RESULTS

0.85 Synergism

Z-138 – Z-138

P<0.3 Strong Synergism

In ER-positive endometrial cancer cells, combination of ARQ 751 with ER antagonists (anastrozole, fulvestrant) 

2 Strong Antagonism

MAVER-1
Jeko-1

values of 0.54, 0.79, and 1.3 nM,

30 ≤1.2; and Antagonistic: CI>1.2.

the Chou-Talalay method, with the following cut-offs applied:

30

2.25 + 0.15) or 0.5% methyl cellulose 400 cP . Anti-PD-1 antibody

SelleckChem. ARQ 751 was prepared in 0.01 M phosphoric acid (pH 7.4). ARQ 531 was prepared in 0.01 M carbonate buffer (pH 9.0) and stored at 4°C.

ARQ 751 and ARQ 531 were synthesized at ArQule, Inc.

Cell culture

Cells were seeded at an optimal number per well in 130 μL of full media in a 96-well plate. All cell lines were grown at 37°C in a humidified atmosphere of 5% CO2.

Reagents

All reagents for in vitro experiments were purchased from Sigma-Aldrich.

MTS Proliferation Assay

Proteins were extracted and resolved from extracts using SDS-PAGE according to manufacturer’s recommendations.

Western Blot Analysis

Reagents

Materials and Methods

MATERIALS AND METHODS

Synergistic effect was observed in 1 CLL and 3 MCL cell lines and additive effects were observed in the other 2 MCL cell lines. The combination study of ARQ 751 with anti-PD-1 or ibrutinib were performed in 1 T319fs*1&V290fs*1 cells.

ARQ 751 and ARQ 531 were synthesized at ArQule, Inc.

ARQ 751 was used at 20 nM and ARQ 531 was used at 0.1 μM for 48 h. Anastrozole was used at 3 μM for 48 h.

Combination of ARQ 751 with ARQ 531, a BTK Inhibitor, Is Superior to the Single Agents

The combination studies of ARQ 751 with anastrozole or fulvestrant were performed in the endometrial cancer cell line Ishikawa.

Combination of ARQ 751 with Ibrutinib

Combination of ARQ 751 with Ibrutinib

Combination of ARQ 751 with ibrutinib was additive in all cell lines except for U266.

Combination of ARQ 751 with ARQ 531, a BTK Inhibitor, Is Superior to the Single Agents

The combination studies of ARQ 751 with anti-PD-1 antibody on syngeneic mouse tumor model

ARQ 751 treatment significantly suppressed T cells and M1 macrophages, resulting in enhanced anti-proliferative activity and increased apoptotic response in vivo.

The combination studies of ARQ 751 with anti-PD-1 antibody on synergistic mouse tumor model

The combination of ARQ 751 with an immune checkpoint inhibitor (anti-PD-1), estrogen receptor (ER) antagonist (anastrozole, fulvestrant), BTK inhibitor (ibrutinib), or AKT inhibitor (ARQ 531) is superior to the single agents.

CONCLUSIONS

Combination of ARQ 751 with anti-PD-1 antibody enhanced anti-proliferative activity in a syngeneic mouse tumor model.

In ER-positive endometrial cancer cells, combination of ARQ 751 with ER antagonists (anastrozole, fulvestrant) acts synergistically with anti-proliferative activity of ARQ 751 and increased apoptotic response.

ARQ 531 enhances anti-proliferative activity of ARQ 751 in prostate cancer cells, accompanied with inhibition of AKT pathway and increased apoptotic response.

Combination of ARQ 751 with the BTK inhibitor, ARQ 531, shows a superior anti-proliferative effect in comparison to single agents in vitro and in prostate xenografts.

A phase I clinical study of ARQ 751 in a molecularly defined patient population is ongoing (NC19276164).