

# **AME100001 for Detection of antibody**

## **internalization-SOP**

### **1. AME100001 incubation with antibodies**

- 1.1 Dissolve AME100001 in freeze-dried powder form according to COA using deionized water.
- 1.2 Mix the tested antibody with AME100001 in a mass ratio of 9:1 (molar ratio 2:1), and dilute the antibody with AME100001 mixture to twice the tested concentration using complete culture medium.
- 1.3 Incubate at room temperature in dark for 1 hour to obtain Ab-AME100001 complex 2X working solution.

### **2. Ab-AME100001 complex incubation with cells**

- 2.1 Collect and wash cells, adjust cell concentration using complete culture medium.  $1-2 \times 10^5$  cells/mL for suspend cells,  $0.5-1 \times 10^5$  cells/mL for adherent cells. Add 100 $\mu$ L cell suspension to each hole of the 96 well plate.
- 2.2 Add 100 $\mu$ L Ab-AME100001 complex 2X working solution to each hole. Incubate in a 5% CO<sub>2</sub> incubator at 37 °C.

### **3. Flow cytometry detection**

After incubation for 18-24 hours, the internalization effect of the antibody was detected by flow cytometry using FITC or AF488 channels.