



Human Intact Parathyroid Hormone (PTH) OneStep ELISA Kit

Catalog number: EK7081

For detection of multiple analytes using one single assay.

This package insert must be read in its entirety before using this product.

For research use only. Not for use in diagnostic procedures.

Human Intact Parathyroid Hormone (PTH) OneStep ELISA Kit

Catalog Number: EK7081

Size: 96 wells/kit

Sample Type: Serum and Plasma

Assay Range: 0 - 1000 pg/ml

Storage: Store the kit at 2°C to 8°C. Keep microwells sealed in a dry bag with desiccants. The reagents are stable until expiration of the kit. Do not expose reagent to heat, sun, or strong light. Avoid multiple freeze-thaw cycles (Ships with gel ice, can store for up to 3 days in room temperature. Freeze upon receiving.)

Introduction

The Bosterbio OneStep ELISA kit is a solid phase sandwich ELISA kit. Instead of incubating with standards, samples and controls and with enzyme conjugated detection antibody separately, the OneStep ELISA kit allows the user to add standards, samples and controls to wells without incubation, and an enzyme conjugate reagent is added into each well immediately. Then incubate standards, samples, controls and enzyme conjugate reagent together. After the excess enzyme conjugate is washed out, the substrate is added into each well. The enzyme catalyzes the substrate yielding a blue color ($A_{max} = 370\text{nm}$ and 652nm) that changes to yellow ($A_{max} = 450\text{nm}$) upon addition of a sulfuric or phosphoric acid stop solution. The intensity of color developed is directly proportional to the concentration of target protein in the samples. A standard curve is generated relating color intensity to the concentration of target protein.

Kit Components

Description	Quantity
1. Microwells coated with Streptavidin	12x8x1
2. PTH Standard 6: 1 Vial (lyophilized)	0.75 ml
3. PTH Controls: 2 Vials (lyophilized)	0.75ml
4. Anti-PTH-Biotin Reagent: 1 Bottle (Ready to use)	7 ml
5. Anti-PTH-HRP Enzyme Conjugate: 1 Bottle (Ready to use)	7 ml
6. Assay Diluent: 1 Bottle (ready to use)	5 ml
6. TMB Substrate: 1 Bottle (Ready to use)	12 ml
7. Stop Solution: 1 Bottle (Ready to use)	12 ml
8. 20X Wash Solution: 1 Bottle	25 ml

Standard Concentrations and example data

	Conc. pg/mL	OD 450 nm
Std 1	0	0.029
Std 2	11.1	0.082
Std 3	33.3	0.178
Std 4	100	0.459
Std 5	300	1.078
Std 6	900	2.535

LIMITATIONS OF THE PROCEDURE

1. Do not use sodium azide as preservative. Sodium azide inhibits HRP enzyme activities.
2. The PTH ELISA kit has exhibited no "high dose hook effect" with high dose spiked samples. However, samples with intact PTH levels greater than the highest calibrator should be diluted and re-assayed for correct value.

Materials Required, but Not Provided

1. Distilled or deionized water
2. Precision pipettes
3. Disposable pipette tips. Multichannel pipettes are recommended in the condition of large amount of samples in the detection.
4. ELISA reader capable of reading absorbance at 450nm
5. Absorbance paper or paper towel
6. Graph paper

STORAGE AND STABILITY

1. Store the kit at 2-8°C
2. Keep microwells sealed in a dry bag with desiccants.
3. The reagents are stable until expiration of the kit.
4. Do not expose reagents to heat, sun, or strong light.

WARNINGS AND PRECAUTIONS

1. For Research Use Only. Not for use in diagnostic procedures.
2. For Laboratory Use.
3. Potential biohazardous materials: The calibrator and controls contain human source components, which have been tested and found non-reactive for hepatitis B surface antigen as well as HIV antibody with FDA licensed reagents. However, there is no test method that can offer complete assurance that HIV, Hepatitis B virus or other infectious agents are absent. These reagents should be handled at the Biosafety Level 2, as recommended in the Centers for Disease Control/National Institutes of Health manual, "Biosafety in Microbiological and Biomedical Laboratories" 1984.
4. Do not pipette by mouth. Do not smoke, eat, or drink in the areas in which specimens or kit reagents are handled.
5. The components in this kit are intended for use as an integral unit. The components of different lots should not be mixed.
6. It is recommended that serum samples be run in duplicate.
7. Optimal results will be obtained by strict adherence to this protocol. Accurate and precise pipetting, as well as following the exact time and temperature requirements prescribed are essential. Any deviation from this may yield invalid data.

SPECIMEN COLLECTION HANDLING

1. Collect blood specimens and separate the serum immediately.
2. Specimens should be stored frozen at (-20°C) for up to one month.
3. Avoid multiple freeze-thaw cycles.
4. Prior to assay, frozen sera should be completely thawed and mixed well.
5. Do not use grossly lipemic specimens.

REAGENT PREPARATION

Standards: Reconstitute the lyophilized standards with 0.75 ml distilled or deionized water. Allow it to remain undisturbed until completely dissolved, and then mix well by gentle inversion(not to foam),prepare the rest standard set (5-2), by 3-fold serial dilution , from standard.#6 as prescribed in the table below: (Mix each tube thoroughly before the next transfer.)

Std No.	Standard. Conc. (pg/mL)	Standard. volume
6	900	reconstitute with 0.75ml of DI water
5	300	0.25ml of Std 6 plus 0.5ml of assay diluent
4	100	0.25ml of Std 5 plus 0.5ml of assay diluent
3	33.3	0.25ml of Std 4 plus 0.5ml of assay diluent
2	11.1	0.25ml of Std 3 plus 0.5ml of assay diluent
1	0	assay diluent only

ASSAY PROCEDURE

1. Allow materials and reagents to reach room temperature
2. Place the desired number of strips in the plate holder.
3. Pipet 25 µl of standards, controls, and samples into the designated wells.
4. Add 50 µl of anti-PTH-Biotin Reagent to all wells.
5. Add 50ul of anti-PTH-HRP Conjugate to all wells.
6. Cover the plate and incubate at room temperature for 90 minutes on a plate shaker (500 - 600rpm).
7. Remove liquid from all wells. Wash wells 4 times with 300 µl of 1X wash buffer. Blot on absorbent paper towels.
8. Add 100 µl of TMB Substrate to all wells.
9. Incubate for 15 minutes at room temperature.
10. Add 50 µl of Stop Solution to all wells. Mix gently.
11. Read absorbance on ELISA Reader at 450nm within 15 minutes after adding the stopping solution. Use 630nm as a reference.

CALCULATION OF RESULTS

1. Assign the concentration for each standard stated on the vial in pg/ml. Plot the data from the standard curve on linear graph paper with the concentration on the X-axis and the corresponding A.U. on the Yaxis.
2. Draw a straight line between 2 adjacent points. This mathematical algorithm is commonly known as the "point-to-point" calculation. Obtain the concentration of the sample by locating the absorbance unit on the Y-axis and finding the corresponding concentration value on the X-axis.

Submit a Product Review to Biocompare.com

Submit a review of this product to Biocompare.com to receive a \$20 Amazon.com gift card! Your reviews help your fellow scientists make the right decisions. Thank you for your contribution.



Human Intact Parathyroid Hormone (PTH) OneStep ELISA Kit

For Research Use Only. Not for use in diagnostic procedures.