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Document # INST-77232-3 10/11
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PC-1™ Chemically Defined Serum-Free Medium

Instructions for Use

Description

PC-1™ Medium is a low-protein, serum-free medium intended for the culture of primary cells and anchorage-dependent cell lines. PC-1™ Medium contains a complete HEPES buffering system with known amounts of insulin, transferrin, fatty acids, and proprietary proteins assembled under strict quality control procedures. PC-1™ Medium is intended for a variety of research and industrial applications and is formulated using defined components for optimal cell growth, while maintaining the lowest possible protein content.

Storage and Stability

PC-1™ Medium is to be stored at 2℃-8℃.The PC-1™ Supplement should be stored at -20℃. When these two components are combined, the resulting PC-1™ Complete Medium is stable for 45 days at 2℃-8℃.

Once thawed, the appropriate volume of one vial of PC-1™ Supplement must be combined with the companion volume of PC-1™ Medium. Partial reconstitution or repeated freezing and thawing of the PC-1™ Supplement is not advised.

Package Contents

77232	PC-1™ 2 x 500 ml Kit:	
	PC-1™ Medium	2 bottles, 500 ml
	PC-1™ Supplement	2 vials, 10 ml each

PC-1™ Complete Medium Preparation

1. Thaw one vial of frozen PC-1™ Supplement at room temperature. Wipe the outer surface of the vial with ethanol and aseptically transfer 2 ml supplement per 100 ml liquid base medium or 10 ml supplement per 500 ml liquid base medium. If preparing the 100 ml kit, there will be 8 ml of excess PC-1™ Supplement which can be discarded.
2. Immediately prior to use, add sterile L-glutamine (final concentration: 2-4 mM) to the PC-1™ Complete Medium prepared in Step 1 above.

Instructions for Use

1. Prior to introducing cells to PC-1™ Complete Medium, the culture vessels containing PC-1™ Complete Medium should be equilibrated in a CO_2 atmosphere (5-7%) at 37℃. This is a precautionary measure to assure that cells, upon introduction to PC-1™ Complete Medium, do not experience temperature and/or pH shock. Pre-warming PC-1™ Complete Medium in a 37℃ waterbath is not recommended.
2. Using your typical method for dispersing monolayers of cells, prepare a cell suspension and harvest by gentle centrifugation (200 x g for 5 minutes). Use of a 0.01% soybean trypsin inhibitor (or other similar protease inhibitor) is recommended at this stage of cell preparation to minimize residual protease activity.
3. Resuspend cells in PC-1™ Complete Medium at a final cell density of 5×10^4 cells/ml. Usually, a seeding density of 5×10^4 cells/ml is appropriate, however, it may be necessary to

determine the optimal seeding density for each specific cell type.

4. Typically, cells in PC-1™ Complete Medium may be plated directly on tissue culture quality plastic surfaces. However, in the case of specific cell types, it may be necessary to determine the importance of pretreating the culture surface with factors such as collagen, fibronectin, poly-D-lysine or laminin.
5. Feeding of cultures may be accomplished by either addition of fresh media to culture vessels or by complete replacement of spent media. When removing spent media, care should be taken not to disturb cells, as they may be loosely attached to the surface of culture vessels. Feeding is typically not necessary prior to 48 hours post-seeding.
6. Monolayers growing in PC-1™ Complete Medium should be monitored closely. When approaching confluency, some monolayers have a tendency to release from the vessel surface, thus making their eventual dispersal more difficult.
7. Confluent monolayers may be passaged using your typical method of enzymatic dispersal. Generally, dilution of cells with PC-1™ Complete Medium prevents further enzymatic activity. However, enzyme inhibitors may become necessary if you desire a minimal culture dilution.

Adaptation of Serum-Grown Cells

Weaning cell lines from serum-containing medium may be required. The following protocol will aid adaptation to a serum-free environment by gradual reduction of serum concentration:

1. Week 1: Reduce the current serum concentration used to 5% fetal bovine serum (FBS) in PC-1™ Complete Medium.
2. Week 2: Reduce the serum concentration to 2.5% FBS in PC-1™ Complete Medium.
3. Week 3: Reduce the serum concentration to 1.25% FBS in PC-1™ Complete Medium.
4. Week 4: Reduce the serum concentration to 0.75% FBS in PC-1™ Complete Medium.

NOTE: Adaptation of cells to PC-1™ Complete Medium containing less than 1% FBS may require more than a 1 week period. Monitor cell growth and proceed to next serum reduction when cells appear to resume normal growth characteristics. The recommended seeding density is 5 x 10^4 cells/ml, however, it may become necessary to seed cells at a higher density until fully adapted to serum-free conditions.

5. Reduce serum concentration to 0.5% FBS in PC-1™ Complete Medium. See note for Step 4.
6. Reduce serum concentration to 0.25% FBS in PC-1™ Complete Medium. See note for Step 4.
7. Eliminate FBS in PC-1™ Complete Medium and culture cells in serum-free PC-1™ Complete Medium.

PC-1™ Liquid Base Medium Has Been Successfully Tested on the Following Cell Lines:

HeLa	Epithelial carcinoma, cervix, human
MRC-5	Embryonal lung, male, human, fibroblast-like
BHK-21	Kidney, Syrian hamster, fibroblast-like
WI-38	Lung, diploid, human
NRK*	Normal rat kidney, epithelial/fibroblast-like
3T3	Embryonic mouse, fibroblast-like
CHO-K1*	Chinese hamster ovary, epithelial-like
HTB-72	Malignant melanoma, human, epithelial-like
HRB-4	Human bladder tumor
WISH	Human amnion, epithelial-like
VERO*	Fibroblast, African green monkey
MDCK	Madin Darby canine kidney, epithelial-like
STO	Transformed mouse fibroblast
HEP2	Transformed larynx, epidermoid carcincoma, human
SIRC	Rabbit cornea
C6	Rat glioma, primary
T9	Rat glioma
ARL6T	Normal rat liver
Human neuroblastoma	
Human foreskin fibroblast	
Human bladder carcinoma	
Human renal papillary collecting tubule (primary)	

Rat dermal fibroblast (primary)
Rat mammary carcinoma (primary)
Baboon (paprocynocephalus) spinal ganglia
Swine testes cell
Bovine kidney
Rat neonatal normal cardiac muscle (primary)
Rat thyroid epithelium (primary)
Human colon epithelium (primary)
Human colon carcinoma (primary)
Rat astrocytes (primary)

*Cell growth evaluations were performed at Hycor. Information for all other cell lines was provided to Hycor from outside sources. Lonza, Inc. has since acquired this product.

Ordering Information

77232	**PC-1™ 2 x 500 ml kit:**	
	PC-1™ Medium	2 bottles, 500 ml
	PC-1™ Supplement	2 vials, 10 ml each

Product Use Statement

THESE PRODUCTS ARE FOR RESEARCH USE ONLY. Not approved for human or veterinary use, for application to humans or animals, or for use *in vitro* diagnostic or clinical procedures.