ChromaDex®

Application Note

As published in "The Handbook of Analytical Methods for Dietary Supplements"

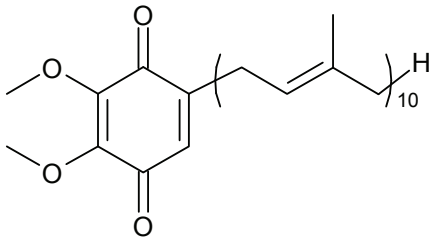
0038 - Coenzyme Q10 by HPLC

Chemical Names: 2-(3,7,11,15,19,23,27,31,35,39-decamethyl-2,6,10,14,18,22,26,30,34,38-tetracontadecaenyl)-5,6-dimethoxy-3-methylbenzo-1,4-quinone

Common Names: Coenzyme Q-199, CoQ10, ubidecarenone, ubiquinone 50

Molecular Weight: 863.34

Chemical Formula: $C_{59}H_{90}O_4$



Solubility: Soluble in ethanol, isopropanol, chloroform, hexane, and other organic solvents.

Other Physical/Chemical Data: UV max = 272 nm (ε= 14,400), 325 nm (ε = 1340);

Uses: Coenzyme Q10 (CoQ10) is a naturally occurring antioxidant. As a dietary supplement, it is used to prevent or to treat congestive heart failure, to delay the onset of Parkinson's syndrome, and to prevent or to treat certain forms of cancer.

Modes of Action:

CoQ10 is a powerful antioxidant that acts as an electron shuttle between flavoproteins and cytochromes in the electron-transport chain. It is the only electron shuttle that is not covalently bonded or tightly bound to a protein. CoQ10 is critical in the oxidation–reduction of FeS in iron–sulfur proteins.

Methods of Analysis

A few methods have been published for the analysis of CoQ10 in dietary supplements; many more have been published for the analysis of CoQ10 in biological samples. Both reversed-phase and normal-phase HPLC with UV detection can be used to quantify CoQ10 in dietary supplements.

CoQ10 is both oxygen- and light-sensitive; therefore, low-actinic glassware and degassed solvents should be used throughout the analysis.

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Method:

Andersson[1] used nonaqueous reversed-phase HPLC to separate CoQ10 from degradation products in raw materials, soy bean oil formulations, and micelle-based preparations. It was found that the nonaqueous reversed-phase system provided better retention time stability and selectivity than normal-phase systems. The method was able to separate CoQ10 (oxidized) from CoQ10 (reduced), coenzyme Q9 (oxidized), coenzyme Q9 (reduced), and ubichromenol.

Sample Preparation:

Prepare soy bean oil capsules containing CoQ10 by transferring the fill material into a 100-mL volumetric flask with 20 mL of hexane, sonicating, and diluting to volume with methanol. Dilute a 2-mL aliquot to 50 mL with methanol and inject into the HPLC system.

For micelle-based preparations, dissolve a 100-mg sample in 100 mL of 20% n-hexane in methanol and sonicate for 5 minutes. Inject this solution.

Chromatography:

Column: Spherisorb ODS-2, 3 µm, 100 × 4.6 mm
Mobile phase: 10% n-hexane in methanol
Flow rate: 1.4 mL/minute
Column temperature: NA
Injection volume: 20 µL
Detection wavelength: 275 nm

Validation Data:

Linearity: 0.2 to 100 mcg/mL
Accuracy: 100.3±1.9% (n = 6) recovery of CoQ10 from spiked placebo capsules, 99.7±2.5% (n = 8) recovery of CoQ10 from spiked micelle vehicle.
Precision: 2.0% RSD (n = 11) for soy bean capsules, 2.6% RSD (n = 6) for micelle-based preparations.
Selectivity: Light stressing followed by HPLC with diode-array detection was used to show separation of CoQ10 from degradation products and related compounds.
Ruggedness: Not determined
Robustness: Not determined
LOD/LOQ: LOD = 2 ng

References:

1. Andersson S. Determination of coenzyme Q by non-aqueous reversed phase liquid chromatography. J Chromatogr A. 1992;606:272–6.

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