SILAC-RPMI (K6R10) Kit

Catalog Number: SM202107

(Version 1.0)

Description

SILAC (*Stable Isotope Labeling with Amino acids in Cell culture*) is a powerful quantitative proteomic method to identify and quantify the relative changes of protein abundance in complex protein samples. This approach utilizes the *in vivo* metabolic incorporation of "heavy" ^{13}C- or/and ^{15}N-labeled amino acids (Lysine, Arginine, etc.) into proteins during cell culture, introducing the specific "MS tag" on proteins in comparison with the unlabeled counterpart, which enables the mass spectrometry (MS) to comprehensively identify, characterize, and quantify the proteins.

Contents

Product	Cat. No.	Contents	Storage
SILAC-RPMI (K6R10) Kit	SM202107	1×500ML (K0R0)	4°C
		1×500ML (K6R10)	4°C
		2×50ML D-FBS (DF-50)	-20°C

SILAC-RPMI (K6R10) Kit is designed to compare two groups of samples using SILAC approach. The formulation of **K6R10** is identical to that of **K0R0** except that **$^{13}C_6$-Lysine (K6)** and **$^{13}C_6$, $^{15}N_4$-Arginine (R10)** replace the **Lysine (K0)** and **Arginine (R0)**. Because of their identity, **K0R0** is the ideal control medium for **K6R10**.

When two groups of cells were cultured in parallel, the additional **6** or **10 Dalton** of "MS tag" would be introduced to the Lysine and Arginine residues respectively on the proteins of cell cultured in **K6R10** medium compared to that of maintained in **K0R0** medium, and the "MS tag" is the key for protein relative quantitation by downstream mass spectrometry analysis.

Usage & Application

1. Before use, add 10% (v/v) of D-FBS (DF-50) into **K0R0** and **K6R10** media (complete media) respectively.
2. Use **K6R10** complete medium to culture cells over six passages, extract proteins,

and check the SILAC labeling efficiency by mass spectrometry.

3. Cryopreservation of **K6R10**-labeled cells with the standard protocol for long-term use.
4. For primary cells and cell lines sensitive to dialyzed FBS, SILAC method is not recommended.

- **Troubleshooting**

Problem	Cause	Solution
1. Cells are not viable or grow poorly. 2. Cell morphological changes.	Probably due to dialyzed FBS. Dialyzed FBS lacks some small molecules important for cell growth.	1. Change cell lines insensitive to dialyzed FBS. 2. Increase the dialyzed FBS up to 15-20%. 3. Try and test the dialyzed FBS from other vendors.