

# Culturing MSC Derived Human Chondrocytes

Catalog Number: SC00A7-500

**Description:** Cells are shipped in vials on dry ice.

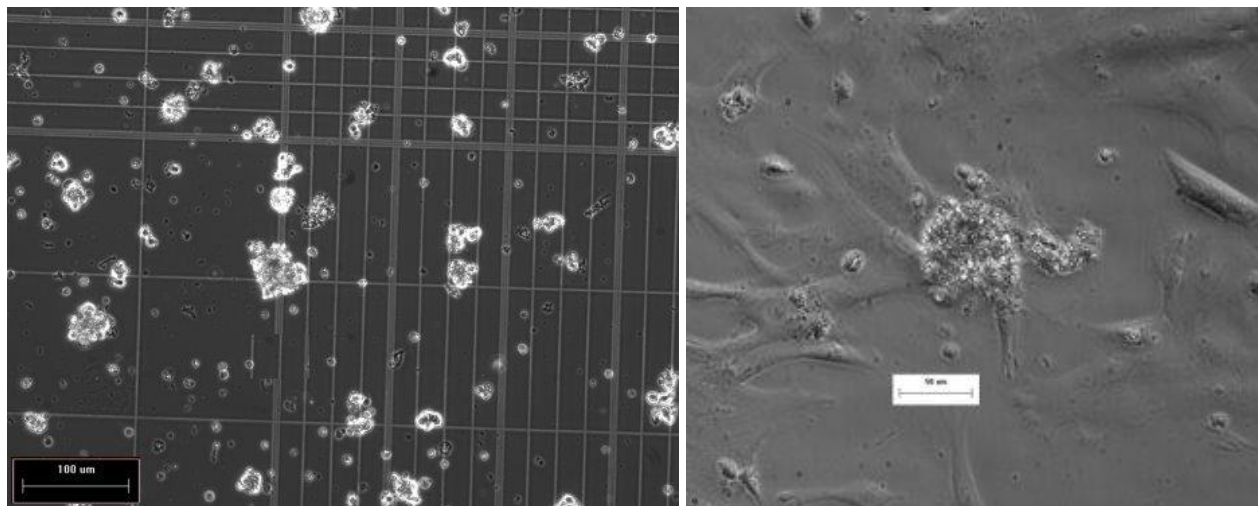
**Storage:** If stored, it is preferable to store in the vapor phase of liquid N<sub>2</sub>. Storage in a -80°C freezer may be used but is likely to result in diminished cell viability proportional to storage time.

## Introduction

We are pleased to be one of the first to offer primary human chondrocytes. There are derived from our human mesenchymal stem cells and designed for the study of cartilage metabolism and related degenerative diseases. We offer both label and unlabeled cells with the number of cells available ranging from 100,000 to 4,000,000. Our desire is to make it easy to “buy and try”. We also want to have a convenient way to ramp up your assays for higher content and through put screening.

All lots of our chondrocytes cells are cultured and characterized in house (includes testing by PCR methods for known viral contaminants and mycoplasma). Cell authentication methods include karyotyping and PCR for actin, cytochrome B and COX1. The multi-cell structures average 20 micrometers in diameter with minimal amounts of non-differentiated MSCs (<10%).

**Note:** Use of these cells requires prior experience in standard methods of mammalian cell culture.



**Images:** **Left:** Native chondrocyte culture. **Right:** Phase contrast image (200x) of chondrocyte and associated cells in day five of culture as described (SC00A7 Protocol). Scale bar = 50 µm. Chondrocyte is darker multicellular structure overlying attached cells, ~ 70 µm in diameter.

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## Protocol

**IMPORTANT:** All steps must be performed using sterile technique in a Laminar Flow Hood.

**Preparation:** These cells require culture in a 37oC, CO2 cell culture incubator, calibrated to 5% CO. Also, a water bath equilibrated to 37oC is needed. Required reagents include 1 x PBS, Fisher Catalog number BP665-1 or equivalent at room temperature or 37oC and chondrocytes culture medium. We provide Chondrocyte Maintenance Medium (Catalog number SC00PC3-1: 500 ml or SC00PC3-2: 100 ml) for optimal chondrocyte maintenance in cell culture. Other commercially available media may be used.

**Note:** Exposure of closed vials containing liquid nitrogen to a 37oC water bath is an explosion hazard! It is essential to ensure that no liquid nitrogen is present in the vial! If liquid nitrogen is present in the vial, allow this to evaporate before proceeding. Please use proper precautions including appropriate gloves to protect skin from exposure to liquid nitrogen, eye protection and other personal protective equipment when transferring vials containing cryopreserved cells from liquid nitrogen to a 37oC water bath.

Provide continuous agitation, e.g., swirling, to the vial while it is submerged in the 37oC water bath. Continue with agitation until the cells are completely thawed and no ice remains within the cell suspension, usually about 1 to 2 minutes. Maximum cell viability is dependent on rapid and complete thawing of frozen cells.

Our products are provided in 0.5 ml (500 l) of cryopreservation medium at a concentration of 500,000 cells/ml. Cell number was determined by dissociation of chondrocytes by enzymatic treatment while intact chondrocytes are provided within the final product vial. The number of chondrocytes contained within the vial is also indicated on the product label. We recommend direct inoculation of cultures from the cell-cryopreservation media suspension without washout of this medium.

Cultures may be established at the density required for a given application. We suggest establishing the initial culture at a plating density at about 5 to 10 or more chondrocytes per well of a 96 well plate (about 25 chondrocytes/cm<sup>2</sup> ). Add the appropriate volume of SC00PC3 culture medium to the plate or flask to be used for culture. Allow cultures to incubate in 5% CO<sub>2</sub> in ambient O<sub>2</sub> in non-TC coated culture plates or flasks at 37oC in a humidified environment. Chondrocyte maintenance medium (SC00PC3) is does not support chondrocyte proliferation but does maintain cellular viability.

**Note:** Should any issues arise while using our cells, our team is here to help troubleshoot any issues. Our cells are backed by our one-time replacement or refund policy. Our recommended protocol including recommended products must be used to be eligible for replacement or refund. Cells that have been refrozen are no longer eligible for refund or replacement.

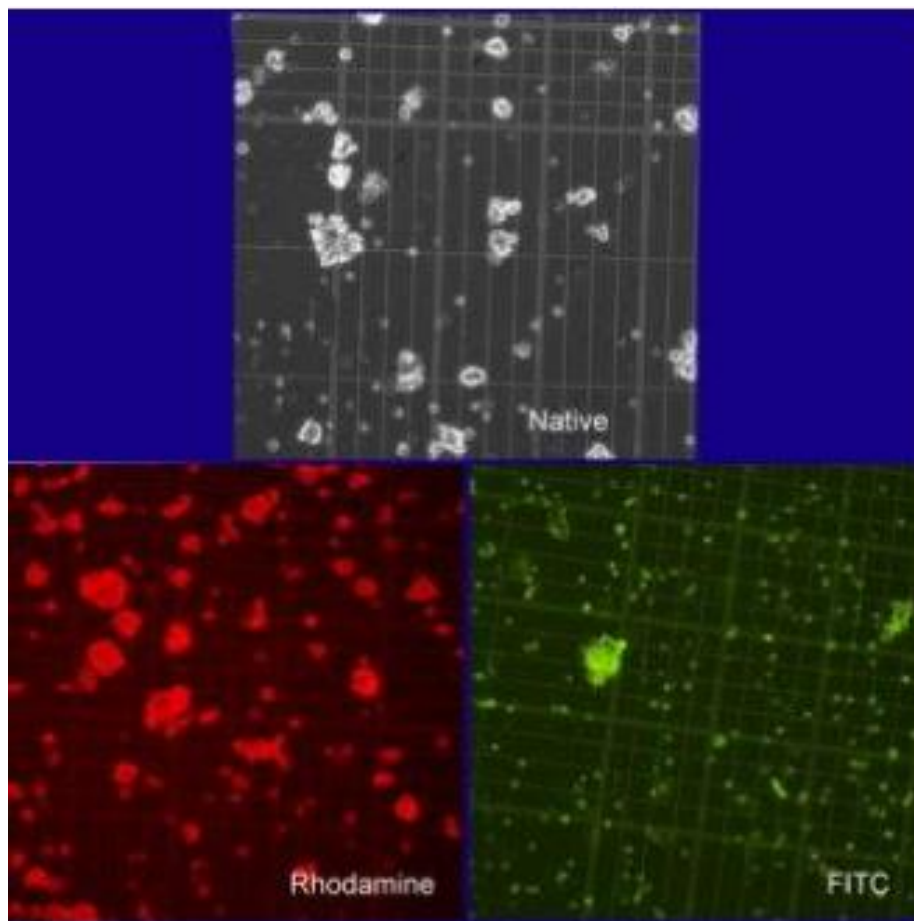
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*Image: Chondrocyte culture*



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