Datasheet for ABIN996930

# Estradiol ELISA Kit

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## Overview

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| Quantity: | 96 tests |
| Target: | Estradiol |
| Reactivity: | Hormone |
| Method Type: | Competition ELISA |
| Detection Range: | 0-1000 pg/mL |
| Minimum Detection Limit: | 0 pg/mL |
| Application: | ELISA |

## Product Details

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| Purpose: | For the quantitative determination of Estradiol (E2) concentration in human serum |
| Sample Type: | Serum |
| Analytical Method: | Quantitative |
| Detection Method: | Colorimetric |
| Specificity: | 100% |
| Sensitivity: | 10 pg/mL |
| Material not included: | 1. Precision pipettes: 25 mL , 50 mL , 100 mL , 200 mL , and 1.0 mL .<br>2. Disposable pipette tips.<br>3. Distilled and deionized water.<br>4. Vortex mixer or equivalent.<br>5. Absorbent paper or paper towel.<br>6. Linear-linear graph paper. |

## Product Details

7. Microtiter plate reader.

## Target Details

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| Target: | Estradiol |
| Abstract: | Estradiol Products |
| Background: | Estradiol (E2) is a C18 steroid hormone with a phenolic A ring. This steroid hormone has a molecular weight of 272.4. It is the most potent natural Estrogen, produced mainly by the ovary, placenta, and in smaller amounts by the adrenal cortex, and the male testes. Estradiol (E2) is secreted into the blood stream where 98 % of it circulates bound to sex hormone binding globulin (SHBG). To a lesser extent it is bound to other serum proteins such as albumin. Only a tiny fraction circulates as free hormone or in the conjugated form. Estrogenic activity is effected via estradiol-receptor complexes which trigger the appropriate response at the nuclear level in the target sites. These sites include the follicles, uterus, breast, vagina, urethra, hypothalamus, pituitary and to a lesser extent the liver and skin. In non-pregnant women with normal menstrual cycles, estradiol secretion follows a cyclic, biphasic pattern with the highest concentration found immediately prior to ovulation.<br>The rising estradiol concentration is understood to exert a positive feedback influence at the level of the pituitary where it influences the secretion of the gonadotropins, follicle stimulating hormone (FSH), and luteinizing hormone (LH), which are essential for follicular maturation and ovulation, respectively. Following ovulation, estradiol levels fall rapidly until the luteal cells become active resulting in a secondary gentle rise and plateau of estradiol in the luteal phase. During pregnancy, maternal serum Estradiol levels increase considerably, to well above the pre-ovulatory peak levels and high levels are sustained throughout pregnancy.<br>Serum Estradiol measurements are a valuable index in evaluating a variety of menstrual dysfunctions such as precocious or delayed puberty in girls and primary and secondary amenorrhea and menopause. Estradiol levels have been reported to be increased in patients with feminizing syndromes, gynaecomastia and testicular tumors. In cases of infertility, serum Estradiol measurements are useful for monitoring induction of ovulation following treatment with, for example, clomiphene citrate, LH-releasing hormone (LH-RH), or exogenous gonadotropins. During ovarian hyperstimulation for in vitro fertilization (IVF), serum estradiol concentrations are usually monitored daily for optimal timing of human chorionic gonadotropin (hCG) administration and oocyte collection. |

## Application Details

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| Comment: | Quality Control:<br>Good laboratory practice requires that controls are run with each calibration curve. A statistically significant number of controls should be assayed - establish mean values and acceptable ranges - assure proper performance. We recommend using Bio-Rad Lyphochek Immunoassay Control Sera as controls. Estradiol EIA kit also provides with internal controls, Level 1 and 2.<br>Limitations of procedure:<br>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 - good laboratory practice.<br>2. The wash procedure is critical. Insufficient washing will result in poor precision and falsely elevated absorbance readings. |
| Sample Volume: | 25 µL |
| Assay Time: | 1.5 h |
| Plate: | Pre-coated |
| Reagent Preparation: | 1. All reagents should be allowed to reach room temperature (18-25 °C) before use.<br>2. All reagents should be mixed by gentle inversion or swirling prior to use. Do not induce foaming.<br>3. Samples with expected testosterone concentrations over 1,000 pg/mL may be quantitated by dilution with diluent available from Diagnostic Automation, Inc. S 31- SeL 2 c 1 500 Estradiol Conc. (pg/ mL) o (A .Q < 0 |
| Sample Preparation: | 1. Serum should be used in the test.<br>2. No special pretreatment of sample is necessary.<br>3. Serum samples may be stored at 2-8 °C for up to 24 hours, and should be frozen at -20 °C or lower for longer periods. Avoid grossly hemolytic (bright red), lipemic (milky), or turbid samples.<br>4. Please note: Samples containing sodium azide should not be used in the assay.<br>PROCEDURAL NOTES 1. Manual Pipetting: It is recommended that no more than 32 wells be used for each assay run. Pipetting of all standards, samples, and controls should be completed within 3 minutes.<br>2. Automated Pipetting: A full plate of 96 wells may be used in each assay run. However, it is recommended that pipetting of all standards, samples, and controls be completed within 3 minutes.<br>3. All standards, samples, and controls should be run in duplicate concurrently so that all conditions of testing are the same. |

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| | 4. It is recommended that the wells be read within 15 minutes following addition of Stop Solution. |
| Calculation of Results: | 1. Calculate the mean absorbance value (A450) for each set of reference standards, controls and samples.<br><br>2. Construct a standard curve by plotting the mean absorbance obtained for each reference standard against its concentration in pg/mL on a linear-linear graph paper, with absorbance values on the vertical or Y axis, and concentrations on the horizontal or X axis.<br><br>3. Use the mean absorbance values for each specimen to determine the corresponding concentration of Estradiol in pg/mL from the standard curve.<br>4. Any values obtained for diluted samples must be further converted by applying the appropriate dilution factor in the calculations.<br>I. Accuracy A statistical study using 80 human serum samples demonstrated good correlation with a commercially available kit as shown below. Comparison between the Diagnostic Automation, Inc. Estradiol EIA and the DRG Estradiol kit provided the following data: N = 80 Correlation coefficient = 0.979 Slope = 0.663 Intercept =<br>1.562 DAI Mean = 169 pg/mL DRG Mean = 113 pg/mL II.<br>Each laboratory should establish its own normal range based on the patient population. Diagnostic Automation, Inc. Estradiol EIA was performed on rando mLy selected outpatient clinical laboratory samples. The results of these determinations are as follows: Males: Females: < 60 pg/mL < 18 pg/mL 30-100 pg/mL 100-400 pg/mL 60-150 pg/mL up to 35,000 pg/mL < 10 pg/mL 8 9<br>1. Secure the desired number of coated wells in the holder.<br><br>2. Dispense 25 mL of standards, specimens and controls into appropriate wells.<br><br>3. Dispense 100 mL of Estradiol-HRP Conjugate Reagent into each well.<br>4. Dispense 50 mL of rabbit anti-Estradiol (E2) reagent to each well.<br>5. Thoroughly mix for 30 seconds. It is very important to mix them completely. 6. Incubate at room temperature (18-25 °C) for 90 minutes. 7. Rinse and flick the microwells 5 times with distilled or deionized water. (Please do not use tap water.) Dispense 100 mL of TMB Reagent into each well. Gently mix for 10 seconds. Incubate at room temperature (18-25 °C) for 20 minutes. 10. Stop the reaction by adding 100 mL of Stop Solution to each well. 1<br>1. Gently mix 30 seconds. It is important to make sure that all the blue color changes to yellow |

## Application Details

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| | color completely. 1<br>2. Read absorbance at 450 nm with a microtiter well reader within 15 minutes. postmenopausal phase ovulating, early follicular late follicular luteal phase pregnant, normal prepubertal children, normal<br>Results of a typical standard run with optical density readings at 450 nm shown in the Y axis against Estradiol concentrations shown in the X axis. Note: This standard curve is for the purpose of illustration only, and should not be used to calculate unknowns. Each laboratory must provide its own data and standard curve in each experiment. |
| Restrictions: | For Research Use only |

## Handling

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| Storage: | 4 °C |
| Expiry Date: | 12-14 months |