

12x12 Virtual Symposium: IL-12 in the Clinic

December 12, 2022

IL-12 IL-12 IL-12 IL-12

Today's Featured Speakers



Jamie Spangler, Ph.D.

Jamie Spangler, Ph.D. is an esteemed researcher whose work has focused on engineering immune molecules such as cytokines, growth factors, and antibodies for targeted treatment of diseases such as cancer and autoimmune disorders. After completing a postdoctoral fellowship at Stanford University School of Medicine, Dr. Spangler launched her independent research group at Johns Hopkins University in July 2017, jointly between the departments of Biomedical Engineering and Chemical & Biomolecular Engineering. Her lab, located in the Translational Tissue Engineering Center at the School of Medicine, applies structural and mechanistic insights to re-engineer existing proteins and design new proteins that therapeutically modulate the immune response. She holds a B.S. in biomedical engineering from Johns Hopkins University and a Ph.D. in biological engineering from MIT.



Jason Luke, M.D.

Jason Luke, M.D. is the Director of the Immunotherapy and Drug Development Center and Associate Professor of Medicine at the University of Pittsburgh. Dr. Luke is an internationally recognized clinical and translational investigator and has been at the forefront of immuno-oncology combination therapies. Dr. Luke's drug development expertise spans first-in-human trials through late-stage clinical studies in melanoma, including lead investigator roles for practice-changing clinical trials of PD-1 inhibitors and immune checkpoint combinations. Dr. Luke is an At-Large Director of SITC and has received numerous awards including the Melanoma Research Foundation Humanitarian Award, Crain's 40 under 40, a DOD Career Development Award, an ASCO Merit Award, and the NCI Cancer Clinical Investigator Team Leadership Award. Dr. Luke received his M.D. degree from Rosalind Franklin University of Medicine and Science/Chicago Medical School, completed Internal Medicine Residency at Boston University Medical Center and Medical Oncology Fellowship at Memorial Sloan-Kettering Cancer Center and Weill Cornell Medical Center.

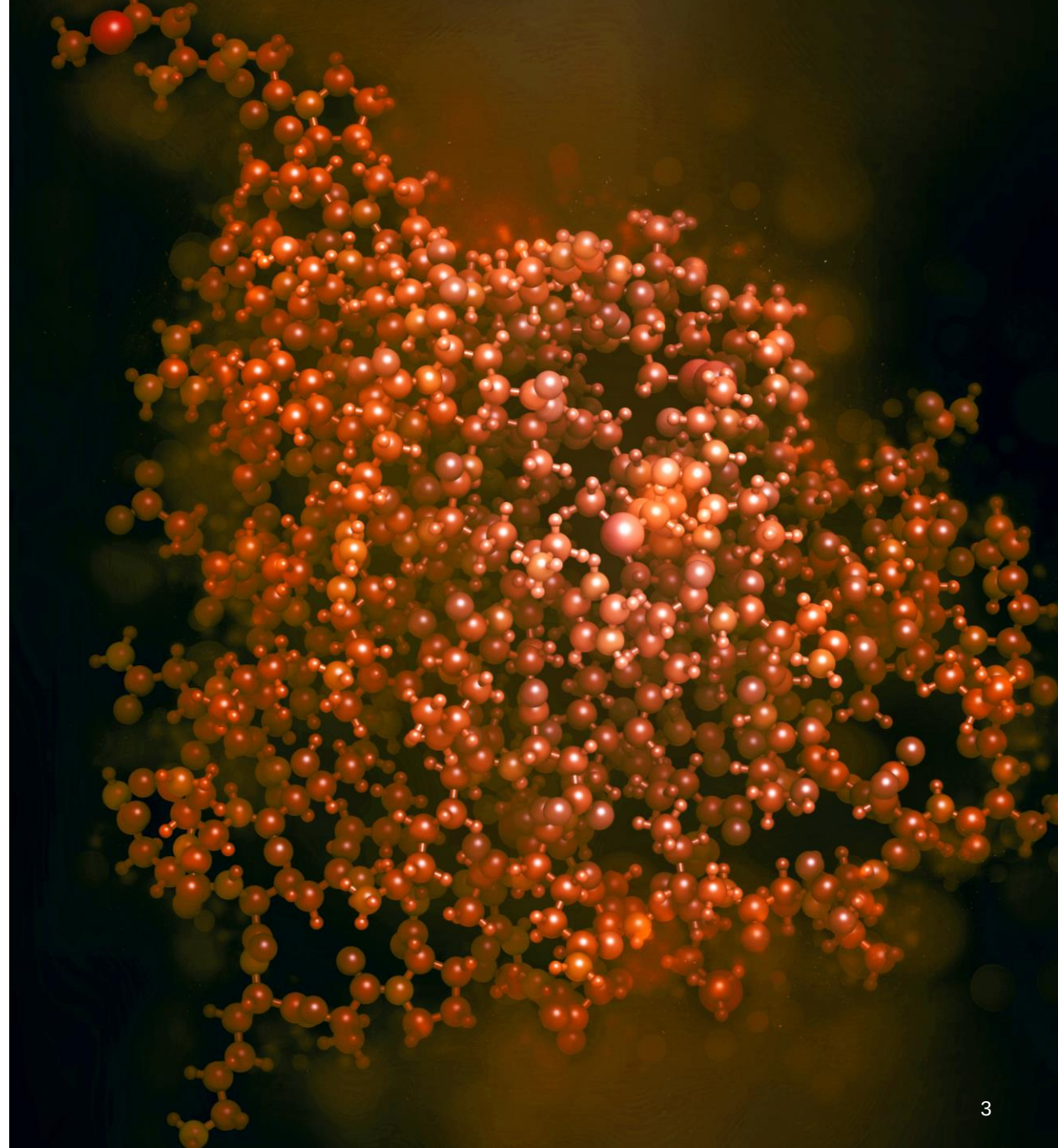
Today's Agenda

1. Unmet need in Solid Tumors

2. Interleukin-12 Overview

3. IL-12 Clinical Landscape

4. Q&A



Unmet Need in Solid Tumors

**"Cold" vs "Hot" Tumors &
Differences in Their TMEs**

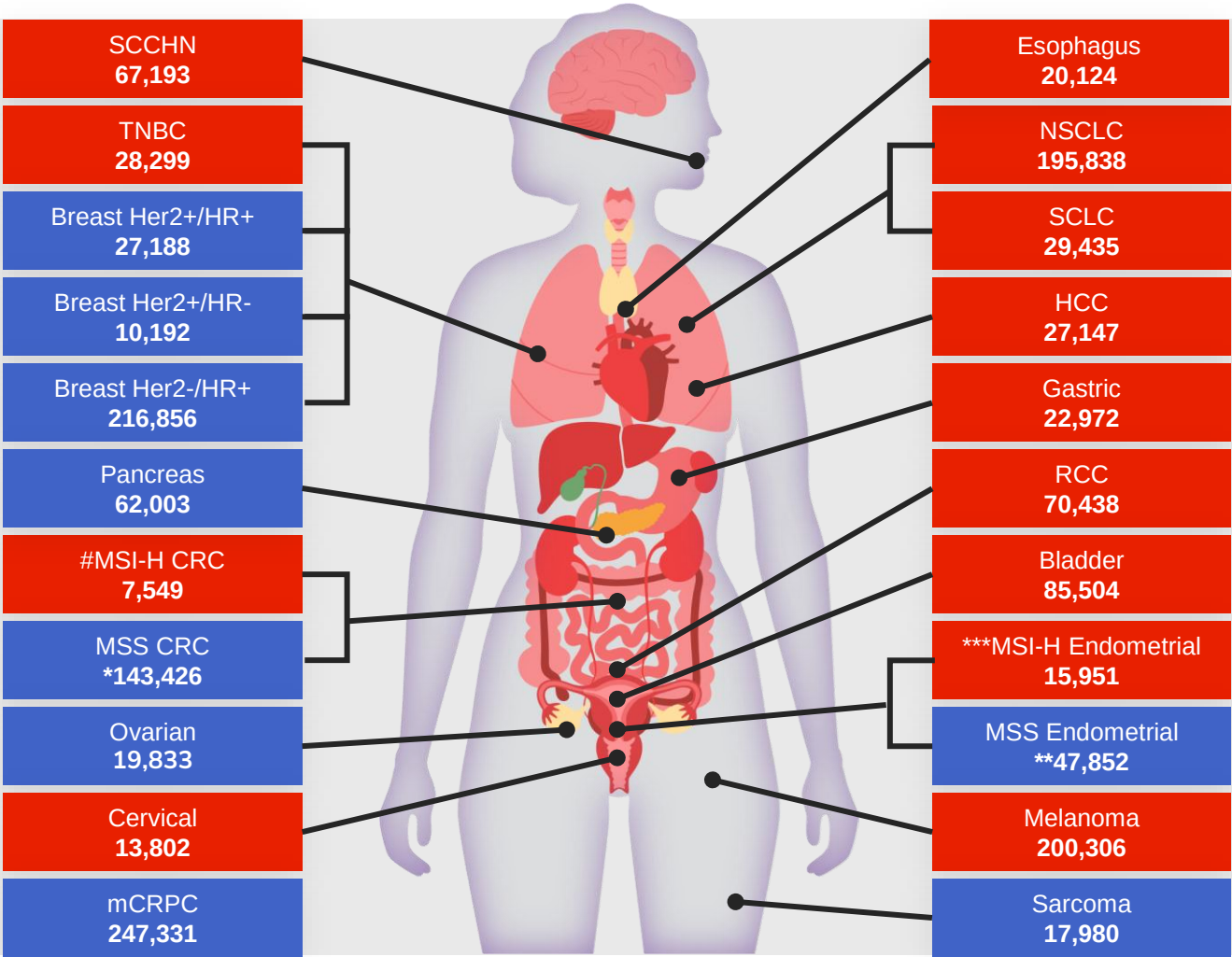
Dr. Jason Luke

Overwhelming Unmet Need Remains: 2022 US Incidence by Tumor Type and Category



Categories assigned based on approval of a checkpoint inhibitor (CPI) in that tumor and relative efficacy

Per Cerner Enviza CancerMPact Epidemiology Database

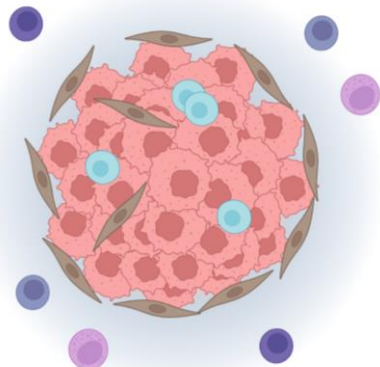


*Estimated by taking incidence CRC patients & removing 5% for MSI-H. # Estimated by taking 5% of incident CRC patients. **Estimated by taking incidence endometrial patients & removing 25% for MSI-H. ***Estimated by taking 25% of incident endometrial patients. CRC: colorectal cancer; HCC: hepatocellular lung cancer; NSCLC: non-small cell lung cancer; SCLC: squamous cell lung cancer; SCCHN: squamous cell cancer of head and neck; TNBC: triple negative breast cancer.

IL-12 can remodel cold tumor microenvironment towards a pro-inflammatory (hot) state that favors anti-tumor immunity

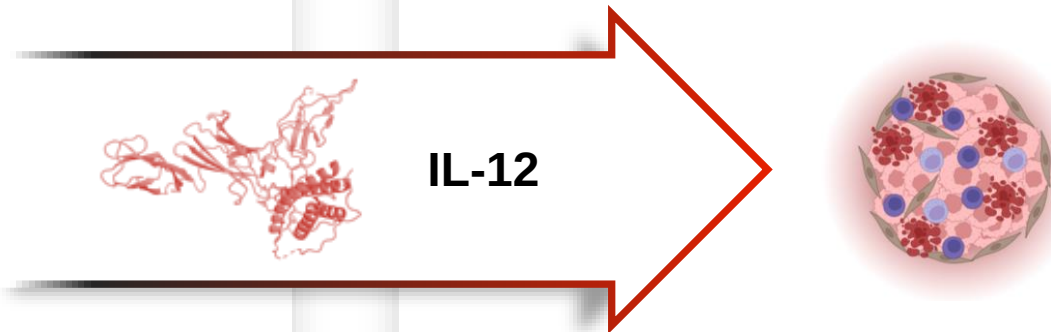
Cold Tumor

- Lack of CD8 T and NK cells within tumor
- Presence of immune suppressive cells (TREGs, MDSCs)
- Poor response to checkpoint inhibitors



Hot Tumor

- Abundant CD8 T and NK cells in tumor
- Pro-inflammatory microenvironment
- Improved prognosis and effective killing of tumor cells with immunotherapy treatment



Cancer cell



Fibroblast



Treg



Dying cancer cell



CD8+ T cell



NK cell

Adapted from "Cold vs Hot Tumors", by BioRender.com, 2022. Retrieved from <https://app.biorender.com/biorender-templates>. Barraondo et al., Clin. Cancer Res., 2018. Nguyen et al., Front. Immunol., 2020.

The Potential of IL-12 as a Therapeutic Agent

- IL-12 has ground-breaking potential as a potent IO therapeutic agent
- Poor tolerability has limited its clinical progress for decades
- No currently approved IL-12 agents

IL-12 Has Highly Compelling Biology for IO Applications



Exquisitely potent stimulator of NK and T cell cytotoxicity and INF γ production



Capable of polarizing CD4 T-cells towards Th1 phenotype, thus driving cellular immunity against infection and cancer



Robust INF γ induction results in broad remodeling of the TME towards a more immune-permissive environment



Demonstrated single agent objective responses in patients, **but poorly tolerated** (MTD <500 nanograms/kg on repeat dosing)

IL-12 Overview

Biology and Clinical Experience to Date

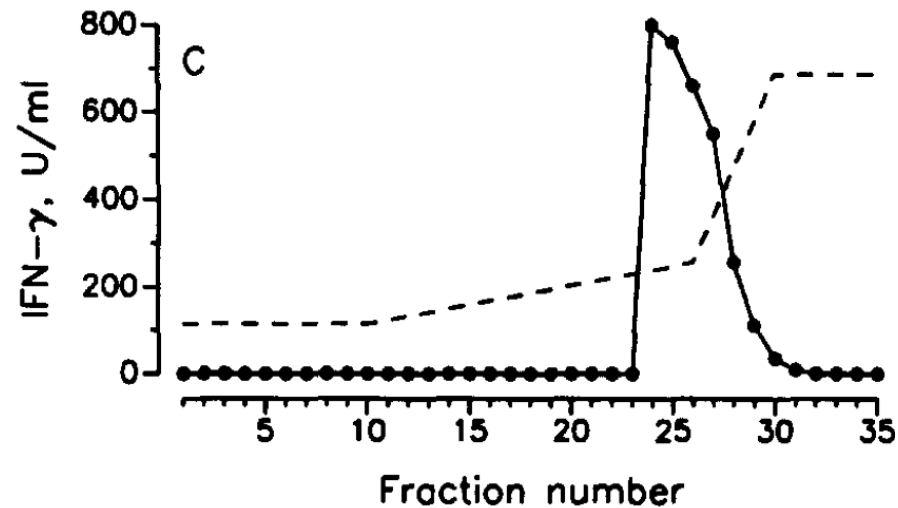
Dr. Jamie Spangler

IL-12: Discovered in 1989 as a potent NK cell stimulator

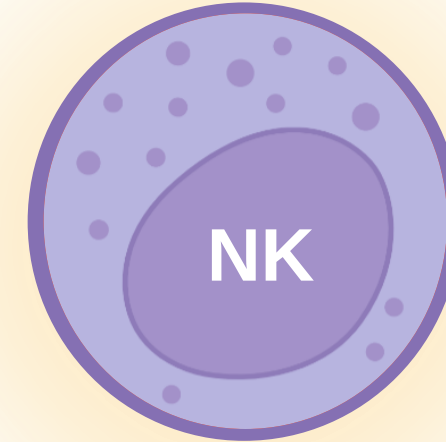
IDENTIFICATION AND PURIFICATION OF NATURAL KILLER CELL STIMULATORY FACTOR (NKSF), A CYTOKINE WITH MULTIPLE BIOLOGIC EFFECTS ON HUMAN LYMPHOCYTES

By MICHIKO KOBAYASHI,*† LORI FITZ,* MARY RYAN,*
RODNEY M. HEWICK,* STEVEN C. CLARK,* SUSAN CHAN,†
ROBERT LOUDON,† FREDERICK SHERMAN,†
BICE PERUSSIA,† AND GIORGIO TRINCHIERI†

From the *Genetics Institute, Inc., Cambridge, Massachusetts 02140; and
†The Wistar Institute of Anatomy and Biology, Philadelphia, Pennsylvania 19104

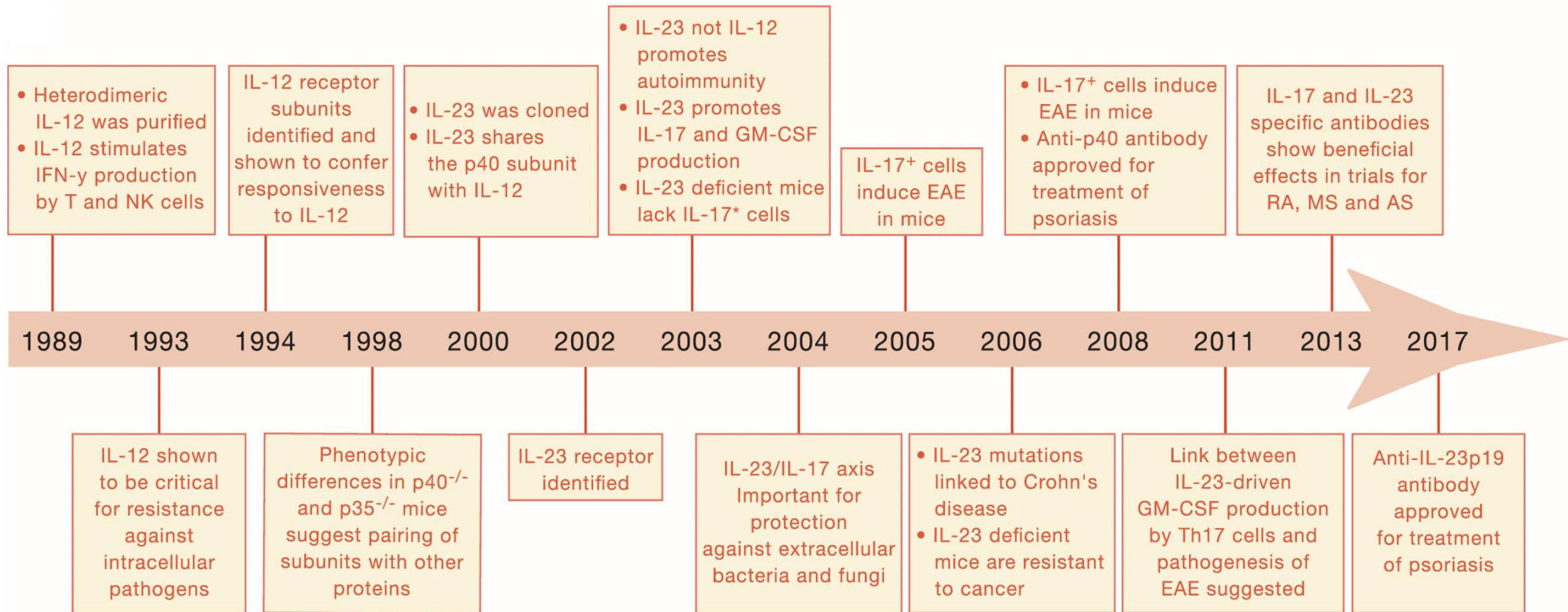


IL-12

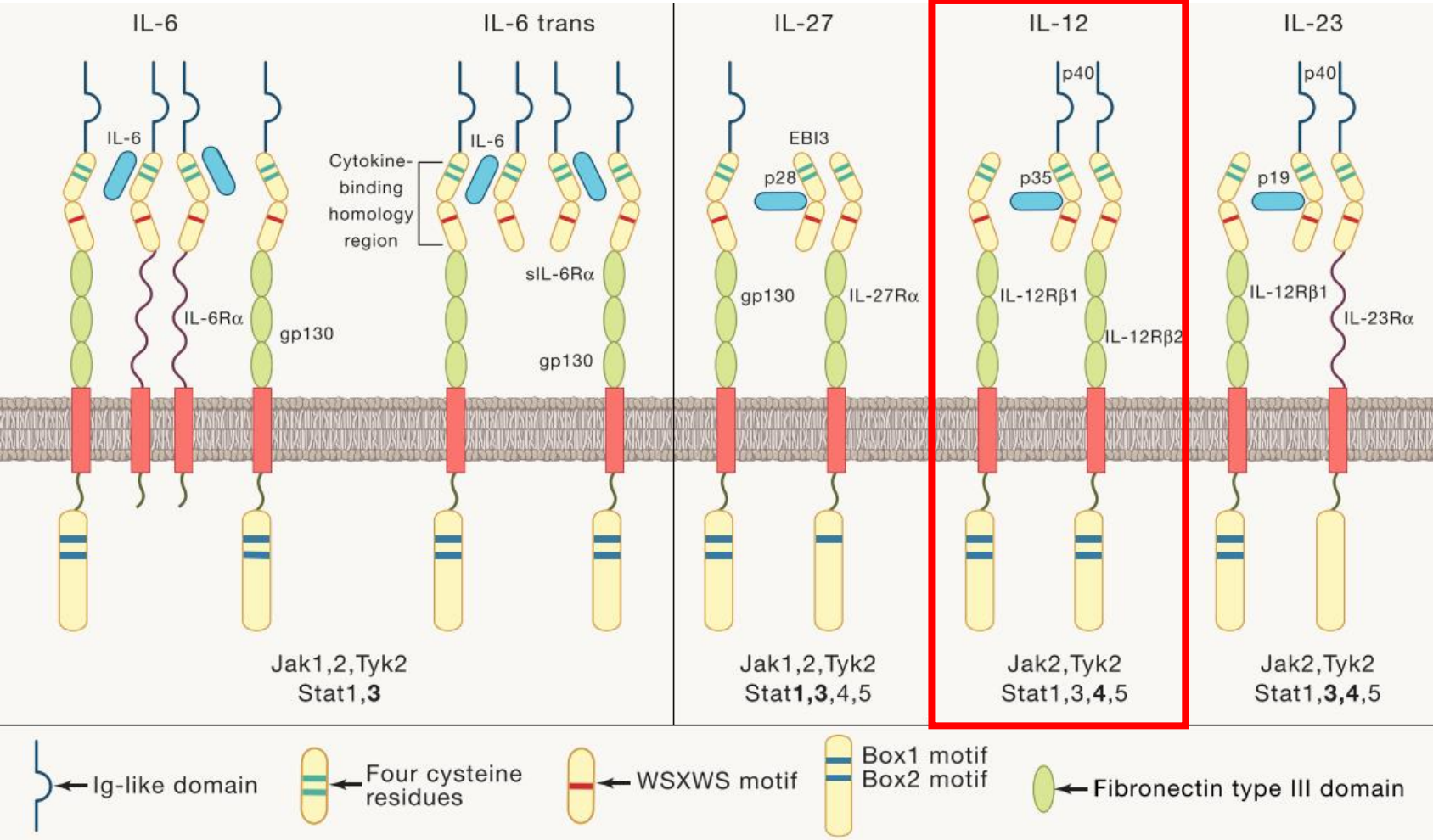


INF-γ

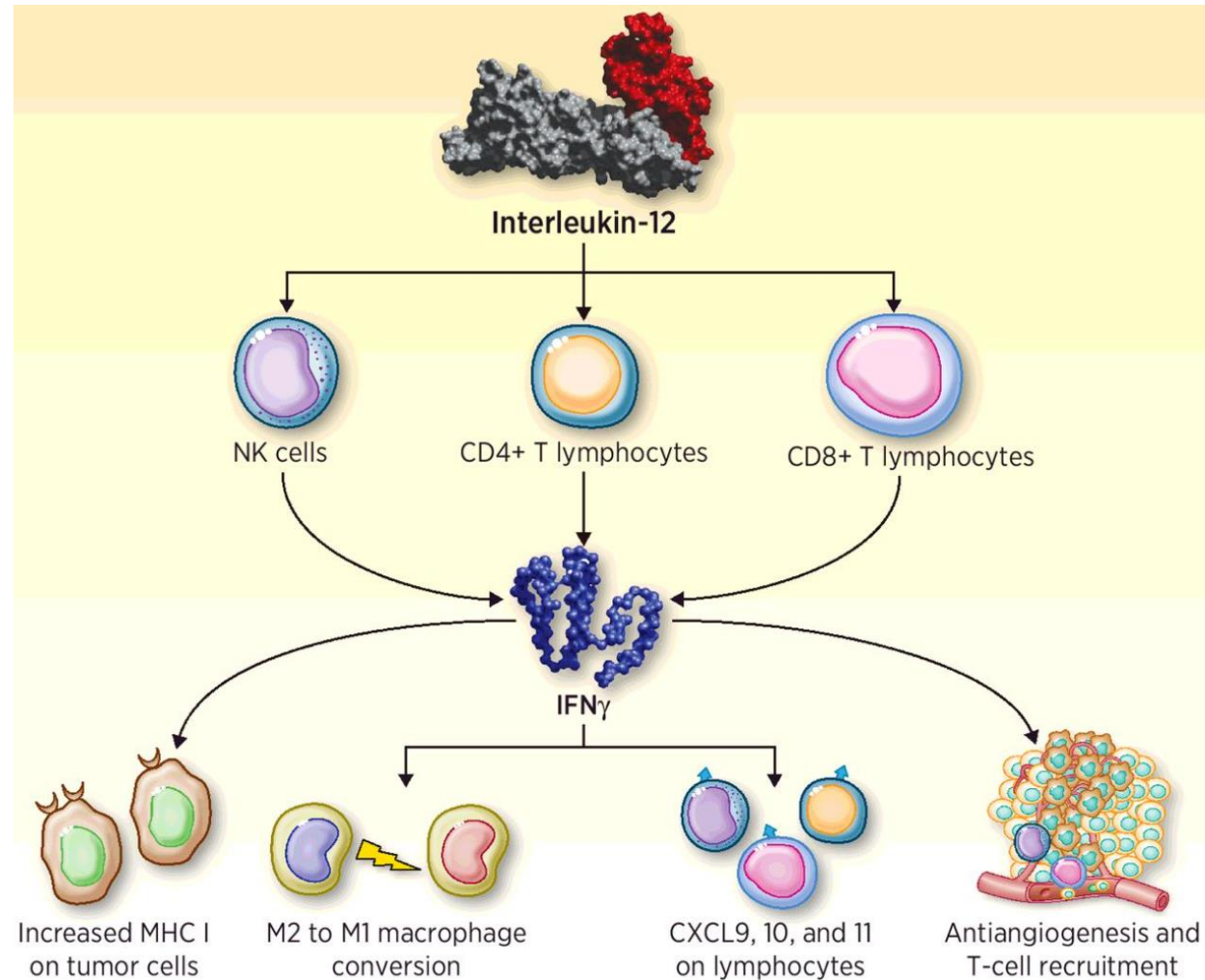
IL-12 Discovery and Development Advances Over Decades



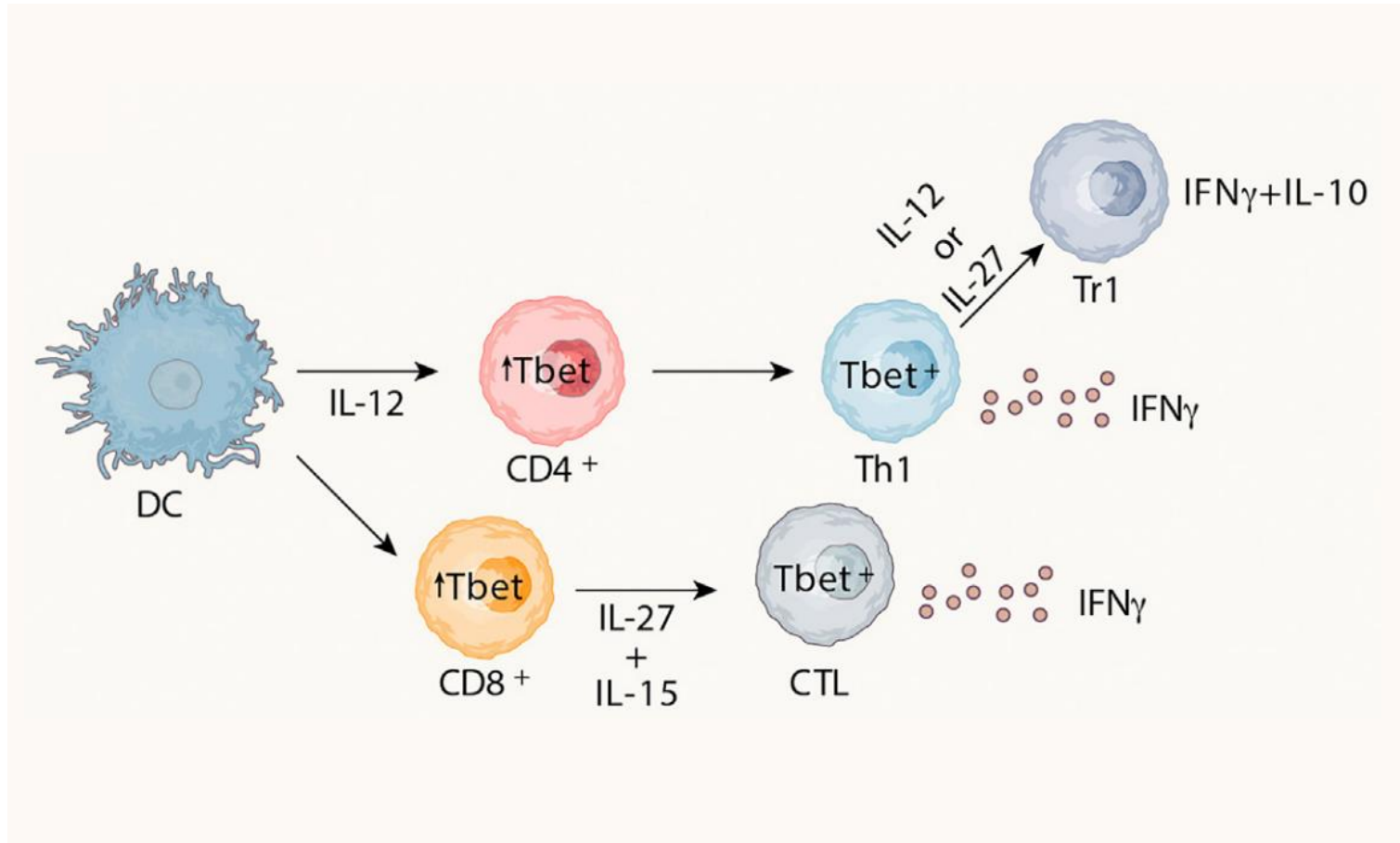
IL-12 is part of the IL-6/IL-12 Family of Cytokines



IL-12 is a heterodimeric cytokine and major driver of IFN- γ signaling



IL-12 Activates Innate and Adaptive Immunity



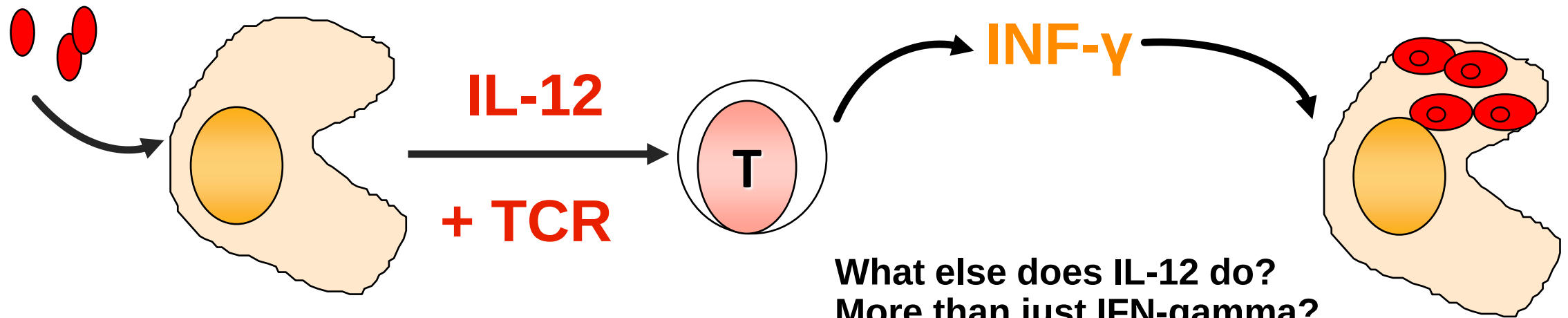
What else does IL-12 do?

- Differentiation, proliferation, cytotoxicity, IL-2 sensitivity and production, IL-10 etc.
- Anti-pathogen
- Anti-cancer responses
- CTL activity

Development of T_H1 CD4⁺ T Cells Through IL-12 Produced by *Listeria*-Induced Macrophages

Chyi-Song Hsieh, Steven E. Macatonia, Catherine S. Tripp, Stanley F. Wolf, Anne O'Garra, Kenneth M. Murphy*

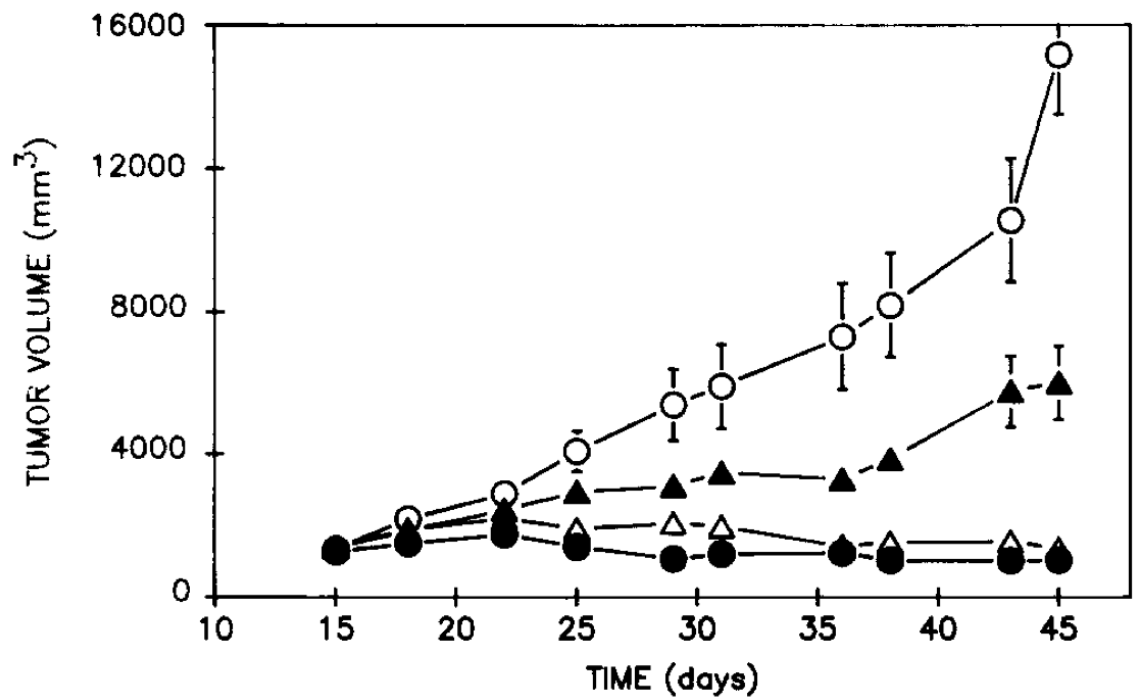
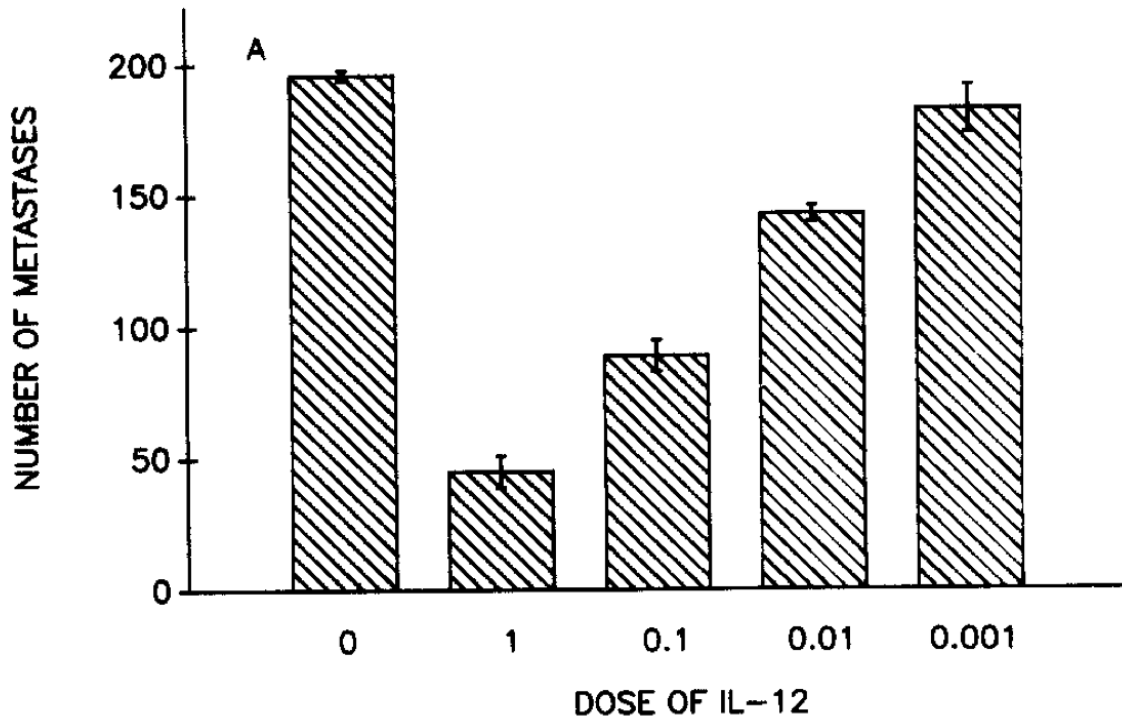
- Anti-pathogen
- Anti-cancer responses
- CTL activity



**What else does IL-12 do?
More than just IFN-gamma?**

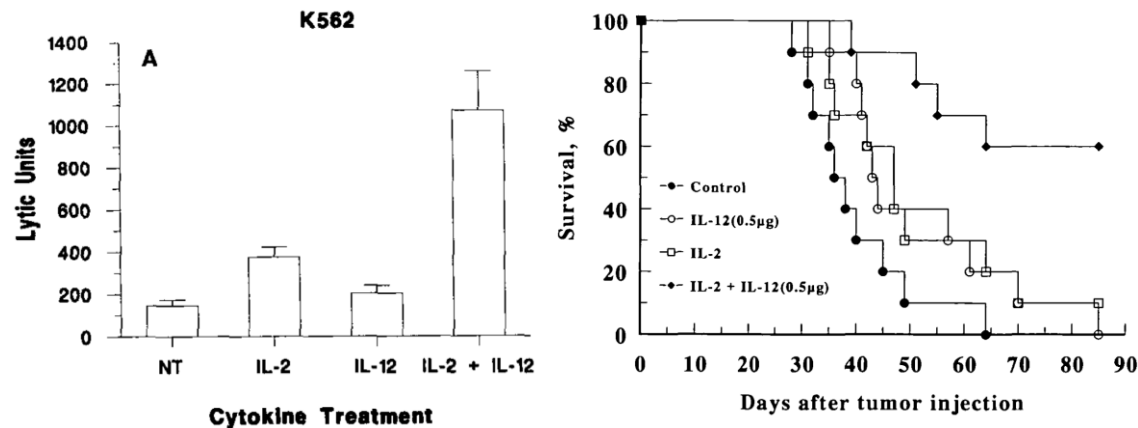
- Differentiation, proliferation, cytotoxicity, IL-2 sensitivity and production, IL-10 etc.

IL-12 Exhibits Anti-tumor Activity

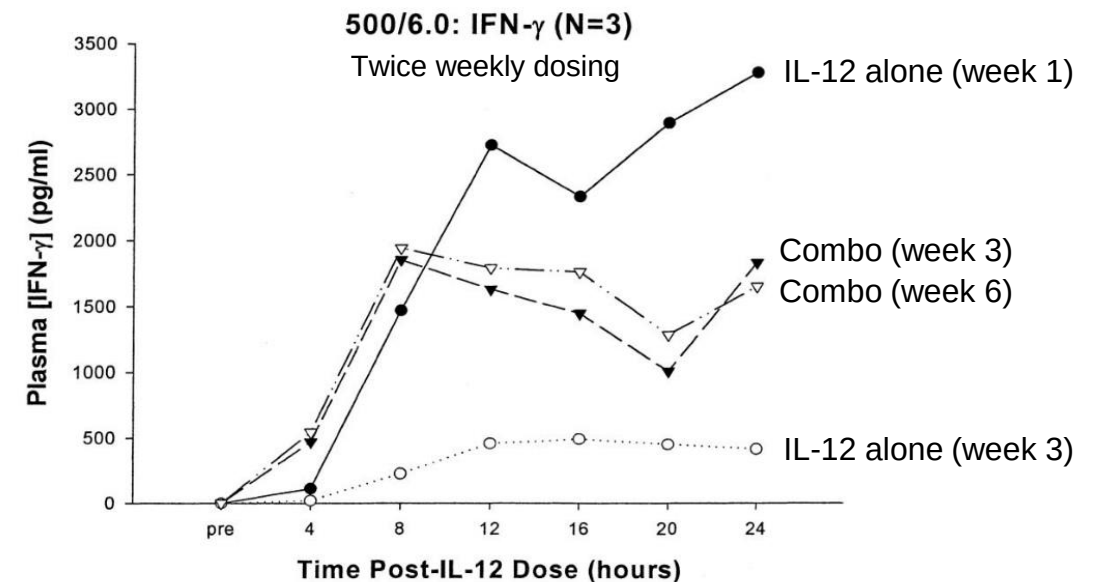


Combinations of IL-2 and IL-12 Potentiate Anti-tumor Immunity

Combination of IL-2 and IL-12 results in enhanced cytotoxicity and anti-tumor activity (pre-clinical data)



Addition of IL-2 to IL-12 reduces negative feedback and results in persistent INF γ production (human clinical data)

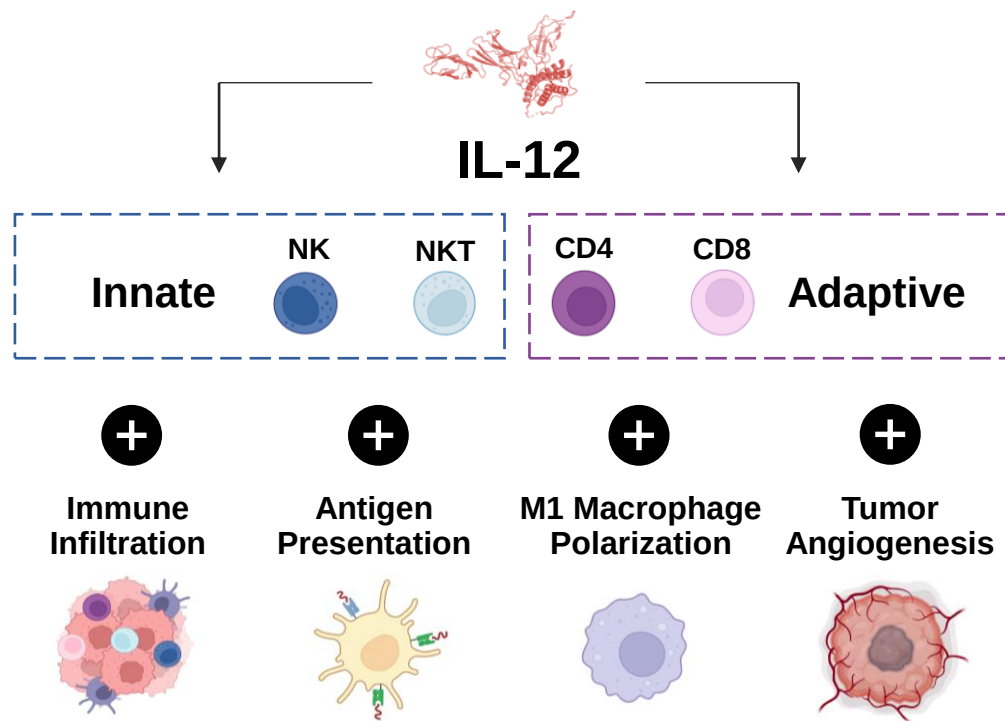


What is the future?

“Surely, if lymphokines prove to be valuable therapeutic agents, new approaches to a physiological type of drug delivery, mimicking normal modes of action, will be needed to improve their efficacy and to avoid the side effects which are almost certain to occur from systemic administration” ([Paul, 1989](#)).

IL-12: Compelling biology for IO applications – but limited without engineering

IL-12: A key cytokine bridging innate and adaptive cellular immunity



IL-12 properties highlight potential in IO, but tolerability remains limiting

- Exquisitely potent stimulator of NK and T cell cytotoxicity and INF γ production.¹⁻²
- Capable of polarizing naïve CD4 T-cells towards Th1 phenotype, thus driving cellular immunity against infection and cancer.¹⁻²
- Robust INF γ induction results in broad remodeling of the TME towards a more immune-permissive environment.¹⁻²
- Demonstrated single agent objective responses in patients, but poorly tolerated (MTD <500 ng/kg).³
- Synergy with TIL and α -PD-1 preclinically.⁴⁻⁵

IL-12 Clinical Landscape

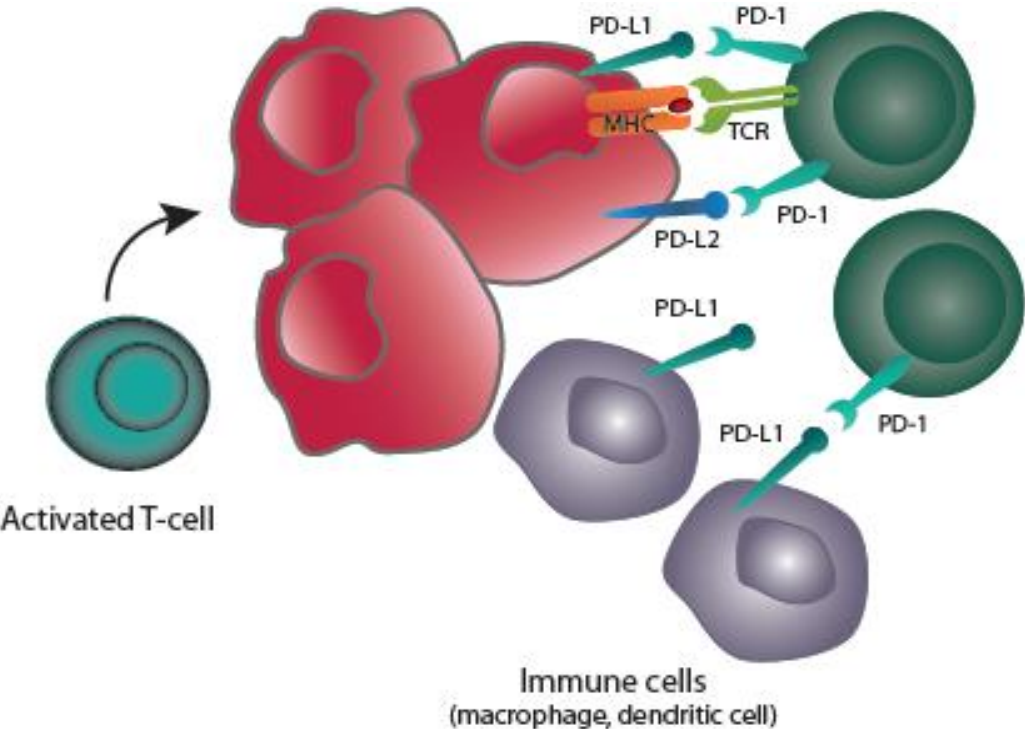
Dr. Jason Luke

IL-12

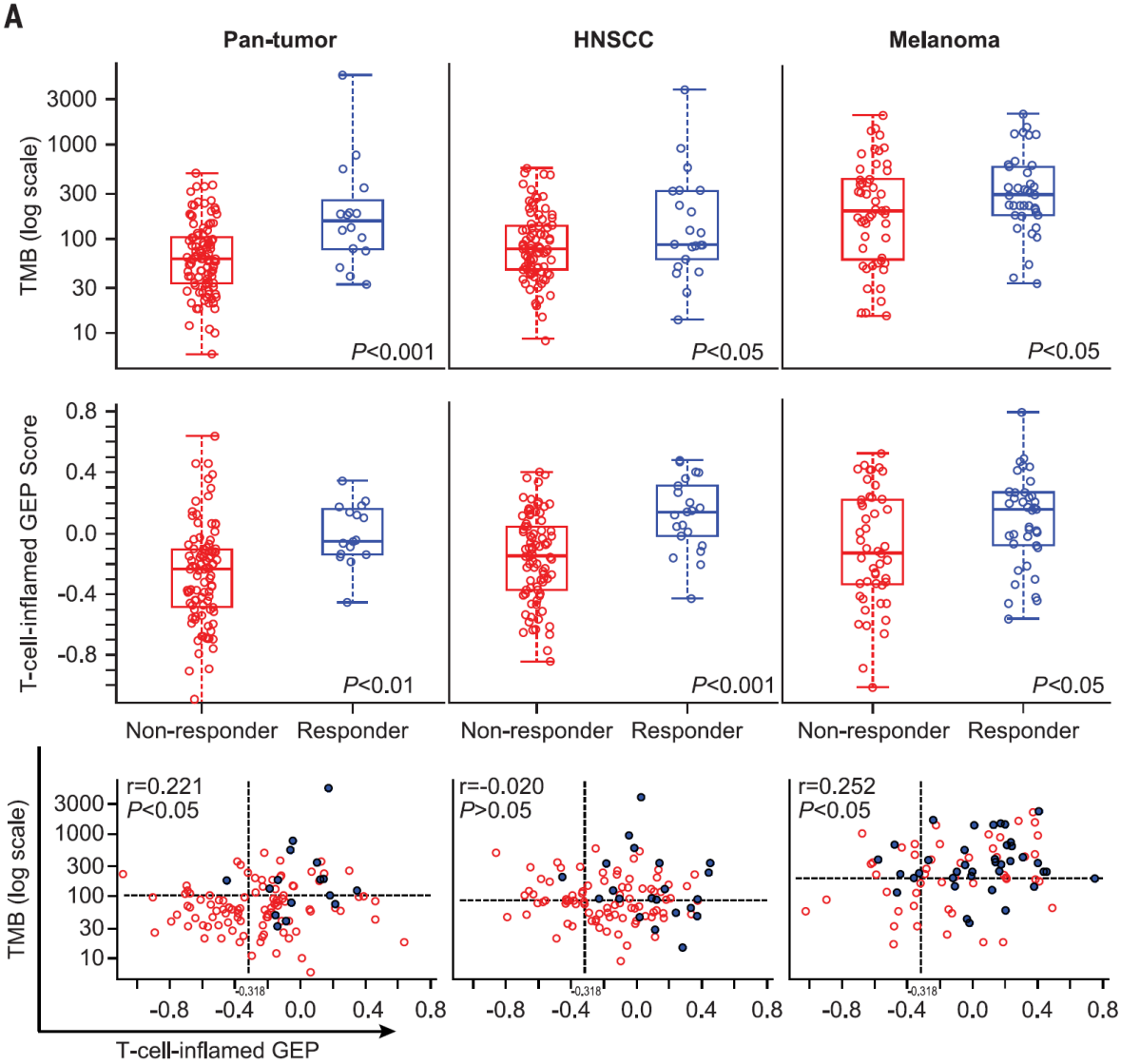
IL-12

IL-12

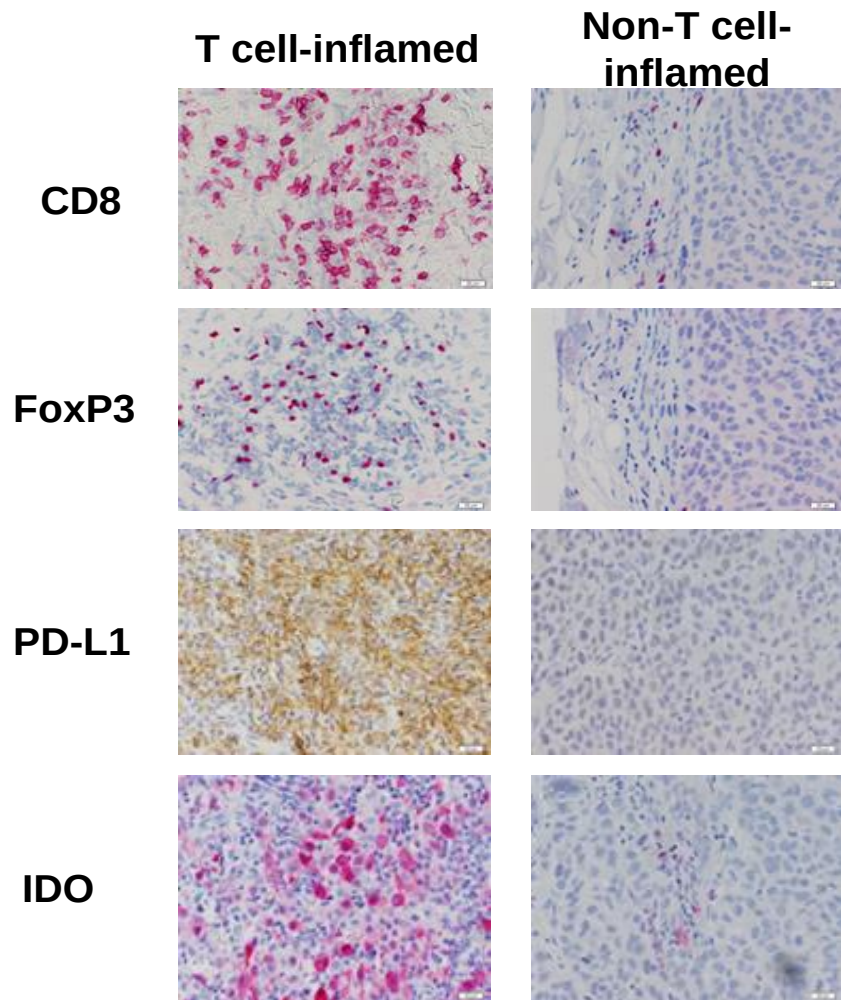
T cell-inflamed gene expression and tumor mutational burden/tumor neoantigenicity predict immunotherapy treatment outcomes



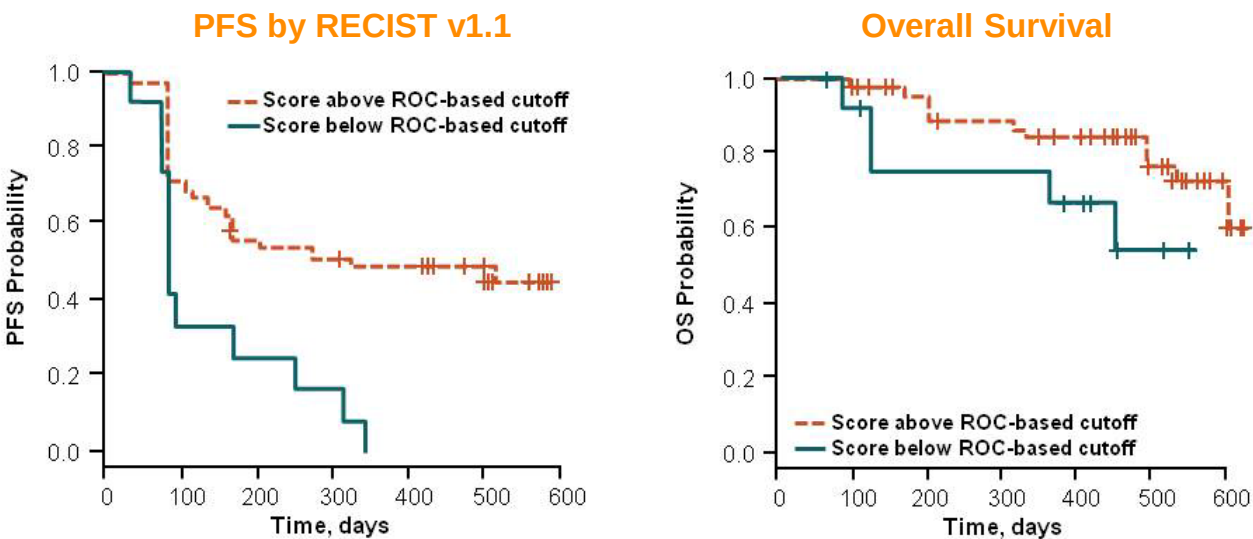
Luke and Ott. *Oncotarget*. 2015; Cristescu et al. *Science* 2018



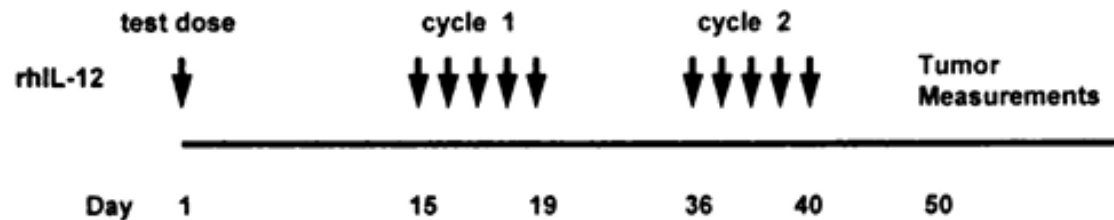
Immunobiology of T cell-inflamed & non-T cell-inflamed tumor microenvironment



PFS and OS in Patients With Melanoma and IFN γ Signature Score Above and Below the Cutoff



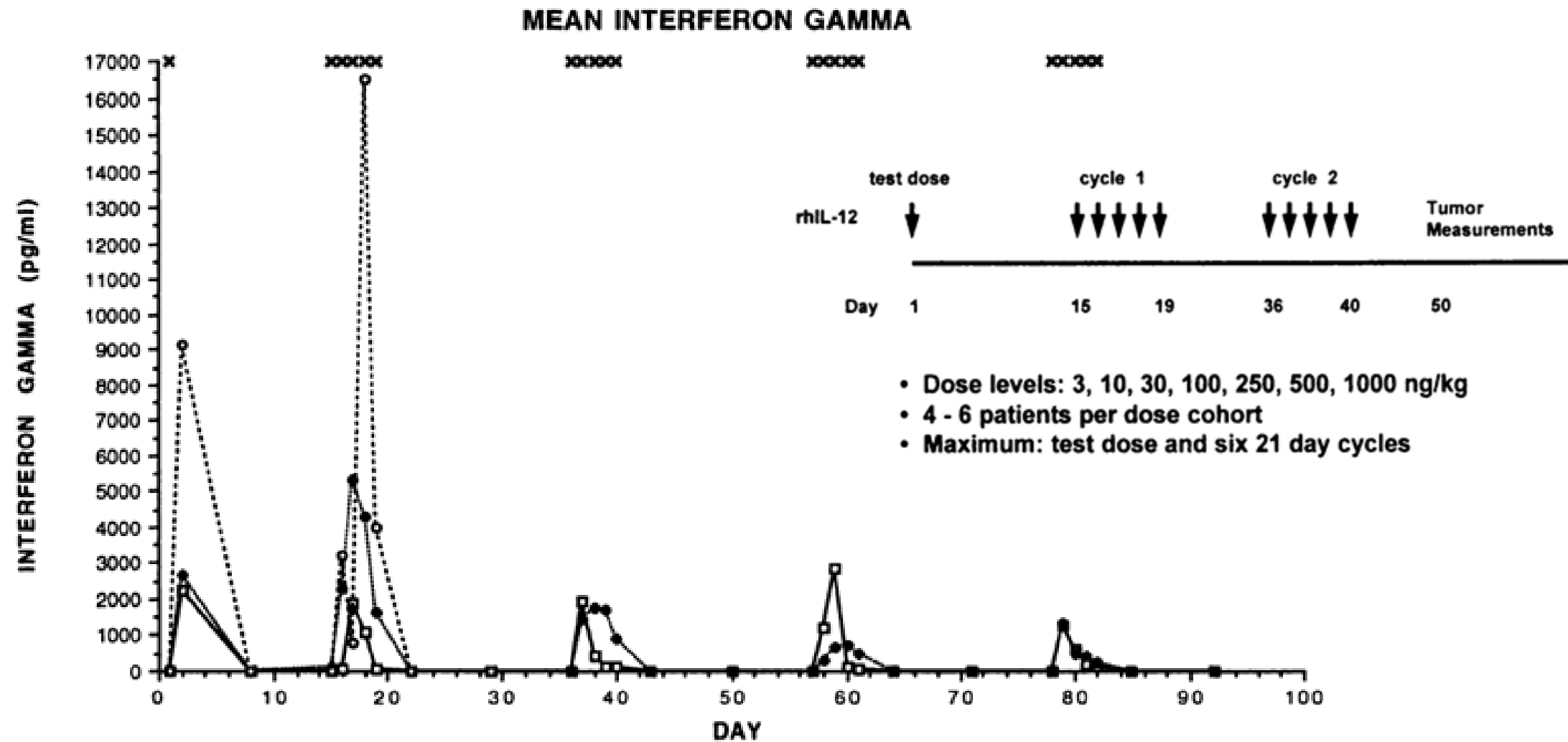
Phase I Study of IV Recombinant Human IL-12 was Highly Toxic



- Dose levels: 3, 10, 30, 100, 250, 500, 1000 ng/kg
- 4 - 6 patients per dose cohort
- Maximum: test dose and six 21 day cycles

- Common toxicities included:
 - Fever/chills
 - Fatigue
 - Nausea
 - Vomiting
 - Headache
- Fever first observed at 3ng/kg dose level, typically occurred 8-12 h after rhIL-12 administration and was incompletely suppressed with non-steroidal drugs.
- Transaminase elevation were dose limiting

Progressive decline in IFN-γ after exposure to IV rhIL-12



Atkins et al. *Clin Can Res.* 1997

Intermittent rhIL-12 to limit IFN- γ suppression

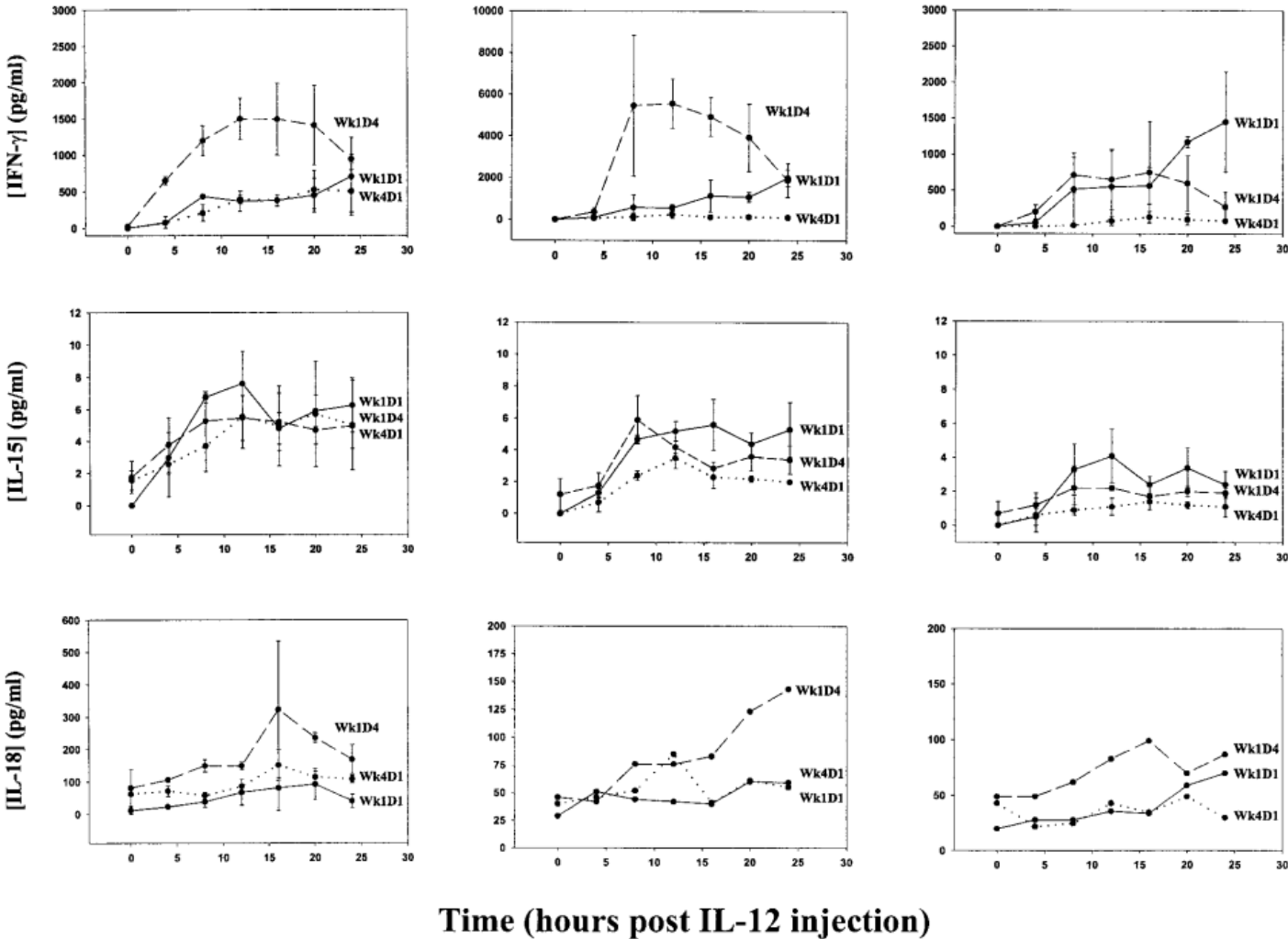
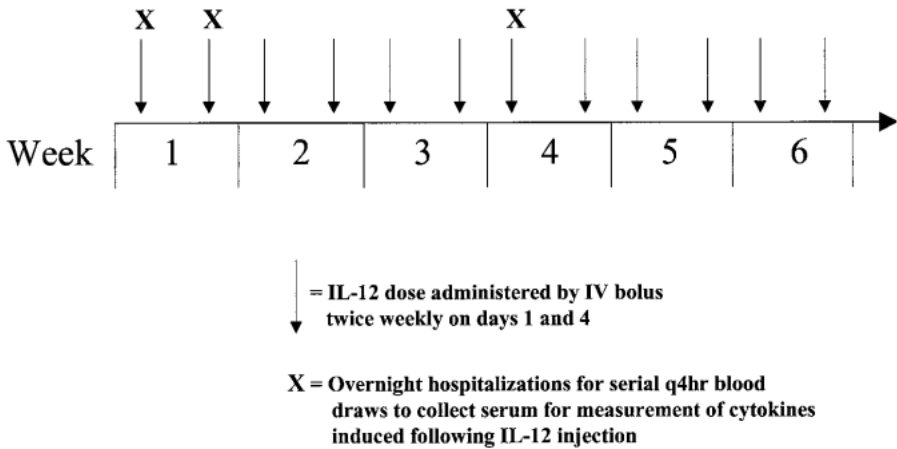
1678 Vol. 6, 1678-1692, May 2000

Clinical Cancer Research

Phase I Trial of Twice-Weekly Intravenous Interleukin 12 in Patients with Metastatic Renal Cell Cancer or Malignant Melanoma: Ability to Maintain IFN- γ Induction Is Associated with Clinical Response¹

Jared A. Gollob,² James W. Mier, Korina Veenstra, David F. McDermott, Daniel Clancy, Marguerite Clancy, and Michael B. Atkins

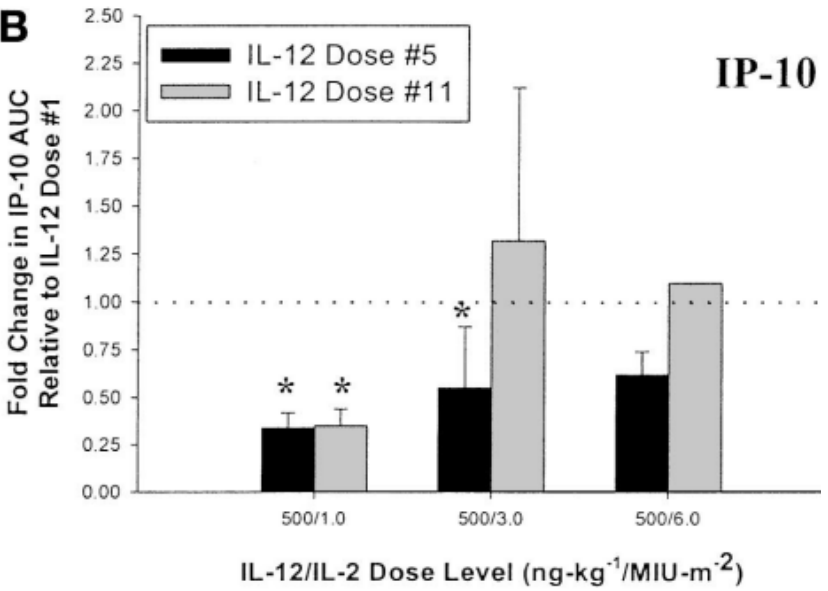
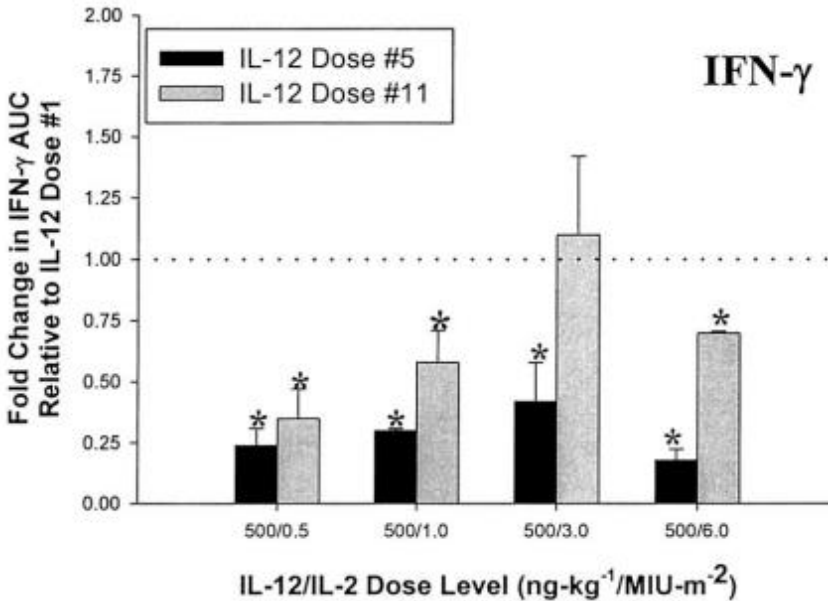
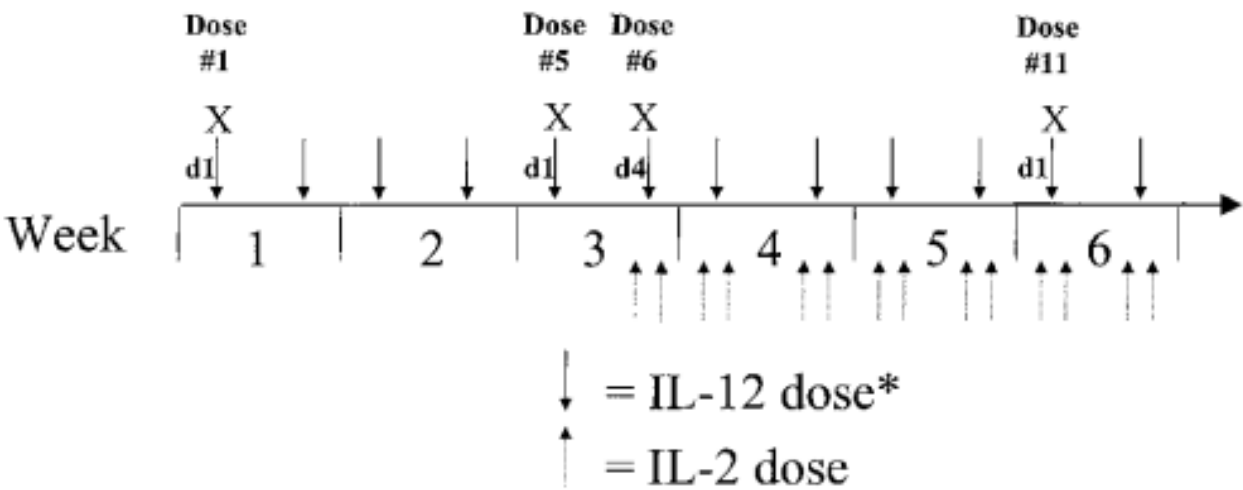
overcome *in vitro* through combined stimulation with IL-12 plus IL-2. These findings show that the chronic administration of twice-weekly i.v. rhIL-12 is well-tolerated, stimulates the production of IL-12 costimulatory cytokines and IFN- γ , and can induce delayed tumor regression. Strategies aimed



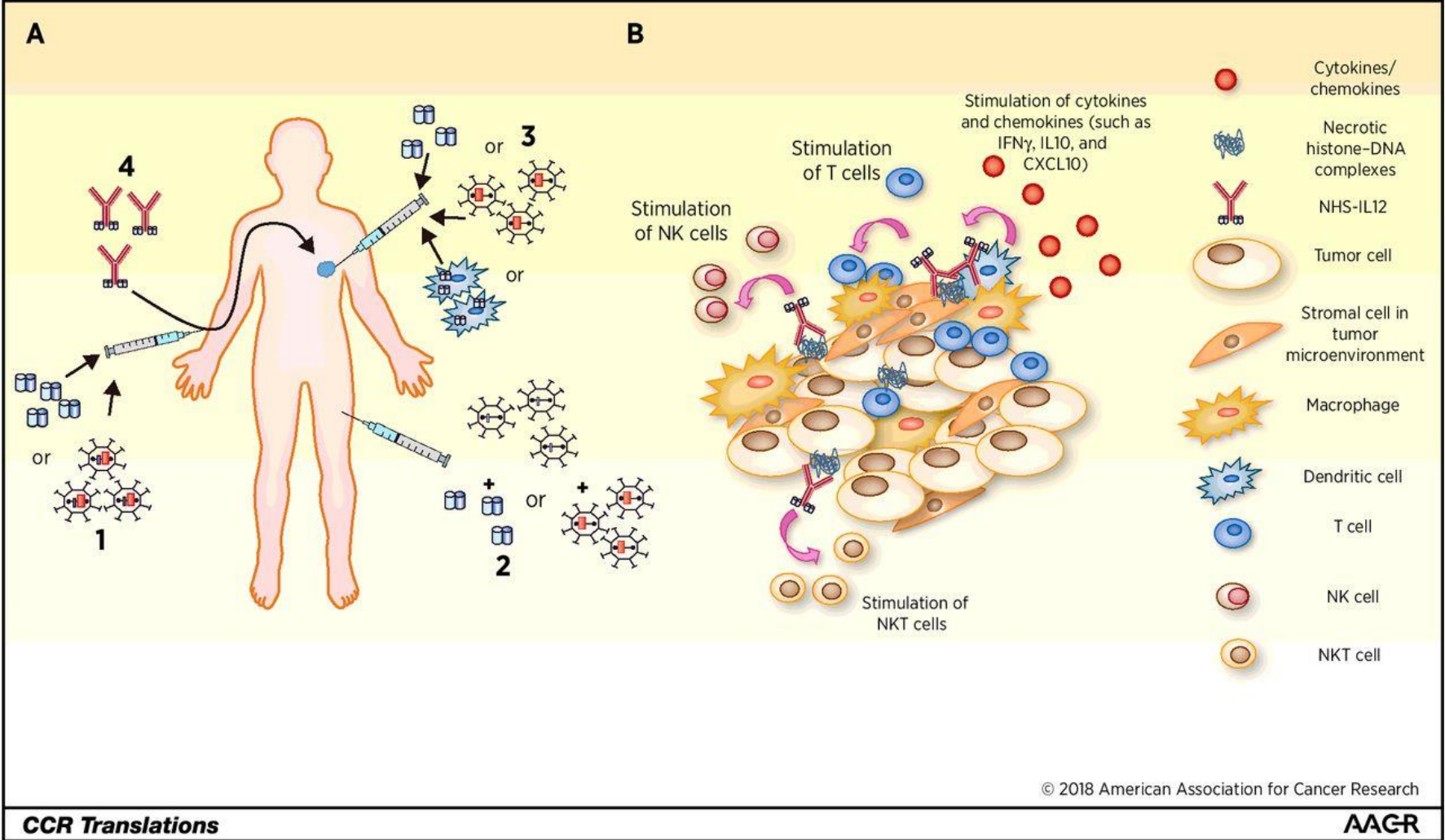
IL-2 + rhIL-12 enhances IFN- γ

Phase I Trial of Concurrent Twice-Weekly Recombinant Human Interleukin-12 Plus Low-Dose IL-2 in Patients With Melanoma or Renal Cell Carcinoma

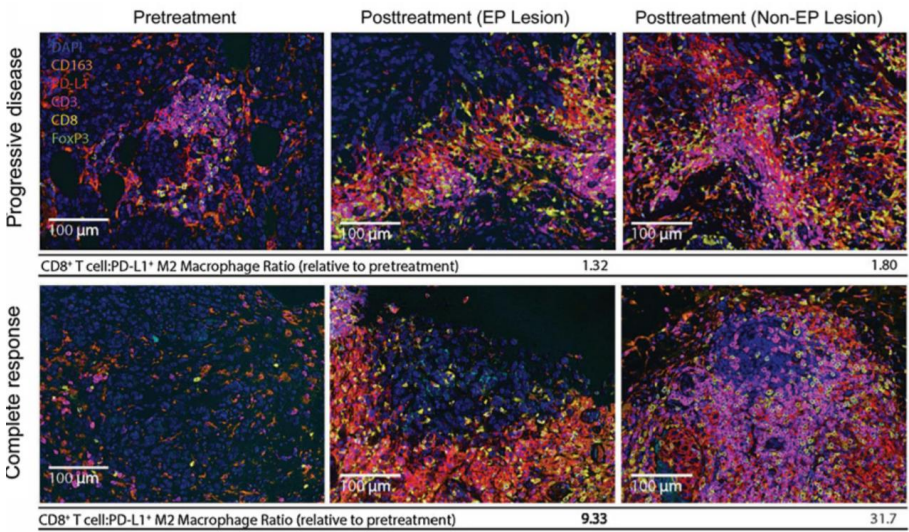
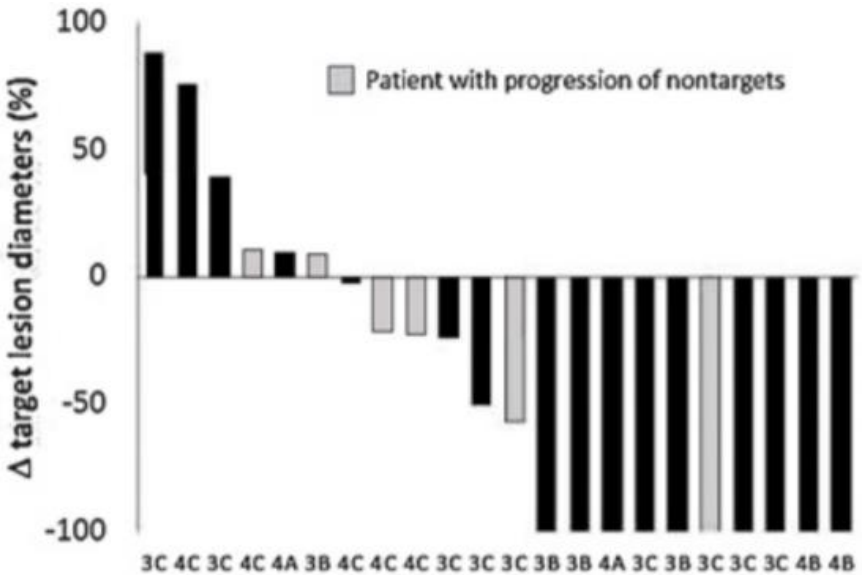
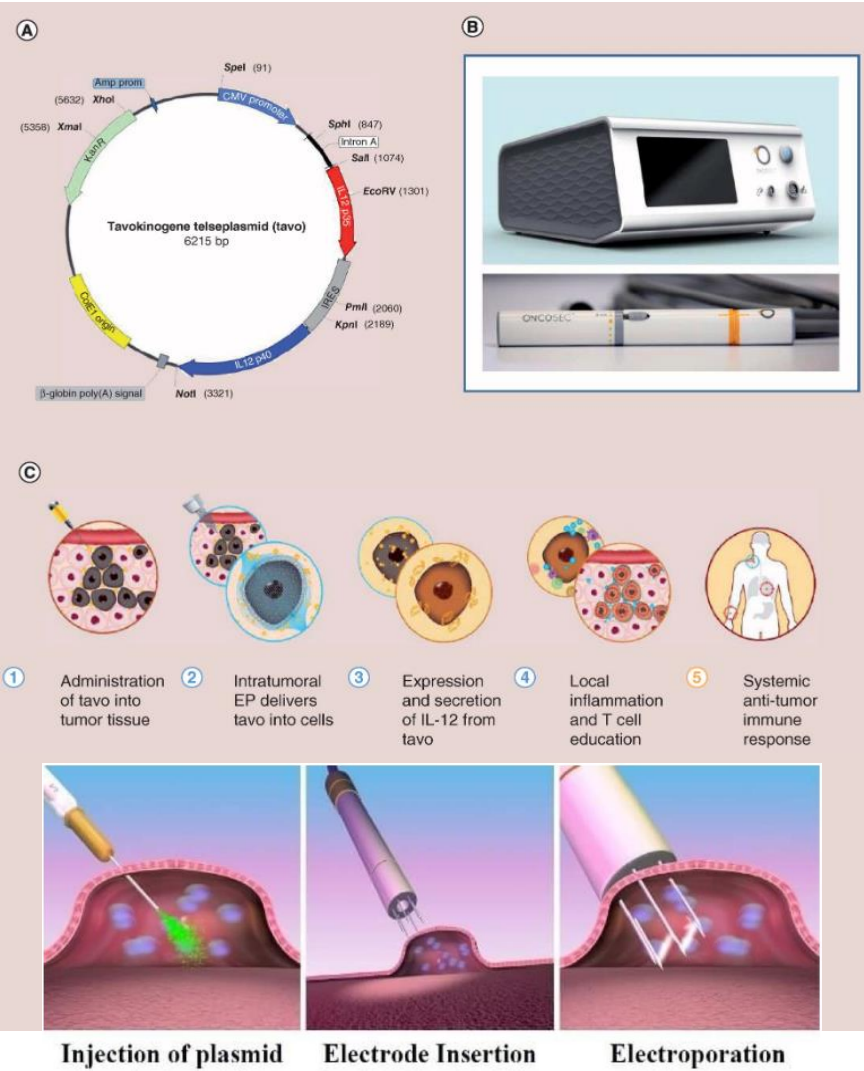
By Jared A. Gollob, Korina G. Veenstra, Robert A. Parker, James W. Mier, David F. McDermott, Daniel Clancy, Linda Tutin, Henry Koon, and Michael B. Atkins



Clinical approaches for delivery of IL-12 and its impact on the tumor microenvironment

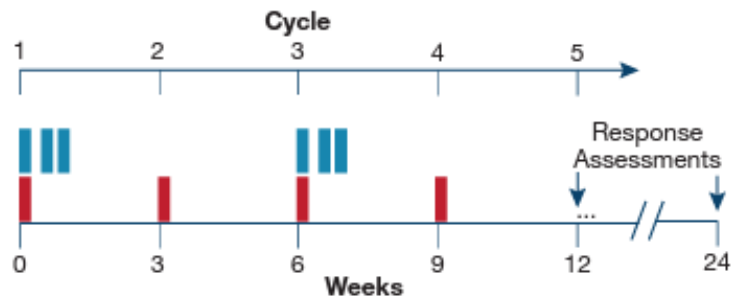


Electroporation of plasmid IL-12 may overcome resistance



TAVO pIL12 + pembro in PD1 refractory melanoma

KEYNOTE-695 Trial Design



IT-TAVO-EP days 1, 5, 8

Pembro 200 mg day



Unresectable or metastatic melanoma treated with anti-PD-1 (alone or in combination) for 12 weeks prior to study entry

- Must have confirmed RECIST v1.1 progression with no intervening therapy prior to study entry
- RECIST v1.1 measurable disease and at least one anatomically distinct lesion accessible for electroporation

Planned Enrollment N=100

- Pembrolizumab 200 mg IV every 3 week
- pIL12 (TAVO) administered to at least one accessible lesion(s) on days 1, 5, and 8 every 6 weeks until no more lesions to treat

All Responses are by RECIST v1.1

Primary Endpoint: ORR by blinded independent review
Secondary Endpoints: Investigator assessed ORR, DOR, PFS, iPFS, iORR, OS

Table 1. Patient Demographics and Disease Characteristics

	N=56			N=56	
Sex			LDH*		
Female	25	(44.6%)	Normal	42	(75.0%)
Male	31	(55.4%)	Elevated >ULN*	11	(19.6%)
			Unknown	3	(5.4%)
Age (years)			Stage		
Median (Range)	66	(30, 86)	III (B,C,D)	9	(16%)
ECOG Performance Status			IV (a,b)	30	(53.6%)
0	35	(62.5%)	IV (c,d)	17	(30.4%)
1	21	(37.5%)	Number of Target + Non Target Lesions at Baseline		
BRAF Status			>3	43	(76.8%)
Mutant (V600E, V600K)	12	(21.4%)	Mean	8.9	(range 1, 169)
Wild-Type or non-V600 mutation	44	(78.6%)	Number of Prior Therapies		
			1 line	28	(50%)
			2-3 lines	13	(23.2%)
			≥ 4 lines	15	(26.8%)
			Median	1.5	
			Mean	2.9	(range 1, 17)
			No. of Pts with Prior Ipilimumab	15	(27%)
			Median time of anti-PD-1 exposure prior to study entry with no more than a 12 week gap in between treatments	5.3	months
			Median time from last dose of anti-PD-1 to study treatment (Cycle 1 Day 1)	1.2	months

*Based on maximum value prior to dosing 2 patients with ≥ 2x ULN

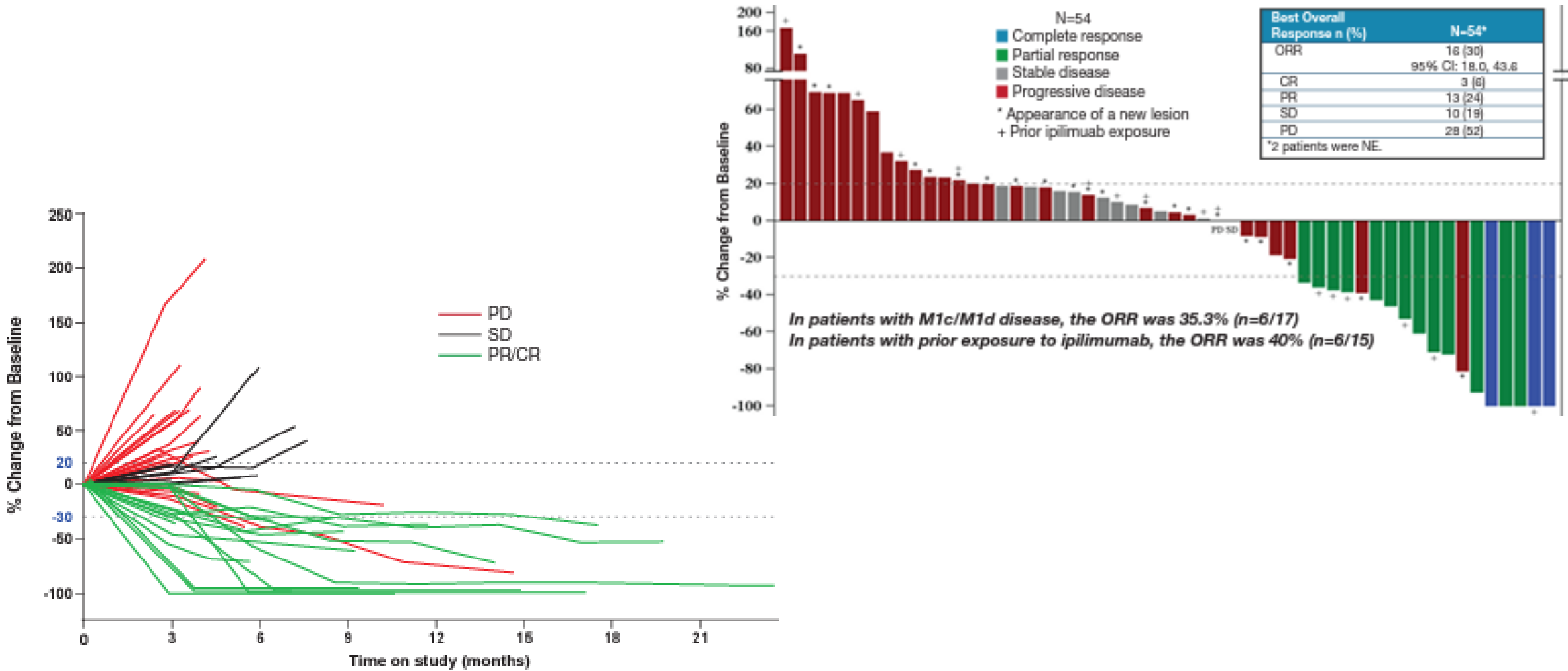
Table 2. Treatment-Related Adverse Events (TRAEs) > 5% of Patients

	All Grades n (%)				
	(N=56)	Grade 1/2	Grade 3	Grade 4	Grade 5
Patients with at least one TRAE	43 (76.8%)	40 (71.4%)	3 (5.4%)	—	—
Fatigue	15 (26.8%)	15 (26.8%)	—	—	—
Procedural pain	13 (23.2%)	13 (23.2%)	—	—	—
Diarrhea	11 (19.6%)	11 (19.6%)	—	—	—
Nausea	6 (10.7%)	6 (10.7%)	—	—	—
Rash	6 (10.7%)	6 (10.7%)	—	—	—
Abdominal pain	4 (7.1%)	4 (7.1%)	—	—	—
Arthralgia	4 (7.1%)	4 (7.1%)	—	—	—
Injection site pain	4 (7.1%)	4 (7.1%)	—	—	—
Myalgia	4 (7.1%)	4 (7.1%)	—	—	—
Pruritus	4 (7.1%)	4 (7.1%)	—	—	—
Administration site reaction	3 (5.4%)	3 (5.4%)	—	—	—
Aspartate aminotransferase increased	3 (5.4%)	3 (5.4%)	—	—	—
Cellulitis	3 (5.4%)	2 (3.6%)	1 (1.8%)	—	—
Dyspnea	3 (5.4%)	3 (5.4%)	—	—	—
Upper respiratory tract infection	3 (5.4%)	3 (5.4%)	—	—	—
Enteritis	1 (1.8%)	—	1 (1.8%)	—	—
Lichen planus	1 (1.8%)	—	1 (1.8%)	—	—

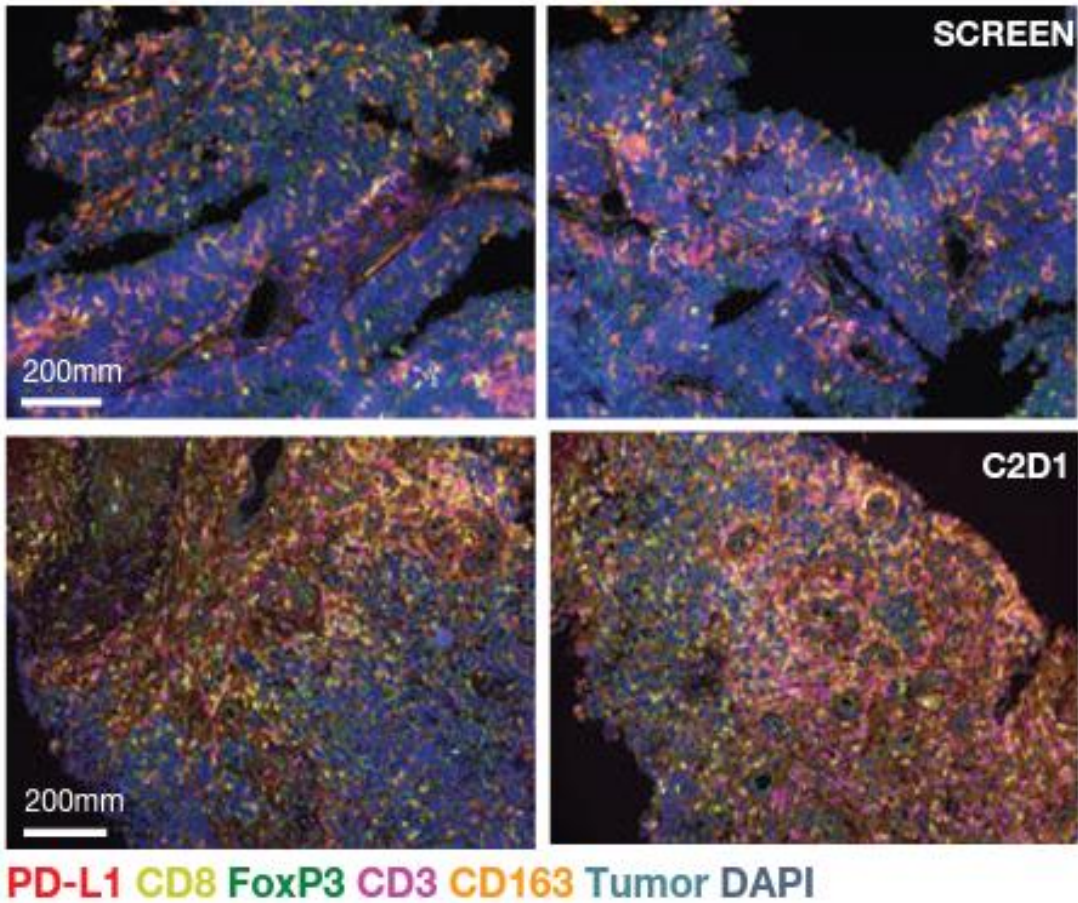
Patients who experienced the same TEAE on more than one occasion (based on the specific category) are counted once in each relevant category (n). Adverse event terms are coded using the Medical Dictionary for Regulatory Activities (MedDRA) Version 19.0.

- The median time for pIL-12 (TAVO™) electroporation treatment was 11 minutes (range 1,76)

TAVO pIL12 + pembro shows durable responses in PD1 refractory melanoma



Patients With Immunologically “Cold” Tumors at Screen Quickly Get “Hot” With a Productive Immune Infiltrate



Cell Type	Screen (cells/mm ²)	C2D1 (cells/mm ²)	Fold-change (C2D1/S)
CD8 ⁺	133.30	738.91	5.54
CD4 ⁺	376.24	260.57	0.69
CD163 ⁺	789.02	1135.15	1.44
FoxP3 ⁺	53.84	35.69	0.66
PD-L1 ⁺	173.01	3655.78	21.13

Novel systemic delivery approaches overcome limitations and provides possible synergies

Existing IL-12 modalities

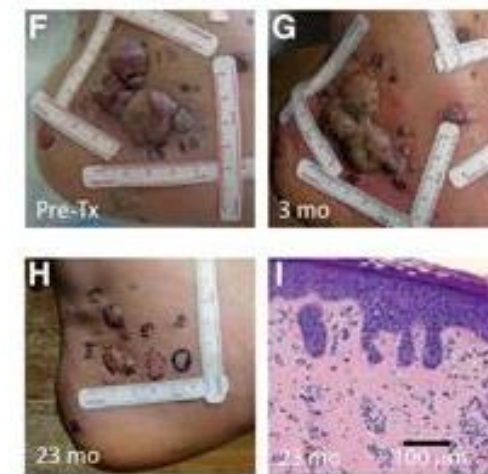
Tavokinogene telseplasmid

- Requires intra-tumoral administration.¹
- Activity observed in melanoma and Merkel cell carcinoma both as single agent and in combination with anti-PD-1.²⁻⁴
- Response is associated with antigen-specific circulating immune responses.⁵

Other approaches: lipid-nanoparticle mRNA encoding IL-12⁶, NDV encoding IL-12⁷ and gene therapy approaches⁸.

Limitations and need for systemic administration

- Local administration is effective in injected lesions, but distant effects are rare.
- Greatest unmet need for IL-12 are often in tumors with inaccessible lesions.



¹Canton DA, Immunotherapy 2017; ²Algazi AP, Clin Cancer Res 2020; ³Algazi AP, Ann Oncol 2020; ⁴Bhatia S, Clin Cancer Res 2020 ⁵Graney SK, Cancer Immunol Res 2020; ⁶Hewitt SL, Clin Cancer Res 2020; ⁷Purroy N, AACR 2022; ⁸Chiocca EA, Sci Transl Med 2019

Examples of current IL-12-based therapies

Intra-tumoral

TAVO

Oncosec Immunotherapies

Systemically active

DF6002

Dragonfly Therapeutics

Tumor-activated

XTX301

Xilio Therapeutics

Conclusions

- IL-12 is a high priority cytokine for clinical development
- Systemic treatments have either not been tolerable or have not demonstrated adequate PK/PD/response to date
- Clinical development of tavokinogene telesplasmid as intra-tumoral approach has demonstrated relevant pharmacodynamics in patients, including in “cold” tumors
- Multiple novel approaches including tumor-activated IL-12 to overcome limitations of intra-tumoral administration and systemic toxicity

Q & A

- Please submit questions via the chat function

Thank you

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