

Datasheet for ABIN1305172

GLP-1 ELISA Kit[Go to Product page](#)**1** Image**1** Publication

Overview

Quantity:	96 tests
Target:	GLP-1
Binding Specificity:	AA 7-36, total
Reactivity:	Human
Method Type:	Sandwich ELISA
Detection Range:	0.06-54 pM/L
Minimum Detection Limit:	0.06 pM/L
Application:	ELISA

Product Details

Purpose:	This ELISA (enzyme-linked immunosorbent assay) kit is produced for the quantitative determination of the total value of glucagon-like peptide-1 (7-36) [GLP-1 (7-36)] and (9-36) [GLP-1 (9-36)] in plasma samples. The primary amino acid sequence of GLP-1 peptide is identical among mammalian species, i.e. rat, mouse, pig, human, etc. This kit is for research purposes only.
Brand:	ED™
Sample Type:	Plasma
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Specificity:	This total GLP-1 assay is a specific measure of GLP-1 (7-36) and (9-36).

Product Details

Cross-Reactivity (Details):	It is expected that this assay does not detect following peptides. GLP-1 (7-36) 100 % GLP-1 (9-36) 100 % GLP-1 (9-37) < 0.1 % GLP-1 (7-37) < 0.1 % GLP-1 (1-36) < 0.1 % GLP-2 < 0.1 % Glucagon < 0.1 %
Components:	1. Streptavidin Coated Microplate One vial containing 12 mL ready-to-use buffer. It should be used only for tracer antibody dilution according to the assay procedures. This reagent should be stored at 2-8 °C and is stable until the expiration date on the kit box.
Material not included:	1. Precision single channel pipettes capable of delivering 25 µL, 50 µL, 100 µL, and 1000 µL etc. 2. Repeating dispenser suitable for delivering 100 µL. 3. Disposable pipette tips suitable for above volume dispensing. 4. Disposable 12 x 75 mm or 13 x 100 glass/plastic tubes. 5. Disposable plastic 100 mL and 1000 mL bottle with caps. 6. Aluminum foil. 7. Deionized or distilled water. 8. Plastic microtiter well cover or polyethylene film. 9. ELISA plate shaker. 10. ELISA multichannel wash bottle or automatic (semi-automatic) washing system. 11. Spectrophotometric microplate reader capable of reading absorbance at 450 nm. 12. DPP-4 Inhibitor.

Target Details

Target:	GLP-1
Alternative Name:	GLP-1 (GLP-1 Products)

Application Details

Comment:	Total GLP-1 ELISA Kit
Assay Time:	4 h
Plate:	Pre-coated
Protocol:	This ELISA is designed, developed and produced for the quantitative measurement of GLP-1 (7-36) and (9-36) in plasma sample. The assay utilizes the two-site sandwich technique with two selected GLP-1 antibodies. This assay used the same assay calibrators and tracer antibodies as the Active GLP-1 (7-36) ELISA (catalog: KT-871). Assay standards, controls and test samples are directly added to wells of a microplate that is coated with streptavidin. Subsequently, a

mixture of biotinylated GLP-1 specific antibody and a horseradish peroxidase (HRP)-conjugated GLP-1 specific antibody is added to each well. After the first incubation period, a sandwich immunocomplex of Streptavidin Biotin-Antibody GLP-1(7-36)/(9-36) HRP-conjugated antibody is formed and attached to the wall of the plate. The unbound HRP-conjugated antibody is removed in a subsequent washing step. For the detection of this immunocomplex, each well is then incubated with a substrate solution in a timed reaction and then measured in a spectrophotometric microplate reader. The enzymatic activity of the immunocomplex bound to GLP-1 (7-36)/(9-36) on the wall of the microtiter well is directly proportional to the amount of Total GLP-1 in the sample.

Reagent Preparation:

(1) Prior to use allow all reagents to come to room temperature. Reagents from different kit lot numbers should not be combined or interchanged.

(2) ELISA Wash Concentrate must be diluted to working solution prior use. Please see REAGENTS section for details.

(3) Reconstitute all standards and controls by adding 1.0 mL of demineralized water to each vial. Allow the standards and controls to sit undisturbed for 10 minutes, and then mix well by gentle vortexing. These reconstituted standards and controls must be stored at - 20 C or below. Do not exceed 3 freeze-thaw cycles.

Sample Collection:

(1) No special preparation of individual is necessary prior to specimen collection. However, fasting sample and non-fasting/glucose induced sample may present great significance for Total GLP-1 level. (2) Both BD[®] P700 Blood Collection and Preservation System (contains a DPP-4 protease inhibitor cocktail) and lavender top Vacutainer[®] EDTA-plasma tube can be used for sample collection. (3) If the Vacutainer[®] EDTA-plasma tube is used for sample collection, it is recommended (but not necessary) to add appropriate amount of DPP-4 inhibitor to the collected EDTA whole blood right after the collection. Refer to DPP-4 inhibitor manufacturer's instruction. Invert tube to mix well and place the tube on ice bath. Centrifuge the tube at 1000 g for 10 minutes in a refrigerated centrifuge. Note: since sample with DPP-4 inhibitor is strongly recommended for measurement of Active GLP-1 (7-36), the DPP-4 inhibitor should be included in sample collection if the same sample will be used for both Active GLP-1 (7-36) and Total GLP-1 measurement. (4) Plasma samples should be stored at 2 ? 8 C if they will be tested within 3 hours of collection. For longer storage, it is recommended to store the plasma sample at -70 C. Aliquot samples before freezing if necessary.

Sample Preparation:

It is recommended to measure patient samples directly (no extraction) with this highly sensitive Total GLP-1 ELISA.

(1) EDTA-plasma samples with or without DPP-IV inhibitor can be directly measured for the Total GLP-1 concentration with this kit.

(2) If a pre-assay sample extraction procedure is desired (i.e. measuring cell/tissue culture supernatant GLP-1 level), we recommend using a solid phase sample extraction procedure with a GLP-1 Sample Extraction Kit (Cat# KT-910, Epitope Diagnostics).

Assay Procedure:

- (1) Place a sufficient number of streptavidin-coated microwell strips/wells (Cat. 10040B) in a holder to run Total GLP-1 standards, controls and unknown samples in duplicate.
- (2) Test Configuration
- (3) Prepare Total GLP-1 Antibody Mixture: mixing Total GLP-1 Tracer Antibody and Total GLP-1 Capture Antibody by 1:21 fold dilution of the Tracer Antibody (30360) and by 1:21 fold dilution of the biotinylated Capture Antibody (30361) with the Tracer antibody Diluent. For each strip, it is required to mix 1 mL of the Tracer Antibody Diluent (30017) with 50 μ L the Capture Antibody and 50 μ L of the Tracer Antibody in a clean test tube.
- (4) Add 100 μ L of standards, controls and test samples into the designated microwell.
- (5) Add 100 μ L of Total GLP-1 Antibody Mixture to each well
- (6) Cover the plate with one plate sealer and incubate plate at 2-8 °C, static for 20 - 24 hours.
- (7) Remove plate sealer. Aspirate the contents of each well. Wash each well 5 times by dispensing 350 μ L of working wash solution into each well and then completely aspirating the contents. Alternatively, an automated microplate washer can be used.
- (8) Add 200 μ L of ELISA HRP Substrate into each of the wells.
- (9) Cover the plate with one plate sealer and also with aluminum foil to avoid exposure to light.
- (10) Incubate plate at room temperature, static for 20 min.
- (11) Remove the aluminum foil and plate sealer. Add 50 μ L of ELISA Stop Solution into each of the wells. Mix gently.
- (12) Read the absorbance at wavelength 450nm/620 nm within 10 minutes in a microplate reader.

Calculation of Results:

1. Calculate the average absorbance for each pair of duplicate test results.
2. Subtract the average absorbance of the STD 1 (0 ng/mL) from the average absorbance of all other readings to obtain corrected absorbance.
3. The standard curve is generated by the corrected absorbances of all standard levels on the ordinate against the standard concentration on the abscissa using point-to-point or log-log paper. Appropriate computer assisted data reduction programs may also be used for the calculation of results. We recommend using Point-to-Point or Quadratic curve fit. The GLP-1 (7-36) concentrations for the controls and test samples are read directly from the standard curve using their respective corrected absorbance.

Assay Precision:

The intra-assay precision was determined by 8 replicates for two control samples in a single assay. A very satisfactory within assay CV% was obtained as indicated below. The inter assay

Application Details

precision was determined by 8 individual assays in different dates with two control samples. A satisfactory between assay CV% was observed as indicated below.

Restrictions: For Research Use only

Handling

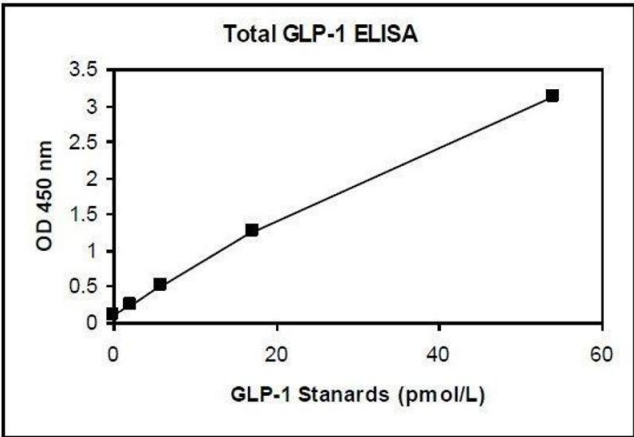
Precaution of Use: The reagents must be used in a professional laboratory environment and are for research use only. Source material (e.g. highly purified bovine serum albumin) of bovine serum was derived in the contiguous 48 United States. It was obtained only from healthy donor animals maintained under veterinary supervision and found free of contagious diseases. Wear gloves while performing this assay and handle these reagents as if they are potentially infectious. Avoid contact with reagents containing TMB, hydrogen peroxide, or sulfuric acid. TMB may cause irritation to skin and mucous membranes and cause an allergic skin reaction. TMB is a suspected carcinogen. Sulfuric acid may cause severe irritation on contact with skin. Do not get in eyes, on skin, or on clothing. Do not ingest or inhale fumes. On contact, flush with copious amounts of water for at least 15 minutes. Use Good Laboratory Practices.

Storage: 4 °C

Publications

Product cited in: Huang, Wang, Wang, Zhang, Deng, Wang, Zhang, Huang: "Poly(ADP-ribose) polymerase 1 is indispensable for transforming growth factor- β induced Smad3 activation in vascular smooth muscle cell." in: **PLoS ONE**, Vol. 6, Issue 10, pp. e27123, (2011) ([PubMed](#)).

Images



ELISA

Image 1.