

INTRODUCTION

The Coenzyme Q, also called Ubiquinone (or Vitamin Q) is an organic molecule and, more precisely, a benzoquinone with a very long isoprenic side chain.

It is ubiquitous in the biological systems and its chemical structure is very similar to that of Vitamin K and Vitamin E.

In living organisms, it is involved in the redox reactions and, depending on the state of oxidation, it can be present in three forms: oxidized, semiquinonic and reduced form.

If coenzyme Q is reduced by acquiring an electron, it forms a radical called **Ubisemiquinone (QH[•])**. By further reduction, the compound becomes a **Ubiquinol (QH₂)**.

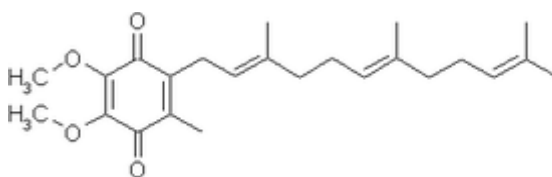


Fig. 1 – Oxidized Form

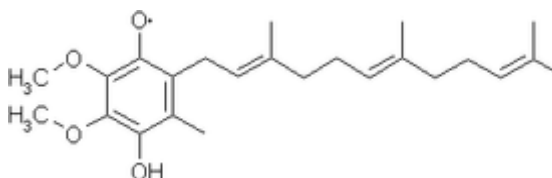


Fig. 2 – Semichinonic Form

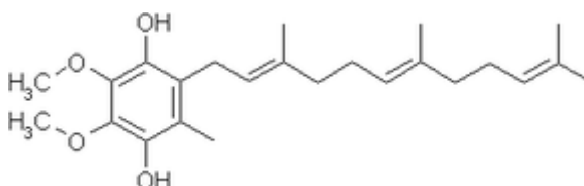


Fig. 3 – Reduced Form

Moreover, the side chains make it very lipophilic, therefore, it is located in the hydrophobic domain of the phospholipid bilayer of all biological membranes. Ubiquinone is found in the membranes of the endoplasmic reticulum, of peroxisomes and lysosomes and inside the mitochondria membrane where it plays an important role in the electron transport chain. Therefore, it influences the ability of cells to produce and use energy.

Various types of Coenzyme Q can be distinguished according to the number of isoprene units in the lateral chain. In human mitochondria the most common type of CoQ is Q10. Higher levels of Q10 could improve the efficiency of the electron transport chain, increasing available energy.

Due to its ability to transfer electrons and act as an antioxidant, Coenzyme Q has been evaluated as a dietary supplement. In particular, CoQ10 has been widely used for the treatment of heart dysfunction, such as heart attack, and breast cancer.

The concentration of coenzyme Q10 tends to decrease with aging; low levels are also recorded in presence of particular chronic diseases, such as those resulting from heart problems, Parkinson's disease, muscular dystrophy, diabetes, cancer and AIDS.

Also, some medications, such as statins used in control hypercholesterolemia, can lower the levels of Coenzyme Q10.

Coenzyme Q10 has also been shown to have beneficial effects on subjects affected by migraine.

Moreover, Q10 is also a major component of the treatment for mitochondrial myopathy and others metabolic disorders.

Recent studies have highlighted how the antioxidant effects of the Q10 enzyme may have benefits on body and brain. Some of these studies indicate how coenzyme Q10 may protect the brain from neurodegenerative diseases, such as Parkinson's disease, as well as the damage caused by ischemia (stroke) to the brain.

Other recent studies show a benefit regarding the chance of survival, after cardiac arrest, if coenzyme Q10 is administered in conjunction with a cool-down body temperature (between 32–34 °C).



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This product fulfills all the requirements of Directive 98/79/EC on in vitro diagnostic medical devices (IVD). The declaration of conformity is available upon request.

N° 008	Coenzyme Q ₁₀ in plasma by UV - FAST	09/2025
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TECHNICAL FEATURES

Principle of the Method:

The **Coenzyme Q₁₀** after be treated with a deproteinization solution, comes stabilized and injected in HPLC.

Recovery : 98%

Sensitivity (LLOD) : 20 µg/l

Minimum concentration analyzable (LLOQ) : 50 µg/l

Linearity : 50 – 40.000 µg/l

Accuracy intra serie (relative error %) :

C1	C2
500 µg/l	1000 µg/l
3,14%	3,26%

Accuracy inter serie (relative error %) :

C1	C2
500 µg/l	1000 µg/l
6,33%	4,62%

Reproducibility intra serie (coefficient of variation %):

C LLOQ	Cm	Cs
50 µg/l	750 µg/l	1500 µg/l
2,61%	0,14%	5,56%

Reproducibility inter serie (coefficient of variation %):

C LLOQ	Cm	Cs
50 µg/l	750 µg/l	1500 µg/l
11,89%	8,15%	6,51%

Coefficient of Correlation R² + Dev Std : 0,9933 ± 0,0009

Normal Range in plasma: 830 – 1430 µg/l

Components of the kit :

Reagent A – Deproteinization Solution, **1 x 130 ml**

Reagent B – Diluting Solution, **1 x 5 ml**

Reagent C – Stabilizing Solution, **1 x 100 ml**

Reagent D – Internal Standard Solution, **1 x 1,1 ml**

Reagent E – Test Solution, **1 x 2 ml**

Plasma Calibrator, **2 x 1 ml**

Reagent M – Mobile Phase, **2 x 500 ml**

All the reagents are ready to use and stable 3 years at 2 – 8 °C.

Supplied separately as Box 3/3, to be stored at – 25 °C / –15 °C

Code Z63016

Packed separately as Box 2/3 – see technical data sheet

Minimum Instrumental equipment required:

Isocratic HPLC System with loop of 20 μ l
Spectrophotometric Detector $\lambda = 275 \text{ nm}$
Chromatograms Recorder

Optional Equipment:

Autosampler
Operational Computer

Plasma Collection Procedure:

Collect 3 ml of blood with **EDTA**. Plasma samples should be stored at - 80 °C. Stable for 3 months. Stable 4 days at 2-8 °C (**at the dark**).

PREANALYTICAL PROCEDURE

Test Solution Preparation:

- 460 µl of **Reagent B – Diluting Solution**
- 20 µl of **Reagent E – Test Solution**
- 20 µl of **Reagent D – Internal Standard Solution**

INJECTION :

- Inject 20 µl of the solution into the chromatographic system.

Verify that the Test Solution has retention time similar to fig. 4. If the Test is all right you can start with the analytical procedure; if not, check the functionality of the analytical system.

Important : Don't use this solution to calibrate!

ANALYTICAL PROCEDURE

STEP 1 : Samples Preparation

Dispense in a tube :

	Calibrator	Control	Sample
Calibrator	400 µl		
Control		400 µl	
Sample			400 µl
Reagent A – Deproteinization Solution	1000 µl	1000 µl	1000 µl
Reagent D – Internal Std Solution	10 µl	10 µl	10 µl

Vortex for 30 sec.

STEP 2 : Centrifuge at 14.000 rpm for 10 minutes

STEP 3 : Take and throw the supernatant

STEP 4 : Add 700 µl of **Reagent C – Stabilizing Solution** and therefore to restore the hurled in solution by means of stick means unrest

Vortex for 60 sec.

STEP 5 : Centrifuge at 14.000 rpm for 10 minutes

STEP 6 : Take 600 µl of clear supernatant

N. B. : the sample so gotten ready is not stable to long, inject it immediately

INJECTION :

- Inject 20 µl of the solution into the chromatographic system.

N.B. : For samples that they present low sensibility it is possible to pull dry, with the aid of a light flow of helium or of nitrogen, the coming supernatant from the STEP 6 and to take again it with 200 of Reagent C – Stabilizing Solution and therefore to proceed the phase of injection.

COENZYME Q₁₀ in PLASMA by UV – Warnings

REAGENT E : TEST SOLUTION

COENZYME Q ₁₀	About 100.000 µg/l
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PARAMETERS FOR SPECTOFOTOMETRIC DETECTOR

λ	275 nm
GAIN	0.001 AUFS

HPLC COLUMN PROTECTION

To save the analytical column Reverse Phase POROSHELL 120 EC-C18 (50 x 4,6 mm, 2,7 µm), the use of Javelin Colfilters (1 x 10 pcs.) cod. S90199511 is obligatory.

HPLC COLUMN CONDITIONING

Install a new analytical C₁₈ POROSHELL 120 EC-C18 (50 x 4,6 mm, 2,7 µm). Disconnect the detector and flux 30 ml of H₂O : Acetonitrile (20 : 80 v/v) et pH>3 and subsequently 30 ml of ml of **H₂O : Acetonitrile (80 : 20 v / v)**, set flow at 1,5 ml / min. **Don't recycle the washing solutions.** Condition the column with the mobile phase at a flow of 1,2 ml / min. and discharge the first 15 ml. Condition further on the column for 15 min. also at recycling phase. Finally inject the Chemical Standard and verify the quality of the HPLC run.
It is NOT possible to make analysis at recycling phase. If Room Tem. is > 20° C is better to preserve the Mobile Phase at 2 – 8° C among runs.

COLUMN CLEANING

Flux a solution made of H₂O : Acetonitrile (20 : 80 v/v) for 30 min and discharge. When you re-use the column, flux 15 ml of H₂O : Acetonitrile (20 : 80 v/v) before a new conditioning with the Mobile Phase.
We recommend washing the Analytic Column at the end of each analytical session. Store the column in H₂O : Acetonitrile (20 : 80 v / v).

PARAMETRI HPLC

LOOP	20 µl
Recommended Flow rate	1,2 ml/min
Pressure	About 100 bar

ACCESSORIES AND CONSUMABLES

CODE	DESCRIPTION	PACKAGING
Z63016	Calibrator in plasma for Coenzyme Q ₁₀	4 x 1 ml
Z63019	Control in plasma for Coenzyme Q ₁₀ - Levels 1 and 2	2 x 5 x 1 ml
Z699975902	Poroshell 120, EC-C18 (50 x 4,6 mm - 2,7 um) Analytical Column	1 Pc
S90199511	Javelin Colfilters	1 x 10 Pcs
S51843550	Clear glass vials with reduced volume from 1,5 ml to 15 ul	1 x 100 Pcs
S51820717	Caps for Clear glass vials with reduced volume from 1,5 ml to 15 ul	1 x 100 Pcs

COENZYME Q₁₀ IN PLASMA BY UV

(Reference Chromatograms)

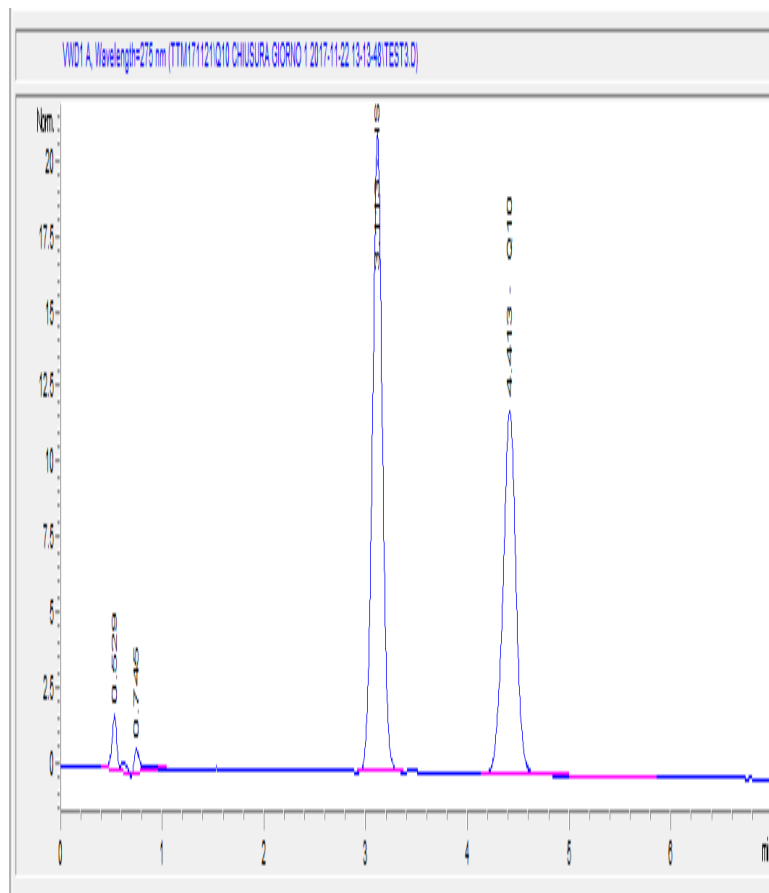


Fig. 4: Test Solution

R.T. 3.11	Internal Standard
R.T. 4.41	Coenzyme Q ₁₀

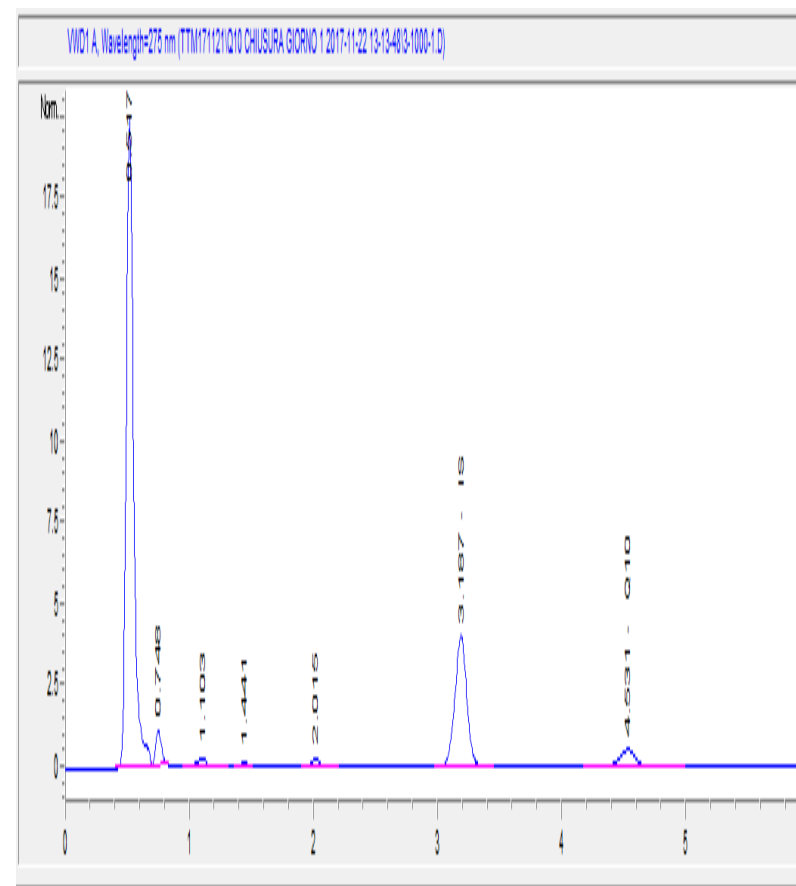


Fig. 5: Plasma Calibrator

R.T. 3.18	Internal Standard
R.T. 4.53	Coenzyme Q ₁₀ 1.000 µg/l

COENZYME Q₁₀ IN PLASMA BY UV (Reference Chromatograms)

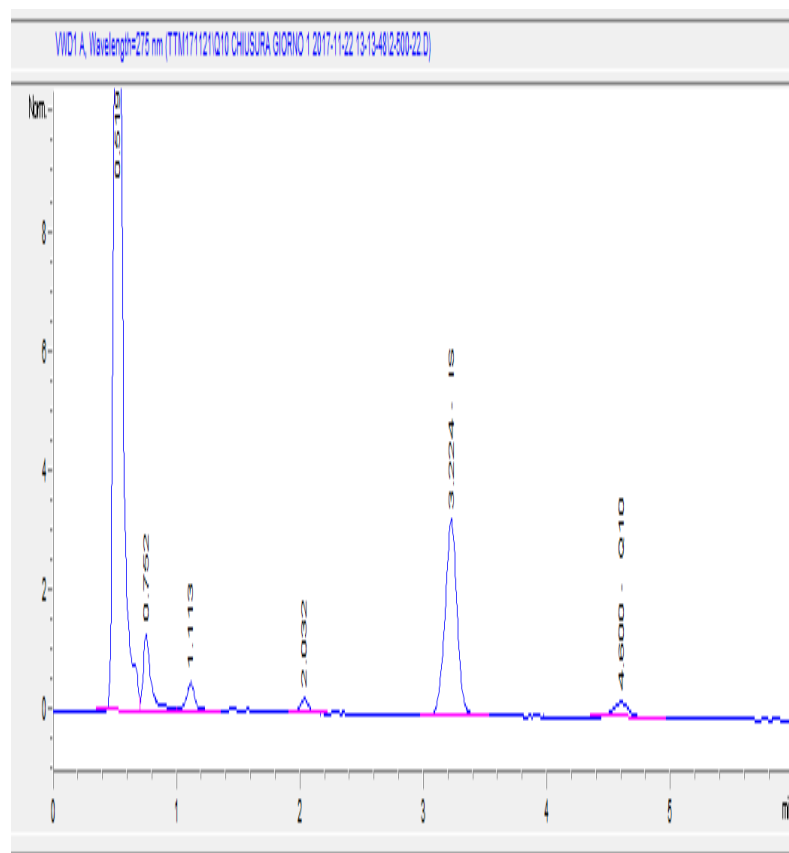


Fig. 6:	Low Control
	R.T. 3.19 Onternal Standard
	R.T. 4.55 Coenzyme Q ₁₀ 500 µg/l

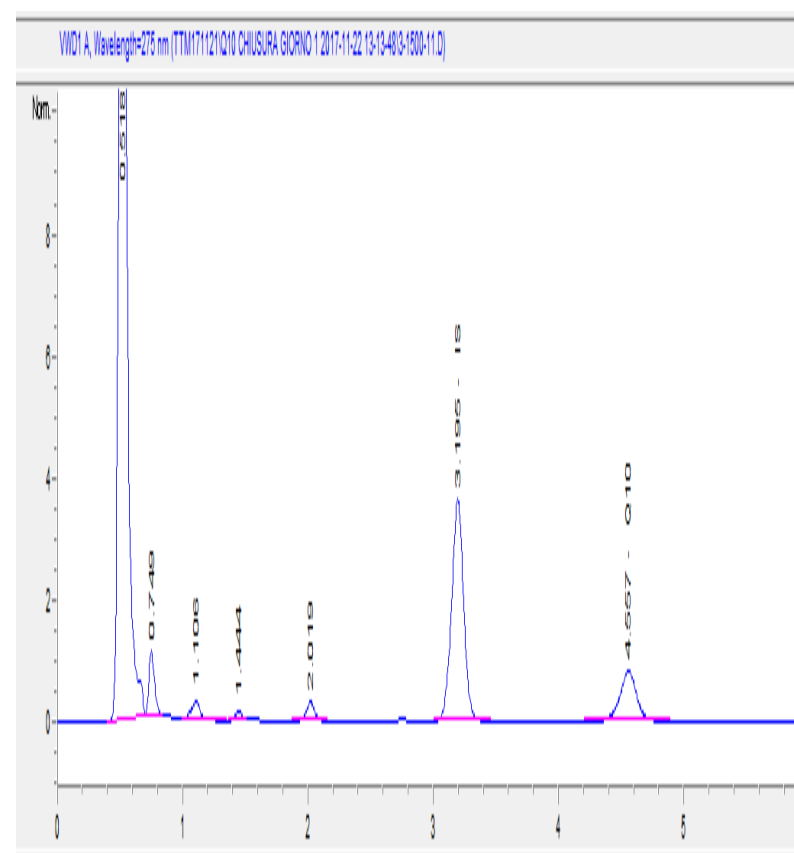


Fig. 7:	High Control
	R.T. 3.22 Internal Standard
	R.T. 4.60 Coenzyme Q ₁₀ 1.500 µg/l