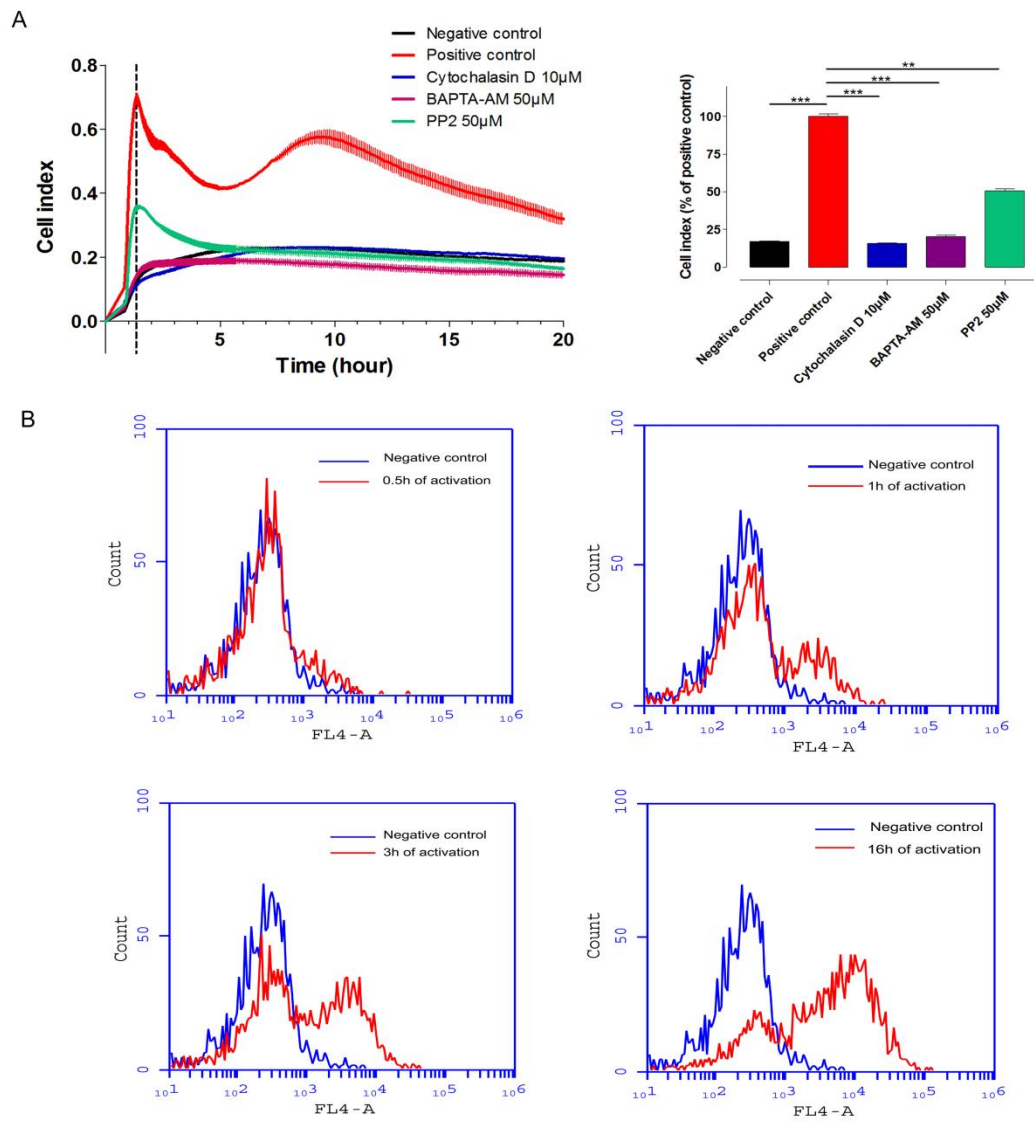


## Supplemental figure legends

**Supplemental Fig. 1.** Dynamic monitoring of PBMCs(peripheral blood mononuclear cells) activation by the xCELLigence system and CD69 expression by FCM. (A) PBMCs separated from healthy donors were preincubated with or without indicated concentrations of cytochalasin D, BAPTA-AM and PP2 prior to seeding onto E-plates in the presence of 1  $\mu\text{g/ml}$  anti-CD3 and anti-CD28 functional antibodies. The resulting CI increase shows that PBMCs were activated by functional antibodies as detected with the xCELLigence system. Cytochalasin D, BAPTA-AM and PP2 could effectively block this response. (B) PBMCs separated from healthy donors were seeded onto 24-well plates in the presence of 1  $\mu\text{g/ml}$  anti-CD3 and anti-CD28 antibodies or 1  $\mu\text{g/ml}$  isotype negative control IgG2a antibody. After incubation for 0.5, 1, 3 or 16 hours, surface expression of CD69 was detected by flow cytometry in the CD4 positive populations. The percentage of CD69 expression increased from 1.75% to 14.79% during the course of incubation.

**Supplemental Fig. 2.**  $Z'$  factor evaluation for the label-free assay.  $Z'$  factor, calculated based on the cell index values obtained in the validation process at the time point indicated by the dotted line. A  $Z'$  factor of 0.56 indicates that the assay system employed meets the technical requirements for high-throughput screening.

Supplemental Fig. 1



Supplemental Fig. 2

