XTX301, a Tumor-Activated Interleukin-12 (IL-12), Demonstrated IL-1 Pharmacology in Patients with Advanced Solid Tumors: Pharmacodynamic Data from First-in-Human Phase 1 Study (NCT05684965)

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SCAN ME

Abstract 567

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BACKGROUND

- IL-12 is a proinflammatory cytokine that has shown promise as an immunotherapy for cancer; however, clinical development of systemically active IL-12 therapies has been limited by severe toxicities

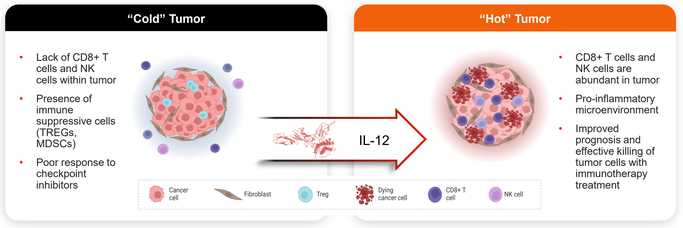
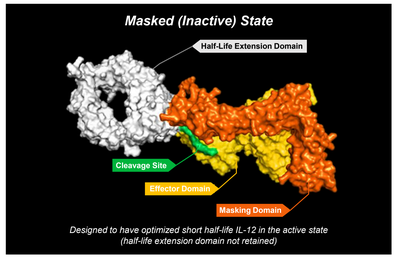


Figure generated using BioRender.com

Efarindodekin alfa (XTX301) is a Tumor-Activated, IL-12 Designed to Overcome the Limitations of Systemic (rh)IL-12

- Efarindodekin alfa is a tumor-activated IL-12 designed to be inactive in non-tumor tissue and when circulating systemically
- In the TME MMPs unmask efarindodekin alfa, thereby localizing activity in the TME and widening the therapeutic index relative to (rh)IL-12
- Once unmasked, efarindodekin alfa potently stimulates anti-tumor immunity and reprograms the TME of poorly immunogenic "cold" tumors towards an inflamed or "hot" state



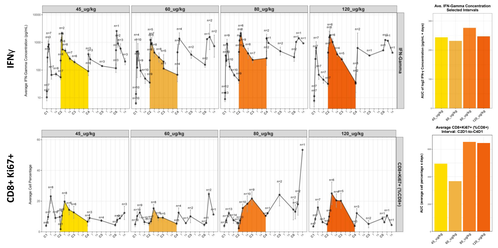
Designed to have optimized short half-life IL-12 in the active state (half-life extension domain not retained)

STUDY DESIGN, METHODS, PATIENT POPULATION

- This Phase 1 dose-escalation study (NCT05684965) evaluated intravenous efarindodekin alfa monotherapy in patients with metastatic solid tumors for safety and tolerability, PK/PD and anti-tumor activity using a standard 3+3 dose escalation design to determine the RP2D and schedule[1]
- Peripheral biomarker analyses from Phase 1 are presented. Serum cytokines were quantified using MSD assays. Flow cytometry was used to evaluate immune cell subsets in peripheral blood. Data from key PD biomarkers IFNγ and CD8+Ki67+ T cells are presented
- Tumor profiling was performed in head and neck cancer patients, including one patient with a confirmed PR. Tumor biopsies obtained pre-treatment and on-treatment were analyzed via IHC and immunofluorescence as indicated. Additionally, FNA samples from matched timepoints were assessed by flow cytometry to characterize the TME

Patient Characteristics and Disposition	Total (n=62)
Age, median (range)	66 (43-83)
Male	37 (60%)
ECOG PS 0	30 (48%)
ECOG PS 1	28 (45%)
ECOG PS 2	4 (6%)
Prior Lines of Anti-Cancer Treatment	**Median: 3 (0-12)**
0-1	6 (10%)
≥2	55 (89%)
Received Immune Checkpoint Inhibitor	50 (81%)
Treatment Status	
Continuing on Treatment	8 (13%)
Discontinued Treatment	54 (87%)
Disease Progression	43 (69%)
Adverse Events	6 (10%)
Other	5 (8%)

SUSTAINED DOSE-DEPENDENT INCREASE IN IFNγ MAINTAINED WITHOUT TACHYPHYLAXIS

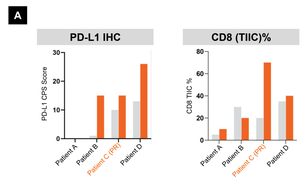


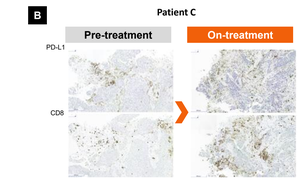
- Efarindodekin alfa induced dose-dependent IFNγ release and CD8+ T cell proliferation, which are established PD biomarkers of IL-12 signaling
- Unlike repeat administration of (rh)IL-12,[2] no tachyphylaxis was observed with efarindodekin alfa repeated dosing
- Pharmacokinetics (n=45) showed a relatively dose-proportional exposure without peripheral cleavage of efarindodekin alfa

Top: Average IFNγ concentration by treatment group (primed). Left: Line graph showing average IFNγ concentration at different visits (x-axis) with respect to treatment groups. Colored areas indicate intervals of interest. Treatment cycles are annotated at the bottom of the plot (Cycle Day). **Right:** Bar plot showing the AUC for the areas of interest highlighted in the graph on the left
Bottom: Average CD8+ Ki67+ T cell percentage by treatment group (primed). Left: Line graph showing average CD8+ Ki67+ T cells (% of CD8s) at different visits/timepoints (x-axis) with respect to treatment groups. Colored areas indicate intervals of interest. Treatment cycles are annotated at the bottom of the plot (cycle day). **Right:** Bar plot showing the AUC for the areas of interest highlighted in the graph on the left

PHARMACODYNAMICS CONSISTENT WITH IL-12 BIOLOGY

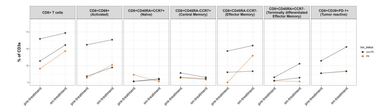
Increased PD-L1 expression and CD8 T cell infiltration in tumors following treatment with efarindodekin alfa





Efarindodekin alfa induced PD-L1 upregulation and CD8+ T cell infiltration across multiple analytical platforms. Tumor biopsies were collected pre-treatment and on-treatment (Cycle 2, Days 2-5) from treated patients. Patient C achieved a confirmed PR by RECISTv1.1
(A) IHC: PD-L1 CPS score and CD8+ TIIC percentage increased on-treatment
(B) Representative images from a patient with a confirmed PR (Patient C) showing PD-L1 upregulation and CD8+ T cell infiltration before and after treatment

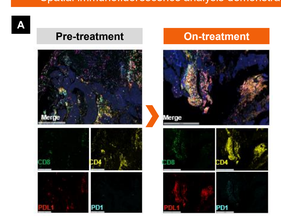
Flowcytometry analysis of FNA showed efarindodekin alfa promoted CD8+T cell infiltration, activation and differentiation to effector memory

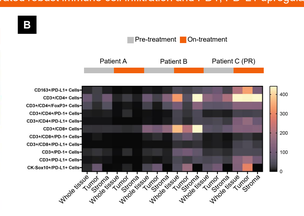


- On-treatment samples showed increased tumor-infiltrating CD8+ T cells with marked upregulation of the CD69 activation marker
- CD8+ T cells exhibited a functional phenotypic shift toward effector memory phenotype
- Additionally, there was an increase in CD8+CD39+PD-1+ T cells, a population associated with tumor reactive T cells

Enhanced T Cell Activation and Functional Phenotypic Shift in Tumor. Flow cytometry analysis of FNA samples collected from tumors pre-treatment and on-treatment (Cycle 2, Days 2-5) demonstrates multiple indicators of enhanced anti-tumor immunity. Patient with confirmed PR (Patient C) is indicated in orange

Spatial immunofluorescence analysis demonstrated robust immune cell infiltration and PD1, PD-L1 upregulation in tumors after treatment





Efarindodekin alfa induced robust immune cell infiltration and PD1, PD-L1 upregulation in tumor. **(A)** A representative image of multiplex immunofluorescence staining of pre-treatment (left) and on-treatment (right) tumor biopsy of a patient with a confirmed PR (Patient C) by RECISTv1.1. **Top panels:** Merged composite images. **Individual channel panels show:** CD8 (green, cytotoxic T-cells), CD4 (yellow, helper T-cells), PD-L1 (red, checkpoint ligand), PD-1 (teal, checkpoint receptor). Efarindodekin alfa resulted in marked increase in T-cell infiltration (CD8+, CD4+ T cells) and upregulation of PD-L1 expression compared with pre-treatment baseline
(B) A comprehensive cell density quantification of multiplex immunofluorescence images across whole tumor tissue, tumor, and stroma, demonstrating robust immune cell infiltration post treatment

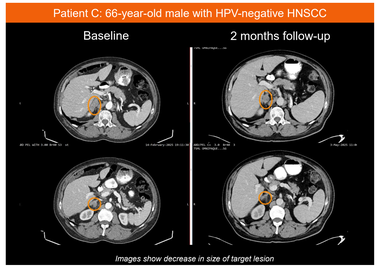
PRELIMINARY ANTI-TUMOR ACTIVITY

Two PRs were reported (one confirmed, one unconfirmed):

1. HPV-negative HNSCC
   - 5 prior lines of therapy, including no response to pembrolizumab-based therapy
   - Confirmed PR (33% decrease in sum of diameters) by RECISTv1.1 accompanied by robust changes in PD biomarkers (Patient C in translational analyses)
2. Uveal melanoma
   - 2 prior lines of therapy
   - Unconfirmed PR (55% decrease in sum of diameters) by RECISTv1.1*

*Patient discontinued treatment due to Grade 3 immune system activation AE that subsequently resolved. There was no evidence of further tumor growth through the 90-day follow-up, suggesting duration of response of at least 2 months

Patient C: 66-year-old male with HPV-negative HNSCC



Images show decrease in size of target lesion

EFARINDODEKIN ALFA WAS WELL-TOLERATED IN PHASE 1, CONSISTENT WITH TUMOR-ACTIVATED DESIGN

TRAE occurring in ≥10% patients	All Doses and Schedules (n=62)		RP2D (n=13)	
	Any Grade n (%)	≥ Grade 3 n (%)	Any Grade n (%)	≥ Grade 3 n (%)
AST increased	19 (31)	3 (5)	3 (23)	0
ALT increased	15 (24)	4 (6)	2 (15)	0
Chills	15 (24)	0	3 (23)	0
Fatigue	15 (24)	1 (2)	5 (38)	0
CRS	13 (21)	2 (3)	1 (8)	0
Nausea	13 (21)	0	1 (8)	0
Mucositis/Stomatitis	12 (19)	2 (3)	3 (23)	0
Pyrexia	11 (18)	0	1 (8)	0
WBC decreased	11 (18)	3 (5)	1 (8)	1 (8)
Headache	10 (16)	0	2 (15)	0
Platelet count decreased	10 (16)	1 (2)	0	0
Neutrophil count decreased	9 (15)	2 (3)	0	0
Lymphocyte count decreased	7 (11)	2 (3)	1 (8)	1 (8)
Vomiting	7 (11)	0	0	0
DLTs	2*		0	
Treatment discontinuation due to TRAE	4 (6)		1 (8)	
Dose reduction due to TRAE	4 (6)		0	

*Both DLTs reported at doses above RP2D

- TRAEs were primarily low grade: one Grade 4 (ANC decreased, resolved) and no Grade 5 TRAEs reported

CONCLUSIONS

- Efarindodekin alfa (tumor-activated IL-12) monotherapy was well-tolerated at doses >100-fold higher than the MTD for (rh)IL-12, and TRAEs were primarily Grade 1 or 2 consistent with tumor-activated design
- IFNγ production was sustained without tachyphylaxis and transformed the TME towards an inflamed state with increased T cell infiltration and differentiation to effector memory
- Two PRs (one confirmed, one unconfirmed) with meaningful changes in PD biomarkers in HNSCC patient with confirmed PR. Phase 2 initiated at the RP2D in selected tumor types
- Promising Phase 1 data support further evaluation of efarindodekin alfa as a monotherapy and in combination (including with immune checkpoint inhibitors) across a range of "cold" and "hot" tumor types

ABBREVIATIONS: AE: adverse event; ALT: alanine aminotransferase; ANC: absolute neutrophil count; AST: aspartate aminotransferase; AUC: area under the curve; CD: cluster of differentiation; CPS: combined positive score; CRS: cytokine release syndrome; DLT: dose limiting toxicity; ECOG PS: Eastern Cooperative Oncology Group Performance Status; FNA: fine needle aspirates; FOXP3: forkhead box protein P3; HNSCC: head and neck squamous cell carcinoma; HPV: human papilloma virus; IFNγ: interferon gamma; IHC: immunohistochemistry; IL-12: Interleukin-12; Ki67: Ki67 IHC; MDSCs: myeloid-derived suppressor cells; MMPs: matrix metalloproteases; MSD: Meso Scale Discovery; MTD: maximum tolerated dose; NK: natural killer; PD-L1: programmed death-ligand 1; PD: pharmacodynamics; PK: pharmacokinetics; RP2D: recommended Phase 2 dose; PR: partial response; RECISTv1.1: Response Evaluation Criteria in Solid Tumors version 1.1; (rh): recombinant human; TIIC: tumor-infiltrating immune cells; TME: tumor microenvironment; TPS: tumor proportion score; TRAE: treatment related adverse events; TREG: regulatory T-cell; WBC: white blood cells.

ACKNOWLEDGEMENTS AND DISCLOSURES: Study sponsored by Xilio Therapeutics, Inc. (Xilio). **REFERENCES.** 1. Knecht ASCO 2023. 2. Atkins et al., 1997 (3)409-417 Clinical Cancer Research

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