# Mouse analog of micvotabart pelidotin, an antibody-drug conjugate targeting extradomain-B of fibronectin, demonstrates anti-tumor efficacy in an immunotherapy-refractory syngeneic head and neck squamous cell carcinoma model

**Anthony B. Rodriguez**, Amanda Facklam, Justin Trickett, Sara Lewandowski, Matthew Iovino, Chuan Shen, Frank Wang, Nicolas Severe, Marsha Crochiere

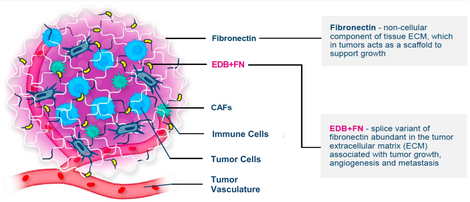
Pyxis Oncology, Boston, Massachusetts, USA

PYXIS ONCOLOGY

Abstract : 4406

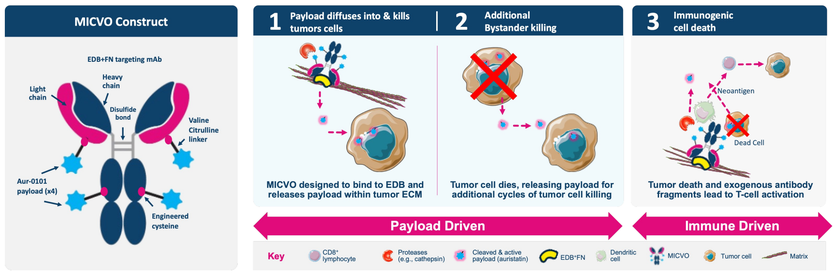
## Background

- Head and neck squamous cell carcinoma (HNSCC) remains a difficult-to-treat disease, and current standard-of-care therapies provide limited long-term survival, underscoring the need for more effective options.

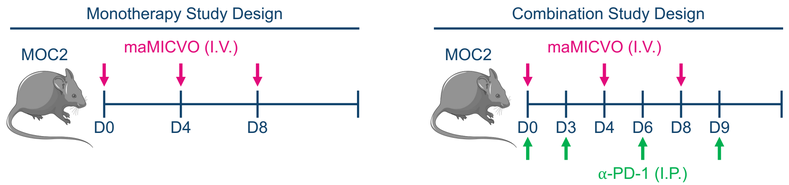



- Micvotabart pelidotin (MICVO) is a first-in-concept antibody-drug conjugate targeting extradomain-B of fibronectin (EDB+FN), a non-cellular extracellular matrix protein abundantly expressed in the stroma of many tumors, including HNSCC, but minimally expressed in normal adult tissues (1).
- Preclinically, MICVO demonstrated broad anti-tumor activity across multiple patient-derived xenograft models, including HNSCC (2), and induced features of immunogenic cell death *in vitro* (3). Similarly, a mouse analog of MICVO (maMICVO) promoted T cell infiltration into tumors and synergized with anti–PD-1 to improve efficacy in immunotherapy-sensitive and -refractory syngeneic triple-negative breast cancer models (4,5).
- This poster aims to characterize the anti-tumor activity of maMICVO and the immune changes it induces to enhance response to anti–PD-1 treatment in an immunotherapy-refractory syngeneic HNSCC model, further supporting the clinical development of MICVO as a monotherapy (NCT05720117) and in combination with pembrolizumab (NCT06795412) for recurrent or metastatic (R/M) HNSCC.

## MICVO: Three-Pronged Mechanism of Action

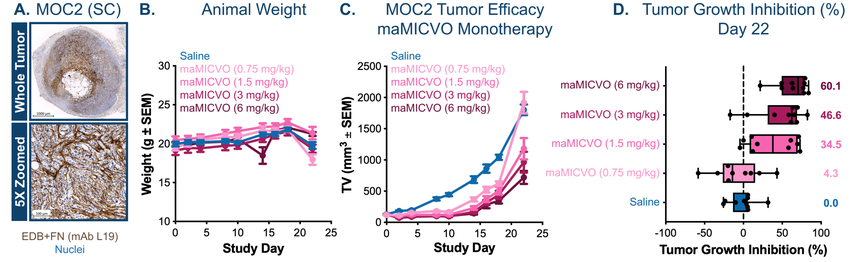



## Methods



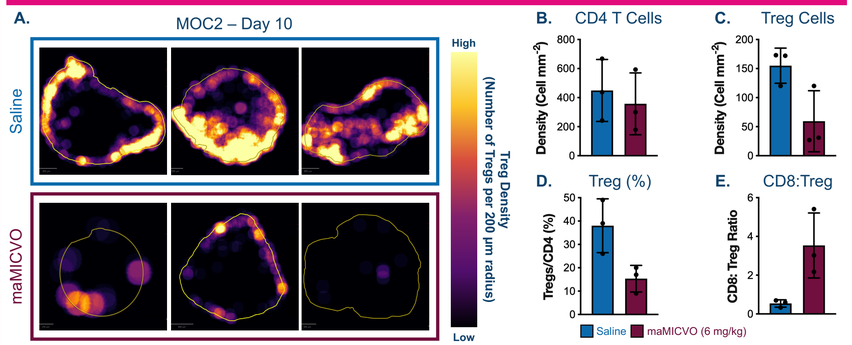

- Six- to eight-week-old female C57BL/6 mice were injected subcutaneously with 0.3 × $10^6$ MOC2 HNSCC cells.
- When tumors reached ~75 mm³ (Day 0), mice were randomized into the following treatment groups: (1) saline; (2) maMICVO; (3) anti-mouse PD-1 (clone RMP1-14); or (4) maMICVO plus anti-mouse PD-1.
- Tumor volume and body weight were monitored to assess efficacy and tolerability, respectively.
- At selected time points, some tumors were collected and processed for immune profiling by flow cytometry and multiplex immunofluorescence (mIF) or for determining EDB+FN expression by immunohistochemistry (IHC).

## maMICVO Induces Dose-Dependent Inhibition of MOC2 Tumor Outgrowth



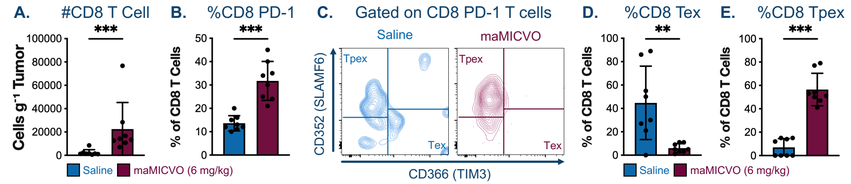

**Mouse analog of MICVO showed dose-dependent inhibition of MOC2 tumor outgrowth.** (**A**) IHC analysis of untreated MOC2 tumors showed EDB+FN protein expression, with most signal localized to the tumor stroma. To evaluate efficacy, MOC2 tumor-bearing mice were treated with saline (control) or escalating doses of maMICVO, as described in Methods. (**B**) All doses of maMICVO were well tolerated, with no significant body weight loss. (**C**) maMICVO monotherapy delayed tumor progression in a dose-dependent manner and (**D**) increased tumor growth inhibition (TGI), with the strongest effect observed at 6 mg/kg. TGI was calculated as the percent reduction in individual tumor volume relative to the mean saline volume at the same time point: TGI (%) = [1 − (treated volume / mean saline volume)] × 100. Data are representative of two independent experiments (n = 10 per group).

## maMICVO Modulates MOC2 Tumors Toward a More Immune-Permissive State



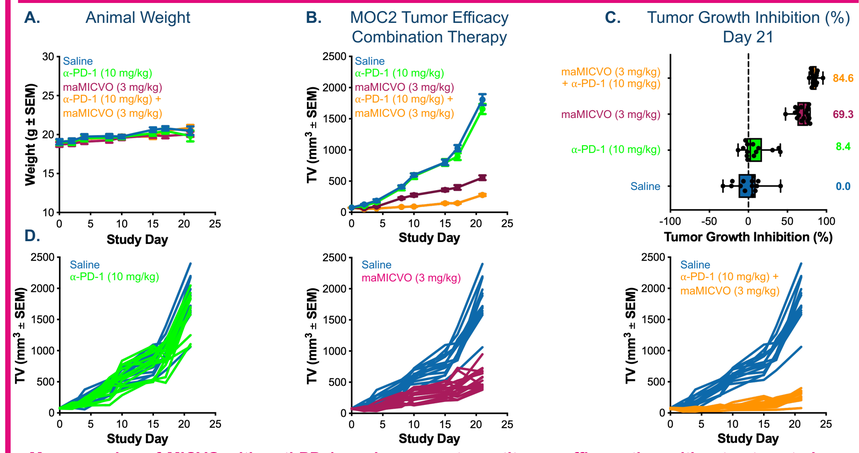

**Mouse analog of MICVO reduces intratumoral Treg burden, promoting a more immune-permissive microenvironment in MOC2 tumors.** MOC2 tumors from saline- or maMICVO-treated mice were collected 48 hours after the third dose and processed for mIF to quantify the density and distribution of CD8 T cells (CD3+ CD8+), CD4 T cells (CD3+ CD8−), and Tregs (CD3+ FoxP3+) by QuPath. (**A**) Representative heat maps showed reduced Treg burden in maMICVO-treated tumors relative to saline controls. QuPath analysis showed (**B**) unchanged CD4 T cell density, but trends toward (**C**) reduced Treg density and (**D**) lower Treg-to-CD4 frequency, accompanied by (**E**) higher CD8:Treg ratios. Data are representative of a single experiment (n = 3 per group).

## maMICVO Increases the Abundance of Anti–PD-1-Responsive CD8 T Cells




**Mouse analog of MICVO increases CD8 T-cell abundance in MOC2 tumors, including a progenitor exhausted subset that is highly responsive to anti–PD-1.** MOC2 tumors from saline- or maMICVO-treated mice were collected 48 hours after the third dose and analyzed by flow cytometry. (**A**) maMICVO increased the number of intratumoral CD8 T cells relative to saline controls. (**B**) The proportion of CD8 T cells expressing PD-1 was also higher with maMICVO treatment. (**C**) Among CD8 PD-1+ T cells, terminally exhausted (Tex) and anti–PD-1-responsive progenitor exhausted (Tpex) subsets were identified. (**D**) Saline-treated tumors were enriched for Tex cells, (**E**) whereas maMICVO-treated tumors were enriched for Tpex cells. Data are representative of two independent experiments (n = 8 per group). P-values by Mann-Whitney *U*-test: **P < 0.01; ***P < 0.001.

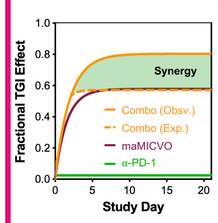
## maMICVO Sensitizes Immunotherapy-Refractory MOC2 Tumors to Anti–PD-1




**Mouse analog of MICVO with anti-PD-1 produces greater antitumor efficacy than either treatment alone.** MOC2 tumor-bearing mice were treated with saline, maMICVO, anti-PD-1, or the combination, as described in Methods. (**A**) All treatments were well tolerated, with no significant body weight loss. (**B**) Anti-PD-1 monotherapy had minimal effect on MOC2 tumor outgrowth, whereas maMICVO monotherapy delayed tumor growth and was associated with greater TGI. (**C**) This effect was further enhanced by combining maMICVO with anti-PD-1, resulting in greater anti-tumor activity and TGI than either monotherapy alone. (**D**) Spider plots confirmed greater tumor control with the combination treatment than with either monotherapy. TGI was calculated as previously described. Data are representative of two independent experiments (n = 14–17 per group).

## maMICVO Synergizes with Anti–PD-1 to Enhance Anti-Tumor Efficacy

MOC2 – Bliss Independence Analysis & Scoring




| | |
|---|---|
| Bliss Score (Mean ± SD): | 0.17 ± 0.11 |
| Max Bliss (Score & Day): | 0.33 (D8) |
| Min Bliss (Score & Day): | 0.01 (D0) |
| Cumulative Bliss (AUC): | 4.82 |
| Positive Bliss Days (%): | 80% |
| Bliss Curve Fit ($R^2$): | 0.9792 |

**Bliss analysis revealed synergy between the mouse analog of MICVO and anti–PD-1, resulting in greater anti-tumor efficacy.** Bliss independence modeling, as previously described (5), was applied to tumor volume data from the study above. The combination treatment exceeded both monotherapies and the predicted additive effect. The shaded green region represents the difference between observed and expected outcomes, indicating the magnitude of synergy. Single-agent effects were defined as fractional tumor growth inhibition, 1 − (T/Control), and the Bliss-expected effect, eA + eB − eA × eB, represented the predicted additive response if both agents acted independently.

## Conclusions

- Monotherapy with maMICVO inhibited MOC2 tumor outgrowth in a dose-dependent manner, further supporting the anti-tumor activity of MICVO in HNSCC.
- Treatment with maMICVO reduced Treg burden and increased the abundance of anti–PD-1-responsive CD8 Tpex cells in MOC2 tumors, further demonstrating MICVO's ability to drive tumor-immune modulation in HNSCC.
- Bliss analysis shows that maMICVO and anti–PD-1 synergize to enhance efficacy in immunotherapy-refractory MOC2 tumors, highlighting MICVO's potential for clinical benefit in patients with HNSCC who do not respond to checkpoint blockade.
- These findings support the clinical development of MICVO as a monotherapy (NCT05720117) and with pembrolizumab for R/M HNSCC (NCT06795412).

### References

(1) Lewandowski S, et al. Cancer Res. 2024 Mar;84(6):2908.
(2) Facklam A, et al. Cancer Res. 2025 Apr; 85(8): 3120
(3) Iovino M, et al. Mol. Cancer Ther. (2025) Oct; 24(10): A112
(4) Rodriguez AB, et al. Cancer Res. 2025 Apr; 85(8): 3137
(5) Rodriguez AB, et al. Mol. Cancer Ther. (2025) Oct; 24(10): A115

***Servier Medical Art for figure design: https://smart.servier.com/***