

Human Lys-63-Specific Deubiquitinase BRCC36 (BRCC3) ELISA Kit

Catalogue No.: abx050248

Human Lys-63-Specific Deubiquitinase BRCC36 (BRCC3) ELISA Kit is an ELISA Kit for the in vitro quantitative measurement of Human Lys-63-Specific Deubiquitinase BRCC36 (BRCC3) concentrations in tissue homogenates, cell lysates and other biological fluids. This gene encodes a subunit of the BRCA1-BRCA2-containing complex (BRCC), which is an E3 ubiquitin ligase. This complex plays a role in the DNA damage response, where it is responsible for the stable accumulation of BRCA1 at DNA break sites. The component encoded by this gene can specifically cleave Lys 63-linked polyubiquitin chains, and it regulates the abundance of these polyubiquitin chains in chromatin. The loss of this gene results in abnormal angiogenesis and is associated with syndromic moyamoya, a cerebrovascular angiopathy. Alternative splicing results in multiple transcript variants. A related pseudogene has been identified on chromosome 5.

Target:	Lys-63-Specific Deubiquitinase BRCC36 (BRCC3)
Reactivity:	Human
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 6 months.
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.
UniProt Primary AC:	P46736 (UniProt , ExpASY)
Gene Symbol:	BRCC3
GeneID:	79184
OMIM:	300617
NCBI Accession:	NP_001018065.1, NM_001018055.2, NP_001229569.1, NM_001242640.1, NP_077308.1, NM_024332.3
HGNC:	24185
KEGG:	hsa:79184
Ensembl:	ENSG00000185515

Datasheet

Version: 1.0.0
Revision date: 27 Jul 2023



String: [9606.ENSPO0000358474](#)

Test Range: 0.156 ng/ml - 10 ng/ml

Standard Form: Lyophilized

Detection Method: Colorimetric

Assay Data: Quantitative

Sample Type: Tissue homogenates, cell lysates and other biological fluids.

Assay Principle: This kit is based on sandwich enzyme-linked immuno-sorbent assay technology. An antibody is pre-coated onto a 96-well plate. Standards, test samples, and biotin-conjugated reagent are added to the wells and incubated. The HRP-conjugated reagent is then added, and the whole plate is incubated. Unbound conjugates are removed using wash buffer at each stage. TMB substrate is used to quantify the HRP enzymatic reaction. After TMB substrate is added, only wells that contain sufficient BRCC3 will produce a blue coloured product, which then changes to yellow after adding the acidic stop solution. The intensity of the yellow colour is proportional to the BRCC3 amount bound on the plate. The Optical Density (OD) is measured spectrophotometrically at 450 nm in a microplate reader, from which the concentration of BRCC3 can be calculated.

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 • 37°C incubator

Not Provided:

- 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

Reagent Preparation: This procedure is provided 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.

- 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: This procedure is provided 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.

- 1) Set standard, test samples and control wells.
- 2) Aliquot 100 µl of diluted standard into the standard wells.
- 3) Aliquot 100 µl of Standard Diluent buffer into control (zero) well.
- 4) Aliquot 100 µl of diluted samples into the sample wells. Incubate for 1 hr at 37 °C.
- 5) Aliquot 100 µl of Detection Reagent A to each well. Incubate for 1 hr 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.

Protocol: This procedure is provided 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.

- Equilibrate the kit components and samples to room temperature (18 - 25 °C) before use. It is recommended to plot a standard curve for each test.
- 1. Set standard, test sample and control (zero) wells on the pre-coated plate respectively, and then, record their positions. It is recommended to measure each standard and sample at least in duplicate.
- 2. Add 100 µL of each standard, control and sample into the appropriate wells. Seal the plate with a cover and incubate for 1 h at 37°C.
- 3. Remove the cover and discard the liquid.
- 4. Add 100 µl of the detection Reagent A working solution to each well. Seal the plate with a cover and incubate for 1 h at 37°C.
- 5. Remove the cover and discard the solution. Wash the plate 3 times with 1X Wash Buffer.
- 6. Add 100 µL of Detection Reagent B working solution into each well, seal and incubate at 37°C for 30 min.
- 7. Discard the solution and wash the plate 5 times with wash buffer as explained in previous step.
- 8. Aliquot 90 µl of TMB Substrate into each well. Seal the plate with a cover and incubate at 37°C for 10-20 min. Avoid exposure to light. The incubation time is for reference use only, the optimal time should be determined by end user. Do not exceed 30 min.
- 9. Add 50 µL of Stop Solution to each well. Read at 450 nm immediately.

Results Calculation: For calculation, average the O.D.450 duplicate readings for each reference standard and each sample and subtract the average control (zero) O.D.450 reading. The standard curve can be plotted as the relative O.D.450 of each reference standard solution (Y) vs. the respective concentration of each standard solution (X). The BRCC3 concentration of the samples can be interpolated from the standard curve.

Note: This product is for research use only.
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 ELISA and CLIA 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.

For Reference Only