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\textsuperscript{®} 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\textsuperscript{™} reagent.

![Sample data generated with MULTI-ARRAY\textsuperscript{®} 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.](image-url)
Lysate Titration

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

<table>
<thead>
<tr>
<th>Lysate (µg)</th>
<th>Positive</th>
<th>Negative</th>
<th>P/N</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Average Signal</td>
<td>StdDev</td>
<td>%CV</td>
</tr>
<tr>
<td>0</td>
<td>49</td>
<td>6</td>
<td>12.4</td>
</tr>
<tr>
<td>0.010</td>
<td>375</td>
<td>15</td>
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<tr>
<td>0.020</td>
<td>717</td>
<td>45</td>
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<tr>
<td>0.039</td>
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<td>46</td>
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<tr>
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<tr>
<td>10</td>
<td>43895</td>
<td>1737</td>
<td>4.0</td>
</tr>
</tbody>
</table>

MSD Advantage

- **Multiplexing:** Multiple analytes can be measured in one well using typical sample amounts of 25 µg/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 [www.mesoscale.com](http://www.mesoscale.com)

References


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