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Datasheet for ABIN956190

beta-Thromboglobulin ELISA Kit

Overview

Quantity:	96 tests
Target:	beta-Thromboglobulin (beta-TG)
Reactivity:	Human
Method Type:	Competition ELISA
Application:	ELISA

Product Details

Purpose:	This -TG ELISA kit is intended for laboratory research use only and not for use in diagnostic or therapeutic procedures.
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Characteristics:	<p>The coated well immunoenzymatic assay for the quantitative measurement of beta-TG utilizes a polyclonal anti-beta-TG antibody and an beta-TG-HRP conjugate. The assay sample and buffer are incubated together with beta-TG-HRP conjugate in pre-coated plate for one hour. After the incubation period, the wells are decanted and washed five times. The wells are then incubated with a substrate for HRP enzyme. The product of the enzyme-substrate reaction forms a blue colored complex. Finally, a stop solution is added to stop the reaction, which will then turn the solution yellow. The intensity of color is measured spectrophotometrically at 450 nm in a microplate reader. The intensity of the color is inversely proportional to the beta-TG concentration since beta-TG from samples and beta-TG-HRP conjugate compete for the anti-beta-TG antibody binding site. Since the number of sites is limited, as more sites are occupied by - TG from the sample, fewer sites are left to bind beta-TG-HRP conjugate. Calibrators of</p>

Product Details

known beta-TG concentrations are run concurrently with the samples being assayed and a calibration curve is plotted relating the intensity of the color (O.D.) to the concentration of beta-TG. The beta-TG concentration is interpolated from this calibration curve.

Components:

Microtiter Plate: 96 wells
Calibrator 1 (0 pg/mL)
Calibrator 2 (100 pg/mL)
Calibrator 3 (250 pg/mL)
Calibrator 4 (500 pg/mL)
Calibrator 5 (1,000 pg/mL)
Calibrator 6 (2,500 pg/mL)
Enzyme Conjugate (1 x 6 mL)
Substrate A (1 x 6 mL)
Substrate B (1 x 6 mL)
Stop Solution (1 x 6 mL)
Wash Buffer (100X concentrate) (1 x 6 mL)
Lysis Buffer Solution (1 x 6 mL)

Note: The lysis buffer solution is used only when the sample is cell culture fluid & body fluid & tissue homogenate, If the sample is serum or blood plasma, then the lysis buffer solution is a superfluous reagent.

Material not included:

1. Microplate reader capable of measuring absorbance at 450 nm.
2. Pipettes and pipette tips.
3. 100 mL and 1 liter graduated cylinders.
4. Calibrated adjustable precision pipettes, preferably with disposable plastic tips. (A manifold multi-channel pipette is desirable for large assays.)
5. 37°C incubator.
6. Absorbent paper.
7. Distilled or de-ionized water
8. Data analysis and graphing software. Graph paper: linear (Cartesian), log-log or semi-log, or log-logit as desired.
9. Tubes to prepare calibrator or sample dilutions.

Target Details

Target:

beta-Thromboglobulin (beta-TG)

Alternative Name:

Thromboglobulin, beta ([beta-TG Products](#))

Application Details

Plate: Pre-coated

Assay Procedure: It is recommended that all Calibrators and Samples be added in duplicate to the Microtiter Plate.

1. Secure the desired number of coated wells in the holder then add 100 μ L of Calibrators or Samples to the appropriate well of the antibody pre-coated Microtiter Plate.
2. Add 50 μ L of Conjugate to each well. Mix well. Mixing well in this step is important. Cover and incubate for 1 hour at 37°C.
3. Wash the Microtiter Plate using one of the specified methods indicated below:
4. Manual Washing: Remove incubation mixture by aspirating contents of the plate into a sink or proper waste container. Fill in each well completely with diluted wash solution, and then aspirate contents of the plate into a sink or proper waste container. Repeat this procedure five times for a total of five washes. After washing, invert plate, and blot dry by hitting plate onto absorbent paper or paper towels until no moisture appears. Note: Hold the sides of the plate frame firmly when washing the plate to assure that all strips remain securely in frame. Complete removal of liquid at each step is essential to good performance.
5. Automated Washing: Wash plate five times with diluted wash solution (350-400 μ L/well/wash) using an auto washer. After washing, dry the plate as above. It is recommended that the washer be set for a soaking time of 10 seconds and shaking time of 5 seconds between each wash.
6. Add 50 μ L Substrate A and 50 μ L Substrate B to each well, subsequently. Cover and incubate for 15 minutes at 20-25°C. (Avoid sunlight).
7. Add 50 μ L of stop solution to each well. Mix well.
8. Read the optical density (O.D.) at 450 nm using a microtiter plate reader immediately.

Calculation of Results:

1. This calibration curve is used to determine the amount of an unknown sample. Construct a calibration curve by plotting the average O.D. (450 nm) for each calibrator on the vertical (Y) axis against the concentration on the horizontal (X) axis, and draw a best fit curve through the points on the graph.
2. First, calculate the mean O.D. value for each calibrator and sample. All O.D. values are subtracted by the mean value of the blank control before result interpretation. Construct the calibration curve using graph paper or statistical software.
3. To determine the amount in each sample, first locate the O.D. value on the Y-axis and extend a horizontal line to the calibration curve. At the point of intersection, draw a vertical line to the X-axis and read the corresponding concentration.
4. Any variation in operator, pipetting and washing technique, incubation time or temperature, and kit age can cause variation in result. Each user should obtain their own calibration curve.

Application Details

5. The sensitivity in this assay is 1.0 pg/mL.

6. This assay has high sensitivity and excellent specificity for detection of -TG. No significant cross- reactivity or interference between -TG and analogues was observed. Note: Limited by current skills and knowledge, it is impossible for us to complete the cross-reactivity detection between -TG and all the analogues, therefore, cross reaction may still exist in some cases.

Restrictions:	For Research Use only
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Handling

Storage:	4 °C
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Storage Comment:	All reagents provided are stored at 4°C. Refer to the expiration date on the label.
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Expiry Date:	The expiry date is stated on the label.
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