Data Sheet

BCRJ Code:	0203
Cell Line:	PC-12
Species:	Rattus norvegicus
Vulgar Name:	Rat
Tissue:	Adrenal Gland
Morphology:	Small Irregularly Shaped Cells
Disease:	Pheochromocytoma
Growth Properties:	Floating Clusters; Few Scattered Lightly Attached Cells.
Sex:	Male
Applications:	This cell line is a suitable transfection host.
Tumor Formation::	Yes, in New England Deaconess Hospital strain rats
Products:	Catecholamines; dopamine; norepinephrine
Biosafety:	1
Addtional Info:	The cells respond reversibly to NGF by induction of the neuronal phenotype when plated on Collagen IV coated culture flasks. The cells do not synthesize epinephrine.
Culture Medium:	RPMI 1640 with 2.0 mM L-glutamine, 4.5 g/L glucose, 1.0 mM sodium pyruvate, heat-inactivated horse serum to a final concentration of 10% and fetal bovine serum to a final concentration of 5%.

Subculturing:	Protocol: Volumes used for this protocol are for a 75cm2 flask; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes. T-75 flasks are recommended for subculturing this product. Transfer cell suspension to centrifuge tube. Centrifuge cells at 180 to 225 xg for 8-15 minutes at room temperature. Remove and discard supernatant leaving cell pellet. Resuspend the cell pellet with 5 mls of fresh medium (or use an appropriate volume of medium which is a multiple of 5 to facilitate the next step). Gently aspirate each 5 ml aliquot of cells 4 or 5 times with a new 20 ml syringe outfitted with a 22g (1½ in.) needle to break up cell clusters. Add appropriate aliquots of the cell suspension to new 75 cm2 flask with 10-15 ml fresh growth medium. Seed flask 5 x 10(5) to 1 x 10(6) viable cells/ml or use subcultivation ratio of 1:2 to 1:4. Place culture vessels in incubator at 37ºC Subculture when cell density reaches between 2-4 x 10(6) viable cells/ml. NOTE: For more information on enzymatic dissociation and subculturing of cell lines consult Chapter 12 in Culture of Animal Cells, a manual of Basic Technique by R. Ian Freshney, 6th edition, published by Alan R. Liss, N.Y., 2010.
Subculturing Medium Renewal:	Every 2 to 3 days
Culture Conditions:	Atmosphere: air, 95%; carbon dioxide (CO2), 5% Temperature: 37°C
Cryopreservation:	95% FBS + 5% DMSO (Dimethyl sulfoxide)

Thawing Frozen Cells:	SAFETY PRECAUTION: Is highly recommend that protective gloves and clothing always be used and a full face mask always be worn when handling frozen vials. It is important to note that some vials leak when submersed in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vessel exploding or blowing off its cap with dangerous force creating flying debris. 1. Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the Oring and cap out of the water. Thawing should be rapid (approximately 2 minutes). 2. Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions. 3. For cells that are sensitive to DMSO is recommended that the cryoprotective agent be removed immediately. Transfer the vial contents to a centrifuge tube containing 9.0 mL complete culture medium and spin at approximately 125 x g for 5 to 7 minutes. 4.Discard the supernatant and Resuspend cell pellet with the recommended complete medium (see the specific batch information for the culture recommended dilution ratio). 5. Incubate the culture in a appropriate atmosphere and temperature (see "Culture Conditions" for this cell line). NOTE: It is important to avoid excessive alkalinity of the medium during recovery of the cells. It is suggested that, prior to the addition of the vial contents, the culture vessel containing the growth medium be placed into the incubator for at least 15 minutes to allow the medium to reach its normal pH (7.0 to 7.6).
References:	Levi A et al. Molecular cloning of gene sequence regulated by nerve growth factor. Science 229:393-395, 1985 Pub Med: 85244666
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ATCC:	CRL-1721