Cell Line Designation: INS-1
AddexBio Catalog No. C0018007
Cell Line Description:

Disease: Insulinoma (Radiation induced cell immortalization)
Origin: Pancreatic Islets
Species: Rattus Norvegicus
Tissue: Pancreas
Properties: Adherent; β cells
Recommended Culture Flask: BD Biosciences Tissue Culture-treated flask
Complete Medium: AddexBio Optimized RPMI-1640 (C0004-02) + 10% FBS + 0.05 mM 2-mercaptoethanol (Gibco 21985023)
Subculture Procedure: 1:2 to 1:3 using 0.25% trypsin or trypsin/EDTA, 5% CO2; 37°C
Medium Renewal: Two to three times weekly.
Freezing Medium: Complete culture medium supplemented with 5% (v/v) DMSO
Additional Information: Additional product and technical information can be obtained from the catalog references and the Addexbio Technical Information site at www.addexbio.com, or by email at customersupport@addexbio.com.

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. laboratory safety is discussed in the following publication: Biosafety in Microbiological and Biomedical Laboratories, 5th ed. HHS Publication No. (CDC) 93-8395. U.S. Department of Health and Human Services, Centers for Disease Control and Prevention. Washington DC: U.S. Government Printing Office; 2007. The entire text is also available online at www.cdc.gov/od/ohs/biosafty/bmbl4/bmbl4toc.htm

Use Restrictions: **These cells are distributed for research purposes only.** Addexbio does not recommend third party distribution of this cell line, as this practice has resulted in the unintentional spreading of contaminated cell lines.

Handling Cells Upon Arrival:

Frozen cells must be thawed immediately upon receipt and grown according to the handling procedures described here in this instruction manual to ensure the best cell viability.
Note: ***Avoid refreezing or repetitive freezing cells upon receipt*** *as it may result in irreversible damage to the cell line.*
Disclaimer: We cannot guarantee cell viability if the cells are not thawed immediately upon receipt and grown according to handling procedures described in this instruction manual.

Handling Procedure for Frozen Cells:

To insure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be

rev. 04/2019
AddexBio Technologies
4907 Morena Blvd, Ste 1408
San Diego, CA 92117 USA
858-348-7819
Fax: 858-538-8847
Email:customersupport@addexbio.com
www.addexbio.com

stored in liquid nitrogen vapor phase and not at -70°C. Storage at -70°C will result in loss of viability.

Safety Precaution:

Addexbio highly recommends 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 O-ring and cap out of 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. Resuspend cell pellet with the recommended complete medium and dispense into a new culture flask (T25). 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 complete growth medium be placed into the incubator for at least 15 minutes to allow the medium to reach its normal pH (7.0-7.6).
4. Incubate the culture at 37°C in a suitable incubator. A 5% CO_2 in air atmosphere is recommended.

References for INS-1 cells:

1. Chang TJ, Tseng HC, Liu MW, Chang YC, Hsieh ML, Chuang LM. Glucagon-like peptide-1 prevents methylglyoxal-induced apoptosis of beta cells through improvingmitochondrial function and suppressing prolonged AMPK activation.Sci Rep. 2016 Mar 21;6:23403. doi: 10.1038/srep23403. PMID: 26997114

rev. 04/2019
AddexBio Technologies
4907 Morena Blvd, Ste 1408
San Diego, CA 92117 USA

858-348-7819
Fax: 858-538-8847
Email:customersupport@addexbio.com
www.addexbio.com

Lot Specific Information Sheet for AddexBio Cat #: C0018007

Lot Number: 0021133

Designation: INS-1 CELLS

Total Cells/mL: >$1x10^5$

Expected Viability: 70.0-75.0%

Ampule Passage #: 8

Dilute Ampule Content: 1:10 (T-25) or 1:15 (T-75)

Volume/Ampule: 1 mL

A T-25 setup at a dilution of 1:10, using culture medium as described in the product information sheet, reaches approximately 40-45% confluence within 24 to 48 hours.

Remarks to INS-1 cell line (Please read before you proceed):

This cell line is not attaching well at the beginning when you thaw the cells or after trypsinized. It may look messy at first. Therefore, please allow more time to let them attach to the flask (usually 24-48 hours without changing the medium). It is normal to observe floating cells the first few days after thawing. This cell line grow slowly. There are times cells may form attached clumps and spread slowly after thawing. After 3 days of thawing, trypsinize the cells (even not expanding) and plate the cells on 2 flasks for them to expand.

Please make sure that sodium pyruvate, HEPES, and mercaptoethanol at the specified concentration are added to the medium for the cells to grow. Missing any of these components will result in failure to attach and grow.

Please make sure you are using a medium that is comparable to our recommended medium as there are many variations of the media. To guarantee the best result, we request that our medium to be used first for cell thawing and recovery until seeds are frozen. You may then switch to your own medium of your choice. Or else warranty will be voided.

rev. 04/2019
AddexBio Technologies
4907 Morena Blvd, Ste 1408
San Diego, CA 92117 USA

858-348-7819
Fax: 858-538-8847
Email:customersupport@addexbio.com
www.addexbio.com