Total Cellular PARylation: From Inquiry to Insight

LysA™ Universal PARylation Assay Kit (#82123) is a sandwich ELISA-based kit designed to analyze the level of total poly ADP-ribosylation (PARylation) present in cellular extracts. The kit contains all the reagents necessary to measure PARylation levels in cell extracts, including a PAR standard for quantitative measurements.

Applications

The assay is *quantitative* and detects differences in protein PARylation levels resulting, for example, from inducing the DNA damage response, or from exposure to PARP inhibitors and inhibitors of PAR erasers such as PARG (poly (ADP-ribose) glycohydrolase).

Principle of the Assay

A 96-well plate is coated with an anti-PAR antibody recognizing PARylated chains. Lysates from cells are added to the coated wells, and PAR (PARylated proteins) present in the cell lysates are captured by the antibody. This is followed by an incubation with an anti-PAR detection antibody, then a secondary HRP-conjugated antibody. Addition of a chemiluminescent HRP substrate provides a luminescence signal that directly correlates with the amount of PAR present in the cell extracts.

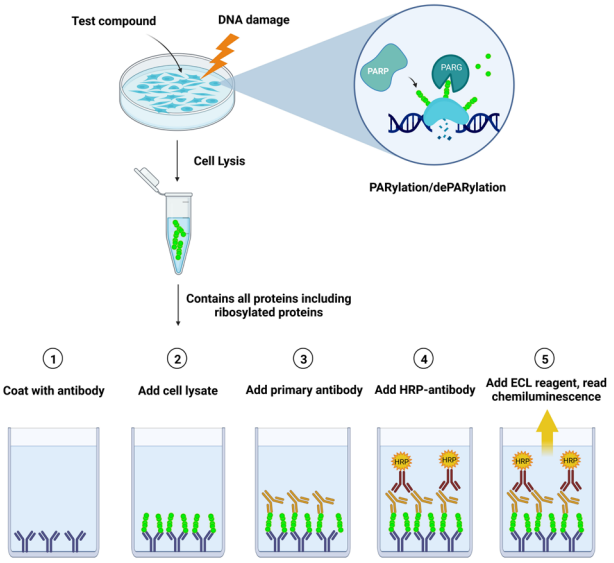
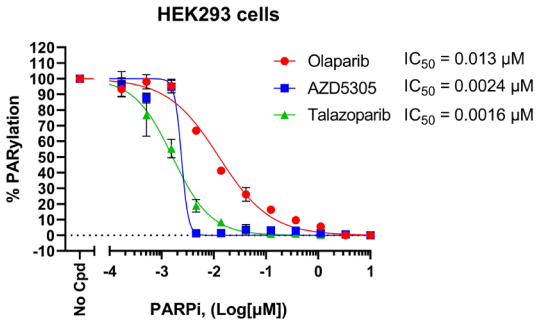
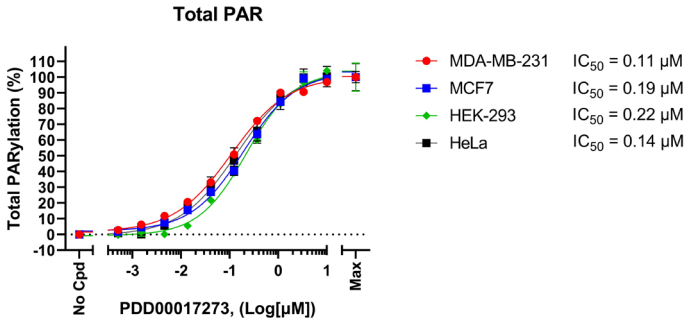


Illustration created with BioRender.com

Advantages

- Measures PARylation from lysates of cells treated with PARP or PARG inhibitors of interest
- Sensitivity ≥100 pM PAR
- Absolute quantification of PAR levels enabled by inclusion of a PAR standard
- Provided with detailed protocols and examples of cellular experiments
- Accessory reagents for cell lysis available, including reagents optimized for this assay kit





Left: *Effect of several PARP inhibitors on PARylation levels in HEK293 cells.* ***Right:*** *Effect of PARG inhibitor PDD00017273 on PARylation levels in various cell lines. Cells were treated with increasing concentrations of inhibitor for 105 minutes and hydrogen peroxide was added for an additional 15 minutes to induce DNA damage. The reaction was stopped and cell extracts were collected. Lysates were analyzed using LysA™ Universal PARylation Assay Kit. Results are expressed as percent of total PARylation in which maximum PARylation is set at 100%.*

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