Chondrogenic Differentiation Small molecule compound Kit-4（Sterile Solution, Without-Alcian blue）

Cat: IK-CHD-4
Storage: -20℃, 6 months

Introduction

This Kit is a small molecule compound kit designed by Solarbio for chondrogenic differentiation. It does not contain TGF-β, and customers need to prepare appropriate TGF-β separately. This Kit selected five classic basic reagents (sterile) that can be used for adipogenic induction, aiming to build a ' one-stop service platform ' for customers, saving time and effort. The products in the library have passed the biosafety detection and product quality detection, with stable and effective performance, small batch difference, good biological activity, and a large number of literatures have been verified in many ways, and the quality is reliable.

Kit Components

Kit components	Size	Storage
Reagent 1　10mM dexamethasone	50μL	-20℃
Reagent 2　25 mg / mL vitamin C	2*300μL	-20℃
Reagent 3　100 mg / mL L-proline	120μL	-20℃
Reagent 4　100 × ITS Supplement	1mL	2-8℃
Reagent 5　1M sodium pyruvate	120μL	-20℃

Note

a. Reagent 1, Reagent 2, Reagent 3, Reagent 4 and Reagent 5 are all sterile solutions, which can be directly added to the medium for use.
b. Vitamin C is more unstable after dissolution. It is recommended to freeze it by self-packaging (avoid repeated freezing and thawing). When changing the liquid, take out a thaw and use it. This Kit gives an additional reagent two.
c. Reagent 4 is an animal-free component with a concentration of 1.0 mg/mL insulin, 0.550 mg/mL transferrin and 0.67 μg/mL sodium selenite.
d. Before use, please immediately centrifuge each tube of small doses of reagents to avoid loss.
e. This product is for scientific research only. Do not use for medicine, clinical diagnosis or treatment, food and cosmetics. Do not store in ordinary residential areas.
f. For your safety and health, please wear a good lab coat and wear disposable gloves and masks.

Product Features

1. This Kit is a universal chondrogenic induction small molecule compound kit without TGF-β,

and customers need to prepare appropriate TGF-β separately.

2. This Kit selected several classic small molecule compounds for chondrogenic induction and was equipped with Alcian blue staining solution.
3. This kit is a ready-to-use Kit, which can be used directly without further preparation or sterilization by customers.
4. The company provides TGF-β of different species and types. Customers can contact our company to purchase and customize the exclusive cartilage Kit.

Protocols *(only for reference)*

The level of chondrogenic differentiation of stem cells varies with cell type, cell donor source, culture conditions, cell passages, cell status and differentiation time. The following methods are for reference only, and customers need to adjust according to the actual induction situation.

Chondrogenic differentiation induction operation (plane induction)

1.Cell differentiation induction

The cells in the logarithmic growth phase were digested and counted. The cells were resuspended in chondrogenic differentiation medium I (containing TGF-β, dexamethasone, vitamin C, L-proline, 1 × ITS Supplement, sodium pyruvate). After centrifugation, the cell density was adjusted to 1.0 ~ 2.0 × 10^7 cells/mL.

A total of 20 μL cell suspension (about 2.0~4.0×10^5 cells) was dropped to the center of the 24-well plate. The cells were cultured at 37°C, 5% CO_2 for 2 ~ 3 h to make the cells adherent.

After 2 ~ 3 h, 1 mL chondrogenic differentiation medium I (containing TGF-β, dexamethasone, vitamin C, L-proline, 1 × ITS Supplement, sodium pyruvate) was supplemented for normal culture. The liquid was changed every 2 ~ 3 days. The cells were induced for 21 ~ 28 days according to the above frequency of medium change, and the morphological changes of the cells were observed.

2. Allixin blue staining identification

Chondrogenic differentiation induction operation (three-dimensional culture)

1. Preparation of stem cells

The cells in the logarithmic growth phase were digested and counted, and 3 × 10^5 cells were transferred to a 15 mL centrifuge tube, and 250 g was centrifuged for 4 min.

The supernatant was discarded and 0.5 mL chondrogenic differentiation medium II (containing dexamethasone, vitamin C, L-proline, 1 × ITS Supplement, sodium pyruvate, without TGF-β) was added. The cells were resuspended and centrifuged at 150 g for 5 min.

The supernatant was carefully discarded, and 0.5 mL of chondrogenic differentiation medium I (containing TGF-β, dexamethasone, vitamin C, L-proline, 1 × ITS Supplement, sodium pyruvate) was added. The cells were resuspended and centrifuged at 150 g for 5 min.

The lid of the 15 mL centrifuge tube was slightly opened and placed in a 37°C, 5% CO_2 culture environment.

2. Cell differentiation induction

After 24 h, the cell precipitation deformation and agglomeration were observed. If there were obvious changes, the bottom of the tube was gently moved, and the cell mass was tried to leave the bottom of the tube, and all the cells were infiltrated in the induction fluid.

The cells were cultured at 37°C, 5% CO_2 for about 21 days, and the freshly prepared chondrogenic differentiation medium I (containing TGF-β, dexamethasone, vitamin C, L-proline, 1 × ITS Supplement, sodium pyruvate) was usually replaced every 2 days. Pay attention to the observation of cell pellet formation and surface smoothness, determine the termination of cell induction time, and staining identification.

3. Allixin blue staining identification

Related Products

IK-LIN-7 Adipogenic induction of small molecule compound Kit-7 (ready-to-use, containing saturated oil red O)

IK-OIN-5 Osteogenic induction of small molecule compound Kit-5 (ready-to-use, containing alizarin red staining solution and 10% CPC).

IK-CHD-3 Cartilage-induced small molecule compound Kit (i.e., containing alixin blue)

IKM1020 10×Protease and Phosphatase Inhibitor Cocktail (Universal type)

IKM1010 100× Protease Inhibitor Cocktail MIX (Universal type)

IKC1032-1 CEPT Cocktail Kit

P02149 Recombinant Human/Mouse/Rat TGF-β3/TGF-beta 3/TGFB3/Transforming Growth Factor β-3

P00121 Recombinant Human TGF-beta 1/TGFB1