1. Obtain collagen IV powder (e.g., collagen IV (#C5533); Sigma Chemical Company, St. Louis, MO).

2. Make a 0.5 mg/ml stock solution by incubating collagen IV powder in 0.025% acetic acid overnight at 4 C.

B. Coating cell culture surfaces

1. Dilute stock collagen IV solution (0.5 mg/ml) to 0.1 mg/ml using 0.025% acetic acid.

2. To coat tissue culture flasks and dishes, pipette 5 ml into the vessel, tip the vessel to coat uniformly.

3. Remove 4 ml of the collagen IV from the vessel and use to coat the next vessel.

4. Let flasks and dishes air dry in a laminar flow sterile hood.

5. To coat 384-well microplates, add 16 µl to each well and tap the plate gently to ensure uniform coverage of each well.

6. Allow the microplates to air dry in a laminar flow sterile hood.

7. Store all coated tissue culture vessels at 4ºC until use.