

Quick Reference Card

Muse® P13K/MAPK Dual Pathway Activation Kit MCH200108

For the investigation of PI3K and MAPK signaling pathways by simultaneous measurement of phosphorylated Akt (pAkt) and phosphorylated ERK (pERK 1/2) in any given cell population For Research Use Only. Not for use in diagnostic procedures.

Storage Conditions

All reagents must be stored at 2-8°C.

Materials Recommended

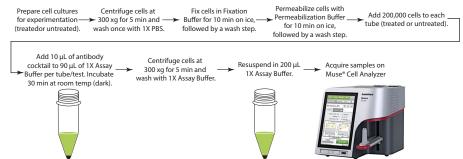
- Guava® Muse® Cell Analyzer
- Test tubes for sample prep and storage
- Tissue culture reagents, i.e., HBSS, PBS w/o Ca²⁺ or Mg²⁺, cell dislodging buffers, etc.
- Micropipettes and tips
- Table top centrifuge
- Mechanical vortex
- Deionized water
- Cells of interest in suspension

 Microcentrifuge tubes with screw caps, 1.5 mL (VWR, Catalogue No. 16466-030, or equivalent)

Kit Components

- 20X Anti-phospho-Akt (Ser473), Alexa Fluor™ 555: (Part No. CS208168), 250 µL
- 20X Anti-phospho-ERK1/2 (Thr202/ Tyr204,Thr185/Tyr187), PECy5: (Part No. CS208167), 250 µL
- 5X Assay Buffer: (Part No. CS202124),
 55 mL
- Fixation Buffer: (Part No. CS202122), 13 mL
- 1X Permeabilization Buffer: (Part No. CS203284), 14 mL

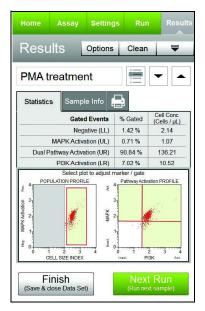
Assay Protocol



Expected Results

Jurkat cells were exposed to 100 ng/mL PMA for 5 minutes to induce a cell signaling cascade response, fixed, permeabilized, and then stained with both antiphospho-ERK 1/2 (Thr202/Tyr204, Thr185/Tyr187) and anti-phospho Akt (Ser473) antibodies in multiplex. Samples were acquired using the Guava® Muse® Cell Analyzer and statistical results are shown.

The statistics captured in this assay show the relative percentages for each population as it is calculated and presented in the dot plot above. Cells which express ERK1/2 (MAPK Activation) can be seen by the data on the top two quadrants of the dot plot, representing about 0.7% and 90.8% of the total cell population, respectively). Alternatively, cells which express Akt (PI3K Activation) can be seen by the data in the two right quadrants of the dot plot, All cells that do not express either Akt or ERK1/2 (e.g., negative) can be calculated and found in the lower left quadrant, 1.4%.



For more information, refer to the comprehensive user's guide for this product, which can be found at: www.luminexcorp.com. (search for Catalogue Code MCH200108)

Related Products

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Muse® EGFR-RTK Activation Dual Detection Kit - MCH200102

Muse® PI3K Activation Dual Detection Kit - MCH200103

Muse® H2A.X Activation Dual Detection Kit - MCH200101

Muse® MAPK Activation Dual Detection Kit - MCH200104

Muse® System Check Kit - MCH100101

Muse® Count & Viability Kit (40mL) - MCH100102

The latest version of Muse® software, which includes all assay modules, as well as the kit user's guide, can be found at www.luminexcorp.com/flowkits.