Imprime PGG, a soluble yeast β-glucan PAMP, synergizes with anti-PD-1 antibody to enhance CD8 T cell anti-tumor immunity


Background

Immune checkpoint inhibitor (CPI) therapy has provided compelling, durable anti-tumor T cell immunity for some cancer patients, particularly those with pre-existing anti-tumor T cell responses. Unfortunately, the majority of patients show little to no pre-existing anti-cancer T cell response and do not benefit from single agent CPI therapy. For these patients, novel therapeutic approaches that initiate or reactivate the anti-cancer T cell responses may be particularly promising as combination agents with CPIs as these would provide the activated T cell population upon which CPIs would act.

Propagated activation of the innate immune system is essential to generate robust immune responses. Central to this process are non-self danger signals known as Pathogen-Associated Molecular Patterns (PAMPs) binding to and signaling via pattern receptor proteins (PRRs) expressed by innate immune effector cells (macrophages, monocytes, dendritic cells, neutrophils). Imprime PGG is a soluble yeast β-1,3/1,6 glucan PAMP currently in multiple clinical trials in combination with pembrolizumab (anti-PD-1). Unlike other PAMPs that must be locally administered to avoid systemic cytokine storms, Imprime has been administered safely by intravenous infusion to >400 human subjects (29 cancer patients). Mechanistically, here we show that Imprime activates innate immune cells by binding the PRR dectin-1, eliciting dendritic cell and monocyte expression of co-stimulatory ligands, recruiting monocyte-derived cells, as well as production of a variety of chemokines and cytokines, including type I interferon. To examine Imprime’s innate modulating effects on CD8 T cell priming, we used an ovalbumin (OVA)-specific OT1 CD8 T cell system. Following OT1 adoptive transfer into congenic C57BL6 recipients, mice were immunized with OVA peptide and Imprime. Co-administration of Imprime increased OT1 expansion compared to peptide alone and induced their differentiation into polyfunctional Tbet+ effector cells. Based on Imprime’s ability to link innate and adaptive immune responses, we asked if Imprime would enhance the anti-tumor efficacy of anti-PD-1 antibody therapy. C57BL6 mice were injected s.c. with the murine adenocarcinoma cell line MC38. Once tumors reached ~20 mm3, mice were randomized to treatment groups: vehicle, Imprime (1.2mg twice weekly), anti-PD-1 antibody (RMP1-14 clone, 100µg twice weekly), or Imprime + anti-PD-1 antibody. While anti-PD-1 treatment alone significantly reduced tumor growth compared to vehicle or Imprime groups, combination treatment provided superior tumor control with a statistically higher frequency of mice completely clearing tumor in the combination group compared to the anti-PD-1 group (4/5 vs 1/5, p < 0.05). As to the immunobiology elicited by treatments, tumors were harvested after two weeks of therapy, and tumors infiltrating leukocytes were stimulated with PMA/ionomycin or tumor-specific peptides. Relative to other treatment groups, CD8 TIL from combination-treated mice had superior frequencies of cells capable of producing IFN-γ, TNFα, or IL-2. Furthermore, mice from these MC38 studies that had previously cleared their tumors were protected against tumor re-challenge in the opposing flank, demonstrating that these mice had durable and protective adaptive immunity. Together, our data demonstrate that Imprime PGG is a unique, clinically relevant PAMP that can be administered systemically to activate key innate immune cell populations and to drive T cell based anti-cancer responses. Consequently, Imprime PGG synergizes with anti-PD-1 antibody treatment to provide anti-tumor CD8 T cells with superior effector functions. We are currently working to extend these mechanistic findings in our ongoing phase III clinical trials.

A general structure of yeast-derived Imprime PGG

Imprime induces a Dectin-1-dependent pro-inflammatory profile

Conclusions

• Imprime binds to a wide variety of professional antigen-presenting cells (APCs).
• Imprime maturation of APCs via Dectin-1, initiating production of localized pro-inflammatory cytokines
• Imprime enhances CD8 T cell expansion and their capacity to produce effector cytokines.
• Imprime synergizes with anti-PD-1 antibody therapy in the murine MC38 tumor model. Combination therapy results in increased clearance of established tumors, increased granulocyte B+C CD8 TIL, and improved TNF-α/IL-2 production by CD8 TIL.
• These data provide rationale for Imprime and checkpoint inhibitor therapy which is currently being tested in clinical trials.

Figure 1. Imprime binds to and activates myeloid lineage cells. Naïve C57BL6 mice were injected with 1.2mg Imprime i.v. and various myeloid populations in skin-draining LNs (sLdNs) were examined 24hrs later for Imprime binding. (A) Identification of DC subsets. Ly6Chi monocytes and neutrophils were first gated out and then the bulk DC population was further subset based on expression of CD103 (CD11c+dir, CCR7−/−, PD1+) (B) Imprime binding profiles 24hrs post injection in the sLdNs and (C) expression levels of CD68, CD40, and PD-L1. (D) Imprime causes Dectin-1-dependent mobilization of Ly6C+ monocytes in sLdNs (E) and upregulation of activation and co-stimulatory molecules.

Figure 2. Imprime induces Dectin-1-dependent production of pro-inflammatory cytokines and chemokines. (A) Naïve WT or Dectin-1−/- C57BL6 mice were injected with PBS or 1.2mg Imprime i.v. and 17hrs later skin-draining lymph nodes were harvested and analyzed for transcriptional changes using the custom QuantGene PLEX. Identified genes were categorized into Activation-associated, Cytokines, Chemokines, Signaling and IFN-stimulated genes. IFNAR1−/- mice were used to confirm the dependence of ISG on type I IFN. (B) Naïve C57BL6 mice were injected with PBS or 1.2mg Imprime i.v. Skin-draining LNs were harvested 24hrs later and cell lysates were analyzed for cytokine and chemokine proteins using the Lumienx platform.