

Iodide Colorimetric Assay Kit

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(Catalog # K2037-100; 100 assays, Store kit at -20°C)

I. Introduction:

Iodide (I^-) is a halide anion and a necessary and limiting substrate for thyroid hormone synthesis. This essential element enables the thyroid gland to produce thyroid hormones, thyroxine (T4) and triiodothyronine (T3). Additionally, Iodide is important in many essential biological processes such as basal metabolism, temperature regulation, intellectual development for children, muscular development, normal heart function, growth of skeleton etc. Food is the main source of daily supply of iodide in humans. Iodide can be found in grains, fish, dairy products, fruits, iodized salts etc. Iodine deficiency affects around two billion people worldwide. Iodide deficiency can cause diseases such as goiter, depression, fatigue, hypothyroidism, intellectual disability, delayed sexual development etc. However, excess intake of Iodide is also harmful and can inhibit the release of thyroid hormones and cause inflammation of the thyroid gland and even thyroid cancer. **BioVision's Iodide Colorimetric Assay Kit** provides a quick, convenient, non-radioactive method to determine Iodide concentration in various sample types including foods, liquid and biological samples. The assay relies on the Iodide ion as a specific catalyst to convert a yellow colored substrate to a colorless product. The reduction in absorbance signal at 330 nm is directly proportional to the Iodide concentration in the samples. Most common ions (eg. Na^+ , Ca^{2+} , Cu^{2+} , K^+ , Mg^{2+} , Fe^{3+} , NH_4^+ , CO_3^{2-} , Br^- , Cl^- , SO_4^{2-} , NO_3^- and PO_4^{3-}) do not interfere with the assay. The kit is simple, rapid, sensitive and can detect as low as 0.2 μM Iodide under the assay conditions.



II. Application:

- Determination of Iodide in different samples such as foods, liquid and biological samples

III. Sample Types:

- Foods (e.g. meats, grains, fruits, dairy products etc.)
- Liquids and biological samples (e.g. urine, saliva and serum)

IV. Kit Contents:

Components	K2037-100	Cap Code	Part Number
Iodide Standard	100 μl	Blue	K2037-100-1
Iodide Substrate	1 vial	Yellow	K2037-100-2
Treatment Reagent	1.1 ml	Red	K2037-100-3
Sample Diluent	90 ml	NM	K2037-100-4
Precipitation Solution	5 ml	NM	K2037-100-5

V. User Supplied Reagents and Equipment:

- 96-well clear flat-bottom plate
- Multi-well spectrophotometer
- Dounce Tissue Homogenizer (BioVision Cat. # 1998)
- ddH₂O

VI. Storage Conditions and Reagent Preparations:

Store the kit at -20°C. The kit components are stable for one year when stored as recommended. Read the entire protocol before performing the experiment.

- Iodide Standard (100 μM):** Warm to room temperature (RT) before use. Stable at 4°C for 2 months.
- Iodide Substrate:** Reconstitute the vial with 1 ml of Sample Diluent to prepare the stock Iodide Substrate solution. Divide into aliquots and store at 4°C. Stable for 2 months at 4°C.
- Treatment Reagent, Sample Diluent & Precipitation Solution:** Ready to use. Warm the bottles to RT before use. Stable for 2 months at 4°C.

VII. Iodide Assay Protocol:

1. Sample Preparation: For Food Samples: Weigh out ~100 mg of the Sample (e.g. meats, grains, fruits and dairy products etc.) and cut it into small pieces (if possible). Transfer the Sample into an eppendorf tube. Add 0.5 ml of Sample Diluent to the tube and homogenize the Sample for 10 min using dounce tissue homogenizer (BioVision Cat. # 1998) and incubate at RT for 10 min. Centrifuge the Sample(s) at 12,000 g for 15 min and collect the clear supernatant. Use 10 μl of the clear supernatant for assay.

For Liquid and Urine Samples: Dilute the Sample 2 - 5 fold using Sample Diluent. Use 10 μl of the diluted Sample for assay.

For Serum Sample: Add 30 μl of Precipitation Solution to 270 μl of Serum in an eppendorf tube and vortex briefly. Centrifuge the Sample at 12,000 g for 15 min and collect the supernatant. Dilute the supernatant 5 fold using Sample Diluent (e.g. 20 μl supernatant in 80 μl of Sample Diluent). Use 10 μl of the diluted supernatant for assay.

2. Standard Curve Preparation: Mix 10 μl of 100 μM Iodide Standard with 990 μl of water to prepare 1 μM Iodide Standard solution. Add 0, 2, 4, 6, 8 and 10 μl of 1 μM diluted Iodide Standard into the desired wells in a 96-well clear flat-bottom plate to generate 0, 0.2, 0.4, 0.6, 0.8 and 1.0 μM of Iodide Standard/well respectively. Adjust the volume of each well to 10 μl using water.

3. Reaction Mix Preparation: Dilute the stock Iodide Substrate 10 fold by adding 200 μl of stock Iodide Substrate solution to 1.8 ml of Sample Diluent (for 20 assays) before performing the assay. Prepare Reaction Mix according to the table below. Make sufficient amount of Reaction Mix to add 90 μl to all assay wells.

Reaction Mix

Diluted Iodide Substrate solution	80 μ l
Treatment Reagent	10 μ l

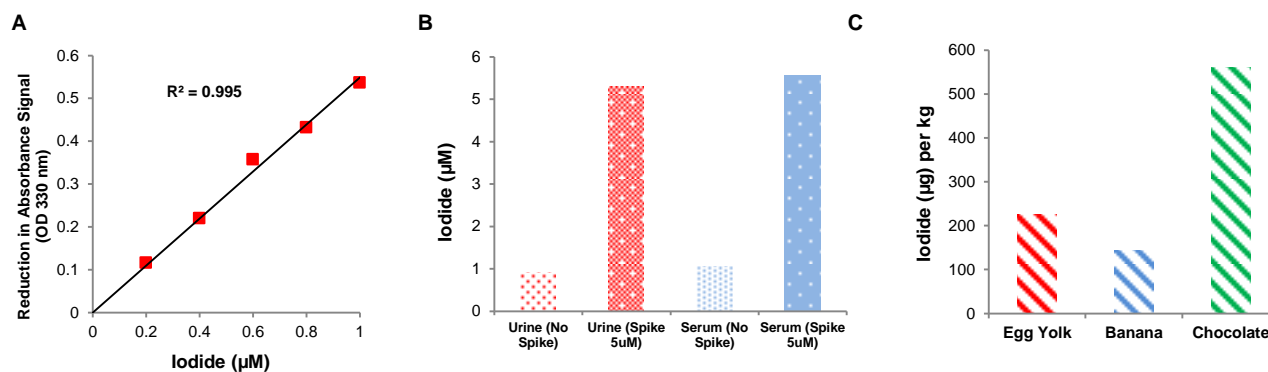
Mix well. Add 90 μ l of Reaction Mix to all the wells containing Standards and Samples.

- Measurement:** Immediately, measure the absorbance (OD) of all wells at 330 nm in kinetic mode at RT for 5 min.
- Calculation:** Subtract the 0 Standard readings from all Standard and Sample readings. For each Standard concentration, subtract the final absorbance (t_2) from the initial absorbance (t_1) to get the reduction in absorbance signal (t_1-t_2). Plot the Iodide Standard Curve by using the reduction in Absorbance Signal ($t_1 - t_2$) values vs Iodide Standard concentrations.

Calculate the Iodide concentration in the Sample (**A**) by applying the reduction in Sample Absorbance Signal values to the Iodide Standard Curve

$$\text{Iodide concentration } (\mu\text{M}) = A \times D$$

Where: A = Iodide concentration from the Standard Curve (μ M)
 D = Sample dilution factor (D = 1, for undiluted samples)



Figures. A. Iodide Standard Curve. **B.** Iodide concentration in urine and serum samples before and after spiking with 5 μ M of iodide. Data shows > 80% recovery under the assay kit conditions. **C.** Iodide in egg yolk, banana and chocolate samples.

VIII. Related Products:

Calcium Colorimetric Assay Kit (K380)
 Magnesium Colorimetric Assay Kit (K385)
 Sodium Assay Kit (Colorimetric) (K391)
 Nickel Colorimetric Assay Kit (K510)

Calcium Assay Kit (Fluorometric) (K409)
 Iron Colorimetric Assay Kit (K390)
 Cobalt Colorimetric Assay Kit (K505)
 Potassium (Serum) Detection Assay Kit (Fluorometric) (K940)

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