

## Stable Isotope Labeling with Amino Acids in Cell Culture (SILAC)

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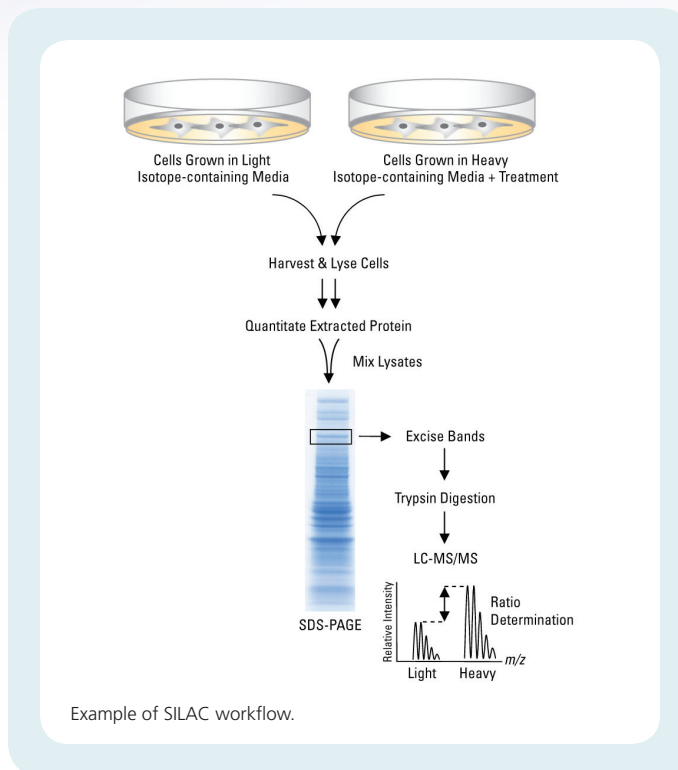
Stable isotope labeling with amino acids in cell culture (SILAC) is a simple and straightforward approach for *in vivo* incorporation of a label into proteins for mass spectrometry (MS)-based quantitative proteomics. SILAC relies on metabolic incorporation of a given "light" (unlabeled) or "heavy" (labeled) form of the amino acid into the proteins. The method relies on the incorporation of amino acids with substituted stable isotopic nuclei (e.g.  $^{13}\text{C}$ ,  $^{15}\text{N}$ ). Thus, in an experiment, two cell populations are grown in culture media that are identical except that one of them contains a "light" and the other a "heavy" form of a particular amino acid (e.g.  $^{12}\text{C}$  and  $^{13}\text{C}$  labeled L-Lysine, respectively). When the labeled analog of an amino acid is supplied to cells in culture instead of the natural amino acid, it is incorporated into all newly synthesized proteins. After a number of cell divisions, each instance of this particular amino acid will be replaced by its isotope-labeled analog. Since there is hardly any chemical difference between the labeled amino acid and the natural amino acid isotopes, the cells behave exactly like the control cell population grown in the presence of normal amino acid. It is efficient and reproducible as the incorporation of the isotope label is 100%. We anticipate that potential applications of SILAC will lead to its use as a routine technique in all areas of cell biology.

### SILAC Highlights:

- **Efficient** –100% label incorporation into proteins of cultured cells
- **Reproducible** – eliminates experimental variability caused by differential sample preparation
- **Flexible** – media deficient in both L-Lysine and L-Arginine, allowing for better proteome coverage through dual amino acid isotope labeling
- **Compatible** – label proteins expressed in a wide variety of mammalian cell lines adapted to grow in DMEM or RPMI 1640 medium, including HeLa, 293T, COS7, U2OS, A549, A431, HepG2, NIH 3T3, Jurkat and others
- **99% Enriched High Quality Reagents** – Stable isotope-labeled amino acids with 99% isotopic enrichment and 98%+ chemical purity

### SILAC Applications:

- Quantitative analysis of relative changes in protein abundance from different cell treatments
- Quantitative analysis of proteins for which antibodies are unavailable
- Protein expression profiling of normal vs. disease cells
- Identification and quantification of hundreds to thousands of proteins in a single experiment



### SILAC Literature References

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