



Clonetics™ Rat Retinal Cell System

R-Ret – Technical Sheet

Introduction

Ready-to-use retinal cells from neonatal Sprague-Dawley rats (post-natal day 3 or 4) are cell suspensions of high quality primary cells (comprised of the seven cell types normally found in retina) prepared by dissection/dissociation without purification, and are ready for immediate culture. Each vial of retinal cells contains $\geq 200,000$ viable cells. This will seed into twenty wells of a 96-well plate or four wells of a 24-well plate using the recommended plating densities and medium. Some cell death will occur during the first few days after plating and debris will be observed - this is normal. After approximately four days in culture, the cells will form a neurite network and by day six to seven, debris will be minimal.

Cell System Components (Sold Separately)

- One normal rat retinal cell product (cryopreserved) (R-Ret-508)
- One Primary Neuron Growth Media BulletKit™ Medium - 200 ml:

Clonetics™ PNGM™ BulletKit™ Medium, (CC-4461) contains 200 ml of Primary Neuron Basal Media (PNBM™ Medium) and the following supplements: L-glutamine, 4.0 ml; Neural Survival Factor-1 (NSF-1), 2.0 ml; and Gentamicin/Amphotericin-B, 0.2 ml.

Additional Required Components (Not Included)

- It is recommended to add 5% heat inactivated FBS to the Primary Neuron Growth Media BulletKit™ Medium for initial plating and short-term culture (up to four days) and change to

serum free medium for long term culture (≥ 5 days)

Recommended Cell Culture Substrates

Primary retinal cells need an appropriate substrate in order to adhere and survive. The preferred substrate is Matrigel. Poly-L-Lysine or Poly-D-Lysine can also be used alone to coat the cell culture plastic ware or cover slips. Coated cell culture plates, dishes, or cover slips can either be purchased from a supplier or prepared immediately prior to use. Protocols for the recommended substrates are available on our website at www.lonza.com.

Characterization of Cells

Each lot of retinal cells is tested using mycoplasma PCR, bio-burden assay and immunohistochemistry, including immunostaining for neuron specific class III β -tubulin (Tuj-1), specific neuronal protein gene product (PGP 9.5), ganglion cell marker, Thy1.1, and GFAP.

Performance

Recommended Seeding Volume after Diluting 0.5 ml Cell suspension in 3.5 ml Fresh Culture Media	Plating Format	Number of Wells to Plate
200 μ l/well	96-well plate	20 wells
1 ml/well	24-well plate	4 wells

Quality Control

The cryopreserved cells are batch-tested for viability based on counts of ganglion cells compared to freshly prepared retinal cells at day seven in culture. The cells test negative for mycoplasma and bacteria. Additional molecular and immunochemical testing for quality is done following conditions that mimic shipping.

Ordering Information

Rat Retinal Cells (Pooled)

Cat. No.	Product	Description
R-Ret-508	Rat retinal cells, cryopreserved	≥ 0.2 x 10 ⁶ viable cells in a 0.5 ml cell suspension

Primary Neuron Growth Media (Sold Separately):

Cat. No.	Product	Description
CC-4461	PNGM™ BulletKit™ Medium	200 ml PNBMTM Basal Medium plus CC-4462 SingleQuots™ Kit to formulate PNGM™ Medium (growth medium)
CC-3256	PNBMTM Basal Medium	Primary neuron basal medium (200 ml)
CC-4462	PNGM™ SingleQuots™ Kit	Formulates 200 ml of PNBMTM Basal Medium to PNGM™ Growth Medium; contains NSF-1, 4.0 ml; L-glutamine, 2.0 ml; GA, 0.2 ml
14-503E	FBS, heat inactivated	100 ml

Product Warranty

Cultures have a finite lifespan *in vitro*.

Lonza guarantees the performance of its cells in the following manner only if Clonetics™ Media and Reagents are used exclusively and the recommend protocols are followed. The performance of cells is not guaranteed if any modifications are made to the complete cell system.

1. Clonetics™ Rat Retinal Cells are assured to be viable and functional when thawed and maintained properly.
2. Clonetics™ Rat Retinal Cells are cryopreserved immediately after isolation without culturing prior to cryopreservation. Routine characterization of retinal cells includes positive immunostaining for neuron specific class III β-tubulin (Tuj-1), specific neuronal protein gene product (PGP

9.5), ganglion cell marker, Thy1.1, and GFAP. Lonza guarantees rat retinal cells will express the markers described when plated out of cryopreservation onto Matrigel.

When placing an order or to contact Scientific Support, please refer to the product numbers and descriptions listed above. For a complete listing of all Clonetics™ Products, refer to the Lonza website or the current Lonza catalog. To obtain a catalog, obtain additional information or want to speak with Scientific Support, you may contact Lonza by web, e-mail, telephone, fax or mail (See page 1 for details).

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

WARNING: Handle as a potentially biohazardous material under biosafety level 1 containment. These cells are not known to contain an agent known to cause disease in healthy adult humans. These cells have not been screened for hepatitis B, human immunodeficiency viruses or other adventitious agents. If you require further information, please contact your site safety officer or Scientific Support.

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