EP-100 sensitizes BRCA wild-type ovarian cancer cells to PARP inhibitor

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Abstract

Objective: EP-100 is a synthetic lyte peptide specific for targeting LHHRH receptors on cancer cells that is being clinically tested for treatment of ovarian cancer. In this study, we aimed to identify combination approaches with EP-100 in ovarian cancer models.

Method: We carried out a series of in vitro (MTT assay, immunoblot analysis, PCR, reverse phase protein array [RPPA], comet assay and immunofluorescence staining) and in vivo (orthotopic mouse model) experiments in ovarian cancer models to determine the biological effects of EP-100 alone and in combination with standard of care drugs.

Results: We first examined the cytotoxic effects of EP-100 alone on 8 ovarian cancer cell lines with variable expression levels of LHHRH receptors and the IC50 values ranged from 0.5 to 3.0 µM. We then tested the effect of combining EP-100 with standard drugs (e.g., paclitaxel, topotecan, luteinizing hormone, and olaparib) on ovarian cancer cells. Of those drugs, we found that combination of EP-100 and olaparib was highly synergistic in drug sensitive and multi-drug resistant ovarian cancer cell lines (HeyA8, HeyA8-MDR, A2780, and A2780CP) and inhibited proliferation in the xenograft mouse model. Furthermore, the nuclear factor of xHAR was significantly increased in combination group of EP-100 and olaparib after 24 h incubation compared with control, EP-100 only, or olaparib alone groups (p < 0.001, ANOVA). In addition, there was increased DNA accumulation in tails using comet assay after treating cells with EP-100 and olaparib for 24 h (p = 0.001, ANOVA). RPPA results identified that PI3K/AKT pathway can be blocked significantly by the combination treatment. The study in HeyA8 xenograft mouse models showed that mice treated with EP-100 and olaparib had the lowest tumor weight (0.48 ± 0.05 g) compared with those treated with vehicle (1.19 ± 0.3 g, p = 0.008), EP-100 alone (0.62 ± 0.28 g, p = 0.006), and olaparib alone (0.50 ± 0.22 g, p < 0.01).

Conclusion: EP-100 has efficacy in preclinical models of ovarian cancer, especially in combination with olaparib. Our findings suggest that combining EP-100 with a PARP inhibitor such as olaparib may be a promising therapeutic strategy for ovarian cancer.

Background

EP-100 is a fusion peptide consisting of the LHHRH ligand and a lyte peptide to specifically target cancer cells that express LHHRH.

LHHRH is a hormonally targeted cancer therapy which is overexpressed in many human tumors whereas it is not expressed or expressed at very low levels in adjacent normal tissues.

Objective

- To evaluate the therapeutic effects of EP-100 on ovarian cancer cells using in vitro studies and orthotopic mouse xenograft models of ovarian cancer.
- To extend the utility of EP-100, identify effective combination therapies with EP-100 in ovarian cancer and explore the potential mechanisms of this combination in ovarian cancer models.

Methods

- Detect cell viability in the presence of EP-100 using MTT assay.
- Western blot analysis to determine protein expression
- RPPA assay to identify the downstream mechanisms of the combination treatment
- Comet assay to detect the DNA damage status
- Immunofluorescence staining to characterize the localization of target protein
- LHHRH-A was depleted by RNAi plasmid transfection

Results

EP-100 sensitizes ovarian cancer cells to the treatment of PARP inhibitor, which can be abrogated upon downregulation of LHHRH-A. The combination is highly synergistic with CI values of < 0.9.

Conclusions

EP-100 can sensitize ovarian cancer cells to treatment with PARP inhibitor and in vivo through enhancing DNA damage accumulation and suppressing PI3K/AKT pathway and inhibition of BRCA1.

References


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FIG. 1: EP-100 sensitizes ovarian cancer cells to PARP inhibitor. EP-100 and olaparib synergize to suppress tumor growth in a BRCA1/2 wild-type ovarian cancer xenograft model. Error bars represent SD.