MitoSciences®

# PARP-1 (cleaved) In-Cell ELISA Kit (IR) (ab110215)

MS Catalog No. MSA43

5 Images URL for this product: http://www.abcam.com/PARP-1-cleaved-In-Cell-ELISA-Kit-IR-ab110215.html

## Product overview

Description

In-Cell ELISA Kits use quantitative immunocytochemistry to measure protein levels or post-translational modifications in cultured cells. Cells are fixed in a 96- or 384-well plate and targets of interest are detected with highly specific, well-characterized monoclonal antibodies, and levels are quantified with IRDyeÂ®-labeled Secondary Antibodies. IR imaging and quantitation is performed using a LI-CORÂ® OdysseyÂ® or AeriusÂ® system.

In-Cell ELISA (ICE) technology is used to perform quantitative immunocytochemistry of cultured cells with a near-infrared fluorescent dye-labeled detector antibody. The technique generates quantitative data with specificity similar to Western blotting, but with much greater quantitative precision and higher throughput due to the greater dynamic range and linearity of direct fluorescence detection and the ability to run 96 samples in parallel. This method rapidly fixes the cells in situ, stabilizing the in vivo levels of proteins, and thus essentially eliminates changes during sample handling, such as preparation of protein extracts. Finally, the cleaved PARP-1 signal can be normalized to cell amount, measured by the provided Janus Green whole cell stain1, to further increase the assay precision.

The assay is designed for use with cultured adherent and suspension cells in a 96-well microplate format. The adherent cells undergoing apoptosis readily detach from a culture plate. The cell detachment often leads to their loss and thus underestimating the proportion of apoptotic cells. This assay was developed to eliminate the loss of apoptotic cells. In addition, this assay is also applicable for suspension cells. Materials and separate protocol sections are provided to achieve efficient attachment of apoptotic adherent cells as well as suspension cells.



Specificity

The PARP-1 (cleaved) In-Cell ELISA Kit (IR) is a highly specific and high-throughput assay for measuring the larger (89 kDa) fragment of poly (ADP-ribose) polymerase 1 (PARP-1) generated by caspase cleavage between Asp214 and Gly215 of human PARP-1. The kit does not recognize full-length PARP-1 or the 24 kDa DNA binding domain fragment.

Tests

2 x 96 well plate

Sample type

Adherent cells, Suspension cells

Tested applications

In-Cell ELISA

Cross reactivity

**Reacts with**

Human

## Properties

Storage instructions

Store at 4Â°C for upto 6 months. Refer to the protocol for further storage instructions.

| Components | 2 x 96 tests |
|---|---|
| 1000X IRDye800-labeled Secondary Antibody | 1 x 24Âµl |
| 100X Triton X-100 | 1 x 0.5ml |
| 10X Blocking Solution | 1 x 15ml |
| 10X Phosphate Buffered Saline | 1 x 100ml |
| 1X Janus Green Stain | 1 x 11ml |
| 400X Tween-20 | 1 x 2ml |
| 500X Primary Antibody | 1 x 42Âµl |

UK office:
Abcam plc
330 Cambridge Science Park Cambridge CB4 0FL UK
Tel: +44 (0)1223 696000
Fax: +44 (0)1223 771600
Email: orders@abcam.com

US office:
Abcam Inc.
1 Kendall Square, Suite B2304 Cambridge, MA 02139-1517 USA
Tel: 617 225 2272
Fax: (617) 849-5829
Email: us.orders@abcam.com

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| | |
|---|---|
| 96-well Assay Plate | 2 x 1unit |
| Plate Seals | 2 x 1unit |

## Applications

| | |
|---|---|
| In-Cell ELISA | In-Cell ELISA: Use at an assay dependent dilution. |

## Images (See the website for higher resolution images of this product)

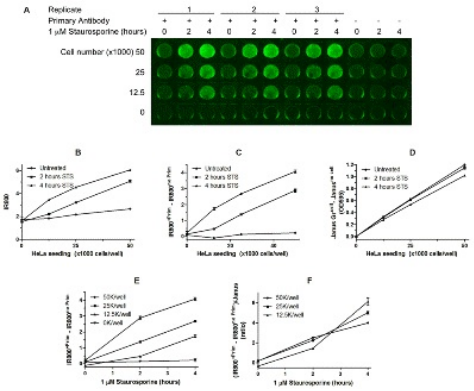

Figure 1. Cell number and treatment time-dependence of PARP-1 cleavage in adherent cells induced to undergo apoptosis.

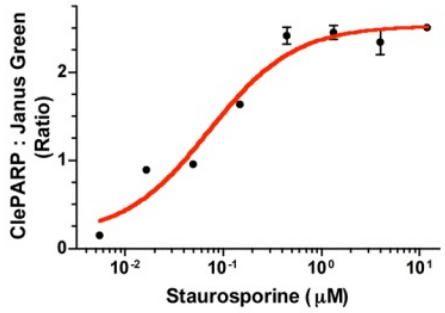


Figure 2. Determination of Staurosporine EC50 of PARP-1 cleavage in HeLa cells. HeLa cells were seeded at 50,000 cells per well, allowed to attach overnight and treated with Staurosporine for 4 hours. Cells were fixed and the plate was analyzed by ICE to measure the cleaved PARP-1 using MSA43. All steps were performed as described in MSA43 Protocol. Mean and standard error of the mean (n=2) is shown. The EC50 of Staurosporine is 0.072 ÂµM (R2=0.954).

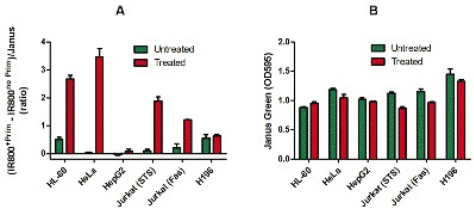


Figure 3. Cell-line dependent induction of PARP-1 cleavage.

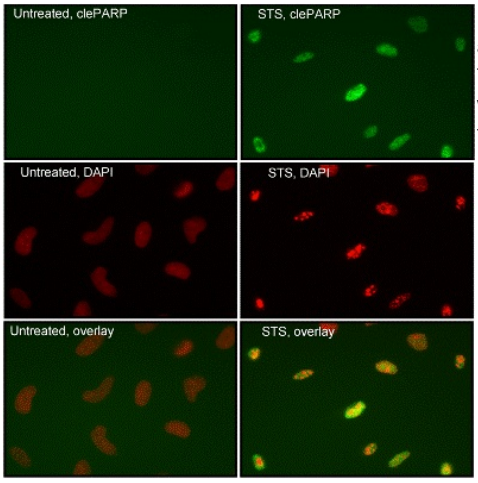


Figure 4. PARP-1 (cleaved) antibody specificity demonstrated by immunocytochemistry. Immunocytochemical analysis of HeLa cells either untreated or treated with 1 ÂµM Staurosporine (STS) for 4 hours to induce apoptosis. The same PARP-1 (cleaved) antibody as in the MSA43 was used in this analysis (in green). Cells well co-stained with DNA stain DAPI (in red). Note that the PARP-1 (cleaved) antibody specifically labels nucleus of only STS-treated cells


UK office:
Abcam plc
330 Cambridge Science Park Cambridge CB4 0FL UK
Tel: +44 (0)1223 696000
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Abcam Inc.
1 Kendall Square, Suite B2304 Cambridge, MA 02139-1517 USA
Tel: 617 225 2272
Fax: (617) 849-5829
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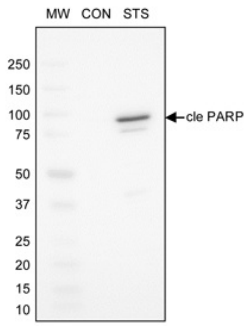


Figure 5. PARP-1 (cleaved) antibody specificity demonstrated by Western Blot. Western blot analysis of HeLa cells either untreated (CON) or treated with 1 ÂµM Staurosporine (STS) for 4 hours to induce apoptosis. The same PARP-1 (cleaved) antibody as in the MSA43 was used in this analysis. Note that the antibody recognize the 89 kDa cleaved PARP-1 and it does not recognize the 113 kDa full length PARP-1

**Our Abpromise to you: Quality guaranteed and expert technical support**

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit http://www.abcam.com/abpromise or contact our technical team.

UK office:
Abcam plc
330 Cambridge Science Park Cambridge CB4 0FL UK
Tel: +44 (0)1223 696000
Fax: +44 (0)1223 771600
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