# Coenzyme Q10 Assay Kit

ARG83425 Coenzyme Q10 Assay Kit can be used to measure Coenzyme Q10 in serum, plasma, tissue extracts, cell lysate, cell culture media and other biological fluids.

Catalog number: ARG83425

Package: 96 wells

# TABLE OF CONTENTS



**MANUFACTURED BY:**

Arigo Biolaboratories Corporation

Address: No. 22, Ln. 227, Gongyuan Rd., Hsinchu City 300, Taiwan

Phone: +886 (3) 562 1738

Fax: +886 (3) 561 3008

Email: info@arigobio.com

## INTRODUCTION

Coenzyme Q10 also known as ubiquinone, is a naturally occurring biochemical cofactor (coenzyme) and an antioxidant produced by the human body, but it can also be obtained from dietary sources such as meat, fish, and some vegetables. CoQ10 is also found in many organisms including animals and bacteria. CoQ10 plays a role in mitochondrial oxidative phosphorylation, aiding in the production of adenosine triphosphate (ATP), which is involved in energy transfer within cells. The structure of CoQ10 consists of a benzoquinone moiety and an isoprenoid side chain, with the "10" referring to the number of isoprenyl chemical subunits in its tail.

## PRINCIPLE OF THE ASSAY

The Coenzyme Q10 Assay Kit can measure Coenzyme Q10 in serum, plasma, tissue extracts, cell lysate, cell culture media and other biological fluids. The increase in absorbance at 620 nm is directly proportional to reactants of the reaction between substrate and Flavonoid.

## MATERIALS PROVIDED & STORAGE INFORMATION

| Component | Quantity | Storage |
|---|---|---|
| Microplate | 1 X 96-well plate | |
| Standard | 1 vial (lyophilized) | -20°C |
| Assay Buffer | 4 X 30 ml (ready to use) | 4°C |
| Reaction Buffer | 15 ml (ready to use) | 4°C |
| Reagent Dye | 3 ml (ready to use) | 4°C |

## MATERIALS REQUIRED BUT NOT PROVIDED

- Microplate reader capable of measuring absorbance at 620 nm
- Pipettes and pipette tips
- Deionized or distilled water

## TECHNICAL HINTS AND PRECAUTIONS

- Wear protective gloves, clothing, eye, and face protection especially while handling blood or body fluid samples.
- Store Standard at -20°C, all other components store at 4°C.
- Briefly spin down the reagents before use.
- It is highly recommended that the standards and samples be assayed in at least duplicates.
- Change pipette tips between the addition of different reagent or samples.

## SAMPLE COLLECTION & STORAGE INFORMATION

The sample collection and storage conditions listed below are intended as general guidelines. Sample stability has not been evaluated.

Tissue samples - Weigh out 0.1 g tissue, homogenize with 1 ml Assay Buffer on ice, centrifuged at 10000g for 10 minutes, take the supernatant into a new centrifuge tube and keep it on ice for detection

Cell and bacteria samples-Collect cell or bacteria into centrifuge tube, discard the supernatant after centrifugation, add 1 ml Assay Buffer for 5×106 cell or bacteria, sonicate (with power 20%, sonicate 3s, interval 10s, repeat 30 times); centrifuged at 10000g for 10 minutes, take the supernatant into a new

centrifuge tube and keep it on ice for detection.

Liquid samples-Add 0.1 ml sample into 0.9 ml Assay Buffer, mix, centrifuged at 10000g for 10 minutes, take the supernatant into a new centrifuge tube and keep it on ice for detection.

## REAGENT PREPARATION

- **Standard:** Add **1 ml** of **Assay Buffer** heat at 40 °C to dissolve standard. The concentration will be 10 µmol /ml. Perform 2-fold serial dilution of the top standards to make the standard curve.

## ASSAY PROCEDURE

Standards and samples should be assayed in at least duplicates.

1. Sample wells: Add **20 µl Sample** into Sample wells.
2. Standard wells: Add **20 µl Standard** into Standard wells.
3. Add **150 µl Reaction Buffer** to each wells.
4. Add **30 µl Reagent Dye** to each wells.
5. Mix well. Incubate at **RT** for **30 min**
6. Read the OD at **620nm**

Summary of Coenzyme Q10 Assay Procedure

| Reagent | Sample | Standard | Blank |
|---|---|---|---|
| Sample | 20 µl | - | - |
| Standard | - | 20 µl | - |
| Distilled water | - | | 20 µl |
| Reagent Buffer | 150 µl | 150 µl | 150 µl |
| Reagent Dye | 30 µl | 30 µl | 30 µl |
| Mix well. Incubate at RT for 30 min<br>Read the OD at 620nm | | | |

## CALCULATION OF RESULTS

1. Calculate the average absorbance values for each set of samples, standard and blank.

2. Calculation:

A. Definition:

$C_{Standard}$: the concentration of Standard, 5 µmol/ml;

$V_{Sample}$: the volume of reaction sample, 20 µl = 0.02 ml;

$V_{Standard}$: the volume of reaction Standard, 20 µl = 0.02 ml;

$V_{assay}$: the volume of Assay Buffer, 1000 µl = 1 ml;

W: the weight of sample, g;

N: the quantity of cell or bacteria, $N \times 10^4$;

V: the volume of liquid sample, 100 µl = 0.1 ml;

B. Formula:

a). According to the weight of sample

Coenzyme Q10 (μmol/g) =

$[(C_{Standard} \times V_{standard}) \times (OD_{Sample} - OD_{Blank})] / [(OD_{Standard} - OD_{Blank}) \times (W \times V_{Sample} / V_{assay})]$

$=5 \times (OD_{Sample} - OD_{blank}) / [(OD_{Standard} - OD_{Blank}) \times W]$

b). According to the cell or bacteria

Coenzyme Q10 ($\mu mol/10^4$) =

$[(C_{Standard} \times V_{standard}) \times (OD_{Sample} - OD_{Blank})] / [(OD_{Standard} - OD_{Blank}) \times (N \times V_{Sample} / V_{assay})]$

$=5 \times (OD_{Sample} - OD_{blank}) / [(OD_{Standard} - OD_{Blank}) \times N]$

c). According to the volume of sample

Coenzyme Q10 (μmol/ml) =

$[(C_{Standard} \times V_{standard}) \times (OD_{Sample} - OD_{Blank})] / [(OD_{Standard} - OD_{Blank}) \times (V_{Sample} \times V / V_{Assay})]$

$=50 \times (OD_{Sample} - OD_{blank}) / (OD_{Standard} - OD_{Blank})$

3. Detection range:

The detection range is from 0.01 μmol/ml - 10 μmol/ml.

4. If the samples have been diluted, the calculated activity must be further converted by the appropriate dilution factor according to the sample preparation procedure as described above.

## EXAMPLE OF TYPICAL RESULT

The following data is for demonstration only and cannot be used in place of data generations at the time of assay. Please note this data is for demonstration only and serial diluted standards are not necessary for this kit.