Design of active targeted PLGA-SO_3 nanoparticles encapsulating PARP and PD-L1 inhibitors for BRCA-mutated breast cancer therapy

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Breast cancer (BC) is the most frequently diagnosed cancer and the second most common cause of cancer mortality in women worldwide. Approximately 15% of all BC are triple negative (TNBC), among them, 30% are BRCA1- or BRCA2-mutated. These tumors are highly aggressive and invasive. Recently, inhibition of poly(ADP-ribose)polymerase-1 (PARP1), a DNA repair enzyme, was shown to induce “synthetic lethality” in BRCA-mutated cancer cells prolonging PFS (Progression Free Survival) . This led to the FDA approval of PARP inhibitors (PARPi) for the treatment of BRCA-mutated BC. Despite their promise, resistance mechanisms to PARPi often develop affecting drug availability, (de)PARylation enzymes, restoration of Homologous Recombination (HR) or restoration of replication fork stability. Moreover, PARPi have been shown to have an impact on cancer-associated immunity, and their combination with immune checkpoint therapy (ICT) has been explored in clinical trials. Therefore, we aimed to rationally-design a nanomedicine combining PARPi with a programmed death-ligand 1 (PD-L1) inhibitor, a small molecule immunosuppressive checkpoint ligand inhibitor developed in our lab. We postulated that co-delivery of these therapeutic agents would result in enhanced therapeutic efficacy in BRCA-mutated cancers. First, we assessed the anti-proliferative effect of several PARPi and selected Talazoparib, as it was shown to be the most potent**.** In addition, as a side effect, we observed increased expression of PD-L1 following treatments with Talazoparib on EMT6 murine BRCA-mutated BC cell line. This was subsequently abrogated following treatment with our PD-L1i small molecule. Furthermore, our PD-L1i small molecule limited EMT6 spheroids proliferation, migration and sprouting when grown in co-culture with activated splenocytes.

Additionally, since the enhanced permeability and retention (EPR) effect varies between tumor types, we synthesized one non-targeted and three targeted NPs with sulfonated moieties that actively target P-Selectin, an adhesion molecule expressed in our 3D EMT6 spheroid models. These nanocarriers were characterized (Figure 1) and evaluated for their biocompatibility and capability to internalize 3D cell culture models to determine the best candidate. The selected candidate demonstrated optimized targeting and tumor internalization features, while retaining their anti-tumor cytotoxic activity. Here, we focus on the rational design and physico-chemico-biological characterization of precision nanomedicines combining PARPi with immunotherapy to treat this aggressive BC. Based on our previous experience with several drug combinations, we hypothesize that the proposed nanocarriers will increase the half-life of the encapsulated drugs, selectively target them to release the active compounds at the tumor site while reducing their side effects following the proposed mechanism (Figure 2). We plan to exploit this nano-based therapy in other BRCA-mutated cancer types such us pancreatic and ovarian.

References:

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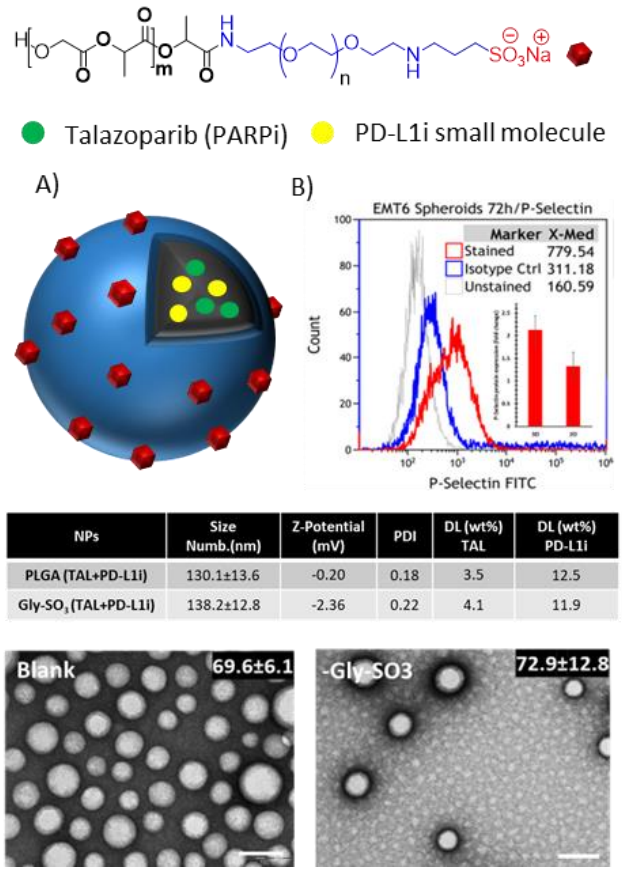

NPs	Size Numb.(nm)	Z-Potential (mV)	PDI	DL (wt%) TAL	DL (wt%) PD-L1i
PLGA (TAL+PD-L1i)	130.1±13.6	-0.20	0.18	3.5	12.5
Gly-SO_3 (TAL+PD-L1i)	138.2±12.8	-2.36	0.22	4.1	11.9

Figure 1. PLGA-PEG-SO_3 NPs. A) Rational design of P PLGA-PEG-SO_3 NPs. B) P-Selectin protein expression in EMT6 cellular models. C) PLGA-PEG-SO_3 NPs physico-chemical characterization: Hydrodynamic radius (Size number); Z-Potential; Polydispersity Index (PDI); Drug Loading (DL); Transmission Electron Microscopy (TEM) Imaging.

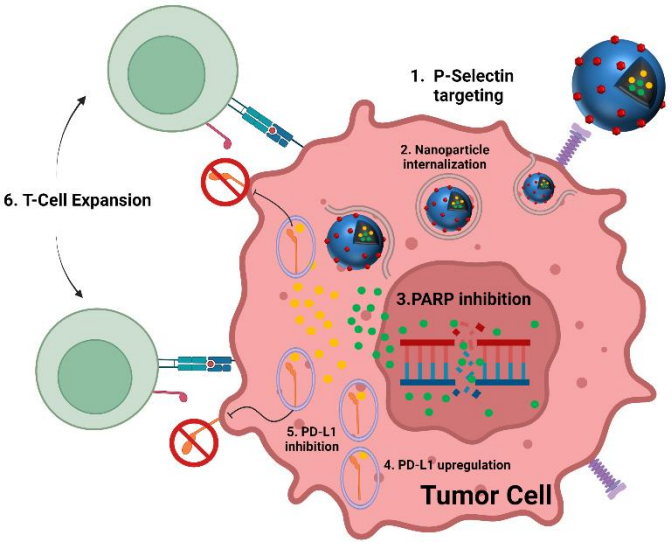

Figure 2. Mechanism of action of PLGA-PEG-SO_3 NPs encapsulating PARP and PD-L1i