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Product No. 19942

19942E-2010-0

DL-Amino Acid Labeling Kit

Features

- This kit is designed to label amino acids prior to HPLC analysis.

- Since the labeling reagent contains a chiral carbon, the labeled amino acids can be optically separated by an achiral column such as C18.
- The detection sensitivity of MS (mass spectrometer) is higher than when used with conventional optical labeling reagents.

Component

Reagents	Volume	Quantity
Labeling agent solution	10 mL	1
Start solution	10 mL	1
Delabeling agent solution for side chain st_1	10 mL	1
Stop solution	10 mL	1

^{*1} Delabeling agent solution for side chain contains 6-Mercapto-1-hexanol [MW 134.24 (C₆H₁₄OS)].

Required equipment

- Sealable glass vessels, such as 1.5 mL glass vials
- Heating device at 50 °C
- Pipettes (for quantitative analysis in which precise weighing is necessary, it is recommended to use glass pipettes or positive-displacement micropipettes)
- Vortex mixer
- Acetonitrile or methanol (for HPLC)

Structural formula of labeling reagent and labeling reaction



Figure 1. Structural formula (a) and schematic diagram (b) of (D)FDLDA (labeling reagent)



Figure 2. Labeling reaction (labeling of tyrosine)

The labeling reagent reacts with amino groups. In addition, some side chains, such as the phenolic hydroxyl group of tyrosine and the thiol group of cysteine, are also labeled. See Table 1 for details.

Protocols

Note regarding sample solutions:

- The total amount of functional groups to be reacted should not be more than 1.0 µmol. If the concentration is higher, dilute or reduce the amount added.
- Neutralize sample solutions before performing the labeling reaction.

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Protocol 1

- 1. Add 100 µL each of sample solution, labeling agent solution, and start solution to a glass vessel, seal the vessel, mix in a vortex mixer for 5 seconds, and then react at 50°C for 2 hours.
- 2. Add 100 µL of the delabeling agent solution for side chain, mix in a vortex mixer for 5 seconds, and then react at 50 °C for 15 minutes.

[This step allows the removal of label groups from the side chains of amino acids (except Lys).]

3. Add 100 µL of stop solution and 500 µL of acetonitrile or methanol, then analyze by HPLC (after filtration, if necessary).

Protocol 2

- 1. Add 100 µL each of sample solution, labeling agent solution, and start solution to a glass vessel, seal the vessel, mix in a vortex mixer for 5 seconds, and then react at 50 °C for 2 hours.
- 2. Add 100 µL of stop solution and 500 µL of acetonitrile or methanol, then analyze by HPLC (after filtration, if necessary).

Amino acids	Protocol 1	Protocol 2	Remarks				
Tyr, Cys	mono	Di	 The mono form of Cys may react with 6-Mercapto-1-hexanol in the delabeling agent solution for side chain during storage. mono: Only the amino group of the a-carbon is labeled. di: In addition to the a-carbon, the functional groups on the side chains are also labeled. 				
Lys	di	di					
His	mono	Mixture of mono and di					
Others	mono	mono					

Table 1. The structure of labeled amino acids by different protocols

Table 2. Kit-derived peaks

Compounds	Molecular weight		Explanations
(D)FDLDA	385.39	(C ₁₆ H ₂₄ FN ₅ O ₅)	Unreacted labeling reagent
(D)FDLDA (Hydrolysed)	383.40	(C16H25N5O6)	Hydrolysate of labeling reagent
(D)FDLDA-S-C₀H12OH	499.63	(C ₂₂ H ₃₇ N ₅ O ₆ S)	Reaction product of labeling reagent with 6-mercapto-1-hexanol

% It is strongly recommended that a blank analysis is performed prior to analyzing your sample.

Cautions

- Wear protective glasses and gloves.

- Wash with copious amount of water if your skin is exposed to reagents.

- Labeled amino acids should be stored in the freezer (-20 °C).

Storage

Avoid extreme heat (store at 15 - 25 °C)

Packing

100 TESTS (Product No.: 19942-74)

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