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# **CRH ELISA Kit**





## Overview

Quantity:	96 tests
Target:	CRH
Reactivity:	Mouse
Method Type:	Competition ELISA
Detection Range:	12.35 pg/mL - 1000 pg/mL
Minimum Detection Limit:	12.35 pg/mL
Application:	ELISA
Product Details	
Purpose:	The kit is a small sample competitive inhibition enzyme immunoassay technique for the in vitro quantitative measurement in various sample types.
Sample Type:	Plasma, Serum
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Specificity:	This assay has high sensitivity and excellent specificity for detection of Corticotropin Releasing Factor.
Sensitivity:	5.18 pg/mL
Grade:	Small Sample
Components:	<ul> <li>Pre-coated, ready to use 96-well strip plate, flat buttom</li> <li>Plate sealer for 96 wells</li> </ul>

- · Reference Standard
- Standard Diluent
- · Detection Reagent A
- · Detection Reagent B
- · Assay Diluent A
- · Assay Diluent B
- Reagent Diluent (if Detection Reagent is lyophilized)

Corticotropin Releasing Factor (CRH Products)

- · TMB Substrate
- · Stop Solution

CRH

- Wash Buffer (30 x concentrate)
- · Instruction manual

# **Target Details**

Alternative Name:

Target:

Background:	CRH, Corticotropin Releasing Hormone, Corticoliberin
Pathways:	Positive Regulation of Peptide Hormone Secretion, Hormone Activity, Negative Regulation of
	Hormone Secretion, cAMP Metabolic Process, Myometrial Relaxation and Contraction, Feeding
	Behaviour
Application Details	
Sample Volume:	25 μL
Assay Time:	3 h
Plate:	Pre-coated
Protocol:	1. Prepare all reagents, samples and standards,
	2. Add 25µL standard or sample to each well.
	Then add 25µL prepared Detection Reagent A immediately.
	Shake and mix. Incubate 1 hour at 37 °C,
	3. Aspirate and wash 3 times,
	4. Add 50µL prepared Detection Reagent B. Incubate 30 minutes at 37 °C,
	5. Aspirate and wash 5 times,
	6. Add 50µL Substrate Solution. Incubate 10-20 minutes at 37 °C,
	7. Add 25µL 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 is not used up all at once, remove only the strips and reagents for the current experiment and

leave the remaining strips and reagents in the desired condition.

- 2. **Standard** Reconstitute the standard with the Standard Diluent, keep it at room temperature for 10 minutes and shake it gently (do not let it foam). Please prepare tubes with Standard Diluent and make a dilution series. Mix each tube thoroughly before the next transfer. The last tube with Standard Diluent is the blank as 0 mg/mL.
- 3. **Detection Reagent A** and **Detection Reagent B** Spin or centrifuge the stock of Detection Reagent A and B briefly before use. Dilute to working concentration (1:100) with Assay Diluent A or B, respectively.
- 4. **Wash Solution** Dilute 10 mL of Wash Solution Concentrate (30x) with 290 mL of deionized or distilled water to make 300 mL of Wash Solution (1x).
- 5. **TMB Substrate** Aspirate the required amount of solution with sterile tip and do not return the residual solution back into the vial.

#### Note:

- 1. Serial dilution directly in the wells is not recommended.
- 2. Prepare standard within 15 minutes before assay. Do not dissolve the reagents directly at 37 °C.
- 3. Detection Reagent A and B are sticky solutions, so pipette them slowly to reduce volume errors.
- 4. Reconstitute Standard or working solutions of Detection Reagent A and B carefully according to instructions, avoiding foaming and mixing gently until crystals are completely dissolved. To minimize inaccuracy caused by pipetting, use small volumes and ensure pipettes are calibrated. It is recommended to aspirate more than 10 μL for one-time pipetting.
- 5. The reconstituted Standard, Detection Reagent A and B can only be used once.
- 6. When crystals have formed in the Wash Solution concentrate (30x), warm it to room temperature and mix gently until the crystals are completely dissolved.
- 7. Contaminated water or preparation containers affect the detection result.

#### Sample Preparation:

- It is recommended to use fresh samples without long storage, otherwise protein degradation
  and denaturation may occur in these samples, leading to false results. Samples should
  therefore be stored for a short period at 2 8 °C or aliquoted at -20 °C (≤1 month) or -80 °C (≤
  3 months). Repeated freeze-thaw cycles should be avoided. Prior to assay, the frozen
  samples should be slowly thawed and centrifuged to remove precipitates.
- If the sample type is not specified in the instructions, a preliminary test is necessary to determine compatibility with the kit.
- If a lysis buffer is used to prepare tissue homogenates or cell culture supernatant, there is a
  possibility of causing a deviation due to the introduced chemical substance. The
  recommended dilution factor is for reference only.
- Please estimate the concentration of the samples before performing the test. If the values
  are not in the range of the standard curve, the optimal sample dilution for the particular
  experiment has to be determined.

Assay Precision:

Intra-assay Precision (Precision within an assay): 3 samples with low, middle and high level of

target were tested 20 times on one plate, respectively.

Inter-assay Precision (Precision between assays): 3 samples with low, middle and high level of target were tested on 3 different plates, 8 replicates in each plate.

CV(%) = SD/meanX100

Intra-Assay: CV < 10%

Inter-Assay: CV < 12%

Restrictions:

For Research Use only

## Handling

Storage:	4 °C/-20 ° C
Storage.	4 6/-20 6

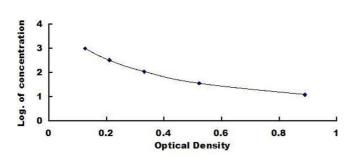
Storage Comment:

- 1. For unopened kit: All reagents should be stored according to the labels on the vials. The Standard, Detection Reagent A, Detection Reagent B, and 96-well Strip Plate should be stored at -20 °C upon receipt, while the other reagents should be stored at 4 °C.
- 2. For opened kits: the remaining reagents must be stored according to the above storage conditions. In addition, please return the unused wells to the foil pouch containing the desiccant and seal the foil pouch with the zipper.

**Expiry Date:** 

6 months

## **Images**



### **ELISA**

Image 1. Typical standard curve