Datasheet for ABIN577656

Corticosterone ELISA Kit

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Overview

Quantity:	96 tests
Target:	Corticosterone (CORT)
Reactivity:	Various Species
Method Type:	Sandwich ELISA
Minimum Detection Limit:	16.9 pg/mL
Application:	ELISA

Product Details

Purpose:	The DetectX® Corticosterone Immunoassay kit is designed to quantitatively measure Corticosterone present in extracted dried fecal samples, serum, plasma and tissue culture media samples.
Brand:	DetectX®
Sample Type:	Serum, Plasma (EDTA), Plasma (heparin), Urine, Fecal, Tissue Culture Medium
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Specificity:	Sample Types validated: Dried Fecal Extracts, Serum, EDTA and Heparin Plasma and Tissue Culture Media
Sensitivity:	18.6 pg/mL
Characteristics:	The Corticosterone Enzyme Immunoassay kit measures corticosterone present in tissue culture, serum, plasma and fecal samples. The kit has been developed for extracted fecal samples and has been validated with multiple animal species. Diluted samples are pipetted into

	a clear microtiter plate coated with an antibody. A corticosterone-peroxidase conjugate is added to the wells. The binding reaction is initiated by the addition of a polyclonal antibody to corticosterone. After an hour incubation the plate is washed and substrate is added. The substrate reacts with the bound corticosterone-peroxidase conjugate. After a short incubation, the reaction is read at 450nm. Corticosterone is a glucocorticoid secreted by the cortex of the adrenal gland. Corticosterone is produced in response to stimulation of the adrenal cortex by ACTH and is the precursor of aldosterone. Corticosterone is a major indicator of stress and is the major stress steroid produced in non-human mammals. Studies involving corticosterone and levels of stress include impairment of long term memory retrieval, chronic corticosterone elevation due to dietary restrictions and in response to burn injuries. In addition to stress levels, corticosterone is believed to play a decisive role in sleep-wake patterns.
Components:	Coated Clear 96 Well plates A clear plastic microtiter plate(s) coated with donkey anti-sheep IgG. 1 or 5 Each Corticosterone Standard Corticosterone at 100,000 pg/mL in a special stabilizing solution. 125 or 625 µL DetectX® Corticosterone Antibody A sheep polyclonal antibody specific for corticosterone. 3 mL or 13 mL DetectX® Corticosterone Conjugate A corticosterone-peroxidase conjugate in a special stabilizing solution. 3 mL or 13 mL Assay Buffer Concentrate A 5X concentrate that must be diluted with deionized or distilled water. 28 mL or 55 mL Dissociation Reagent 1 mL or 5 mL Dissociation Reagent is to be used only with Serum and plasma samples. Wash Buffer Concentrate A 20X concentrate that should be diluted with deionized or distilled water. 30 mL or 125 mL TMB Substrate 11 mL or 55 mL Stop Solution A 1M solution of hydrochloric acid. CAUSTIC. 5 mL or 25 mL Plate Sealer 1 or 5 Each
Material not included:	Distilled or deionized water. Repeater pipet with disposable tips capable of dispensing 25 µL, 50 µL and 100 µL. Colorimetric 96 well microplate reader capable of reading optical density at 450 nm. Software for converting raw relative optical density readings from the plate reader and carrying out four parameter logistic curve (4PLC) fitting.

Target Details

Target:	Corticosterone (CORT)
Alternative Name:	Corticosterone (CORT Products)
Target Type:	Hormone
Background:	Corticosterone (C H O , Kendall's Compound 'B') is a glucocorticoid secreted by the cortex of 21 30 4 the adrenal gland. Corticosterone is produced in response to stimulation of the adrenal cortex by ACTH and is the precursor of aldosterone. Corticosterone is a major indicator of stress and is the major stress steroid produced in non-human mammals. Studies involving corticosterone and levels of stress include impairment of long term memory retrieval1, chronic corticosterone elevation due to dietary restrictions2 and in response to burn injuries3. In addition to stress levels, corticosterone is believed to play a decisive role in sleep-wake patterns4,5

Application Details

Application Notes:	This assay has been validated for serum, EDTA and heparin plasma, urine samples and for tissue culture samples. It has also been validated for dried fecal extract samples. Samples containing visible particulate should be centrifuged prior to using. Moderate to severely hemolyzed samples should not be used in this kit. Corticosterone can be assayed in other sample types by using one of the extraction protocols available on our website at: www.ArborAssays.com/resources/lit.asp. Corticosterone is identical across all species and we expect this kit may measure corticosterone from sources other than human. The end user should evaluate recoveries of corticosterone in other samples being tested.
Plate:	Pre-coated
Protocol:	This kit measures total corticosterone in serum and plasma and in extracted fecal samples. A corticosterone stock solution is provided to generate a standard curve for the assay and all samples should be read off the standard curve. We provide protocols on page 8 to prepare assay standards from 5,000 to 78.125 pg/mL or from 10,000 to 78.125 pg/mL. Please choose the standard range that fits your sample concentrations most appropriately. Standards or diluted samples are pipetted into a clear microtiter plate coated with an antibody to capture sheep antibodies. A corticosterone-peroxidase conjugate is added to the standards and samples in the wells.

	The binding reaction is initiated by the addition of a polyclonal antibody to corticosterone to each well. After an hour incubation the plate is washed and substrate is added. The substrate reacts with the bound corticosterone-peroxidase conjugate. After a short incubation, the reaction is stopped and the intensity of the generated color is detected in a microtiter plate reader capable of measuring 450nm wavelength. The concentration of the corticosterone in the sample is calculated, after making suitable correction for the dilution of the sample, using software available with most plate readers.
Reagent Preparation:	Allow the kit reagents to come to room temperature for 30 minutes. We recommend that all standards and samples be run in duplicate to allow the end user to accurately determine corticosterone concentrations. Ensure that all samples have reached room temperature and have been diluted as appropriate prior to running them in the kit. assay Buffer Dilute Assay Buffer Concentrate 1:5 by adding one part of the concentrate to four parts of deionized water. Once diluted this is stable at 4 °C for 3 months. Wash Buffer Dilute Wash Buffer Concentrate 1:20 by adding one part of the concentrate to nineteen parts of deionized water. Once diluted this is stable for 3 months at room temperature. Standard Preparation Label test tubes as #1 through #8. Pipet 450 µL of Assay Buffer into tube #1 and 250 µL into tubes #2 to #8. the corticosterone stock solution contains an organic solvent. Prerinse the pipet tip several times to ensure accurate delivery. Carefully add 50 µL of the corticosterone stock solution to tube #1 and vortex completely. Take 250 µL of the corticosterone solution in tube #1 and add it to tube #2 and vortex completely. Repeat the serial dilutions for tubes #3 through #8. The concentration of corticosterone in tubes 1 through 8 will be 10,000, 5,000, 2,500, 1,250, 625, 312.5, 156.25, and 78.125 pg/mL. Use all Standards within 2 hour of preparation. Std 1 Std 2 Std 3 Std 4 Std 5 Std 6 Std 7 Std 8 assay Buffer (µL) 450 250 250 250 250 250 250 250 addition Stock Std 1 Std 2 Std 3 Std 4 Std 5 Std 6 Std 7 Vol of addition (µL) 50 250 250 250 250 250 250 250 Final Conc (pg/ mL) 10,000 5,000 2,500 1,250 625 312.5 156.25 78.125 ® 8 EXPECT ASSAY ARTISTRY
Sample Preparation:	Serum and plasma samples need to be treated with the supplied Dissociation Reagent. Addition of this reagent will yield the total corticosterone concentration in serum or plasma. Dissociation

	reagent is to be used only with Serum and Plasma samples. Serum and Plasma Samples Allow the Dissociation Reagent to warm completely to room Temperature before use. We suggest pipetting 5 µL of Dissociation Reagent into 1 mL Eppendorf tubes. Add 5 µL of serum or plasma to the Dissociation Reagent in the tube, vortex gently and incubate at room temperature for 5 minutes or longer. Dilute with 490 µL of diluted Assay Buffer. This 1:100 dilution can be diluted further with diluted Assay Buffer. Final serum and plasma dilutions should be ≥ 1:100. noTe: Dissociation reagent is to be used only with Serum and Plasma samples. Urine Samples Urine samples should be diluted ≥ 1:20 with the diluted Assay Buffer prior running in the assay. Please see our Urinary Creatinine Detection kits, K002-H1 and K002-H5, for assays to measure urine creatinine which can be used to allow normalization of corticosterone in a random urine specimen. Dried Fecal Samples: We have a detailed Extraction Protocol available on our website at: www.ArborAssays.com/resources/lit.asp. The ethanol concentration in the final diluted Assay Buffer dilution added to the well should be <5 % .
Assay Procedure:	1. Use the plate layout sheet on the back page to aid in proper sample and standard identification. Determine the number of wells to be used and return unused wells to the foil pouch with desiccant. Seal the ziploc plate bag and store at 4ºC. 2. Pipet 50 µL of samples or standards into wells in the plate. 3. Pipet 75 µL of Assay Buffer into the non-specific binding (NSB) wells. 4. Pipet 50 µL of Assay Buffer into wells to act as maximum binding wells (Bo or 0 pg/mL). 5. Add 25 µL of the DetectX® Corticosterone Conjugate to each well using a repeater pipet. 6. Add 25 µL of the DetectX® Corticosterone Antibody to each well, except the NSB wells, using a repeater pipet. 7. Gently tap the sides of the plate to ensure adequate mixing of the reagents. Cover the plate with the plate sealer and shake at room temperature for 1 hour. If the plate is not shaken signals bound will be approximately 45 % lower. 8. Aspirate the plate and wash each well 4 times with 300 µL wash buffer. Tap the plate dry on clean absorbent towels. 9. Add 100 µL of the TMB Substrate to each well, using a repeater pipet. 10. Incubate the plate at room temperature for 30 minutes without shaking. 11. Add 50 µL of the Stop Solution to each well, using a repeater or a multichannel pipet. 12. Read the optical density generated from each well in a plate reader capable of reading at 450 nm. 13. Use the plate reader's built-in 4PLC software capabilities to calculate corticosterone concentration for each sample.
Calculation of Results:	Average the duplicate OD readings for each standard and sample. Create a standard curve by reducing the data using the 4PLC fitting routine on the plate reader, after subtracting the mean OD's for the NSB.

Application Details

	The sample concentrations obtained, calculated from the %B/B0 curve, should be multiplied by the dilution factor to obtain neat sample values.
Restrictions:	For Research Use only

Handling

Precaution of Use:	As with all such products, this kit should only be used by qualified personnel who have had laboratory safety instruction. The complete insert should be read and understood before attempting to use the product. The antibody coated plate needs to be stored desiccated. The silica gel pack included in the foil ziploc bag will keep the plate dry. The silica gel pack will turn from blue to pink if the ziploc has not been closed properly. This kit utilizes a peroxidase-based readout system. Buffers, including other manufacturers Wash Buffers, containing sodium azide will inhibit color production from the enzyme. Make sure all buffers used for samples are azide free. Ensure that any plate washing system is rinsed well with deionized water prior to using the supplied Wash Buffer as prepared on Page 8. The Stop Solution is acid. The solution should not come in contact with skin or eyes. Take appropriate precautions when handling this reagent.
Storage:	4 °C
Storage Comment:	All components of this kit should be stored at 4°C until the expiration date of the kit.

Publications

Product cited in:	Khanna, Patwardhan, Yang, Li, Cai, Ji, Chew, Dorame, Bellampalli, Schmoll, Gordon, Moutal, Vanderah, Porreca, Ibrahim: "Development and Characterization of An Injury-free Model of Functional Pain in Rats by Exposure to Red Light." in: **The journal of pain**, Vol. 20, Issue 11, pp. 1293-1306, (2020) (PubMed). Quinn, McCalla, He, Xu, Cidlowski: "Silencing of maternal hepatic glucocorticoid receptor is essential for normal fetal development in mice." in: **Communications biology**, Vol. 2, pp. 104, (2019) (PubMed). Bauman, Buban, Russell, Handa, Wu: "Isoflavones Alter Hypothalamic-Pituitary-Adrenal Axis

Publications

Response Following Photoperiod Alteration." in: **Neuroscience**, Vol. 406, pp. 268-277, (2019) (PubMed).

Ramírez-Rosas, Velázquez, Verdugo-Díaz, Pérez-Armendáriz, Juárez-Oropeza, Paredes-Carbajal: "Subchronic stress effects on vascular reactivity in C57BL/6 strain mice." in: **Physiology & behavior**, Vol. 204, pp. 283-289, (2019) (PubMed).

Kott, Mooney-Leber, Brummelte: "Developmental outcomes after gestational antidepressant treatment with sertraline and its discontinuation in an animal model of maternal depression." in: **Behavioural brain research**, Vol. 366, pp. 1-12, (2019) (PubMed).

There are more publications referencing this product on: Product page

Images

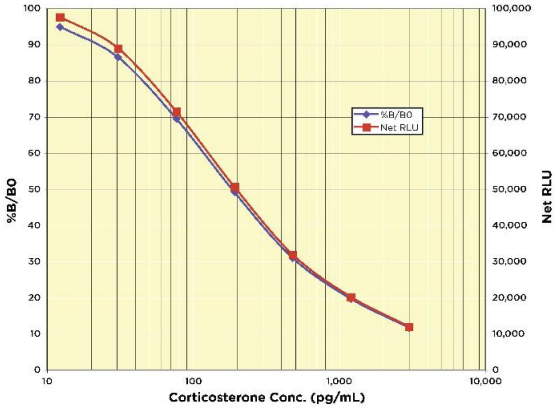

Image 1.

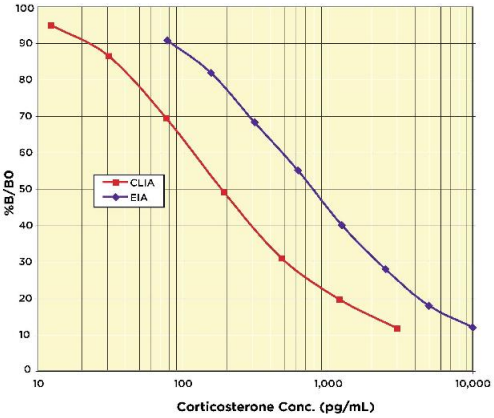

Image 2.

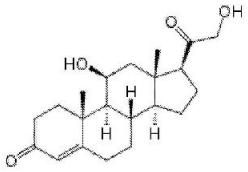

Image 3.

Please check the product details page for more images. Overall 4 images are available for ABIN577656.