The antitumoral activity of TLR7 ligands is corrupted by the microenvironment of pancreatic tumors

AUTHORS

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Purpose

Toll like receptors (TLRs) are key players in the innate immune system, and recent studies have suggested that TLRs may impact pancreatic adenocarcinoma (PDAC), a disease with no cure. We aim to better understand the mechanism of TLRs in PDAC biology, to open door for clinical applications.

Experimental Design:

We used novel TLR2 (CL419), TLR7 (CL347) TLR2+TLR7 (CL553) ligands that form positively charged liposomes and proved highly efficient for the inhibition of experimental melanoma. Experiments were performed in murine and human pancreatic cancer cell lines. Cell transfection efficacy was measured by FACS. Cell proliferation was measured by MTS, cell counting, and non-invasively using the Incucyte Zoom. In vivo studies were performed in NSG and syngenic mouse models of PDAC. Transcriptomic studies were performed using Nanostring.

Results:

We found that TLRs expression is heterogeneous in pancreatic cancer cells. In vitro, CL347 strongly inhibits the proliferation of murine and human pancreatic cancer cells in the micromolar range, as compared to CL419 and CL553. CL347 induces cell death by apoptosis but fails to eradicate cell populations. Sublethal doses of ligands transfected PDAC cell lines with low efficacy, failed to induce IFN response nor to sensitize tumor cells to chemotherapy following therapeutic genes delivery. In vivo, CL347 treatment significantly delayed the growth of very aggressive experimental murine tumors with 80% of NSG mice surviving, while placebo-treated mice died within few days. In vivo, TLR7 ligands strongly induced pro-inflammatory genes and antitumoral cytokines. Surprisingly, CL347 antitumoral effect was annihilated in syngenic models, as mice receiving CL347 developed massive tumors and ascites and died prematurely as compared to control animals.
Transcriptomic signatures advocate for the presence of tumor-promoting macrophages following CL347 treatment.

**Conclusion:**
Taken together, we demonstrate for the first time that TLR ligands have great potential to inhibit PDAC cell proliferation and growth in vivo. However, TLR7 ligands may also recruit tumor-promoting macrophages that drive pancreatic fibrogenesis and tumorigenesis. Thus, TLR7 agonists-based therapies must be carefully considered for PDAC management and should be further combined with agents that antagonize the accumulation of alternatively activated macrophages at these lesions.

**References:** (Times New Roman, font11)