

## SILAC-RPMI (K6R6) Kit

Catalog Number: SM202106 (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" <sup>13</sup>C- or/and <sup>15</sup>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 (K6R6) Kit	SM202106	1×500ML (K0R0)	4°C
		1×500ML (K6R6)	4°C
		2×50ML D-FBS (DF-50)	-20°C

SILAC-RPMI (K6R6) Kit is designed to compare two groups of samples using SILAC approach. The formulation of <u>K6R6</u> is identical to that of <u>K0R0</u> except that <sup>13</sup>C<sub>6</sub>-Lysine (K6) and <sup>13</sup>C<sub>6</sub>-Arginine (R6) replace the Lysine (K0) and Arginine (R0). Because of their identity, <u>K0R0</u> is the ideal control medium for <u>K6R6</u>.

When two groups of cells were cultured in parallel, the additional **6 Dalton** of "MS tag" would be both introduced to the <u>Lysine</u> and <u>Arginine</u> residues on the proteins of cell cultured in <u>K6R6</u> medium compared to that of maintained in <u>K0R0</u> 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 K6R6 media (complete media) respectively.
- 2. Use <u>K6R6</u> complete medium to culture cells over six passages, extract proteins, and check the SILAC labeling efficiency by mass spectrometry.



- 3. Cryopreservation of <u>K6R6</u>-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	Probably due to	1.	Change cell lines	
	or grow poorly.	dialyzed FBS. Dialyzed		insensitive to dialyzed	
2.	Cell morphological	FBS lacks some small		FBS.	
	changes.	molecules important for	2.	Increase the dialyzed	
		cell growth.		FBS up to 15-20%.	
			3.	Try and test the	
				dialyzed FBS from	
				other vendors.	