

Vitamin D2 (Ergocalciferol) ELISA Kit

Catalogue No.: abx575377

Vitamin D2 (Ergocalciferol) ELISA Kit is an ELISA Kit for the in vitro quantitative measurement of Vitamin concentrations in serum, plasma and other biological fluids.

Target:	Vitamin D2 (Ergocalciferol)
Reactivity:	General
Tested Applications:	ELISA
Recommended dilutions:	Optimal dilutions/concentrations should be determined by the end user.
Storage:	Shipped at 4 °C. Upon receipt, store the kit according to the storage instruction in the kit's manual.
Validity:	The validity for this kit is at least 6 months. Up to 12 months validity can be provided on request.
Stability:	The stability of the kit is determined by the rate of activity loss. The loss rate is less than 5% within the expiration date under appropriate storage conditions. To minimize performance fluctuations, operation procedures and lab conditions should be strictly controlled. It is also strongly suggested that the whole assay is performed by the same user throughout.
Test Range:	3.125 ng/ml - 200 ng/ml
Sensitivity:	1.88 ng/ml
Standard Form:	Lyophilized
Detection Method:	Colorimetric
Assay Type:	Competitive
Assay Data:	Quantitative
Sample Type:	Serum, plasma and other biological fluids.

Kit Components:

The kit components listed are for reference only. The product manual may differ slightly. The product should be used as stated on the product manual included and delivered together with the product.

- Pre-coated 96-Well Microplate
- Standard
- Standard Diluent Buffer
- Wash Buffer
- Detection Reagent A
- Detection Reagent B
- Diluent A
- Diluent B
- TMB Substrate
- Stop Solution
- Plate Sealer

Material Required But Not Provided:

- 37°C incubator
- Multi and single channel pipettes and sterile pipette tips
- Squirt bottle or automated microplate washer
- 1.5 ml tubes
- Distilled water
- Absorbent filter papers
- 100 ml and 1 liter graduated cylinders
- Microplate reader (wavelength: 450 nm)
- ELISA Shaker

Sample Collection/Preparation:

- **Serum:** Samples should be collected into a serum separator tube. Coagulate the serum by leaving the tube undisturbed in a vertical position overnight at 4°C or at room temperature for up to 60 minutes. Centrifuge at approximately 1000 × g for 20 min. Analyze the serum immediately or aliquot and store at -20°C or -80°C.
- **Plasma:** Collect plasma using heparin or EDTA as an anticoagulant. Centrifuge for 15 minutes at 1000 × g within 30 minutes of collection. Assay immediately or aliquot and store at -20°C or -80°C. Avoid hemolysis and high cholesterol samples.
- **Tissue homogenates:** The preparation of tissue homogenates will vary depending upon tissue type – this is just an example. Rinse tissues with ice-cold PBS to remove the excess of blood. Weigh before homogenization. Finely mince tissues and homogenize with a tissue homogenizer on ice in PBS and sonicate the cell suspension. Centrifuge the homogenates at 5000 × g for 5 min and collect the supernatant. Assay immediately or aliquot and store at -20°C.

Reagent Preparation:

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- 1) Standard: Prepare the standard with the recommended volume of Standard Diluent Buffer, to make the standard solution. Then use the Standard Diluent buffer to carry out serial dilutions of the standard solution, as instructed in the Protocol.
- 2) Wash Buffer: Dilute the concentrated Wash Buffer with distilled water, as instructed in the Protocol.
- 3) Detection Reagent Preparation: Calculate the total volume of working solution required. Dilute Detection Reagent A and Detection Reagent B with Diluent A and Diluent B, respectively, at 1:100.

Assay Procedure:

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- 1) Set standard, test samples and control wells.
- 2) Aliquot 50 µl of diluted standard into the standard wells.
- 3) Aliquot 50 µl of Standard Diluent buffer into the control (zero) well.
- 4) Aliquot 50 µl of diluted samples into the sample wells.
- 5) Immediately aliquot 50 µl of Detection Reagent A to each well. Incubate for 45 mins at 37 °C.
- 6) Wash 3 times.
- 7) Aliquot 100 µl of Detection Reagent B to each well. Incubate for 30 mins at 37 °C.
- 8) Wash 5 times.
- 9) Aliquot 90 µl of TMB Substrate to each well. Incubate for 10-20 mins at 37 °C.
- 10) Aliquot 50 µl of Stop Solution.
- 11) Measure the OD at 450 nm.

Assay Precision:

Intra-assay Precision (Precision within an assay): 3 samples with low, medium and high levels of Vitamin D2 (Ergocalciferol) were tested 20 times on one plate, respectively.

Inter-assay Precision (Precision between assays): 3 samples with low, medium and high levels of Vitamin D2 (Ergocalciferol) were tested on 3 different plates, 8 replicates in each plate.

$$CV (\%) = (\text{Standard Deviation} / \text{mean}) \times 100$$

Intra-Assay: CV<10%

Inter-Assay: CV<10%

Note:

THIS PRODUCT IS FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES.

The range and sensitivity is subject to change. Please contact us for the latest product information. For accurate results, sample concentrations must be diluted to mid-range of the kit. If you require a specific range, please contact us in advance or write your request in your order comments.

Please note that our kits are optimised for detection of native samples, rather than recombinant proteins. We are unable to guarantee detection of recombinant proteins, as they may have different sequences or tertiary structures to the native protein.