A systemically delivered, clinically-ready, attenuated Salmonella Typhimurium strain ACTM-838 alters the tumor microenvironment and provides durable anti-tumor activity in vivo

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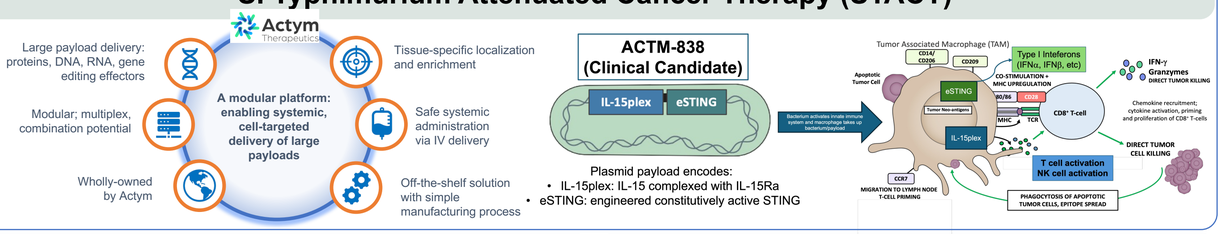
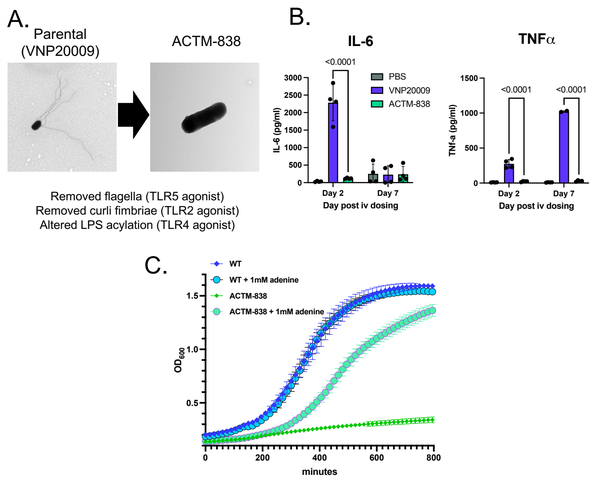
Actym's Systemic Gene Therapy Delivery Platform: S. Typhimurium Attenuated Cancer Therapy (STACT)

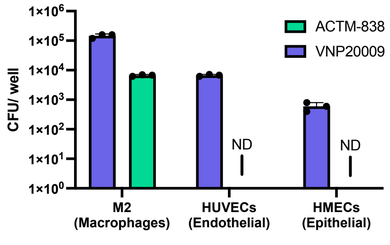


1. Strain engineering generates bacterial chassis dependent on adenosine for growth while reducing systemic inflammation



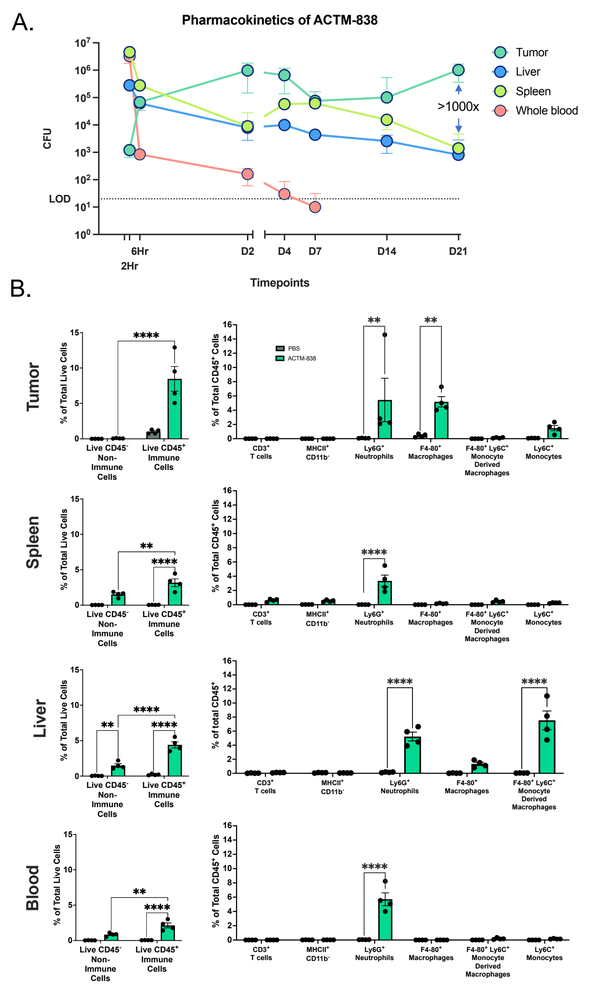
A. Modifications to the parental strain VNP20009 (a clinically tested Salmonella strain) to create ACTM-838 include removal of flagella, curli fimbriae, and LPS acylation modifications resulting in an attenuated strain without visible flagella by microscopy.
B. These modifications to create ACTM 838 result in reduced systemic inflammatory response in vivo. Data not shown: Additional cytokines analyzed, IP-10, KC, MCP-1, exhibited kinetics similar to IL-6 whereas IL-10 exhibited similar kinetics to TNFα.
C. Modifications to the purM gene result in purine auxotrophic ACTM-838, such that bacteria only proliferate in the presence of purines (which are enriched in the TME).

2. Strain engineering of the bacterial chassis increases drug safety by restricting internalization to phagocytic cells



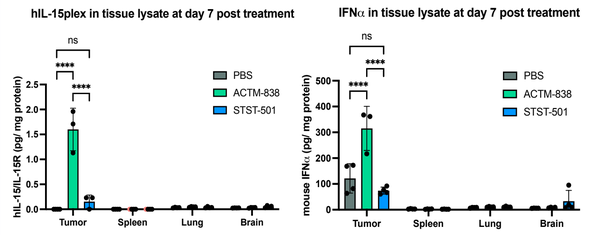
In contrast to VNP20009 (parental strain), in vitro internalization of ACTM-838 is restricted to phagocytic macrophages and not epithelial and endothelial cells.

3. As a result of chassis modifications, I.V. administered ACTM-838 localizes/enriches in the tumor and is internalized only by phagocytic cells



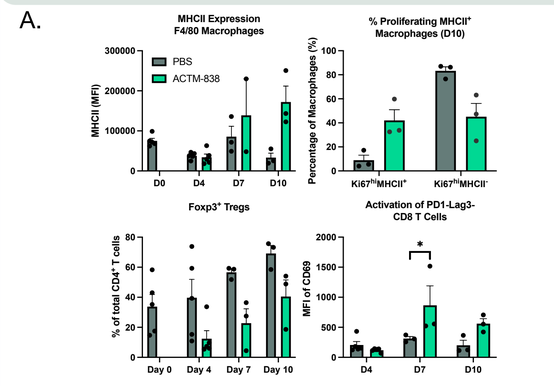
A. ACTM-838 administered in vivo enriches in tumor in orthotopic EMT6 tumor model.
B. Phagocytic cells of the tumor, spleen, liver, and blood analyzed by intracellular staining demonstrate internalization of ACTM-838.

4. Delivery of IL-15plex and eSTING payload by ACTM-838 is observed in tumor tissue and not in normal tissues



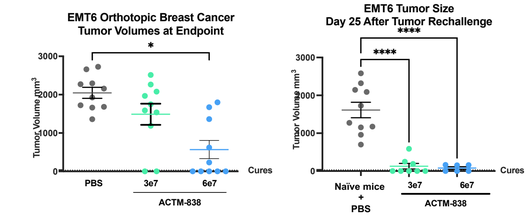
IL-15 (payload, left) and IFNα (cytokine downstream of eSTING payload, right) are detected in tumors (ACTM-838 colonized) and not in normal tissues in an in vivo EMT6 model. Tissues were collected at day 7 post-treatment and payload was measured by ELISA.

5. ACTM-838 chassis and encoded IL-15plex and eSTING reprogram the TME to enable an anti-tumor immune response



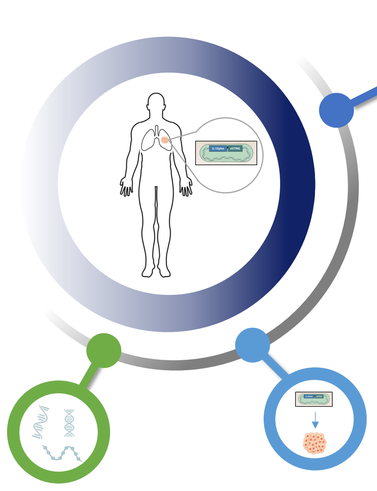
A. Immune cell profiling by flow cytometry in EMT6 tumors reveals increased macrophage MHCII expression, increased proliferating antigen-presenting macrophages, decreased Tregs, and increased activation of PD1-Lag3- CD8 T cells.
B. RNAseq analysis indicates that ACTM-838 induces broad changes within the TME, increasing lymphoid infiltration, myeloid activation, antigen presentation, and stromal remodeling.

6. ACTM-838-mediated changes in the TME lead to in vivo efficacy, durable anti-tumor immunity, and an additive effect in combo with anti-PD-1



A. EMT6 tumor-burdened (orthotopic) BALB/c mice treated with ACTM-838 experience tumor control (left panel). Memory response is observed when cured mice are rechallenged with tumor (right panel).
B. EMT6 tumor-burdened (orthotopic) BALB/c mice treated with a low dose of ACTM-838 and low dose of anti-PD-1 experience greater tumor control than mice treated with either ACTM-838 or anti-PD-1 as single agents, respectively.

7. Summary: Preclinical data supports the initiation of an ACTM-838 phase 1 clinical trial



BACTERIAL STRAIN ENGINEERING
- Genetic modifications enable systemic IV dosing with safe toxicity profile
- Tailored to tissue-specific metabolite dependencies

TUMOR LOCALIZATION
- TME-specific extracellular colonization, and phagocytic APC-specific uptake
- Low or no enrichment in healthy tissues

IN VIVO EFFICACY
- Plasmid payload delivered to and expressed at the tumor site
- Reprogramming of TME to an anti-tumor inflammatory profile
- In vivo efficacy and durable anti-tumor immunity
- Additive effect on tumor control observed with anti-PD-1

Phase 1 ACTM-838 trial initiation mid-2024