Mitotic Assay Kits
determine the proportion of cells undergoing mitosis in a population

Active Motif’s Mitotic Assay Kits provide a simple, accurate method to determine the percentage of cells undergoing mitosis within a given population. When chromosomes condense during mitosis, histone H3 is phosphorylated at serines 10 and 28. These sites, therefore, are reliable markers for detecting cells undergoing mitosis. Each of our kits uses as its mitosis marker a monoclonal antibody (clone HTA28) specific to the phosphorylated serine 28 residue of histone H3. Paclitaxel is included so you can treat your cells to generate a high-mitotic reference population. Each kit also provides a method for normalizing cell number.

Active Motif offers 3 different kits for assaying mitosis, which differ primarily in their readout. The Mitotic Index Assay Kit is a fluorescent method that also includes propidium iodide. This enables you to count your cells and very accurately determine the percentage of cells in a population undergoing mitosis. In contrast, the Mitotic Assay Kits use either colorimetric or chemiluminescent detection to determine the fold induction of mitosis, making them ideal for testing the effect of various treatments on cellular division.

**Fluorescent Mitotic Index Assay Kit Advantages**

- Secondary antibody with Chromeo™ 488 for exceptionally bright detection
- Highly specific monoclonal phospho-Histone H3 (Ser28) clone HTA28
- High-content and high-throughput compatible method, easily adaptable to any plate format
- Propidium iodide included to visualize total cell population
- Whole cell analysis with no cell lysis steps

**Contents & Storage**

5 x 96 rxns each of Chromeo™ 488 Goat anti-Rat IgG, anti-phospho Histone H3 (Ser28), propidium iodide, paclitaxel and RNase A. Store all reagents at -20°C. All reagents are guaranteed stable for 6 months from date of receipt when stored properly.

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<th>Product</th>
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<td>Mitotic Index Assay Kit</td>
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**Figure 1: Mitotic Index Assay results with fluorescent microscopy.** HeLa cells were treated with 1 µM paclitaxel for 6 hours, Phospho-histone H3 (Ser 28) detected with Chromeo 488 Goat anti-Rat IgG (green) in cells co-labeled with propidium iodide (red). Cells with both green and red signals appear yellow.
Mitotic Assay Kits (Colorimetric & Chemi)
determine the proportion of cells undergoing mitosis in a population

The chemiluminescent and colorimetric Mitotic Assay Kits provide a simple method to test the effect of various treatments on cellular division. These kits utilize the same highly specific mitosis marker antibody as the fluorescent Mitotic Index Assay, but offer an alternative colorimetric or chemiluminescent readout, enabling you to choose the assay format that best suits your needs.

Colorimetric & Chemi Mitotic Assay Kit Advantages

- Save time by growing, stimulating and assaying cells directly in 96-well plates
- Cell-based assay eliminates the need for extracts and Western blotting
- Highly specific monoclonal phospho-Histone H3 (Ser28) clone HTA28
- Paclitaxel provided to create high-mitotic reference populations
- Crystal Violet stain included to normalize cell numbers after treatment
- Less than 3 hours of hands-on time

![Figure 1: Fold induction of mitosis in HeLa cells.](image)

HeLa cells were seeded at 20,000 cells per well and assayed using the Colorimetric (A) and Chemiluminescent (B) Mitotic Assay Kits. For a reference mitosis population, cells were treated with 1 µM paclitaxel for 6 hours. Results are the average from eight wells.

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<tr>
<td>Mitotic Assay Kit (Chemi)</td>
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Contents & Storage
96-well plates for culturing cells, primary antibody, HRP-conjugated secondary antibody, Quenching Solution, 1X Antibody Blocking Buffer, 1X Antibody Dilution Buffer, 10X PBS, 10% Triton X-100, 1% SDS Solution, Developing & Stop Solutions and Crystal Violet Cell Quantification Solution. Storage conditions vary from room temperature to -20°C, see manual for details. All reagents are guaranteed stable for 6 months when stored properly.