**STRO-002, an anti-FolRa ADC, Demonstrates Immune-Modulating Properties and Potentiates PD-L1 Blockade**

**Millicent Embry**, **Sihong Zhou**, **Christine Cheng**, **Janice Yu**, **Vinita Ahmad**, **Xiaofan Li**, **Jeff Hanson**, **Cuong Tran**, **Gang Yin**, **Shamin Ahmad**, **Krishna Bajjuri**, **Vinita De Almeida**, **Mark Lusher**, and **Trevor Hallam**

**Sutro Biopharma**, South San Francisco, CA, USA

* Contributed equally. ** Corresponding author: vdealmeida@sutrobio.com

---

**Introduction**

- Folate receptor alpha (FolRα) is a cell-surface protein overexpressed in ovarian and endometrial cancer, representing an attractive target for therapeutic development.
- We have used a platform based on Sutro’s proprietary XpressCell-Free™ (XpressCF+) technology to synthesize single-domain (V)H antibodies using cell-free expression systems. The active antibodies can be affixed to a small toxin payload using Sutro’s proprietary LinkerX™ technology.
- **STRO-002** is a monoclonal antibody conjugate of a single-domain VHH (aFolRα) with a prodrug payload of monomethyl auristatin (MMAE) and a bispecific single-domain payloads targeting PD-L1 (aGFP-SC209).
- The active metabolite of STRO-002, aFolRα-ADC, is a tubulin-targeting cancer cell toxin with potent activity against cancer cell lines expressing high levels of FolRα.

**Results**

- **Generation of STRO-002 and STRO-002 using Sutro’s proprietary Xpress Cell-Free (XpressCF+) system.**

**Cell-Free Antibody Synthesis And Site-Specific Conjugation**

**In vivo methods**

- Human KB, human Igrov-1, or murine MC38-hFolR α cells were implanted subcutaneously into the right flank of female nude mice. SCID mice in C57Bl/6 background, respectively. Treatment schedule was initiated when tumors were established in vivo. The aGFP-SC209 conjugate was administered intravenously at 10 mg/kg in a 5-day cell proliferation assay.

**Figure 1: Flow cytometry assay.**

- Cells were cultured with human PBMCs.
- CD86+ MFI of CD14+ Cells

**Combination Treatment of STRO-002 and Avelumab Significantly Increased Infiltration of CD8+ T Cells in MC38-hFolRa Tumors**

- Animals bearing MC38-hFolRa tumors were treated with a single dose of vehicle, 10 mg/kg STRO-002, 20 mg/kg Avelumab or the combination of both. Tumors were measured by caliper every three days and FPS sections stained for CD8+ T cells by immunohistochemistry.
- Representative images of CD8 staining (brown) with nuclei counterstain (blue) are shown on the left. The percentage of CD8+ cells (left) was quantified using the HCS Analysis software. As indicated by the IHC data, there was a significant increase in CD86+ T cell infiltration into the tumor microenvironment.

**Summary**

- Our studies demonstrate that in addition to its potent cytotoxic activity, STRO-002 demonstrates a complementary mechanism of action involving engagement of the host immune system to potentiate protective anti-tumor immunity after tumor re-challenge.
- IHC analysis revealed a significant increase in CD8+ T cell infiltration in the tumors of animals treated with a combination of STRO-002 and Avelumab.
- Our data suggest that STRO-002 mediated induction of CD8 and upregulation of PD-1 expression contribute to the added benefit observed with combination of STRO-002 and Avelumab.

**Acknowledgments**

- This work was supported by the National Cancer Institute (R01CA233453) and the National Institute of Biomedical Imaging and Bioengineering (U01EB026398). The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.