Human Free Prostate Specific Antigen (f-PSA) OneStep ELISA Kit

Catalog number: EK7079

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.

BOSTER
antibody and ELISA experts

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3942 B Valley Ave, Pleasanton, CA 94566
888-466-3604 | support@bosterbio.com | www.bosterbio.com

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Catalog Number: EK7079

Size: 96 wells/kit

Sample Type: Serum and Plasma

Sensitivity: 1 ng/ml

Assay Range: 1-25 ng/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.

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 (Amax = 370nm and 652nm) that changes to yellow (Amax = 450nm) 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. f-PSA Standard: 6 vials (ready to use)	0.5 ml
3. f-PSA Conjugate Reagent: 1 bottle (ready to use)	12 ml
4. TMB Substrate: 1 bottle (ready to use)	12 ml
5. Stop Solution: 1 bottle (ready to use)	12 ml
6. 20X Wash concentrate: 1 bottle	25 ml

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

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

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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. This test kit is designed for Research Use Only.
5. Do not pipette by mouth. Do not smoke, eat, or drink in the areas in which specimens or kit reagents are handled.
6. The components in this kit are intended for use as an integral unit. The components of different lots should not be mixed.
7. It is recommended that serum samples be run in duplicate.
8. 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 AND HANDLING

1. Collect blood specimens and separate the serum immediately
2. Typically, specimens may be stored refrigerated at (2-8°C) for 5 days. If storage time exceeds 5 days, store 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

Prepare 1X Wash buffer by adding the contents of the bottle (25 ml, 20X) to 475 ml of distilled or deionized water. Store at room temperature (20-25°C).

ASSAY PROCEDURE

Prior to assay, allow reagents to stand at room temperature. Gently mix all reagents before use.

1. Place the desired number of coated strips into the holder.
2. Pipette 50 ul of f-PSA standards, control and patient's sera to selected wells.
3. Add 100 ul of conjugate reagent to all wells
4. Mix the content of the plate, gently, for 30 seconds
5. Cover the plate and incubate for 60 minutes at room temperature (20-25°C).
6. Remove liquid from all wells. Wash wells three times with 300 ul of 1X wash buffer. Blot on absorbent paper towels.
7. Add 100 ul of TMB substrate to all wells
8. Incubate for 15 minutes at room temperature.
9. Add 50 ul of stop solution to all wells. Shake the plate gently to mix the solution.
10. Read absorbance on ELISA Reader at 450 nm within 15 minutes after adding the stopping solution.

CALCULATION OF RESULTS

1. Calculate the average absorbance values (A450) for each set of reference standards, control, and samples.
2. Construct a standard curve by plotting the mean absorbance obtained for each reference standard against its concentration in ng/ml on linear graph paper, with absorbance on the vertical (y) axis and concentration on the horizontal (x) axis.
3. Using the mean absorbance value for each sample, determine the corresponding concentration of fPSA in ng/ml from the standard curve.

LIMITATION OF THE TEST

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1. Reliable and reproducible results will be obtained when the assay procedure is carried out with a complete understanding of the package insert instructions and with adherence to good laboratory practice.
2. The wash procedure is critical. Insufficient washing will result in poor precision and falsely elevated absorbance readings.
3. Sodium azide inhibits the activity of enzyme conjugate; hence do not use samples with Na-Azide preservative.

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