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Datasheet for ABIN457061
PIVKA-II ELISA Kit

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Overview

Quantity:	96 tests
Target:	PIVKA-II
Reactivity:	Human
Method Type:	Sandwich ELISA
Detection Range:	0.312-20 ng/mL
Minimum Detection Limit:	0.312 ng/mL
Application:	ELISA

Product Details

Purpose:	For the quantitative determination of human protein induced vitamin K absence or antagonist-II (PIVKA-II) concentrations in serum, plasma, ascitic fluid, tissue homogenates.
Sample Type:	Plasma, Serum, Tissue Homogenate
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Specificity:	This assay has high sensitivity and excellent specificity for detection of human PIVKA-II.
Cross-Reactivity (Details):	Limited by current skills and knowledge, it is impossible for us to complete the cross-reactivity detection between the target antigen and all analogues for other species. Therefore, cross reaction may still exist.
Sensitivity:	0.078 ng/mL
Components:	• Assay plate (12 × 8 coated Microwells)

Product Details

- Standard (freeze dried)
- Biotin-antibody (100 × concentrate)
- HRP-avidin (100 × concentrate)
- Biotin-antibody Diluent
- HRP-avidin Diluent
- Sample Diluent
- Wash Buffer (25 × concentrate)
- TMB Substrate
- Stop Solution
- Adhesive Strip (for 96 wells)
- Instruction manual

Material not included:	• Microplate reader capable of measuring absorbance at 450nm, with the correction wavelength set at 540nm or 570nm.
	• An incubator which can provide stable incubation conditions up to 37°C ± 0.5°C.
	• Squirt bottle, manifold dispenser or automated microplate washer.
	• Absorbent paper for blotting the microtiter plate.
	• 100mL and 500mL graduated cylinders.
	• Deionized or distilled water.
	• Pipettes and pipette tips.
	• Test tubes for dilution.

Target Details

Target:	PIVKA-II
Alternative Name:	Protein Induced Vitamin K Absence or Antagonist-II (PIVKA-II) (PIVKA-II Products)

Application Details

Application Notes:	• The supplier is only responsible for the kit itself, but not for the samples consumed during the assay. The user should calculate the possible amount of the samples used in the whole test. Please reserve sufficient samples in advance.
	• Samples to be used within 5 days may be stored at 2-8°C, otherwise samples must be stored at -20°C (≤ 1 month) or -80°C (≤ 2 months) to avoid loss of bioactivity and contamination.
	• Grossly hemolyzed samples are not suitable for use in this assay.
	• If the samples are not indicated in the manual, a preliminary experiment to determine the validity of the kit is necessary.
	• Please predict the concentration before assaying. If values for these are not within the range of the standard curve, users must determine the optimal sample dilutions for their particular experiments.
	• Tissue or cell extraction samples prepared by chemical lysis buffer may cause unexpected ELISA results due to the impacts of certain chemicals.

Application Details

- Owing to the possibility of mismatching between antigens from another resource and antibodies used in this supplier's kits (e.g., antibody targets conformational epitope rather than linear epitope), some native or recombinant proteins from other manufacturers may not be recognized by this supplier's products.
- Influenced by factors including cell viability, cell number and cell sampling time, samples from cell culture supernatant may not be recognized by the kit.
- Fresh samples without long time storage are recommended for the test. Otherwise, protein degradation and denaturalization may occur in those samples and finally lead to wrong results.

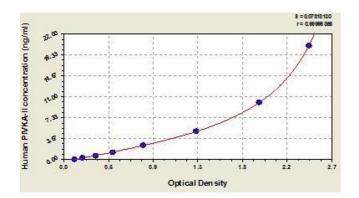|	
Comment:	Detection wavelength: 450 nm Information on standard material: Depending on the antigen to be detected, standards can be either native or recombinant protein. The recombinant proteins are being expressed in CHO cells in most cases. Please inquire for more information. The formulation of auxiliary material in the standard is considered proprietary information, however it does not contain any poisonous substance. Proclin 300 (1:3000) is used as preservative. Information on reagents: In most cases the stop solution provided is 1 N H2SO4. The formulation of wash solution is proprietary information. None of the components contain (sodium) azide, thimerosal, 2-mercaptoethanol (2-ME) or any other poisonous materials. For the sandwich method kits, the sample diluent, antibody diluent, enzyme diluent and standard all contain BSA. Information on antibodies: The antibodies provided in different kits vary in regards to clonality and host. Some antibodies are affinity purified, some are Protein A
Sample Volume:	100 μL
Assay Time:	1 - 4.5 h
Plate:	Pre-coated
Protocol:	This assay employs the quantitative sandwich enzyme immunoassay technique. Antibody specific for PIVKA-II has been pre-coated onto a microplate. Standards and samples are pipetted into the wells and any PIVKA-II present is bound by the immobilized antibody. After removing any unbound substances, a Horseradish Peroxidase (HRP)-conjugated antibody specific for PIVKA-II is added to the wells. Following a wash to remove any unbound reagent, a

substrate solution is added to the wells and color develops in proportion to the amount of PIVKA-II bound in the initial step. The color development is stopped and the intensity of the color is measured.

Sample Collection:	• **Serum**: Use a serum separator tube (SST) and allow samples to clot for two hours at room temperature or overnight at 4 °C before centrifugation for 15 minutes at 1000 × g. Remove serum and assay immediately or aliquot and store samples at -20 °C or -80 °C. Avoid repeated freeze-thaw cycles. • **Plasma**: Collect plasma using EDTA or heparin as an anticoagulant. Centrifuge for 15 minutes at 1000 × g at 2-8 °C within 30 minutes of collection. Assay immediately or aliquot and store samples at -20 °C or -80 °C. Avoid repeated freeze-thaw cycles. • **Tissue Homogenates**: Rinse 100 mg tissue with 1× PBS, homogenize in 1mL of 1× PBS and store overnight at -20 °C. After two freeze-thaw cycles to break the cell membranes, centrifuge the homogenates for 5 minutes at 5000 × g, 2-8 °C. Remove and assay the supernate immediately. Alternatively, aliquot and store samples at -20 °C or -80 °C. Centrifuge the sample again after thawing before the assay. Avoid repeated freeze-thaw cycles.
Calculation of Results:	Average the duplicate readings for each standard and sample and subtract the average zero standard optical density. Create a standard curve by reducing the data using computer software capable of generating a four parameter logistic (4-PL) curve fit. As an alternative, construct a standard curve by plotting the mean absorbance for each standard on the x-axis against the concentration on the y-axis and draw a best fit curve through the points on the graph. The data may be linearized by plotting the log of the target antigen concentration versus the log of the O.D. and the best fit line can be determined by regression analysis. This procedure will produce an adequate but less precise fit of the data. If samples have been diluted, the concentration read from the standard curve must be multiplied by the dilution factor.
Assay Precision:	Intra-assay precision (precision within an assay): Three samples of known concentration were tested twenty times on one plate to assess precision. Inter-assay precision (precision between assays): Three samples of known concentration were tested in twenty assays to assess precision. • Intra-assay: CV% less than 8% • Inter-assay: CV% less than 10%
Restrictions:	For Research Use only

Handling

Precaution of Use:	The Stop Solution provided with this kit is an acid solution. Wear eye, hand, face and clothing protection when using this material.

Handling Advice:	• The kit should not be used beyond the expiration date on the kit label. • Do not mix or substitute reagents with those from other lots or sources. • If samples generate values higher than the highest standard, dilute the samples with Sample Diluent and repeat the assay. • Any variation in Sample Diluent, operator, pipetting technique, washing technique, incubation time/temperature and kit age can cause variation in binding. • This assay is designed to eliminate interference by soluble receptors, binding proteins and other factors present in biological samples. Until all factors have been tested in the Immunoassay, the possibility of interference cannot be excluded.

Storage:	4 °C/-20 °C
Storage Comment:	For unopened kit: All the reagents should be kept according to the labels on vials.
Expiry Date:	6 months

Publications

Product cited in:	Cullere, Lauterbach, Tsuboi, Mayadas: "Neutrophil-selective CD18 silencing using RNA interference in vivo." in: **Blood**, Vol. 111, Issue 7, pp. 3591-8, (2008) (PubMed). Varga, Balkow, Wild, Stadtbaeumer, Krummen, Rothoeft, Higuchi, Beissert, Wethmar, Scharffetter-Kochanek, Vestweber, Grabbe: "Active MAC-1 (CD11b/CD18) on DCs inhibits full T-cell activation." in: **Blood**, Vol. 109, Issue 2, pp. 661-9, (2007) (PubMed). Watts, Beurskens, Martin-Padura, Ballantyne, Klickstein, Brenner, Lee: "Manifestations of inflammatory arthritis are critically dependent on LFA-1." in: **Journal of immunology (Baltimore, Md. : 1950)**, Vol. 174, Issue 6, pp. 3668-75, (2005) (PubMed). Barlow, Langston, Matthews, Chidlow, Kevil: "CD18 deficiency protects against multiple low-dose streptozotocin-induced diabetes." in: **The American journal of pathology**, Vol. 165, Issue 6, pp. 1849-52, (2004) (PubMed). Sakurai, Taguchi, Anand, Ambati, Gragoudas, Miller, Adamis, Ambati: "Targeted disruption of the CD18 or ICAM-1 gene inhibits choroidal neovascularization." in: **Investigative ophthalmology & visual science**, Vol. 44, Issue 6, pp. 2743-9, (2003) (PubMed).

Images

ELISA

Image 1. Typical standard curve