6044 Cornerstone Court West, Suite E
San Diego, CA 92121
Tel: 1.858.829.3082
Fax: 1.858.481.8694
Email: info@bpsbioscience.com

Data Sheet

Activated DNA (5x)

Catalog # 80605
Lot #: 150707 **Conc.:** 5x

DESCRIPTION: Activated DNA for PARP assay

APPLICATIONS: Study enzyme kinetics, and screen small molecular inhibitors of PARP for drug discovery and HTS applications

SUPPLIED AS: Aqueous buffer solution

QUANTITY: 500 µl

STORAGE: -20°C or -80°C

6044 Cornerstone Court West, Suite E
San Diego, CA 92121
Tel: 1.858.829.3082
Fax: 1.858.481.8694
Email: info@bpsbioscience.com

Assay Protocol:

Materials: Blocking buffer; Streptavidin-HRP, HRP substrate(s); Histone mixture; Activated DNA (BPS catalog number 80605); Assay buffer (BPS catalog number 80600); PARP Substrate (BPS catalog number 80601).

Step 1: Coat 50 µl of histone solution to a 96 well plate (VWR catalog no. 62409-300)

1. Dilute 5x histone mixture 1:5 with PBS.
2. Add 50 µl of histone solution to each well and incubate at 4ºC overnight.
3. Wash the plate three times with PBST buffer (1x PBS containing 0.05% Tween 20).
4. Block the wells by adding 150 µl of Blocking buffer to every well. Incubate at room temperature for 30 min.
5. Wash plate three times with PBST buffer as above.

Step 2: Ribosylation reaction

1. Run the reaction in a total volume of 50 µl containing 100-300 ng of PARP, 2.5 µl of 10x assay mixture, 2.5 µl of 10x PARP buffer, and 5 µl of activated DNA at room temperature for 1 hour.
2. Wash plate three times with PBST buffer as above.

Step 3: Detection

1. Dilute Streptavidin-HRP 1:500 in Blocking buffer.
2. Add 50 µl of diluted Streptavidin-HRP to each well, incubate for 30 min. at room temperature.
3. Wash three times with PBST buffer as above.
4. Just before use, mix on ice 50 □l HRP chemiluminescent substrate A and 50 µl HRP chemiluminescent substrate B and add 100 µl per well.

Step 4: Immediately read sample in a microtiter-plate reading chemiluminescence.