

# Tumor-Activated Anti-CTLA-4 Monoclonal Antibody, XTX101, Demonstrates Monotherapy and Anti-PD-1 Combination Benefit in Preclinical Models

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## BACKGROUND

- The clinical benefit of CTLA-4 blockade to cancer patients has been well established
- However, efficacy of current therapies is limited by dose limiting toxicity arising from systemic immune activation
- XTX101** is a tumor-selective anti-CTLA-4 mAb that is designed to improve upon the therapeutic index of existing anti-CTLA-4 therapies by overcoming potency and tolerability limitations
- XTX101** is designed for improved potency
  - Higher affinity binding to the target CTLA-4
  - Enhanced Fc effector function
- XTX101** is designed to have reduced peripheral immune activity
  - CDR sequences are masked while in circulation
  - Activated by protease-dependent release of the masks
  - Selectively active in the tumor microenvironment and avoids toxicity associated with systemic immune activation

### Ipilimumab data strongly validate potential for improved $\alpha$ -CTLA-4 mAb

Ipilimumab Melanoma Randomized Phase 3 Study<sup>1</sup>

Dose	Median OS	Adverse Events: gr 3/4 irAEs/disconts.
Standard Approved Dose 3 mg/kg	11.5 mo	14% / 19%
10 mg/kg	15.7 mo	30% / 31%

**Conclusion:** Treatment with higher dose resulted in increased overall survival (OS) but also increased AEs and discontinuations

- Ipilimumab shows preliminary evidence of promising antitumor activity in a range of tumor types, outside of the approved indications<sup>2,3,4</sup>
- Patients with high affinity Fc $\gamma$ R polymorphisms have shown improved clinical responses to ipilimumab<sup>5,6</sup>
- Ipilimumab is more active when combined with nivolumab, including increased rate of irAEs<sup>7</sup>
- The therapeutic potential of ipilimumab monotherapy or in combination with anti-PD-1 is curtailed by dose limiting systemic immune toxicities

Xilio's approach is to combine tumor selectivity and enhanced potency of anti-CTLA-4 treatment to achieve an improved therapeutic index (TI)

Sources: <sup>1</sup> Ascierto PA, Lanctot Oncol. (2017); <sup>2</sup> Beer TM, J. Clin. Oncol. (2017); <sup>3</sup> Hellmann MD, NEJM (2019); <sup>4</sup> Kao HF, Head Neck. (2019); <sup>5</sup> Arce-Vargas F, Cancer Cell (2018); <sup>6</sup> Quezada SA, Clin. Cancer Res. (2019); <sup>7</sup> Lebba C, J. Clin. Oncol. (2019)

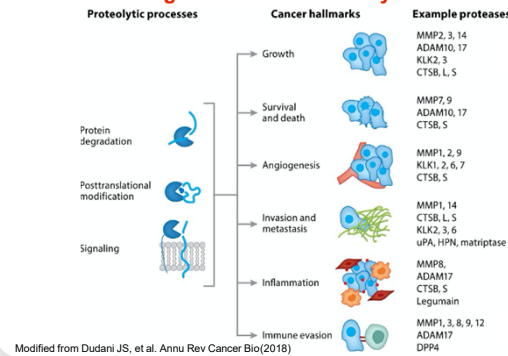
## BACKGROUND

### XTX101: Affinity and Fc enhancements combined with tumor selectivity for optimized TI



- $\alpha$ -CTLA-4 mAb
- Fc mutations for enhanced Fc $\gamma$ R binding and ADCC
- Improved affinity over ipilimumab
- Tumor-selectivity
  - Increased potency through improved affinity and enhanced ADCC to deplete Tregs
  - Improved tolerability by adding tumor-selectivity
  - Combining increased potency and improved tolerability to maximize opportunity for improved TI

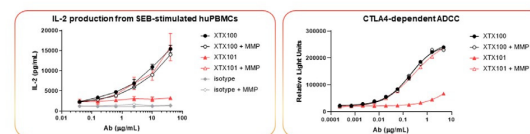
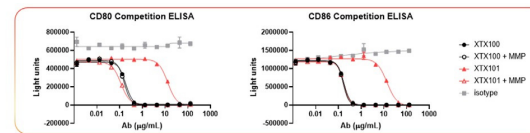
### Tumors are proteolytically active environments that we leverage for tumor selectivity of XTX101



Modified from Dudani JS, et al. Annu Rev Cancer Bio(2018)

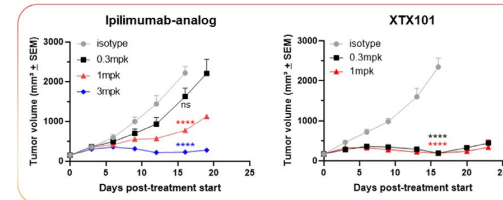
## RESULTS

### After proteolytic activation, XTX101 inhibits binding of CTLA-4 to its cognate ligands CD80 and CD86, and mediates cellular activity in PBMC and ADCC assays



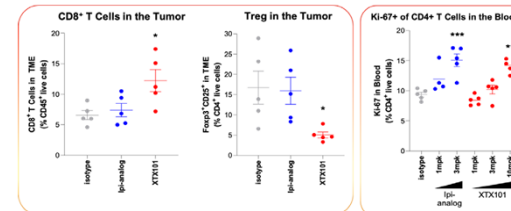
## RESULTS

### XTX101 is more potent than ipilimumab-analog



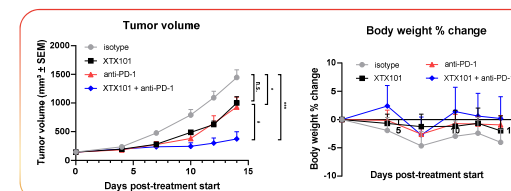
- XTX101 drives potent tumor growth inhibition (TGI), superior to ipilimumab-analog
- A dose of 3 mg/kg of ipilimumab-analog is required to achieve similar efficacy and CR rate as XTX101 at 0.3 mg/kg, suggesting XTX101 has 10x higher potency
- MB49 cells were inoculated subcutaneously into C57BL/6-huCTLA-4 mice. Mice were dosed single-dose i.v. A two-way ANOVA with Bonferroni's multiple comparisons post-test was performed to determine the statistical significance of treatment vs. isotype (\*P<0.05; \*\*P<0.01; \*\*\*P<0.001; \*\*\*\*P<0.0001).

### XTX101 demonstrates potent intratumoral pharmacodynamic (PD) response, superior to ipilimumab-analog



- XTX101 shows Treg depletion in the tumor and no effect on peripheral Treg levels
- A dose of 3 mg/kg of ipilimumab-analog is required to achieve similar peripheral PD as XTX101 at 10 mg/kg, consistent with better tolerability of XTX101 in mice
- MB49 cells were inoculated subcutaneously into C57BL/6-huCTLA-4 mice. Mice were dosed single-dose i.v. A one-way ANOVA with Bonferroni's multiple comparisons post-test was performed to determine the statistical significance of treatment vs. isotype (\*P<0.05; \*\*P<0.01; \*\*\*P<0.001).

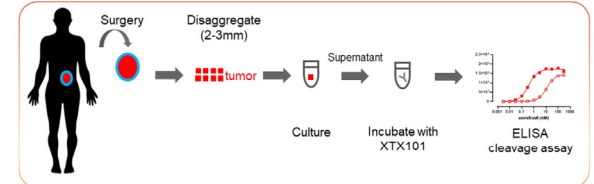
### Significant combination benefit observed with XTX101 and anti-muPD-1



- Significant TGI observed in mice treated with XTX101 and anti-PD-1 combination, and no significant decrease of body weight was observed.
- MC38 cells were inoculated subcutaneously into the right flank of C57BL/6-huCTLA-4 mice. Therapy started when the tumors reached ~144 mm<sup>3</sup>. Mice were dosed i.v. with 0.3 mg/kg of XTX101 on day 0; i.p. with 10 mg/kg of anti-PD-1 on day 0, 3, 6. A two-way ANOVA with Dunnett's multiple comparisons post-test was performed to determine the statistical significance of treatment between groups (n.s.=not significant; \*p<0.05; \*\*p<0.01; \*\*\*p<0.001).

## RESULTS

### Broad activation of XTX101 across human tumors in ex vivo assay



Cancer type	Melanoma	RCC	Ovary	Bladder	Colon	NSCLC	Breast	Liver	Total
% Tumors that activate XTX101	71%	57%	58%	67%	91%	67%	75%	67%	66%
Sample size	7	30	12	6	11	9	4	6	85

- Broad potential for tumor selective XTX101 activity leads to opportunities in many indications
- Given the broad activation of XTX101 across diverse tumor types, Companion Diagnostic (CDX) biomarker for protease activity or expression likely not required

## CONCLUSION

- The clinical benefit of CTLA-4 blockade is well established, but remains limited due to toxicities arising from systemic immune activation
- XTX101 is a potent, tumor-selective anti-CTLA-4 mAb with the potential to significantly expand the TI relative to ipilimumab and therefore potentially reduce toxicity while enhancing efficacy
- Functional characterization of XTX101 confirmed protease-dependent activity
  - XTX101 binds to CTLA-4 with high affinity in a protease-dependent manner
  - XTX101 exhibits protease dependent inhibition of CD80/86 binding to CTLA-4
  - Protease-dependent activation of XTX101 overcomes CTLA-4 inhibition of T cell activation
  - Enhanced effector function of XTX101 confers potent protease & CTLA-4 binding dependent ADCC activity
- Tumor-selective activity of XTX101 was demonstrated by *in vivo* studies
  - XTX101 demonstrates 10x improvement in potency in murine tumor growth inhibition studies
  - XTX101 exhibited enhanced Treg depletion in tumors
  - XTX101 showed reduced peripheral immune activation by comparison to ipilimumab-analog
  - Significant benefit observed with XTX101 and anti-PD-1 combination, inducing increased TGI without an increase in toxicity
- XTX101 is activated broadly across multiple tumor subtypes based on *ex vivo* studies in fresh human explant tumor tissue
- These data support evaluation of XTX101 in clinical studies