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AccQ•Tag Ultra Derivatization Kit

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I. INTRODUCTION

The AccQ•Tag[™] Ultra Derivatization Kit is a component of the UPLC[®] Amino Acid Analysis Application Solution. This application solution is an integrated combination of instrumentation, derivatization chemistry, separation column and eluents, methods, and software. Analysts are assured of accurate and precise amino acid analyses with the complete application solution. The use of the AccQ•Tag Ultra Derivatization Kit without the rest of the application solution is not supported as an amino acid analysis method because there are significant risks of inaccurate qualitative and quantitative analysis.

The AccQ•Tag Ultra Derivatization Kit differs from the AccQ•Tag Derivatization Kit (P/N WAT052880). Although the specified chemical composition is the same for both kits, the quality control tests for The AccQ•Tag Ultra Derivatization Kit are executed with UV detection. In contrast, the quality control tests for The AccQ•Tag Derivatization Kit are executed with fluorescence detection. Substituting one kit for the other may result in unexpected interferences that may compromise sensitivity or introduce ambiguity into peak identification.

II. ACCQ•TAG ULTRA OVERVIEW

The UPLC Amino Acid Analysis Application Solution is pre-column derivatization and analysis technique. To perform this analysis:

- 1. Use AccQ•Tag Ultra Reagent Powder (US Patent 5,296,599 and European Patent EP 0 533 200 B1) to derivatize the amino acids.
- Separate the derivatives using the properly configured ACQUITY UPLC System.
- The reversed phase separation occurs on the Waters AccQ•Tag Ultra, 2.1 x 100 mm, 1.7 μm column. The derivatives are eluted with the specified dilution of Waters AccQ•Tag Ultra Eluent A and Waters AccQ•Tag Ultra Eluent B.
- 4. Quantitate the derivatives with UV detection.
- 5. Process the data using Empower® software.

For complete details of the instrument configuration and of the separation and analysis methods, consult the UPLC Amino Acid Analysis Application Solution System Guide.

III. CONTENTS OF KIT

The AccQ•Tag Ultra Derivatization Kit contains five sets of the derivatizing reagents used for the UPLC Amino Acid Analysis Application Solution. Each set of reagents includes one vial each of:

- AccQ•Tag Ultra Borate Buffer Vial 1. The buffer is added to the samples to ensure the optimum pH for derivatization.
- AccQ•Tag Ultra Reagent Powder Vial 2A. The reagent powder is the 6-aminoquinolyl-N-hydroxysuccinimidyl carbamate (AQC) derivatizing reagent. It is shipped dry for maximum stability.
- AccQ•Tag Ultra Reagent Diluent Vial 2B. This diluent, acetonitrile, is used reconstitute the reagent for derivatization.

IV. SAMPLE PREPARATION

- I. Reconstitute AccQ•Tag Ultra Reagent Powder.
 - a. Preheat a heating block to 55 °C.
 - b. Tap vial 2A (AccQ•Tag Ultra Reagent powder) to ensure all reagent is at bottom of vial.
 - c. Draw 1 mL of AccQ•Tag Ultra Reagent Diluent from vial 2B.
 - d. Transfer to vial 2A.
 - e. Vortex for 10 seconds.
 - f. Heat on top of heating block, vortex occasionally, until the powder dissolves. Do not heat longer than 15 minutes.
- II. Prepare 100 pmol/µL calibration standard for protein and peptide hydrolysates.
 - a. Transfer the Amino Acid Hydrolysate Standard (P/N WAT088122) from the supplied ampule to a vial appropriate for storage in a freezer.
 - b. Dilute Amino Acid Hydrolysate Standard to appropriate concentration by taking 40 μL of standard and adding it to 960 μL of water.

- III. Derivatize standards and samples.
 - a. Use a micropipettor to deliver 70 μL of AccQ•Tag Ultra Borate Buffer (Vial 1) to a Total Recovery Vial (P/N 186000384C).
 - b. Use a micropipettor to add 10 μL of the calibration standard or dissolved sample amino acids to the vial. Vortex for several seconds.
 - c. Use a micropipettor to add 20 μL of reconstituted AccQ+Tag Reagent to the vial.
 - d. Cap vial and vortex for several seconds.
 - e. Let stand for one minute at room temperature.
 - f. Heat in heating block for 10 minutes at 55 °C.
 - g. Remove from heating block with forceps and place in instrument for analysis. The resulting standard has a concentration of 10 pmol/µL.

Representative Chromatogram: Protein and Peptide Hydrolysates (10 pmol)



V. STORAGE AND STABILITY

- The AccQ•Tag Ultra Derivitization Kit may be stored at room temperature. Please refer to the lot specific expiration date. Under no circumstances should the kits be used over the expiration date stated on the label.
- Reconstituted AccQ•Tag Ultra Reagent can be stored at room temperature in a dessicator with dry dessicant for up to one week.

Note: AccQ•Fluor[™] Ultra Reagent reacts with atmospheric water. Seal the vial tightly. Do not refrigerate because of the risk of condensation.

Derivatized amino acid samples may be stored at room temperature for up to one week.

VI. ORDERING INFORMATION

Description	Part Number
AccQ•Tag Ultra Derivatization Kit, 250 analyses	186003836
AccQ•Tag Ultra Column, 2.1 x 100 mm, 1.7 μm	186003837
AccQ•Tag Ultra Eluent A Concentrate, 1 L	186003838
AccQ•Tag Ultra Eluent B, 1 L	186003839
Amino Acid Hydrolysate Standard, 10 x 1 mL	WAT088122
Sample Tubes, 6 x 50 mm, 72/pkg	WAT007571
Total Recovery Vials, 100/pkg	186000384C



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