

QuantiChrom™ Cobalt Assay Kit (DCBT-100)

Quantitative Colorimetric Cobalt Determination at 485nm

DESCRIPTION

Cobalt is an essential micronutrient for all multicellular organisms as the active center of cobalamins, such as vitamin B-12, which is essential for plants and animals. Cobalt is also a micronutrient for bacteria, algae, and fungi. Despite being an important micronutrient, excess cobalt in the environment can be deleterious to life, leading to cardiomyopathy in humans and necrosis in plants.

Simple, direct and automation-ready procedures for measuring cobalt concentrations find wide applications in research and environmental monitoring. BioAssay Systems' cobalt assay kit is designed to measure total cobalt directly in aqueous samples without any pretreatment. The intensity of the color, measured at 485nm, is directly proportional to the cobalt concentration in the sample.

KEY FEATURES

Sensitive and accurate. Linear detection range 1.4 μM to 200 μM cobalt in 96-well plate assay.

Simple and high-throughput. The procedure involves addition of a single working reagent with immediate color development. Can be readily automated as a high-throughput assay for thousands of samples per day.

Improved reagent stability and versatility. The optimized formulation has greatly enhanced reagent stability. Cuvette or 96-well plate assay.

APPLICATIONS

Direct Assays: cobalt in aqueous samples.

KIT CONTENTS (100 tests in 96-well plates)

Detection Reagent: 1.0 mL

Cobalt Standard: 0.8 mL 200 μM Co²⁺

Storage conditions. The kit is shipped at room temperature. Store Detection Reagent at -20°C. Shelf life: 6 months after receipt.

Precautions: reagents are for research use only. Normal precautions for laboratory reagents should be exercised while using the reagents. Please refer to the Material Safety Data Sheet for detailed information.

PROCEDURES

Note: (1) Metal chelators (e.g. EDTA) interfere with this assay and should be avoided in sample preparation. (2) Samples should be clear and free of precipitates or turbidity. If not, centrifuge or filter to clarify samples prior to assay. (3) The metal ions Fe²⁺, Cu²⁺, Zn²⁺, Mn²⁺, Mg²⁺, Al³⁺, Mo²⁺, Pb²⁺ and W²⁺ exhibit less than 10% interference in this assay. Ni²⁺ concentrations >10% of Co²⁺ concentration should be avoided. Excess Ca²⁺ may cause precipitation to occur.

Procedure using 96-well plate:

1. Standards. Directly transfer 200 μM Cobalt Standards into a clear flat bottom 96-well plate as follows.

No	Standard + H ₂ O	Vol (μL)	Co ²⁺ (μM)
1	90 μL + 0 μL	90	200
2	54 μL + 36 μL	90	120
3	27 μL + 63 μL	90	60
4	0 μL + 90 μL	90	0

Transfer 90 μL sample into a clear flat bottom 96-well plate.

2. Add 10 μL of Detection Reagent to each well. Tap plate to mix.
3. Read optical density at 485nm within 10 min at room temperature.

Procedure using cuvette:

1. Prepare standards as in 96-well assay with appropriate volumes for the cuvettes. Set up centrifuge tubes labeled Standards and Samples. Transfer standards and samples to tubes.
2. Add **Detection Reagent** to all tubes at a ratio of 1:10. Mix by vortexing.
3. Transfer to cuvettes and read OD at 485nm within 10 minutes.

CALCULATION

Subtract OD of "0 μg/dL Co" from all other standard OD values and plot the OD against standard Cobalt concentrations. Determine the slope using linear regression fitting. Cobalt concentration of the sample is calculated as

$$[\text{Co}^{2+}] = \frac{\text{OD}_{\text{SAMPLE}} - \text{OD}_{\text{BLANK}}}{\text{Slope}} \quad (\mu\text{M})$$

Where OD_{BLANK} is OD values of the water blank (Standard #6), or Sample Blank, if a sample blank is used.

MATERIALS REQUIRED, BUT NOT PROVIDED

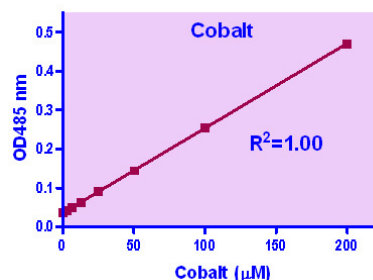
Pipetting devices and accessories.

Procedure using 96-well plate:

Clear bottom 96-well plates (e.g. Corning Costar) and plate reader.

Procedure using cuvette:

Cuvettes and spectrophotometer for measuring OD at 485nm.



Standard Curve in 96-well plate assay

REFERENCES

1. Palit, S.; Sharma, A.; Talukder, G. (1994). "Effects of cobalt on plants". *Bot. Rev.* **60** (2): 149–181.
2. Alexander, Carl S. (1972). "Cobalt-beer cardiomyopathy". *The American Journal of Medicine.* **53** (4): 395–417.

