Antibody Internalization Detection

Standard Operating Procedure (SOP)

Cat. No.	Description
AME100005	DiTag™ Eribulin IgG labeling reagent

1. AME100005 incubation with antibodies

1.1 Dissolve lyophilized AME100005 powder with ddH_2O according to COA.

1.2 Prepare a 4X working solution of the tested antibody in cell culture medium. To begin, a concentration of 1μg/ml is recommended for testing purposes, thus resulting in a 4μg/ml working solution.

1.3 Prepare a 4X working solution of AME100005 in cell culture medium. The tested antibody and AME100005 were mixed at a mass ratio of 9:1 (molar ratio of 2:1).

1.4 Mix equal volumes of the 4X working solutions of the tested antibody and AME100005. Incubate the mixture at 37℃ for 1 hour to obtain the Ab-AME100005 complex at a 2X working solution concentration.

2. Incubation of Ab-AME100005 Complex with Cells

2.1 Collect and wash cells, adjusting the cell concentration using complete culture medium based on the growth rate of cells. A cell concentration of 1×10^5 cells/mL is recommended. Add 50μL of the cell suspension into each well of a 96 well plate.

2.2 Add 50μL of the Ab-AME100005 complex (2X working solution) to each well. For adherent cells, add the complex after cell adhesion. Incubate the plate in a 5% CO_2 incubator at 37℃.

3. Detection of Killing Effect

After incubating for 2-3 days, assess the killing effect of the tested antibody using the CCK8 or CTG method following the manufacturer's protocols.