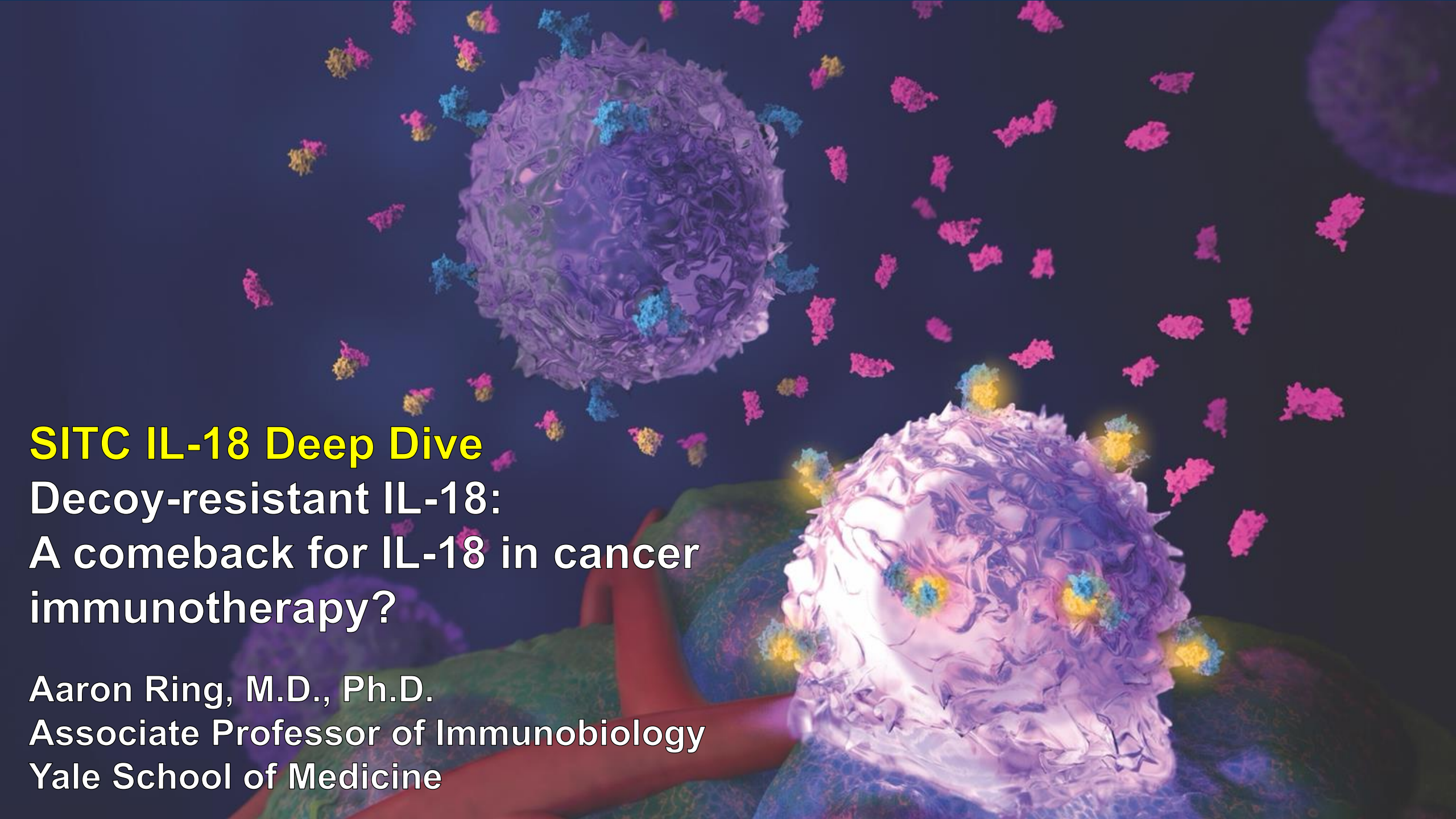


SITC IL-18 Deep Dive

Decoy-resistant IL-18: A comeback for IL-18 in cancer immunotherapy?

Aaron Ring, M.D., Ph.D.
Associate Professor of Immunobiology
Yale School of Medicine

Competing Interests

Simcha Therapeutics, Founder and Director

Forty Seven Inc./Gilead Sciences: Patent royalties

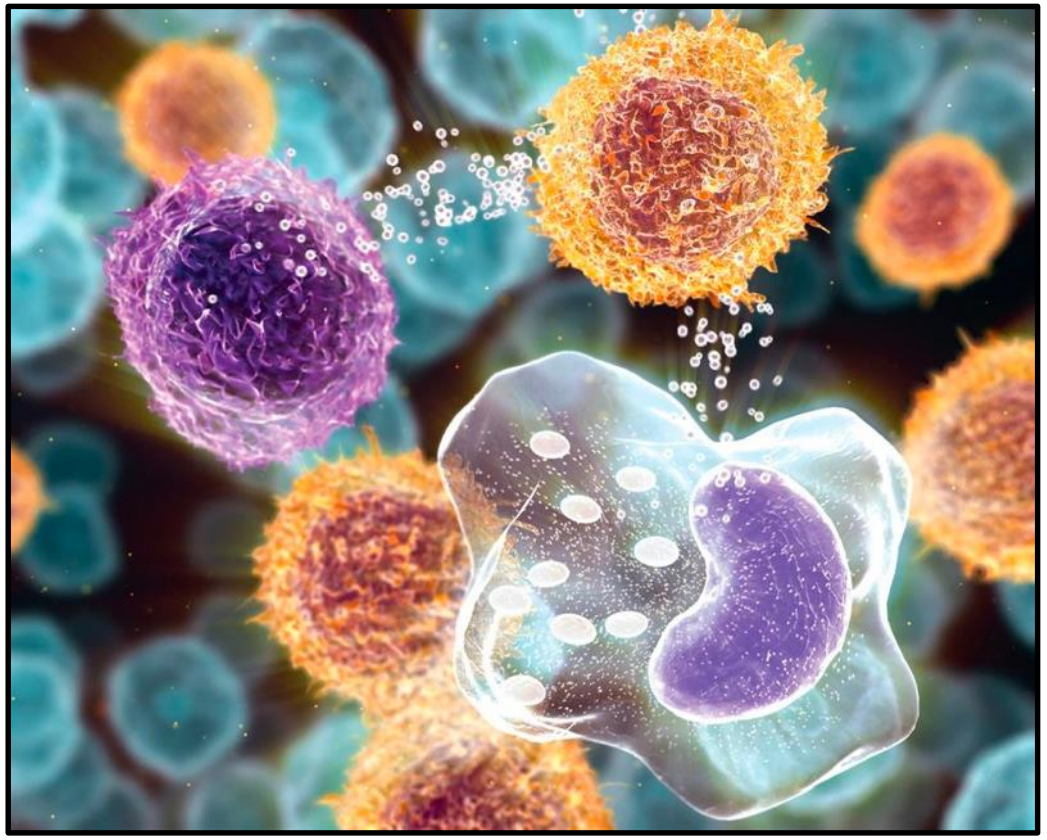
ALX Oncology: Co-Founder

Medicenna Therapeutics: Patent royalties

Seranova Bio: Founder and Director

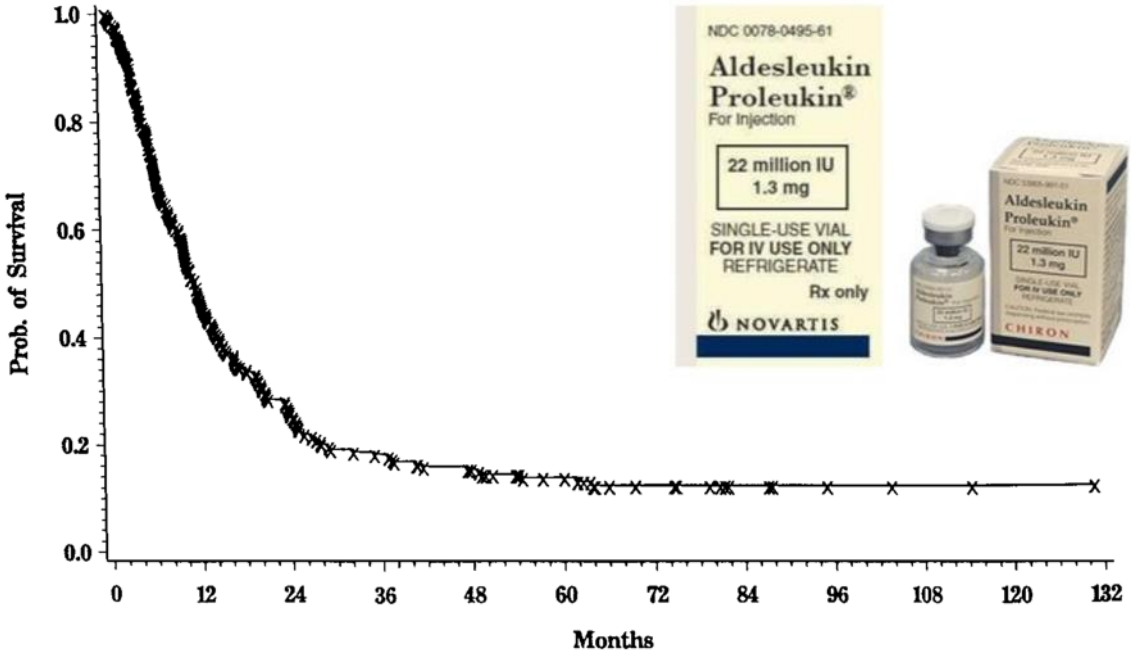
Stipple Bio: Co-Founder and Director

Cytokines: The first immune-targeting drugs that could cure



High-Dose Recombinant Interleukin 2 Therapy for Patients With Metastatic Melanoma: Analysis of 270 Patients Treated Between 1985 and 1993

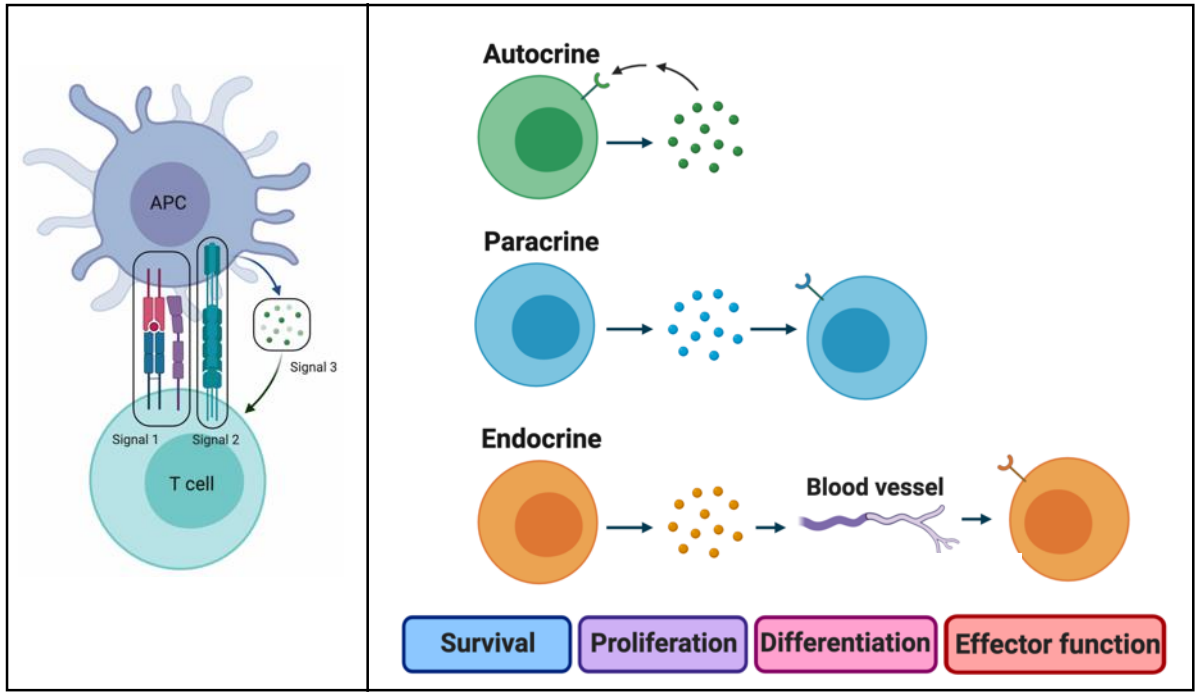
By Michael B. Atkins, Michael T. Lotze, Janice P. Dutcher, Richard I. Fisher, Geoffrey Weiss, Kim Margolin, Jeff Abrams, Mario Sznol, David Parkinson, Michael Hawkins, Carolyn Paradise, Lori Kunkel, and Steven A. Rosenberg



Atkins et al., Cancer J Sci Am., 2000

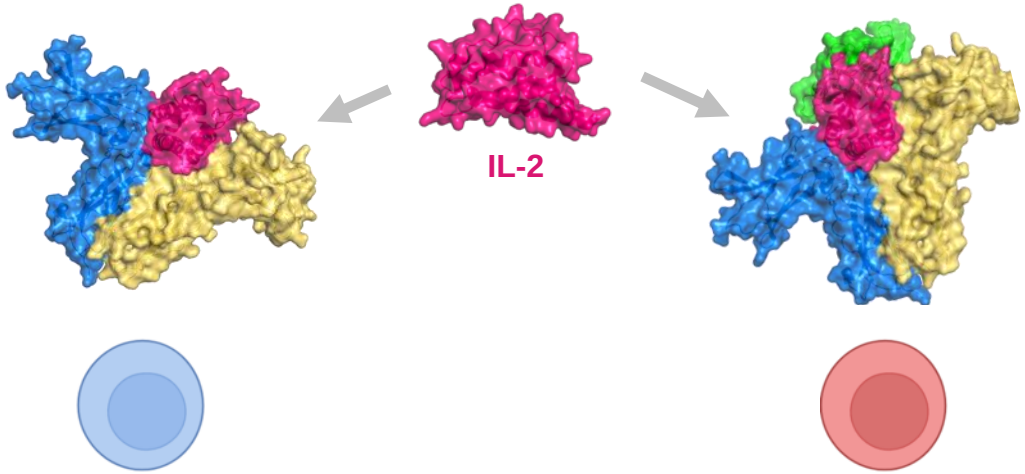
The promise and problem with cytokines

Potent control of immune physiology



Limitations of natural biology

"Pleiotropic"/paradoxical functions

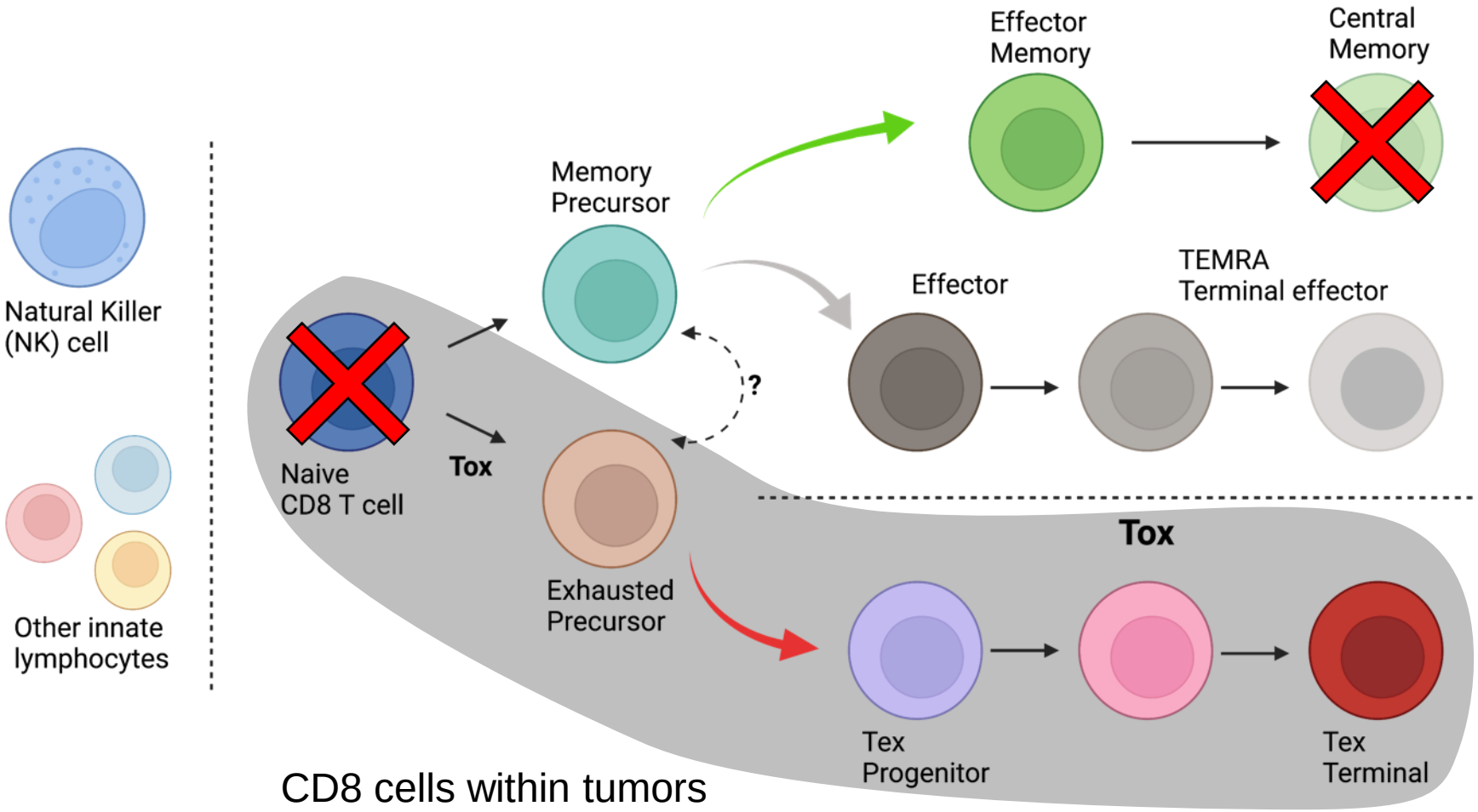


Anti-tumor
Cytotoxic T cells

Pro-tumor
Immunosuppressive Treg

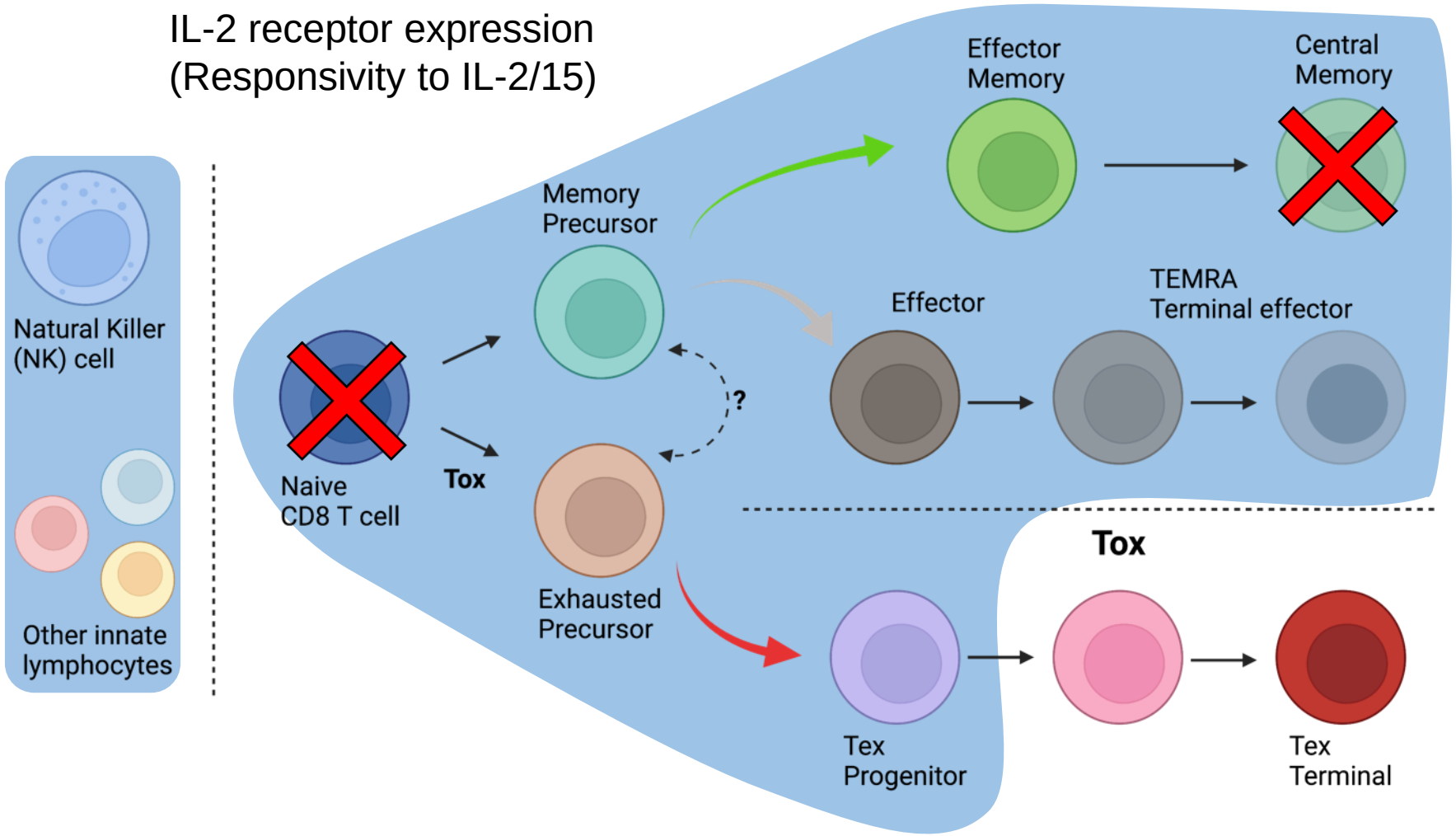
Can we avoid pleiotropy w/ more tumor-selective cytokines?

CD8-centric view of cytotoxic lymphocytes:



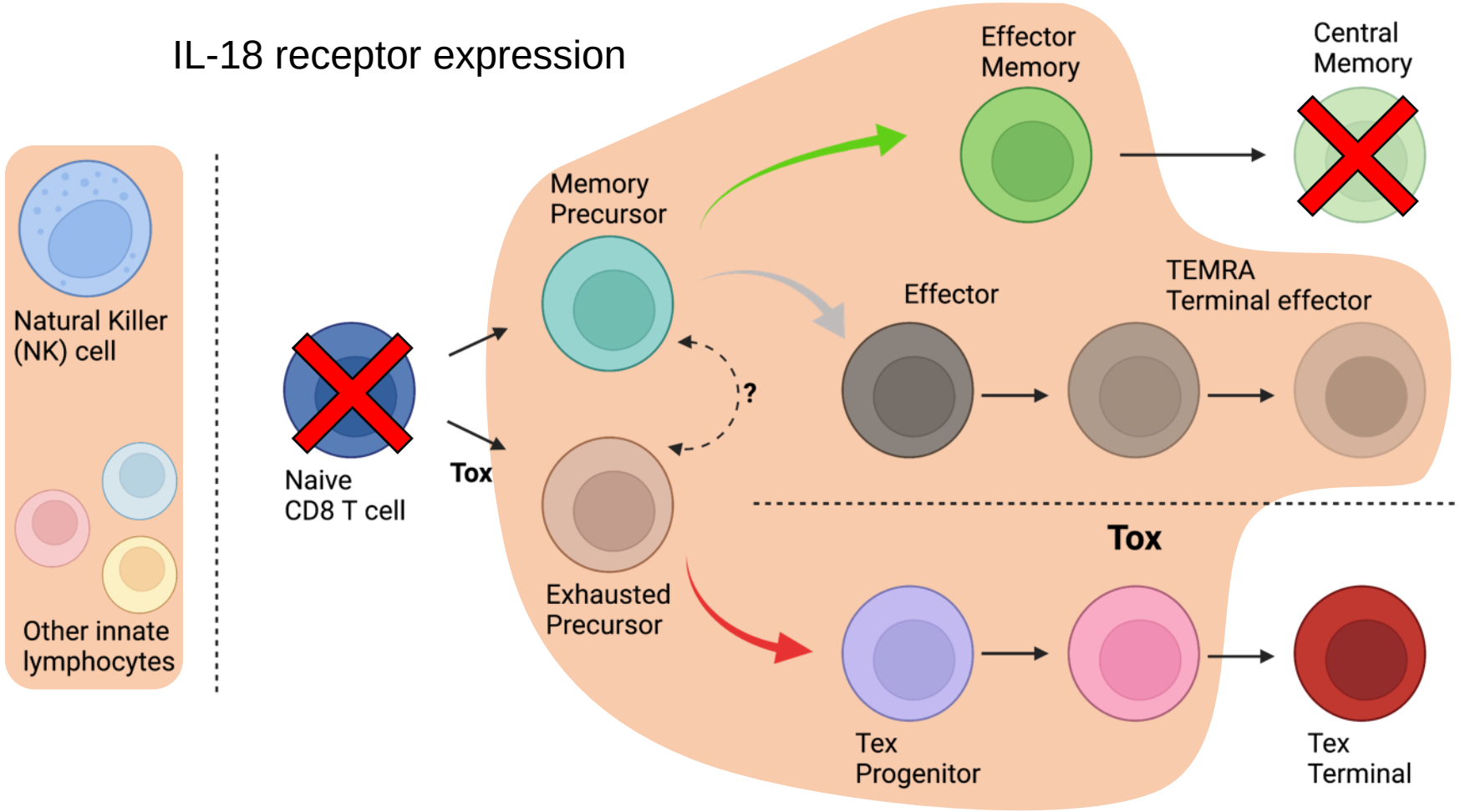
CD8 cells within tumors

Can we avoid pleiotropy w/ more tumor-selective cytokines?



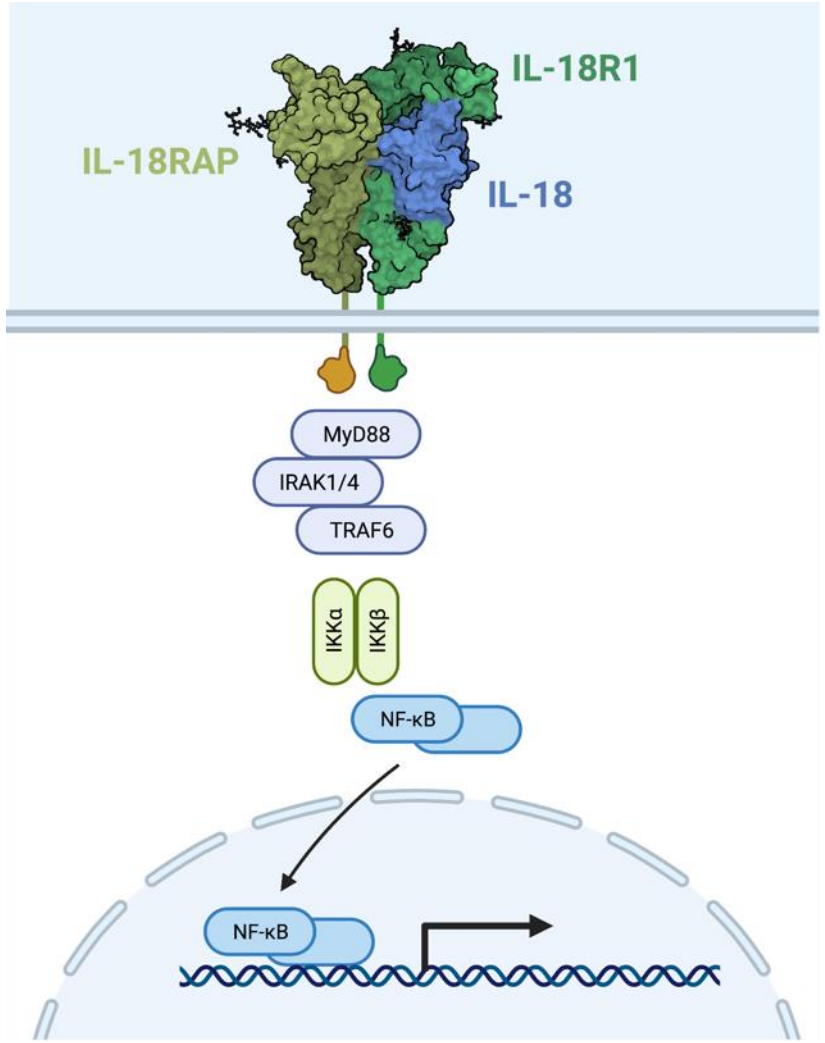
Slide adapted from John Wherry

Can we avoid pleiotropy w/ more tumor-selective cytokines?



Slide adapted from John Wherry

IL-18 delivers a powerful message to the right cells in the TME

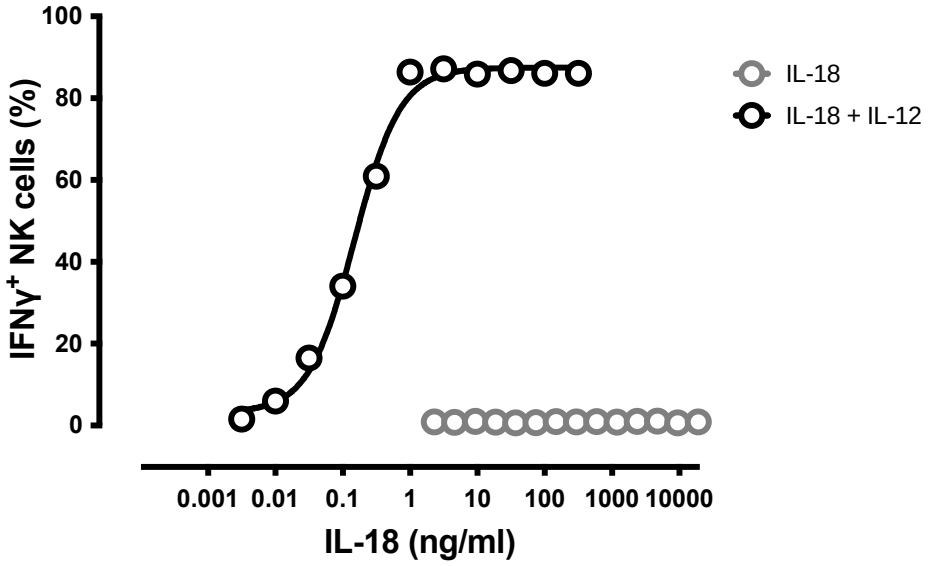


- Member of the IL-1 family of cytokines (‘alarmins’)
- Activates complementary signaling (MyD88) to most other immunotherapeutic agents against other cytokine/immunoreceptors
- Stimulates diverse anti-tumor effector cells of both adaptive and innate immune system: NK cells, antigen-experienced CD8 & T_H1, & MΦ
- Inhibits/alters Treg function

➢ Suggests broad utility in both immunogenic and “cold” tumors

IL-18: An amplifier, not an “on” switch

IL-18 does **not** induce IFNγ on resting splenocytes, but greatly amplifies the effect of IL-12



Zhou and Ring, unpublished

In a screen of CD8 stimulation elicited by 1,849 cytokine combinations, **IL-18 was in 6 of the top 10** combinations.

Table 1. Summary of the most potent cytokine combinations capable of triggering IFNγ production by virus-specific effector and memory T cells

	Effector		Memory	
Cytokine combination	% IFNγ+ unsorted	% IFNγ+ CD8 sorted	% IFNγ+ unsorted	% IFNγ+ CD8 sorted
→ IL-12 + IL-18	76.6	77.5	31.6	41.3
IL-12 + TNFα	67.4	47.9	29.7	24.5
IL-12 + IL-33	53.3	36.9	9.3	12.2
→ IL-2 + IL-18	52.7	46.9	4.9	10.0
IL-2 + IL-12	52.0	28.1	8.5	12.3
IL-12 + IL-15	44.0	8.9	12.6	7.6
→ IL-10 + IL-18	36.9	32.6	1.3	2.6
→ IL-18 + IL-21	34.7	33.7	2.8	5.9
→ IL-18 + IFNβ	31.6	28.5	13.7	17.1
→ IL-15 + IL-18	29.8	25.4	1.8	3.1

Freeman et al., *PNAS*, 2012

rIL-18 therapy: Remarkably well tolerated for a cytokine

A Dose-Escalation Study of Recombinant Human Interleukin-18 Using Two Different Schedules of Administration in Patients with Cancer

Michael J. Robertson,[1,2] John M. Kirkwood,[3] Theodore F. Logan,[2] Kevin M. Koch,[4] Steven Kathman,[4] Lyndon C. Kirby,[4] William N. Bell,[4] Linda M. Thurmond,[4] Jill Weisenbach,[2] and Mohammed M. Dar[4]

	Group A (daily × 5)			Group B (weekly)			
	Dose level (μg/kg)						
	100	500	1,000	100	1,000	2,000	
	n = 3	*n* = 3	*n* = 3	*n* = 4	*n* = 3	*n* = 3	*N* = 19
Chills	2/0	3/0	3/0	3/0	2/0	3/0	16/0
Fever	2/0	3/0	1/1	3/0	2/1	2/0	13/2
Headache	1/1	3/0	3/0	2/0	1/0	—	10/1
Fatigue	1/0	2/0	2/0	2/0	1/0	2/0	10/0
Pain (back/extremity)	3/0	5/0	1/0	—	1/0	—	10/0
Pruritus/pruritic rash	1/0	3/0	2/0	2/0	—	1/0	9/0
Nausea	2/0	2/0	2/0	1/0	1/0	—	8/0
Pain	1/0	2/0	2/0	2/0	—	1/0	8/0
Myalgia/arthralgia	2/0	1/0	1/0	—	1/0	3/0	8/0
Anorexia	1/0	2/0	—	1/0	2/0	1/0	7/0
Vomiting	—	1/0	1/0	2/0	1/0	1/0	6/0
Diarrhea	1/0	1/0	2/0	1/0	1/0	—	6/0
Cough	1/0	1/0	2/0	—	1/0	1/0	6/0
Dizziness	1/0	2/0	—	1/0	1/0	1/0	6/0
Insomnia	2/0	2/0	1/0	—	—	—	5/0
Injection site/skin reaction	1/0	—	3/0	1/0	—	—	5/0
Abdominal pain/distension	3/0	—	1/0	—	—	1/0	5/0
Chest pain	1/0	—	—	1/0	1/0	2/0	5/0
Rash/urticaria	1/0	2/0	1/0	—	—	1/0	5/0
Paresthesia/hypoesthesia	1/0	1/0	1/0	—	—	1/0	4/0
Hypotension	1/0	1/0	—	—	0/1	1/0	3/1
Hypertension	—	—	2/0	—	—	—	2/0

*No grade 4 adverse events were reported.

Common laboratory abnormalities (all cycles, worst toxicity grade by patient)

Laboratory abnormality	No. patients with grade 1-2/3-4* adverse events						All cohorts
	Group A (daily × 5)			Group B (weekly)			
	Dose level (μg/kg)						
	100	500	1,000	100	1,000	2,000	
	n = 3	*n* = 3	*n* = 3	*n* = 4	*n* = 3	*n* = 3	*N* = 19
Lymphopenia	1/2	0/2	2/1	0/4	0/3	0/2	3/14
Anemia	2/0	2/0	3/0	3/1	3/0	1/0	14/1
Leukopenia	2/0	1/0	2/0	3/0	2/0	1/0	11/0
Neutropenia	1/0	1/0	2/0	1/0	1/1	—	6/1
Hyperglycemia	1/0	2/1	2/1	2/1 †	2/0	1/0	10/3
Hypoalbuminemia	—	2/0	2/0	2/0	2/0	3/0	11/0
Elevated alkaline phosphatase	2/0	1/0	1/1	1/0	2/0	2/0	9/1
Elevated AST	2/0	—	2/0	2/0	—	2/0	8/0
Elevated creatinine	1/0	1/0	—	3/0	2/0	1/0	8/0
Hyponatremia	1/0	1/0	—	1/0	1/0	1/0	5/0
Elevated ALT	1/0	—	1/0	—	—	2/0	4/0
Elevated bilirubin	—	—	1/0	1/0	1/0	1/0	4/0

Abbreviations: ALT, alanine aminotransferase; AST, aspartate aminotransferase.
*All grade 3 with the exception of single episode of grade 4 hyperglycemia.
† Grade 4 hyperglycemia.

Conclusions: rhIL-18 can be given in biologically active doses by either weekly infusions or daily infusions for 5 days repeated every 28 days to patients with advanced cancer. Toxicity was generally mild to moderate, and a maximum tolerated dose of rhIL-18 by either schedule was not determined.

Robertson et al., *Clinical Cancer Research* 2008

IL-18 is powerful in a test tube, but a dud in the clinic

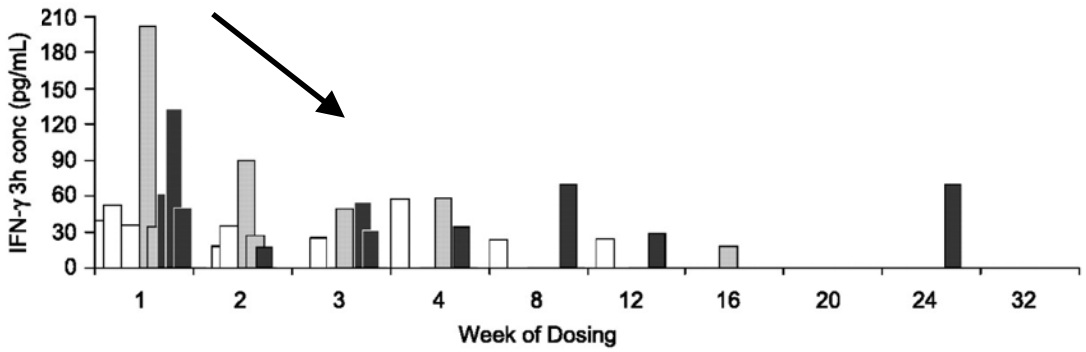
A Phase 2, Randomized Study of SB-485232, rhIL-18, in Patients With Previously Untreated Metastatic Melanoma

Ahmad A. Tarhini, MD[1], Michael Millward, MD[2], Paul Mainwaring, MD[3], Richard Kefford, MD[4], Ted Logan, MD[5], Anna Pavlick, MD[6], Steven J. Kathman, MD[7], Kevin H. Laubscher, MD[8], Mohammed M. Dar, MD[9], and John M. Kirkwood, MD[1]

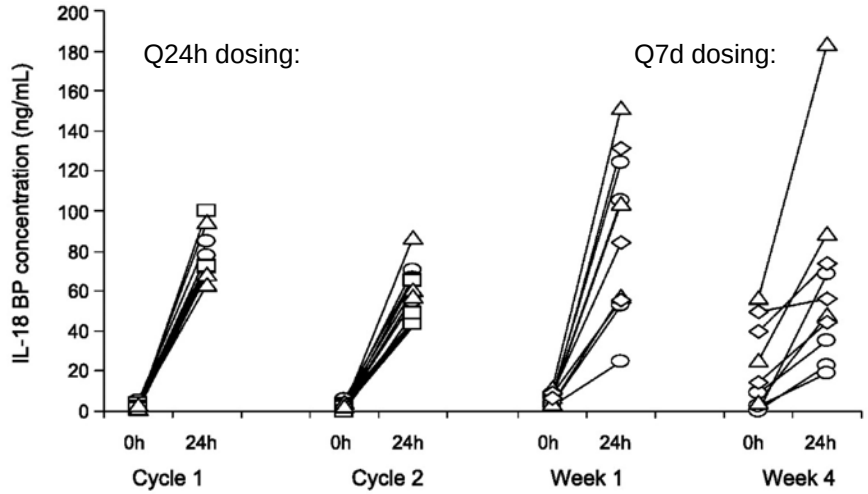
"Among 63 subjects evaluable for response, [only] 1 achieved a partial response... Due to the low apparent level of clinical efficacy, the study was terminated at the end of stage 1."

Tarhini et al., *Cancer*, 2009

IL-18 pharmacodynamic activity wanes with repeated dosing



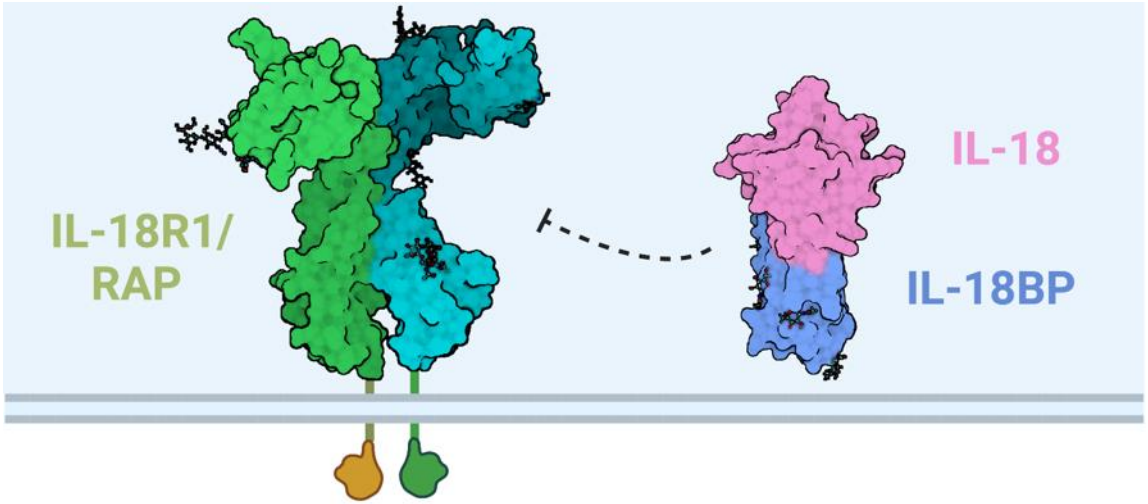
Corresponding to a massive systemic upregulation of IL-18BP:



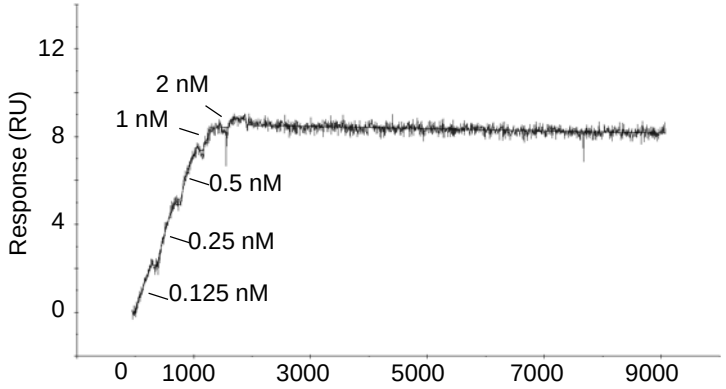
Robertson et al., *Clinical Cancer Research*, 2006
Robertson et al., *Clinical Cancer Research*, 2008

IL-18BP is a secreted immune checkpoint

IL-18BP is an ultra potent soluble decoy receptor that antagonizes IL-18

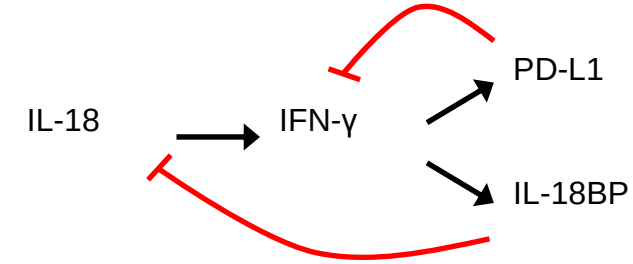


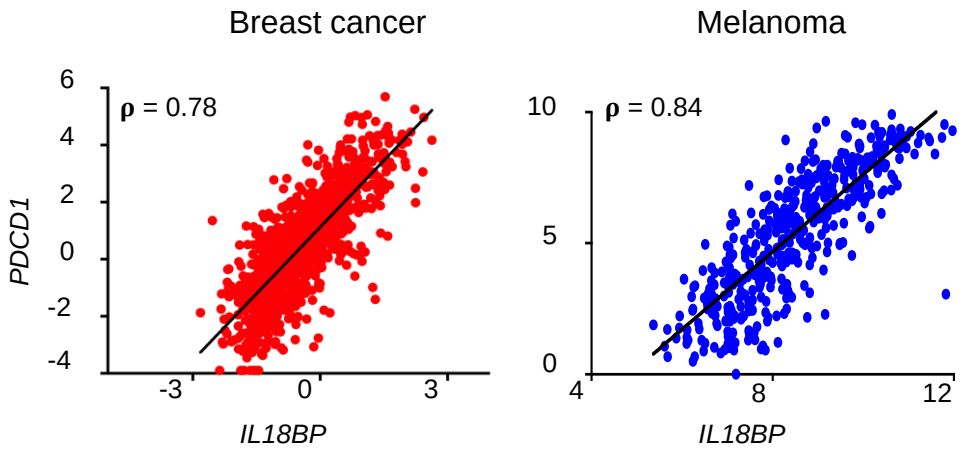
IL-18:IL-18BP Kinetic Binding (SPR)



K_D = 1.1 pM

IL-18BP is regulated by IFNγ, akin to other immune checkpoints





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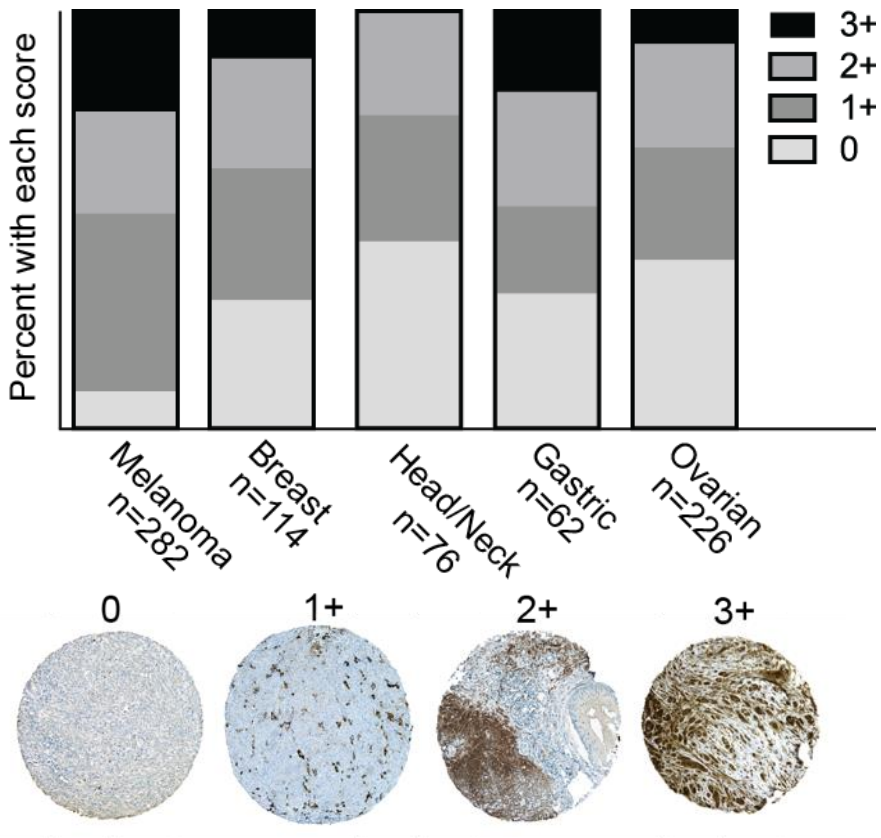
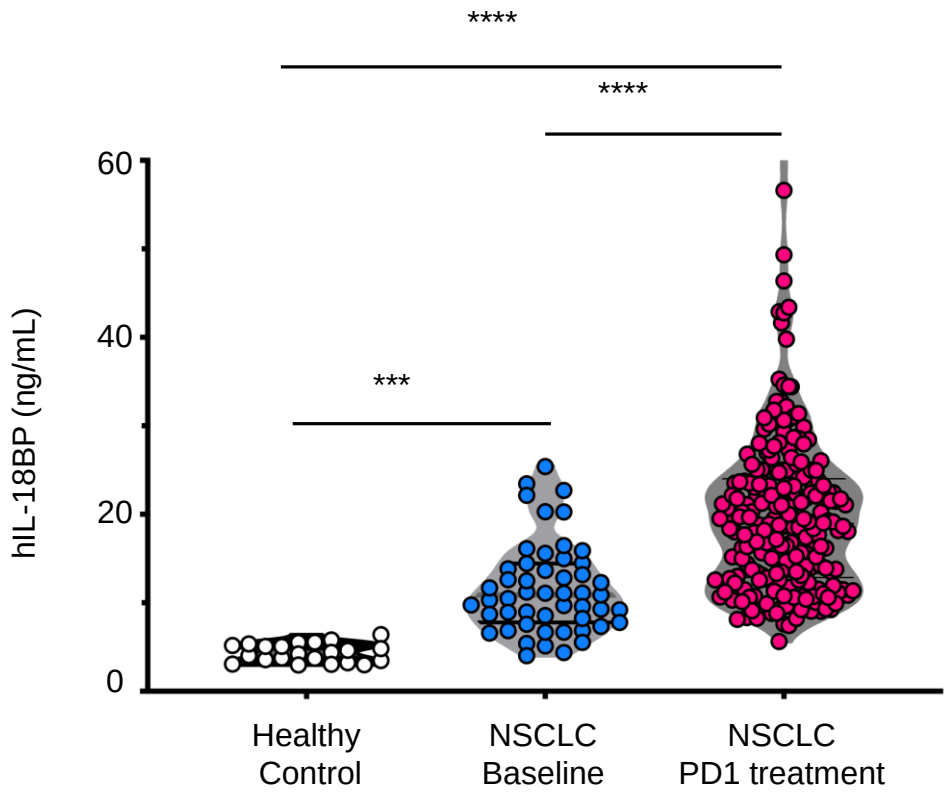
Zhou et al., *Nature*, 2020

IL-18BP is prevalent in the TME and in cancer patient plasma

IL-18BP tumor IHC



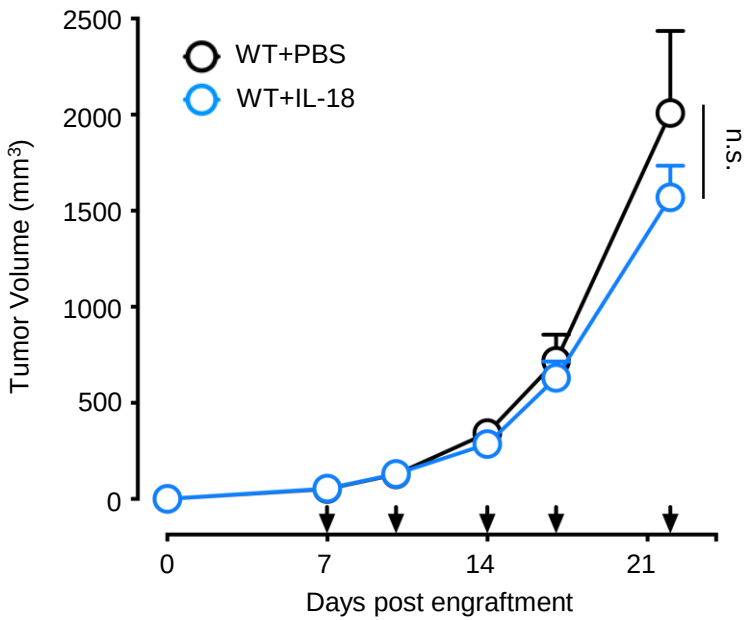
IL-18BP plasma ELISA



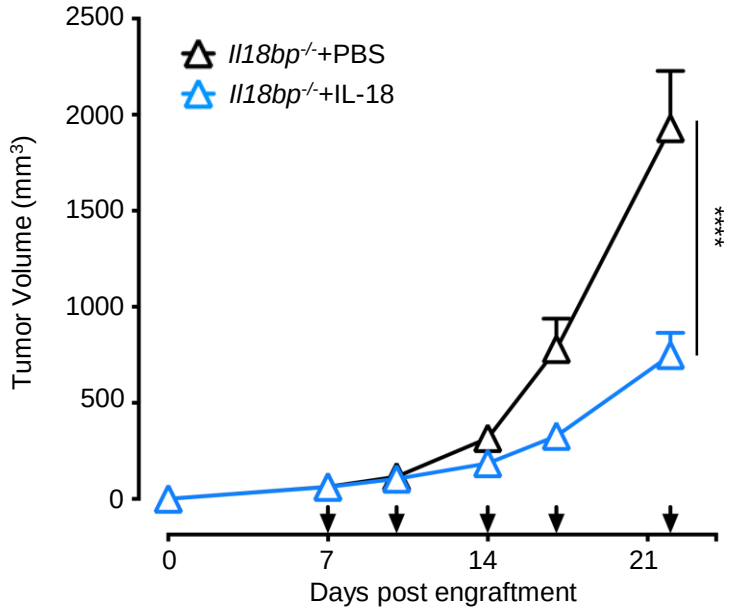
Zhou et al., *Nature*, 2020

IL-18BP is a barrier to effective rIL-18 immunotherapy

rIL-18 is ineffective in treating established MC38 tumors in WT mice

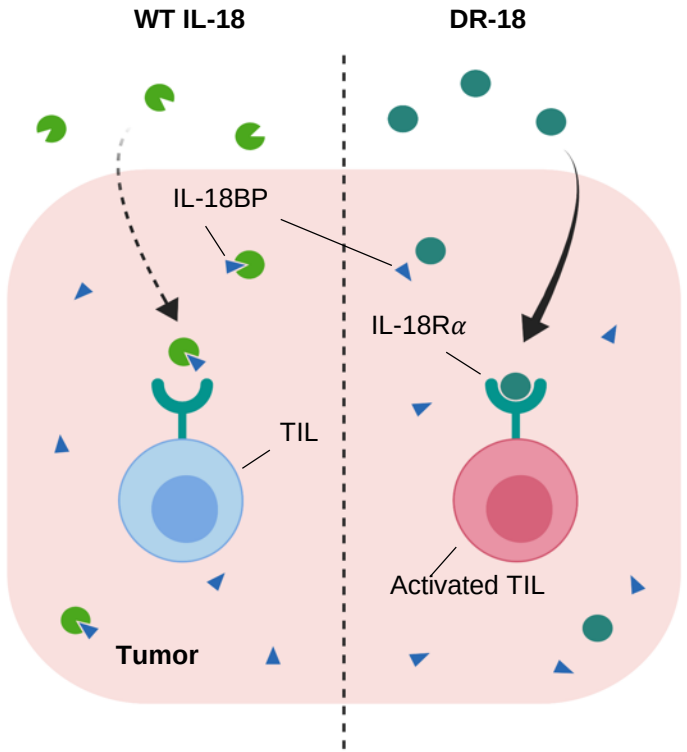


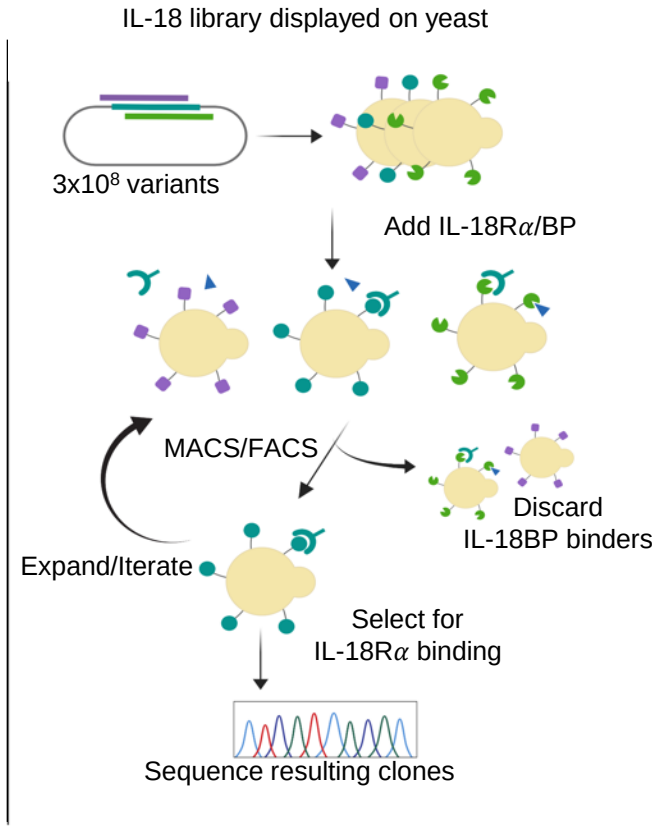
Knockout of IL-18BP unmasks monotherapeutic activity of rIL-18

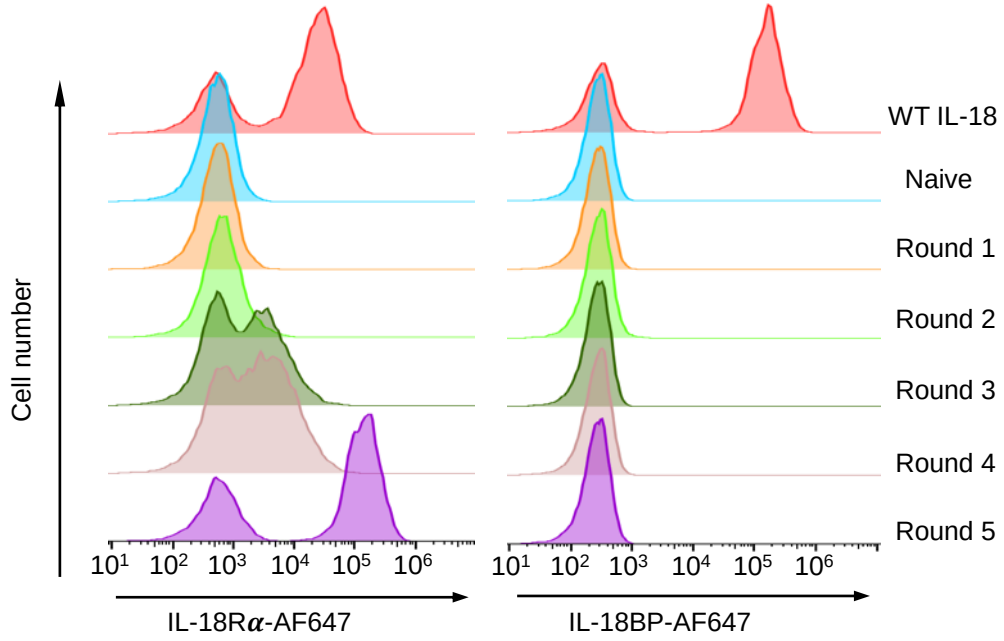


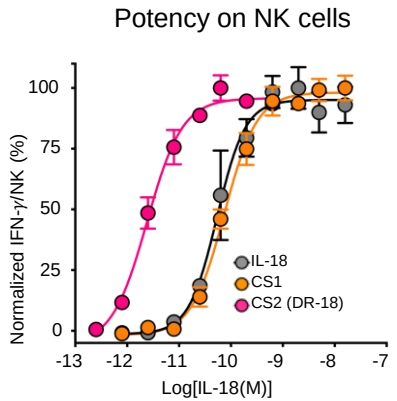
Zhou et al., *Nature*, 2020

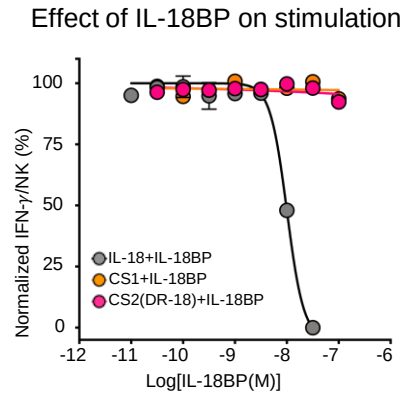
Engineering ‘decoy-resistant’ IL-18 (DR-18)











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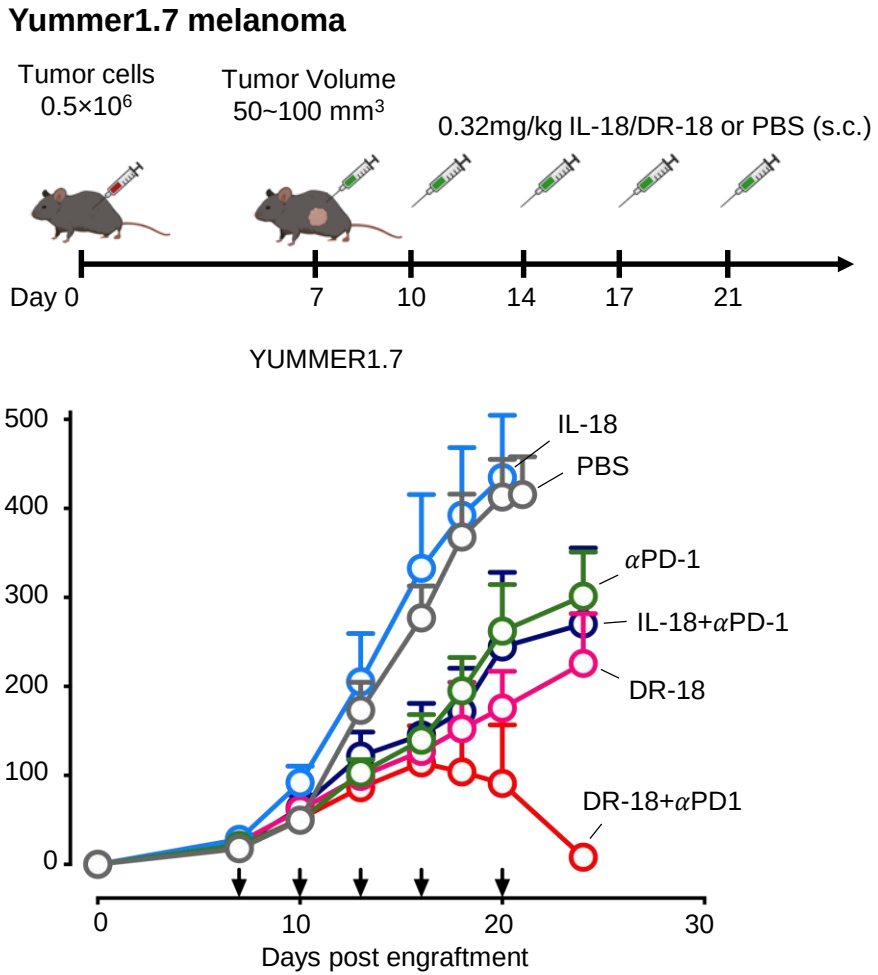
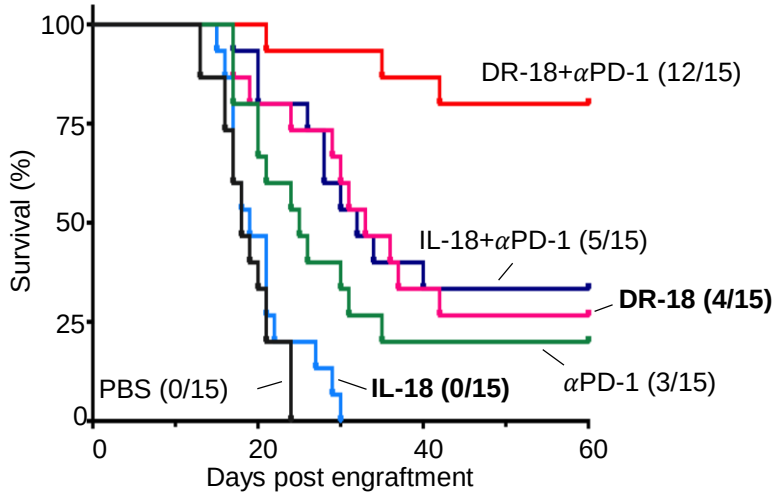
Zhou et al., *Nature*, 2020

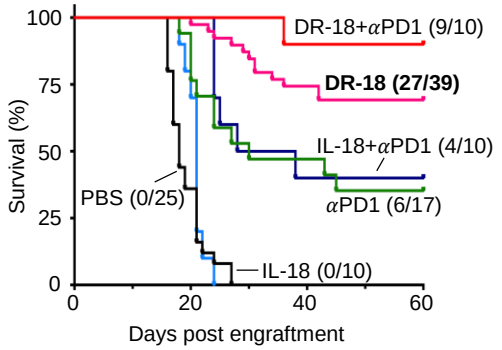
DR-18 is effective by itself and in combination with anti-PD-1



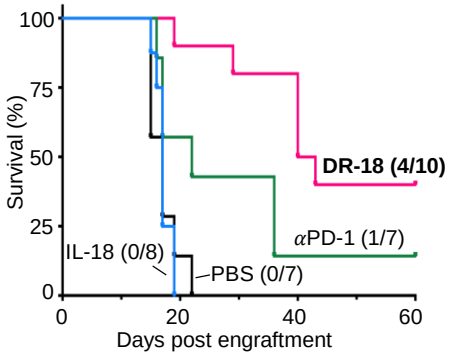


DR-18 efficacy is generalizable to numerous syngeneic tumor models:

MC38



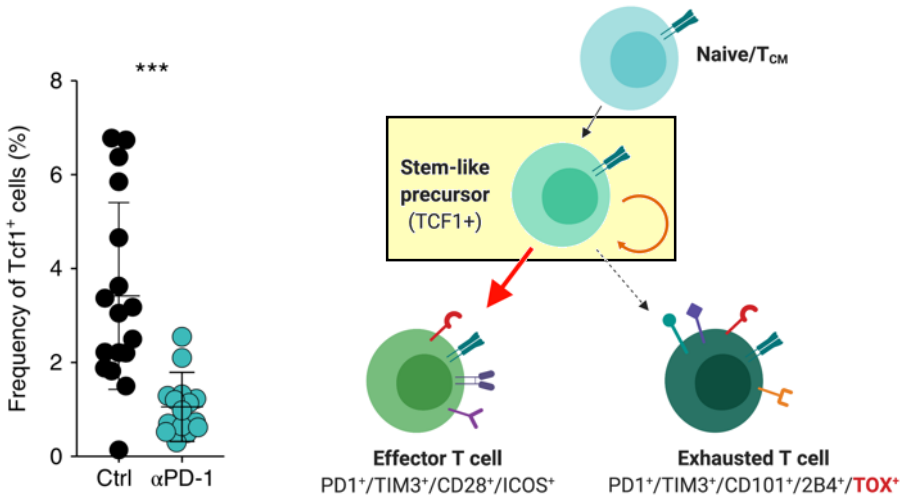
CT26



Zhou et al., *Nature*, 2020

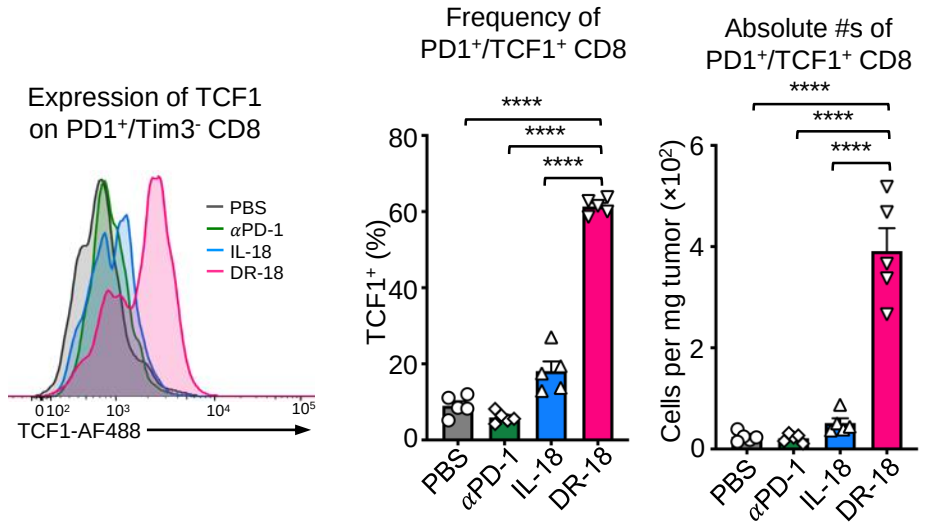
DR-18 expands precursor TCF1⁺ CD8 cells

TCF1+ precursor CD8 TIL are key targets of immunotherapy, but are not replenished by anti-PD-1 therapy

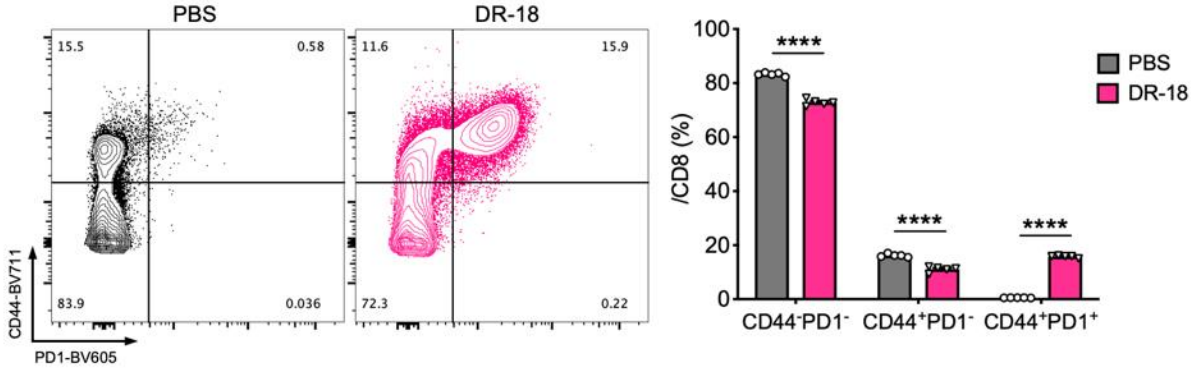


Miller et al., Nature Immunology, 2019

DR-18 treatment expands precursor TCF1+ CD8 >10-fold **in the TME**



And **in the dLN**:



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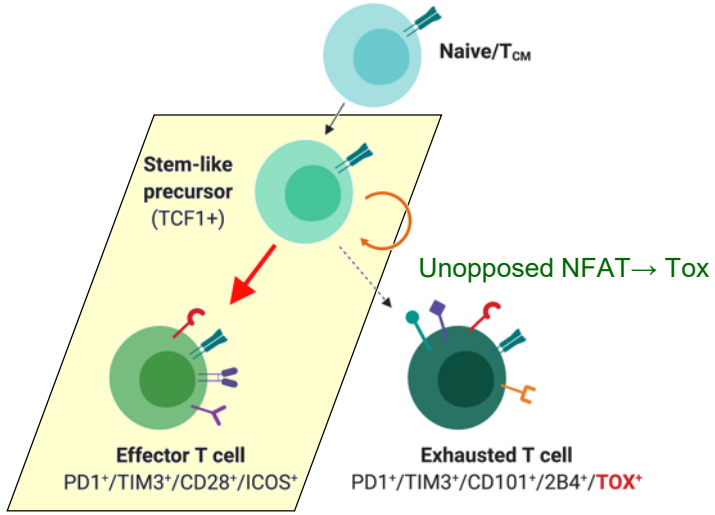
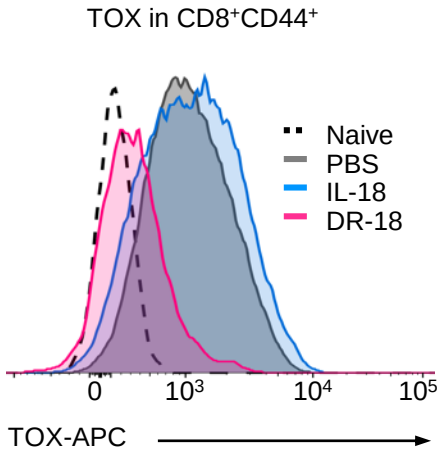
Zhou et al., *Nature*, 2020

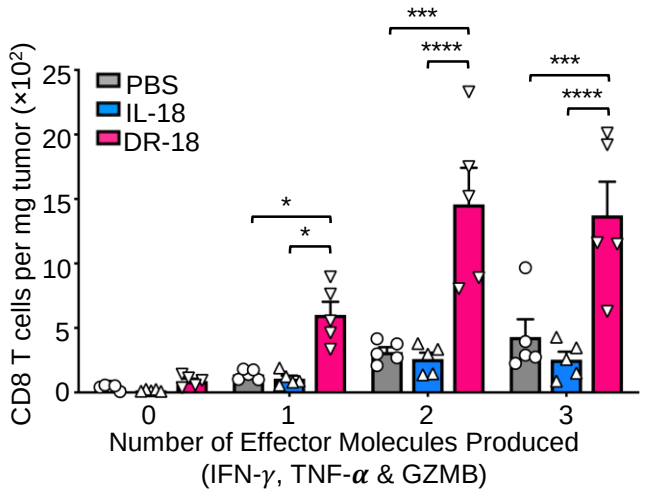
DR-18 skews CD8 precursor differentiation to Teff from Tex

TOX is a master regulator of T cell exhaustion



DR-18 suppresses TOX expression & promotes polyfunctional T_{EFF} CD8 cells





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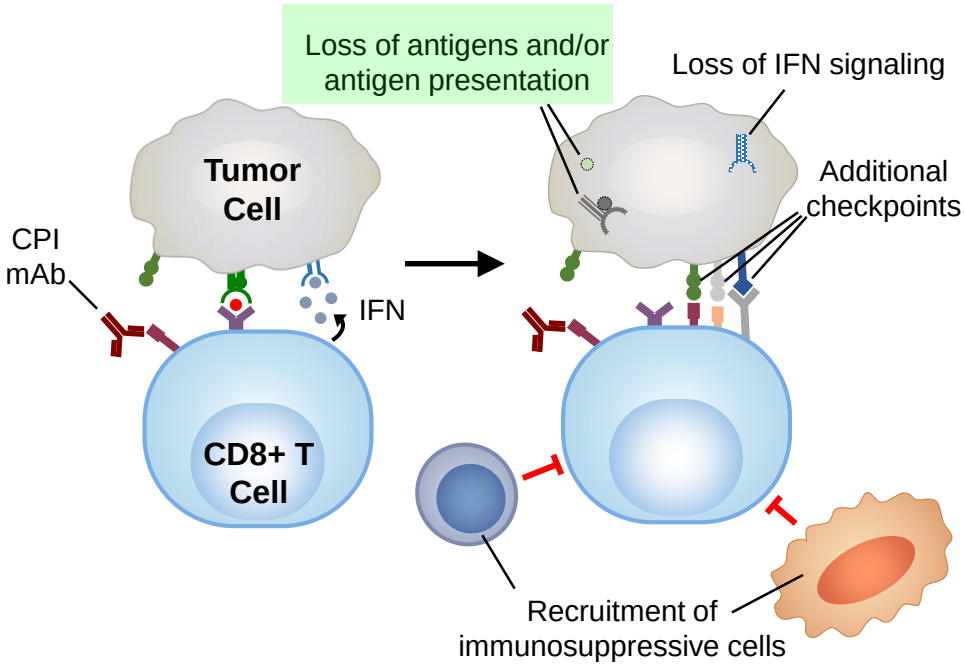
Zhou et al., *Nature*, 2020

IL-18 agonism in ICI resistance

Tapping into innate immunity in the setting of MHC class I loss

Mechanisms of resistance to immune checkpoint blockade:

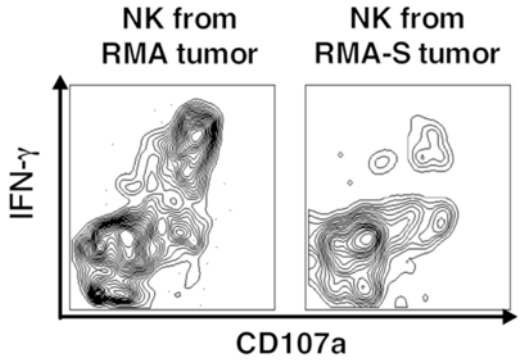


Total MHC class I loss is a common phenomenon:

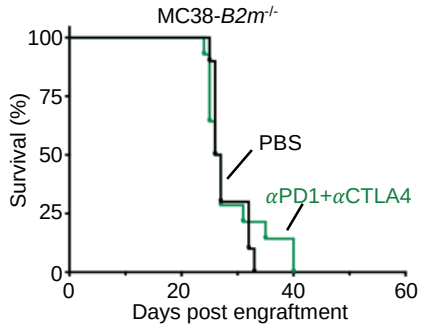
Colorectal cancer	Laryngeal cancer	Cervical cancer	Bladder cancer	Prostate cancer	Breast cancer	Renal cancer	Melanoma cell lines
14%	11%	10%	25%	55%	52%	–	18%

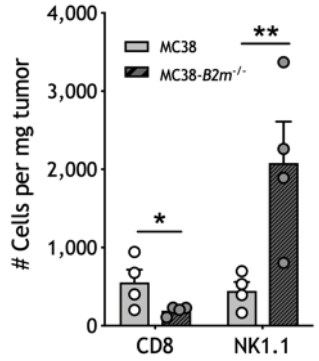
Tumor Immunology and Immunotherapy, Chapter 5. 2014

NK cells infiltrate into MHC-I deficient tumors, but they are highly dysfunctional



Ardolino et al., JCI, 2014





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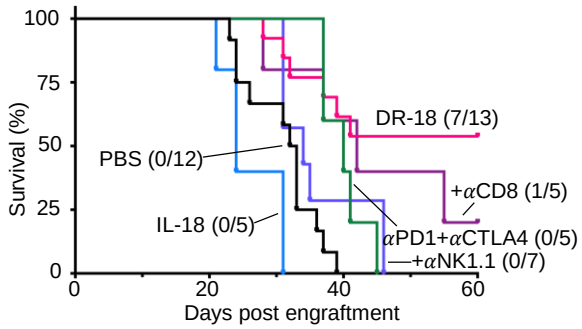
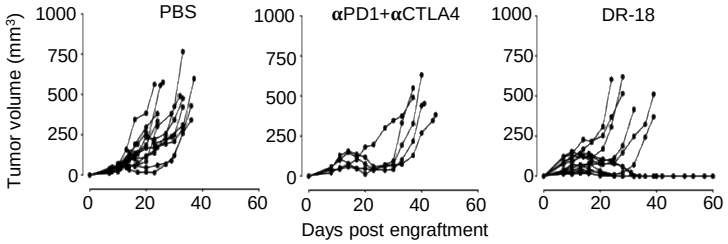
Ardolino et al., *Journal of Clinical Investigation,* 2014

DR-18 is highly active in ICI-resistant tumors that lack MHC-I

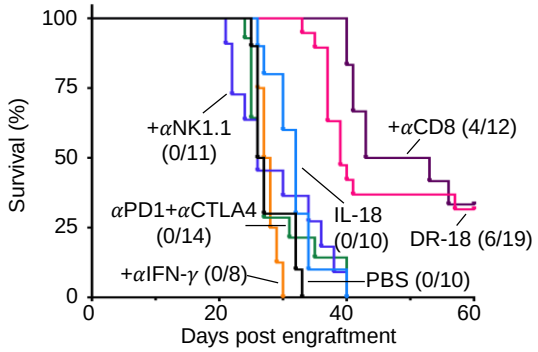
Activity requires NK but not T cells

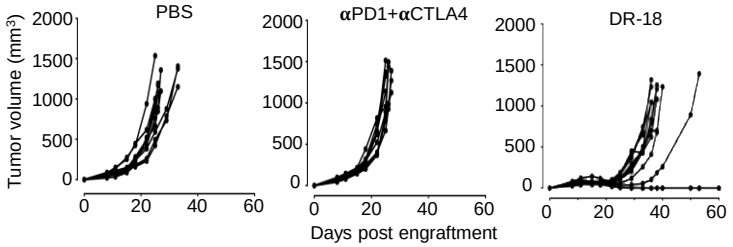
YUMMER1.7-*B2m*$^{-/-}$



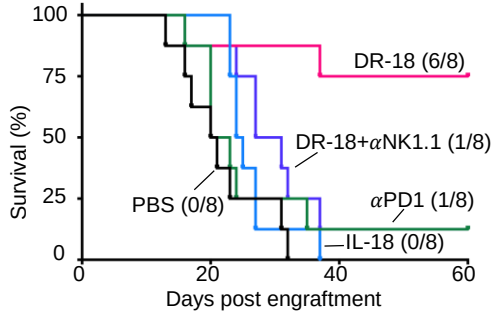


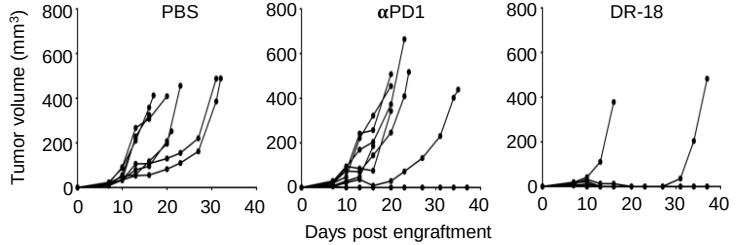
MC38-*B2m*$^{-/-}$





RMA-S





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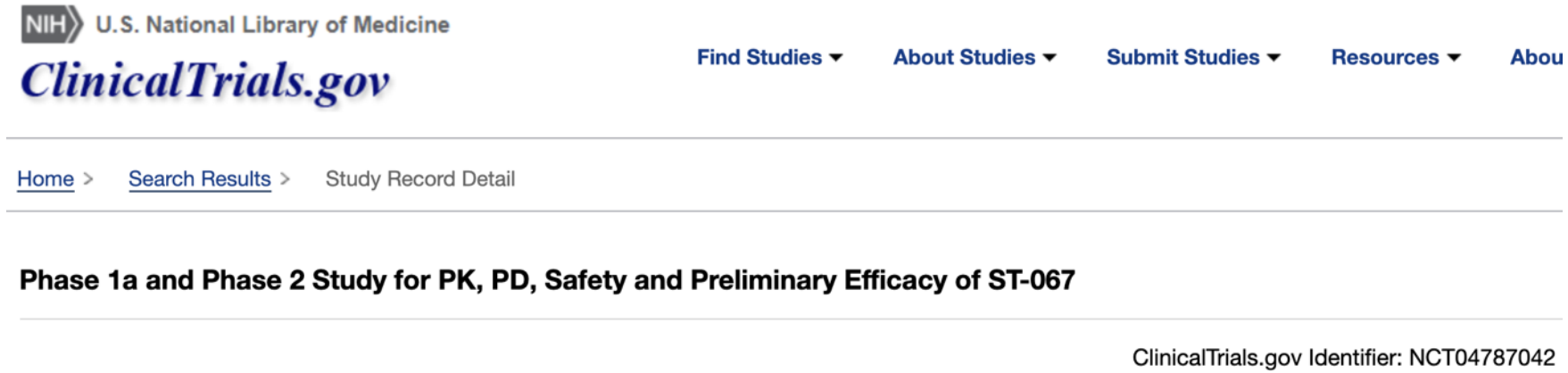
Zhou et al., *Nature*, 2020

Conclusions and ongoing studies

- The IL-18 receptor marks key anti-tumor TIL populations in tumors
- IL-18BP is a secreted immune checkpoint and barrier to effective IL-18 immunotherapy
- DR-18 overcomes IL-18BP and exhibits potent anti-tumor activity in immunogenic and ICI-resistant tumors
- DR-18 expands TCF1^{+} precursor cells and skews their differentiation to a highly polyfunctional Teff population
- DR-18 enhances NK cell maturation and function in MHC-class I deficient tumors



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