

WST-8 Cell Proliferation Assay Kit (treatment)

Introduction

<https://www.dojindo.com/EUROPE/products/CK04/>

Materials

- › Cell counting Kit-8 #CK04-11
- › 96well Plates
- › Culture Medium
- › Plate Reader
- › SMPD1 inhibitor ARC39 (S1078, Selleckchem)
- › KrasG12D inhibitor MRTX1133 (E1051, Selleckchem)

Procedure

Assay Protocol

1. Seed cells in a 96-well plate at a density of 5×10^3 cells/well in 50 μ l of culture medium.
2. After an overnight incubation, 50 μ l of the respective medium containing increasing concentrations of the SMPD1 inhibitor ARC39 (diluted in PBS) and/or KrasG12D inhibitor MRTX1133 (diluted in 1% DMSO) were added to the cells. (Vehicles for MRTX1133 treatment were subjected to 1% DMSO)
3. After 24, 48, and 72 hours (for ARC39 treatment of KPC1050) or only 72 hours (for all other treatments), 10 μ l of WST-8 solution was added, and plates were incubated for 2 h at 37°C with 5% CO₂.
4. During the incubation time, WST-8 is reduced extracellularly in a process requiring an intermediate electron carrier in viable cells and is converted into a water-soluble formazan product.
5. The final formazan product was quantified by measuring the OD_{450nm} using SpectraMax® Plus 384 Microplate Reader.
6. The measured OD_{450nm} values were considered to be proportional to the number of viable cells.
7. For statistical analysis, OD_{450nm} measurements were blank-corrected and normalized to control at 72 h.