MSD® Phospho-Aurora A (Thr288) Assay Whole Cell Lysate Kit

For quantitative determination in human whole cell lysate samples

Alzheimer's Disease BioProcess Cardiac

Cell Signaling

Clinical Immunology
Cytokines
Hypoxia
Immunogenicity
Inflammation
Metabolic
Oncology
Toxicology
Vascular

Catalog Numbers

Phospho-Aurora A (Thr288) Whole Cell Lysate Kit				
Kit size				
1 plate	K150JCD-1			
5 plates	K150JCD-2			
20 plates	K150JCD-3			

Phospho-Aurora A (Thr288) Whole Cell Lysate Set					
200 μ g	C10JC-1				

Ordering information

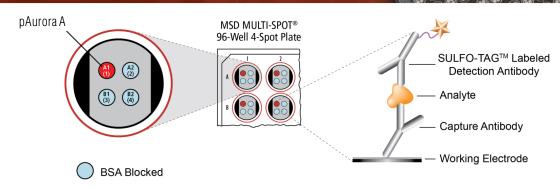
MSD Customer Service Phone: 1-301-947-2085 Fax: 1-301-990-2776 Email: CustomerService@ mesoscale.com

Company Address

MESO SCALE DISCOVERY® A division of Meso Scale Diagnostics, LLC. 9238 Gaither Road Gaithersburg, MD 20877 USA

www.mesoscale.com®

For Research Use Only. Not for use in diagnostic procedures.



Aurora A, also known as serine/threonine-protein kinase 6, is a 72 kDa member of a family of mitotic serine/threonine protein kinases. It is involved in cell cycle progression through mitosis and meiosis and checkpoint regulation. Aurora A is most frequently associated with the centrosomes and the spindle microtubules, and helps regulate centrosome maturation, bipolar spindle formation and maturation, as well as chromosome segregation.¹⁻³

Aurora A's activity is controlled by its phosphorylation state and it is phosphorylated on S51, T288, and S342.⁴ Phosphorylation of S51 seems to regulate destruction of Aurora A, and phosphorylation at T288 appears to be required for Aurora A to be able to transform tissue culture cells and promote tumor formation.¹ A few substrates of Aurora A kinase are p53, MBD3, and BRCA1.⁵ Aurora A is an oncogene and its chromosomal region is often amplified in cancers of the breast, colon, pancreas, ovaries, bladder, liver, and stomach.⁵ There has been much interest in the development of Aurora kinase inhibitors for the treatment of cancer.

The MSD Phospho-Aurora A (Thr288) Assay is available on 96-well 4-Spot plates. This datasheet outlines the performance of the assay.

Typical Data

Representative results for the Phospho-Aurora A (Thr288) Assay are illustrated below. The signal and ratio values provided below are example data; individual results may vary depending upon the samples tested. Western blot analyses of each lysate type were performed with phospho-Aurora A (Thr288) and total Aurora A antibodies and are shown below for comparison. Growing HeLa cells (negative) were treated with nocodazole (0.2 mg/mL; 19 hours) and calyculin A (50 nM for the final 30 minutes) (positive). Whole cell lysates were added to MSD MULTI-SPOT® 4-Spot plates coated with anti-total Aurora A antibody on one of the four spatially distinct electrodes per well. Phosphorylated Aurora A was detected with anti-phospho-Aurora A (Thr288) antibody conjugated with MSD SULFO-TAG™ reagent.

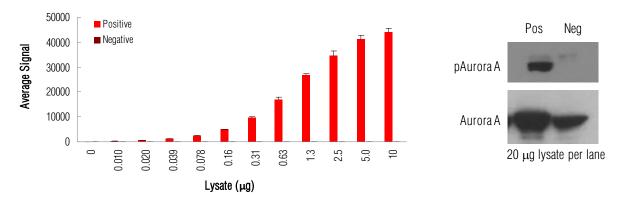


Fig. 1: Sample data generated with MULTI-ARRAY® Phospho-Aurora A (Thr288) Assay. Increased signal is observed with the titration of pAurora A positive cell lysate. The Phospho-Aurora A (Thr288) Assay provides a quantitative measure of the data obtained with the traditional Western blot.





MSD Phosphoprotein Assays

Lysate Titration

Data for positive and negative HeLa cell lysates using the MULTI-ARRAY Phospho-Aurora A (Thr288) Assay are presented below.

Lysate	Positive			Negative			D/N
(μg)	Average Signal	StdDev	%CV	Average Signal	StdDev	%CV	P/N
0	49	6	12.4	38	5	13.4	
0.010	375	15	3.9	49	6	13.1	7.7
0.020	717	45	6.2	52	6	12.0	14
0.039	1347	46	3.4	47	3	5.3	28
0.078	2637	22	0.8	43	2	4.7	61
0.16	4974	144	2.9	56	8	14.0	89
0.31	9734	309	3.2	62	8	12.6	158
0.63	17032	943	5.5	55	7	11.9	310
1.3	26868	643	2.4	58	7	11.8	466
2.5	34701	1742	5.0	49	2	4.4	715
5.0	41318	1370	3.3	74	8	10.8	558
10	43895	1737	4.0	82	4	4.9	537

MSD Advantage

- \blacktriangleright **Multiplexing:** Multiple analytes can be measured in one well using typical sample amounts of 25 μ g/well or less without compromising speed or performance
- Large dynamic range: Linear range of up to five logs enables the measurement of native levels of biomarkers in normal and diseased samples without multiple dilutions
- Minimal background: The stimulation mechanism (electricity) is decoupled from the signal (light)
- > Simple protocols: Only labels near the electrode surface are detected, enabling no-wash assays
- Flexibility: Labels are stable, non-radioactive, and conveniently conjugated to biological molecules
- High sensitivity and precision: Multiple excitation cycles of each label enhance light levels and improve sensitivity

For a complete list of products, please visit our website at <u>www.mesoscale.com</u>

References

- 1. Crane R, Gadea B, Littlepage L, Wu H, Ruderman JV. Aurora A, meiosis and mitosis. Biol Cell. 2004 Apr;96(3):215-29.
- 2. Blagden SP, Glover DM. Polar expeditions--provisioning the centrosome for mitosis. Nat Cell Biol. 2003 Jun;5(6):505-11.
- 3. Goepfert TM, Brinkley BR. The centrosome-associated Aurora/IpI-like kinase family. Curr Top Dev Biol. 2000;49:331-42.
- 4. Littlepage LE, Wu H, Andresson T, Deanehan JK, Amundadottir LT, Ruderman JV. Identification of phosphorylated residues that affect the activity of the mitotic kinase Aurora-A. Proc Natl Acad Sci U S A. 2002 Nov 26;99(24):15440-5. Epub 2002 Nov 6.
- 5. Carvajal RD, Tse A, Schwartz GK. Aurora kinases: new targets for cancer therapy. Clin Cancer Res. 2006 Dec 1;12(23):6869-75.

MESO SCALE DISCOVERY, MESO SCALE DIAGNOSTICS, WWW.MESOSCALE.COM, MSD, MSD (DESIGN), DISCOVERY WORKBENCH, QUICKPLEX, MULTI-ARRAY, MULTI-SPOT, SULFO-TAG, SECTOR, SECTOR HTS, SECTOR PR, 4-SPOT (DESIGN) and SPOT THE DIFFERENCE are trademarks and/or service marks of Meso Scale Diagnostics, LLC. @ 2011 Meso Scale Diagnostics, LLC. All rights reserved.

For Research Use Only. Not for use in diagnostic procedures.

