Datasheet for ABIN1873286

Ubiquitin ELISA Kit

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Overview

Quantity:	96 tests
Target:	Ubiquitin (Ub)
Reactivity:	Human, Mouse, Rat, Cow, Horse, Rabbit, Dog, Pig
Method Type:	Competition ELISA
Detection Range:	4.94 ng/mL - 400 ng/mL
Minimum Detection Limit:	4.94 ng/mL
Application:	ELISA

Product Details

Purpose:	The kit is a competitive inhibition enzyme immunoassay technique for the in vitro quantitative measurement of ubiquitin in serum, plasma, tissue homogenates and other biological fluids.Due - the 100% homology of the sequence among different species, the kit can be used - detect human, rat, mouse, porcine, rabbit, equine, bovine, canine samples.
Sample Type:	Plasma, Serum
Detection Method:	Colorimetric
Specificity:	This assay has high sensitivity and excellent specificity for detection of Ubiquitin (Ub).
Cross-Reactivity (Details):	No significant cross-reactivity or interference between Ubiquitin (Ub) and analogues was observed.
Sensitivity:	2.14 ng/mL
Characteristics:	This assay employs the competitive inhibition enzyme immunoassay technique. A monoclonal antibody specific to ubiquitin has been pre-coated onto a microplate. A competitive inhibition

Product Details

	reaction is launched between biotin labeled ubiquitin and unlabeled ubiquitin (Standards or samples) with the pre-coated antibody specific to ubiquitin. After incubation the unbound conjugate is washed off. Next, avidin conjugated to Horseradish Peroxidase (HRP) is added to each microplate well and incubated. The amount of bound HRP conjugate is reverse proportional to the concentration of ubiquitin in the sample. After addition of the substrate solution, the intensity of color developed is reverse proportional to the concentration of ubiquitin in the sample.
Components:	• Pre-coated, ready to use 96-well strip plate, flat buttom • Plate sealer for 96 wells • Reference Standard • Standard Diluent • Detection Reagent A • Detection Reagent B • Assay Diluent A • Assay Diluent B • Reagent Diluent (if Detection Reagent is lyophilized) • TMB Substrate • Stop Solution • Wash Buffer (30 x concentrate) • Instruction manual

Target Details

Target:	Ubiquitin (Ub)
Abstract:	Ub Products
Pathways:	Mitotic G1-G1/S Phases, Ubiquitin Proteasome Pathway

Application Details

Application Notes:	• Limited by the current condition and scientific technology, we cannot completely conduct the comprehensive identification and analysis on the raw material provided by suppliers. So there might be some qualitative and technical risks to use the kit. • The final experimental results will be closely related to validity of the products, operation skills of the end users and the experimental environments. Please make sure that sufficient samples are available. • Kits from different batches may be a little different in detection range, sensitivity and color developing time. • Do not mix or substitute reagents from one kit lot to another. Use only the reagents supplied by manufacturer.

Application Details

	• Protect all reagents from strong light during storage and incubation. All the bottle caps of reagents should be covered tightly to prevent the evaporation and contamination of microorganism. • There may be some foggy substance in the wells when the plate is opened at the first time. It will not have any effect on the final assay results. Do not remove microtiter plate from the storage bag until needed. • Wrong operations during the reagents preparation and loading, as well as incorrect parameter setting for the plate reader may lead to incorrect results. A microplate plate reader with a bandwidth of 10nm or less and an optical density range of 0-3 O.D. or greater at 450 ± 10nm wavelength is acceptable for use in absorbance measurement. Please read the instruction carefully and adjust the instrument prior to the experiment. • Even the same operator might get different results in two separate experiments. In order to get better reproducible results, the operation of every step in the assay should be controlled. Furthermore, a preliminary experiment before assay for each batch is recommended. • Each kit has been strictly passed Q.C test. However, results from end users might be inconsistent with our in-house data due to some unexpected transportation conditions or different lab equipments. Intra-assay variance among kits from different batches might arise from above factors, too. • Kits from different manufacturers for the same item might produce different results, since we have not compared our products with other manufacturers.
Comment:	Information on standard material: The standard might be recombinant protein or natural protein, that will depend on the specific kit. Moreover, the expression system is E.coli or yeast or mammal cell. There is 0.05% proclin 300 in the standard as preservative. Information on reagents: The stop solution used in the kit is sulfuric acid with concentration of 1 mol/L. And the wash solution is TBS. The standard diluent contains 0.02 % sodium azide, assay diluent A and assay diluent B contain 0.01% sodium azide. Some kits can contain is BSA in them. Information on antibodies: The provided antibodies and their host vary in different kits.
Sample Volume:	50 µL
Assay Time:	2.5 h
Plate:	Strips
Protocol:	1. Prepare all reagents, samples and standards, 2. Add 50µL standard or sample to each well.

Then add 50µL prepared Detection Reagent A immediately.
Shake and mix. Incubate 1 hour at 37 °C,
3. Aspirate and wash 3 times,
4. Add 100µL prepared Detection Reagent B. Incubate 30 minutes at 37 °C,
5. Aspirate and wash 5 times,
6. Add 90µL Substrate Solution. Incubate 10-20 minutes at 37 °C,
7. Add 50µL Stop Solution. Read at 450 nm immediately.

Reagent Preparation:

1. Bring all kit components and samples to room temperature (18-25 °C) before use. If the kit will not be used up in one time, please only take out strips and reagents for present experiment, and leave the remaining strips and reagents in required condition.
2. Standard - Reconstitute the Standard with 1.0 mL of Standard Diluent, kept for 10 minutes at room temperature, shake gently(not to foam). The concentration of the standard in the stock solution is 400 ng/mL. Please prepare 5 tubes containing 0.6 mL Standard Diluent and produce a triple dilution series according to the picture shown below. Mix each tube thoroughly before the next transfer. Set up 5 points of diluted standard such as 400 ng/mL, 133.33 ng/mL, 44.44 ng/mL, 14.81 ng/mL, 4.94 ng/mL, and the last EP tubes with Standard Diluent is the blank as 0 ng/mL.
3. Detection Reagent A and Detection Reagent B - If lyophilized reconstitute the Detection Reagent A with 150µL of Reagent Diluent, kept for 10 minutes at room temperature, shake gently (not to foam). Briefly spin or centrifuge the stock Detection A and Detection B before use. Dilute them to the working concentration 100-fold with Assay Diluent A and B, respectively.
4. Wash Solution - Dilute 20 mL of Wash Solution concentrate (30x) with 580 mL of deionized or distilled water to prepare 600 mL of Wash Solution (1x).
5. TMB substrate - Aspirate the needed dosage of the solution with sterilized tips and do not dump the residual solution into the vial again.

Note:

1. Making serial dilution in the wells directly is not permitted.
2. Prepare standard within 15 minutes before assay. Please do not dissolve the reagents at 37 °C directly.
3. Detection Reagent A and B are sticky solutions, therefore, slowly pipette them to reduce the volume errors.
4. Please carefully reconstitute Standards or working Detection Reagent A and B according to the instruction, and avoid foaming and mix gently until the crystals are completely dissolved. To minimize imprecision caused by pipetting, use small volumes and ensure that pipettors are calibrated. It is recommended to suck more than 10µL for one pipetting.
5. The reconstituted Standards, Detection Reagent A and Detection Reagent B can be used only once.
6. If crystals have formed in the Wash Solution concentrate (30x), warm to room temperature and mix gently until the crystals are completely dissolved.
7. Contaminated water or container for reagent preparation will influence the detection result.

Application Details

Assay Precision:	**Intra-assay Precision (precision within an assay):** Three samples with low, medium and high levels of the target antigen were tested twenty times on one plate, respectively. **Inter-assay Precision (precision between assays):** Three samples with low, medium and high levels of the target antigen were tested on three different plates, eight replicates in each plate. **CV (%)** = SD/mean X 100 • Intra-assay: CV less than 10 % • Inter-assay: CV less than 12 %
Restrictions:	For Research Use only

Handling

Precaution of Use:	The Stop Solution suggested for use with this kit is an acid solution. Wear eye, hand, face, and clothing protection when using this material.
Handling Advice:	The stability of ELISA kit is determined by the loss rate of activity. The loss rate of this kit is less than 5 % within the expiration date under appropriate storage condition. To minimize extra influence on the performance, operation procedures and lab conditions, especially room temperature, air humidity, incubator temperature should be strictly controlled. It is also strongly suggested that the whole assay is performed by the same operator from the beginning to the end.
Storage:	4 °C/-20 °C
Storage Comment:	• For unopened kit: All the reagents should be kept according to the labels on vials. The Standard, Detection Reagent A, Detection Reagent B and the 96-well strip plate should be stored at -20°C upon receipt while the others should be at 4°C. • For opened kit: When the kit is opened, the remaining reagents still need to be stored according to the above storage condition. Besides, please return the unused wells to the foil pouch containing the desiccant pack, and reseal along entire edge of zip-seal. Note: It is highly recommended to use the remaining reagents within 1 month provided this is within the expiration date of the kit. • For ELISA kit, 1 day storage at 37°C can be considered as 2 months at 4°C, which means 3 days at 37°C equaling 6 months at 4°C.
Expiry Date:	6 months