

NEONATAL CARDIOMYOCYTE ISOLATION SYSTEM

Tissue Dissociation/Cell Isolation





The Worthington Neonatal Cardiomyocyte Isolation System has been introduced to provide researchers with a reliable, convenient, and consistent cell isolation system. By utilizing purified rather than crude enzyme preparations, it has been possible to minimize the lot-to-lot variation. In addition, Worthington use-tests the kits by isolating cardiomyocytes from neonatal rat hearts to assure performance, reliability and consistent yield of viable cells.

The kit has been formulated in conjunction with Dr. Ronal MacGregor¹. The method is based on that described by Toraason², et al. (1988) in which the minced tissue is incubated overnight with purified trypsin at 4°C. As pointed out by Toraason, this step reduces the hands-on time required to harvest cells compared to the time involved in sequential incubations in warm trypsin or collagenase. Also, purified collagenase is utilized to maximize yield and viability. The kit includes five single-use vials each of purified trypsin, collagenase and trypsin inhibitor supplied along with Hank's Balanced Salt Solution, Leibovitz L-15 culture media, Falcon Cell Strainers and detailed instructions.

Description	Code	Catalog No.	Size	Price
Neonatal Cardiomyocyte Isolation System	NCIS	LK003300 LK003303	1 Kit 3 Kits	\$288.00 785.00
Individual Components				
Trypsin Vials	TRLSNK	LK003220 LK003225	1 vi 5 vi	\$ 14.00 48.00
Collagenase Vials	CLSPANK	LK003240 LK003245	1 vi 5 vi	\$ 32.00 130.00
Inhibitor Vials	SICNK	LK003230 LK003235	1 vi 5 vi	\$ 15.00 45.00
Hank's Buffer	HBSS	LK003210	500 ml	\$ 58.00
L-15 Media Powder	L15NK	LK003250	1 x 1L	\$ 30.00
Cell Strainers	CELSTRNK	LK003265	5 ea	\$ 28.00

Description and Package Contents

The package contains sufficient materials for five separate tissue dissociations, each containing up to twelve hearts. For larger or smaller tissue samples prepare proportionate volumes of reagents at each step and combine them in the same ratio as described in the protocol.

Phone: 800.445.9603 • 732.942.1660 • Fax: 800.368.3108 • 732.942.9270

Vial #1: HBSS, 1 bottle, 500 ml Sterile calcium and magnesium-free Hank's Balanced Salt Solution (CMF HBSS), pH 7.4. The solution is used for reconstituting the contents of Vials #2 and #3 in addition to serving as the medium for the dissociation.

Vial #2: Trypsin Vial, 5 vials, 1,000 μg each Worthington Trypsin (Code: TRLS), 3X crystallized, dialyzed against 1mM HCl, filtered through 0.22μm pore size membrane, and lyophilized. Before use, reconstitute with 2ml CMF HBSS (Vial #1) and swirl gently to dissolve contents. Store at 2–8°C.

Vial #3: Inhibitor Vial, 5 vials, 2,000 μg each Worthington Soybean Trypsin Inhibitor (Code: SIC), a 0.22μm pore size membrane filtered, lyophilized powder. This material is 0.22 micron membrane filtered and lyophilized in autoclaved vials. A vial reconstituted with 1 ml of HBSS or equivalent yields a solution of 2 mg/ml of trypsin inhibitor, Code SIC. Before use, reconstitute with 1ml CMF HBSS (Vial #1) and swirl gently to dissolve contents. Store at 2–8°C.

References

- 1. MacGregor, Ronal R, Klein, Robert M, Bansal, David D.: Secretion of Plasminogen Activator Activity from Neonatal Rat Heart Cells is Regulated by Hormones and Growth Factors, Ann N Y, *Acad Sci.*, 752: 331-42, 1995.
- 2. Toraason, Mark, Luken, Mary E., Breitenstein, Michael, Krueger, John A., and Biagini, Raymond E.: Comparative Toxicity of Allylamine and Acrolein in Cultured Myocytes and Fibroblasts from Neonatal Rat Heart, *Toxicology*, *56*, 107 1988.

Vial #4: Collagenase Vial, 5 vials, 1,500 Units each Worthington Purified Collagenase (Code: CLSPA), a 0.22μm pore size membrane filtered, lyophilized powder which has been chromatographically purified. It contains less than 50 caseinase units per milligram and is composed of two separable but very similar collagenases. Before use, reconstitute with 5ml Leibovitz L-15 Media (prepared as described below) and swirl gently to dissolve contents. Store at 2–8°C.

Pouch Containing Leibovitz L-15 Media Powder: 1x1 L Reconstitute entire contents of pouch by cutting open top of envelope and pouring contents into beaker containing 800ml of cell culture grade water. Rinse pouch 2 - 3 times with additional 100ml. Bring total volume to 1 liter and filter through a 0.22µm pore size filter. Store at 2-8°C.

The kit also includes 5 Cell Strainers (Falcon), and a card correlating phenol red color with pH for checking the pH of balanced salt solution and culture medium.

For a listing of up-to-date enzyme and biochemical citations, go to: http://Worthington-Biochem.com/index/manual.html

Related Products

Cell Isolation Optimizing System • Collagenase • Elastase • Hyaluronidase • Neutral Protease (Dispase®) • Papain Papain (Neural) Dissociation System • Hepatocyte Isolation System • STEMxyme® 1 & 2 Collagenase/Neutral Protease Blends • Trypsin