Caspase activity assays

PeerJ

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1 Works for me

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EXTERNAL LINK

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THIS PROTOCOL ACCOMPANIES THE FOLLOWING PUBLICATION

Kusunoki M, Hayashi M, Shoji T, Uba T, Tanaka H, Sumi C, Matsuo Y, Hirota K, Propofol inhibits stromatoxin-1-sensitive voltage-dependent K channels in pancreatic β-cells and enhances insulin secretion. PeerJ doi: 10.7717/peerj.8157

BEFORE STARTING

The levels of caspase 3/7 activity were assessed using an Apo-ONE® Homogeneous Caspase-3/7 Assay Kit (Promega), according to the manufacturer's protocols.

1 Cells were seeded into 96-well plates and incubated overnight.

2 The following day, cells were treated with reagents for varying lengths of time.

3 100 µl of Apo-ONE® Caspase-3/7 Reagent was added to each well.

4 Cells were incubated at room temperature for 1 h.

5 The luminescence of each well was measured using an EnSpire® Multimode Plate Reader (PerkinElmer, Waltham, MA, USA).

6 Caspase activity was then calculated by comparing the levels of luminescence of the treated cells with those of the control cells (incubated without drugs), with the latter defined as 100%.

Assays were performed in triplicate at least twice. Data were expressed as mean ± standard deviation (SD).