



RESEARCH PRODUCTS

¹³C-Enriched Carbon Sources for Solid State NMR of Proteins

• BioExpress[®] 1000 • Glycerol • Glucose

Tailoring ¹³C Enrichment of Proteins for Solid State NMR

For many years, the use of highly enriched, uniformly isotopelabeled recombinant proteins has enabled the study of protein structure and dynamics by NMR spectroscopy. This approach has been relatively easy to perform since uniformly ¹³C-labeled carbon sources such as ¹³C₆ glucose have been readily available. Unfortunately, uniform ¹³C labeling of highly enriched proteins is not conducive to some NMR experiments (such as solid state NMR experiments using stationary aligned samples) because strong, homonuclear ¹³C-¹³C dipole-dipole coupling can act to broaden ¹³C resonances very significantly, sometimes so severely that they are beyond detection. An effective approach to alleviating this problem is to use proteins containing spatially isolated ¹³C sites. Such sparsely labeled proteins can either be produced using uniform, random, fractionally enriched carbon sources or specific ¹³C-labeled carbon sources.

Uniform Fractionally 13 C-Enriched Bacterial Cell Growth Media and Glucose (${}^{13}C_s$)

CIL is also pleased to offer fractionally ¹³C-enriched BioExpress[®] 1000, as well as fractionally enriched glucose, for those researchers that use minimal essential media instead of a ready-made rich culture medium.

Catalog No.	Description	Amount
CGM-1000-CN-25-S	BioExpress [®] 1000 (¹³ C, 25%; ¹⁵ N, 98%)	10 mL (10x)
CGM-1000-CN-25	BioExpress [®] 1000 (¹³ C, 25%; ¹⁵ N, 98%)	100 mL (10x)
CGM-1000-CN-35-S	BioExpress [®] 1000 (¹³ C, 35%; ¹⁵ N, 98%)	10 mL (10x)
CGM-1000-CN-35	BioExpress [®] 1000 (¹³ C, 35%; ¹⁵ N, 98%)	100 mL (10x)
CLM-1396-25-1	Glucose (13C, 24-25%)	1 g

The number of adjacent ¹³C sites in most amino acid residues can be minimized by using uniform fractionally ¹³C-enriched bacterial cell growth media or glucose [$^{13}C_6$]. Opella et al.¹ have reported optimal results using BioExpress[®] media with fractional ¹³C enrichments between 25% and 35%.



Specific ¹³C-Labeled Carbon Sources

CIL offers the following specific ¹³C-labeled substrates for use in the microbial expression of proteins labeled with isolated ¹³C sites.

Catalog No.	Description	Amount
CLM-1397	Glycerol (2-13C, 99%	1g
CLM-1857	Glycerol (1,3-13C, 99%)	1g
CLM-746	Glucose (2-13C, 99%)	1g
CLM-441	Sodium bicarbonate (¹³ C, 99%)	1g

Glycerol $[1,3^{-13}C_2]$ and glycerol $[2^{-13}C]$ have been shown to give rise to complementary labeling patterns. When choosing the appropriate specific ¹³C-labeled carbon source, the ¹³C enrichment of the expressed protein can be tailored to highlight regions of interest. Sodium bicarbonate (¹³C) should be added to growths using 2-¹³C glycerol.

Advantages

Glycerol (2-13C)

- labels alpha carbons
- increases overall labeling of carbons
- significantly lower levels of carbonyl and aliphatic sidechain carbons

Glycerol (1,3-13C₂)

• creates reduced intensity in the α -carbon region.¹

Glucose (2-13C)

- minimizes adjacent ¹³C pairs in most residues
- reported to be more useful than specifically labeled glycerol in detecting backbone ¹³C resonances.¹

Reference

1. Filipp, F.V.; Sinha, N.; Jairam, L.; Bradley, J.; Opella, S.J. 2009. Labeling strategies for ¹³C-detected aligned-sample solid-state NMR of proteins, J Magn Reson, 201, 121-130.