A Multifaceted Genetically Engineered Whole-Cell Vaccine Platform Generates Potent, Broad, and Persistent Anti-Tumor Immunity Across Multiple Cancer Indications

Miguel Lopez-Lago[1], Pravin Kesarwani[1], Vikas Bhardwaj[1], Xiaoyi Zheng[1], Ying Liu[1], Sagarika Pachhal[1], Patience Cournoo[1], Renee Cortez[1], George Woodfield[1], Charles L. Wiseman[1], William V. Williams[1]

[1]*BriaCell Therapeutics Corp. Philadelphia, PA*

BriaCell

SITC 2026 - **# 863**

BACKGROUND AND OBJECTIVES

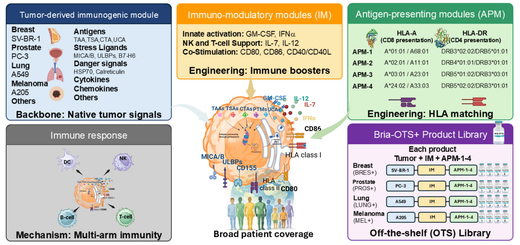
BACKGROUND

Tumor heterogeneity, immune evasion, and impaired immune responses limit the effectiveness of cancer immunotherapies. Allogeneic whole cell cancer vaccines provide an off-the-shelf source of diverse tumor antigens, but they may lack the immunostimulatory signals needed to generate coordinated and sustained anti-tumor responses. Bria-OTS+ is a modular genetically engineered whole cell vaccine platform, integrating diverse tumor antigens, immunostimulatory cytokines, costimulatory molecules, and multiple HLA alleles to promote antigen presentation and coordinated activation of innate and adaptive immune effectors

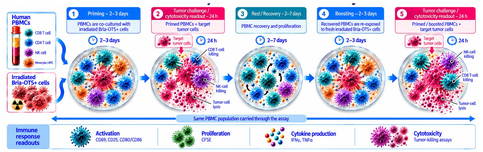
OBJECTIVE

To characterize the potency, breadth, and functional persistence of Bria-OTS+ immune responses across breast and prostate cancer models.

METHODS

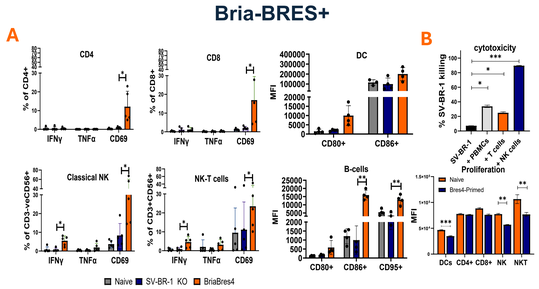


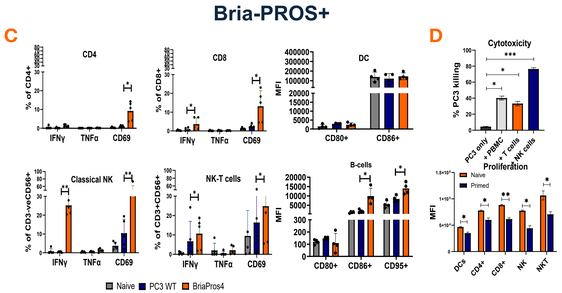
Modular Engineering of Bria-OTS+ Cell Lines: Integrating Tumor immunogenetic factors, Immune Modulators, and HLA Diversity: *Bria-OTS+ is built from three functional modules that together create a modular, immune-educating vaccine platform. The Tumor-derived Antigen Modules (TAM) provide a broad repertoire of tumor-associated and stress-induced antigens to ensure antigenic diversity. The Immuno-modulatory Modules (IM) recruit, prime, and activate immune effectors. The Antigen-Presentation Modules (APM) introduce diverse HLA-A and HLA-DRB3/4/5 alleles, enabling semi-allogeneic antigen display and optimized T-cell recognition. Four pre-manufactured Bria-OTS+ cell lines, each carrying two HLA-A and two HLA-DRB3/4/5 alleles, together provide > 99 % predicted HLA matching across populations, personalizing the cell lines thus enabling broad patient applicability and coordinated activation of dendritic, T, B, and NK cells*



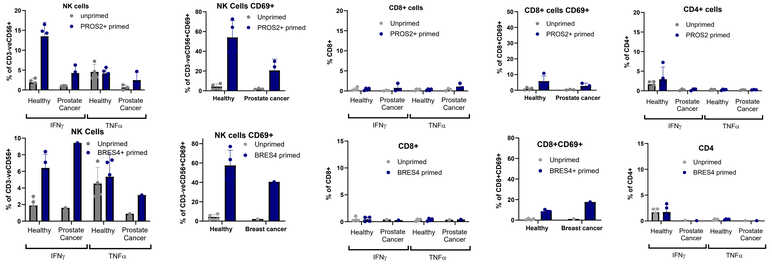
In vitro vaccination assay using Bria-OTS+ cells. *Human peripheral blood mononuclear cells (PBMCs) were evaluated in a sequential prime–rest–boost assay to assess immune activation, proliferation, cytokine production, and tumor-cell killing. The assay consisted of five stages: (1) Priming (2–3 days): PBMCs were co-cultured with irradiated Bria-OTS+ cells to induce T-cell and NK-cell activation through direct cellular interactions, co-stimulatory signaling, and cytokine production. (2) Initial cytotoxicity (24 h): Primed PBMCs were tested against target tumor cells to assess early cytotoxic activity. (3) Rest/recovery (2–7 days): The remaining primed PBMCs were maintained without additional Bria-OTS+ stimulation to allow immune-cell recovery and proliferation. (4) Boosting (2–3 days): PBMCs were re-exposed to fresh irradiated Bria-OTS+ cells to evaluate secondary immune activation and functional responsiveness. (5) Post-boost cytotoxicity (24 h): Boosted PBMCs were challenged with target tumor cells to assess their retained or enhanced killing capacity. Immune responses were characterized by activation markers (CFSE-based proliferation, cytokine production, and tumor-cell cytotoxicity.*

RESULTS

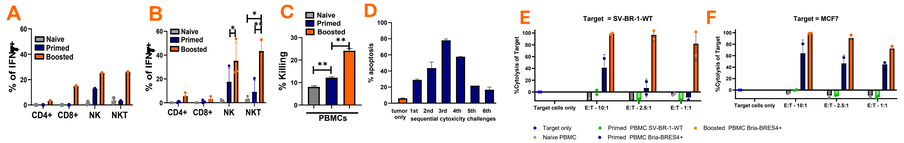




Bria-OTS+ Primes Broad Innate and Adaptive Immune Activation: *PBMCs were co-cultured with Bria-OTS+ cells during the priming phase (3 days) of the in vitro vaccination assay. Bria-BRES4+(A–B) and Bria-PROS4+ (C–D) induced strong early activation marked by IFN-γ/TNF-α production and CD69 upregulation across CD4+, CD8+, NK, and NK-T cells. Dendritic and B cells exhibited increased CD80/CD86 expression, indicating enhanced antigen-presenting potential. Primed PBMCs showed elevated proliferation and cytotoxic activity against parental tumor targets, confirming the potency of Bria-OTS+ in initiating robust innate and adaptive immune responses.*

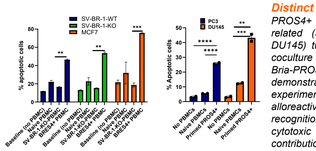


Bria-OTS+ induces innate and adaptive immune activation in PBMCs from patients with breast and prostate cancer. ***(A) Bria-PROS+ and (B) Bria-BRES+*** *were evaluated for their ability to activate immune cells in PBMCs from healthy donors and patients with prostate or breast cancer, respectively. PBMCs were primed with the corresponding engineered cellular immunotherapy platform, and immune responses were assessed by intracellular IFNγ and TNFα production and CD69 expression in classical NK cells, CD8+ T cells, and CD4+ T cells. Both platforms induced robust NK-cell activation and cytokine production in healthy donor PBMCs, with responses retained in cancer patient-derived PBMCs, although generally at lower frequencies. CD69 induction was also observed in CD4+ and CD8+ T cells, supporting activation of both innate and adaptive immune compartments. These findings demonstrate that Bria-OTS+ retains immunostimulatory activity in PBMCs from patients with cancer.*



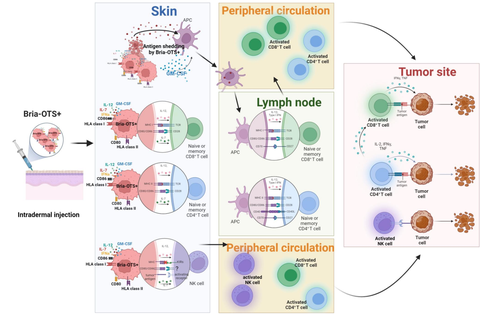
Bria-OTS+ Priming and Boosting Enhance Immune Recall, Cytotoxic Potency, and Sustained Antitumor Function. *PBMCs were primed with Bria-OTS+ cells, rested, and subsequently boosted to evaluate immune recall and functional cytotoxicity. (A–B)* ***Immune recall:*** *Secondary stimulation with (A) Bria-BRES+ or (B) Bria-PROS+ enhanced IFNγ production, particularly in NK and NK-T cells, demonstrating increased effector responsiveness following boosting. (C)* ***Cytotoxic recall:*** *Bria-BRES4+ boosting enhanced PBMC-mediated target-cell apoptosis compared with naïve or primed PBMCs. (D)* ***Sequential killing:*** *Bria-PROS+-primed PBMCs maintained cytotoxic activity across repeated tumor-cell challenges, supporting sustained effector function. (E–F)* ***Enhanced cytotoxic potency following boosting:*** *Real-time cell analysis (RTCA) quantified cytolysis of (E) SV-BR-1-WT and (F) MCF7 target cells after 48 hours of coculture with naïve, parental SV-BR-1-WT-primed, Bria-BRES4+-primed, or Bria-BRES4+-boosted PBMCs. Bria-BRES4+ priming increased cytotoxic activity relative to parental-cell priming, while boosting further enhanced cytolysis across the tested effector-to-target (E) ratios. Notably, boosted PBMCs retained substantial cytotoxic activity even at an E ratio of 1:1, demonstrating enhanced functional potency under limiting effector conditions. Together, these complementary assays demonstrate enhanced immune recall and cytotoxic potency following boosting, alongside sustained killing capacity across sequential tumor-cell challenges..*

RESULTS



Broad Tumor-Cell Recognition and Cytotoxicity Across Distinct Cancer Models. *PBMCs primed with Bria-BRES4+ or Bria-PROS4+ exhibited enhanced cytotoxic activity against both parental or related (SV-BR-1-WT, SV-BR-1-KO, PC3) and heterologous (MCF7, DU145) tumor-cell targets. Target-cell apoptosis was quantified following coculture with naïve or Bria-OTS+-primed PBMCs. Both Bria-BRES4+ and Bria-PROS4+ priming increased tumor-cell killing across the tested models, demonstrating broad functional cytotoxicity. Given the allogeneic experimental setting, these responses may involve HLA-dependent alloreactivity, NK-cell recognition pathways, and tumor-associated antigen recognition. These findings support the capacity of Bria-OTS+ to induce cytotoxic responses against diverse tumor targets, although the relative contributions of individual recognition mechanisms remain to be established.*

PROPOSED MECHANISM OF ACTION



Bria-OTS+ Proposed Mechanism of Action: *Bria-OTS, when injected intradermally, directly activates both naive and previously exposed (memory) T-cells, as well as natural killer (NK) cells. Concurrently, professional antigen-presenting cells (APCs) process the Bria-OTS+ antigens. These APCs then migrate to regional lymph nodes, where they prime T-cells against tumor antigens. The activated T-cells and NK cells subsequently travel to the tumor site, where they trigger a robust anti-tumor immune response.*

CONCLUSIONS

Bria-OTS+ is a modular, genetically engineered whole-cell vaccine platform that integrates antigen breadth, multi-lineage immune activation, and functional persistence to overcome key limitations of cancer immunotherapy.

- **Broad immune activation:** Bria OTS+ induces activation of T-cells, NK cells and other immune populations, supporting coordinated innate and adaptive immune responses across models.
- **Functional anti-tumor immunity:** Bria-OTS+ primed PBMCs exhibit enhanced cytotoxicity against multiple tumor cell targets, including parental or heterologous breast and prostate cancer cell lines.
- **Activity in cancer patient PBMCs:** Bria-OTS+ induces measurable NK- and T-cell activation despite reduced immune responsiveness compared with healthy donors.
- **Persistent effector function:** Priming and boosting enhance immune recall and cytotoxic activity, with sustained tumor-cell killing demonstrated through sequential challenge and complementary real-time growth inhibition assays.
- **Modular therapeutic potential:** Integration of tumor antigens, immunostimulatory signals, and diverse HLA alleles supports further development of Bria-OTS+ as an off-the-shelf cancer vaccine platform.