



Animal Model Workshop

Choosing Kidney Cancer Targets and Models Wisely

William G. Kaelin, Jr., M.D.

Sidney Farber Professor of Medicine

Dana-Farber Cancer Institute and Harvard Medical School

Investigator

Howard Hughes Medical Institute

September 11-13, 2025

**Session 2: Challenges and Specifics of
Creating Good Model**



KidneyCancerAssociation® IN PARTNERSHIP WITH

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Kidney Cancer Program



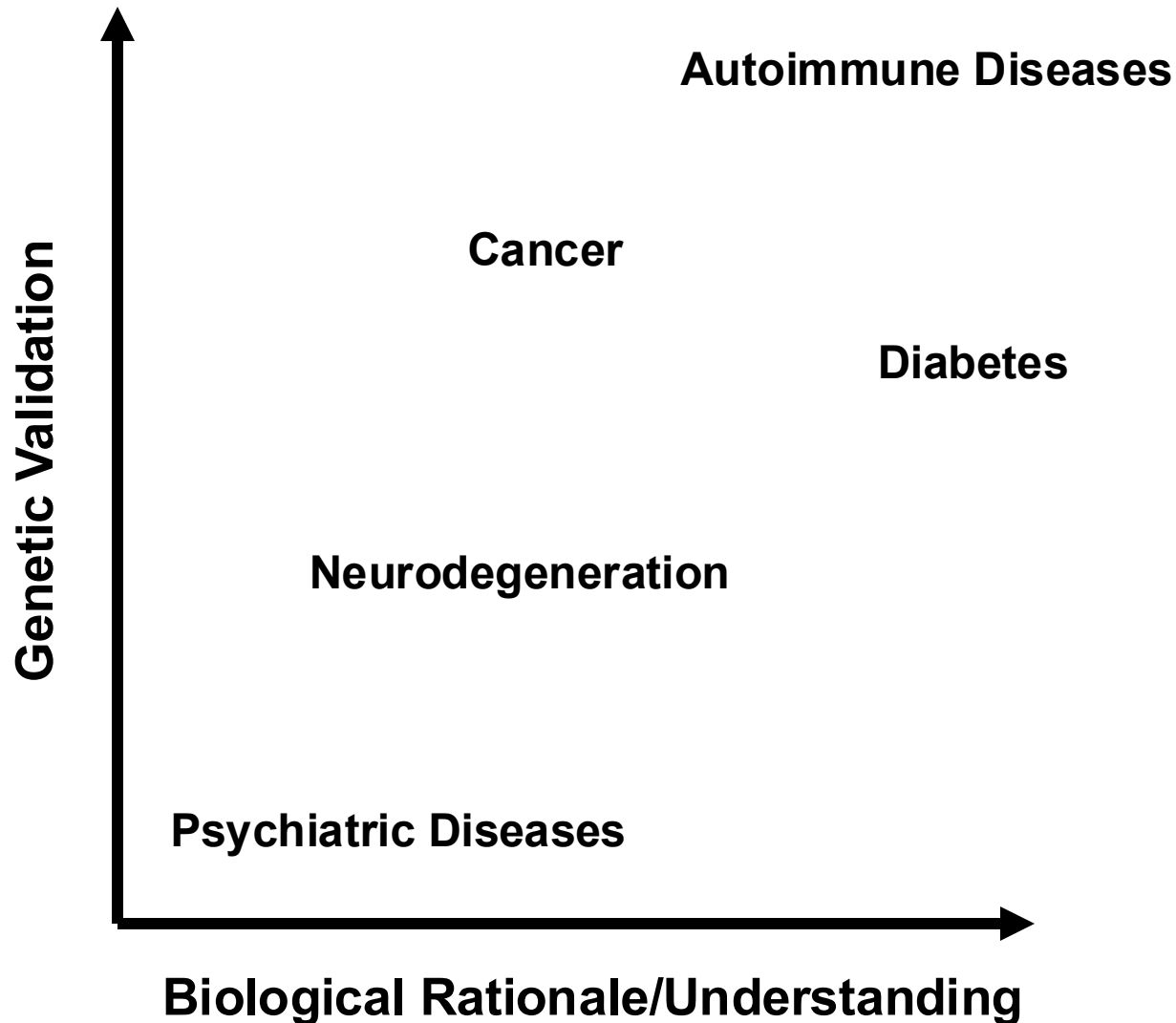
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Disclosure

- I have a financial interest in **Fibrogen**, Inc., which is developing prolyl hydroxylase inhibitors for the treatment of anemia and ischemic diseases
- I am on **Lilly** Board of Directors
- I am a cofounder of **Tango Therapeutics**, Inc., which is developing cancer drugs based on synthetic lethality
- I have a financial interest in HIF2 inhibitor **Belzutifan** (being developed by Merck)
- I am on **LifeMine Therapeutics** Board of Directors
- I am on **Circle Pharma** Scientific Advisory Board
- I advise **Nextech Invest** and **Casdin Capital**

Drug Development Success Increases with High Quality Targets (Genetically Validated and Biology Understood)



The support of human genetic evidence for approved drug indications

Matthew R Nelson¹, Hannah Tipney², Jeffery L Painter¹, Judong Shen¹, Paola Nicoletti³, Yufeng Shen^{3,4}, Aris Floratos^{3,4}, Pak Chung Sham^{5,6}, Mulin Jun Li^{6,7}, Junwen Wang^{6,7}, Lon R Cardon⁸, John C Whittaker² & Philippe Sanseau²

RESEARCH ARTICLE

Are drug targets with genetic support twice as likely to be approved? Revised estimates of the impact of genetic support for drug mechanisms on the probability of drug approval

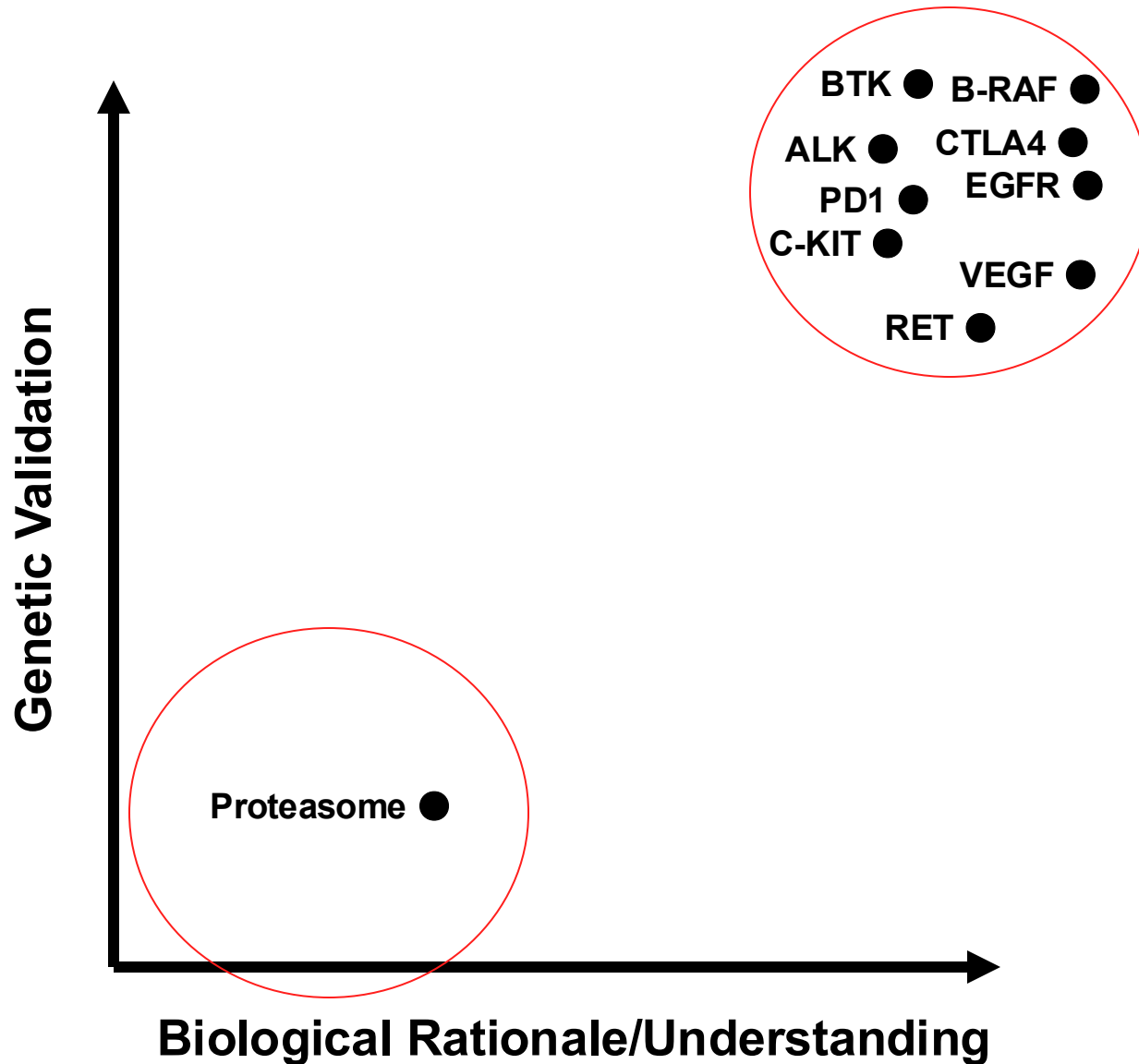
Emily A. King *, J. Wade Davis, Jacob F. Degner

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Targets of Some Recent Cancer Approvals



Genetic Validation of CTLA-4

Immunity



Volume 3, Issue 5, November 1995, Pages 541-547

Article

Loss of CTLA-4 leads to massive lymphoproliferation and fatal multiorgan tissue destruction, revealing a critical negative regulatory role of CTLA-4

Elizabeth A. Tivol *, Frank Borriello *, A.Nicola Schweitzer *, William P. Lynch *, Jeffrey A. Bluestone †, Arlene H. Sharpe *

REPORTS

Lymphoproliferative Disorders with Early Lethality in Mice Deficient in *Ctla-4*

Paul Waterhouse, Josef M. Penninger, Emma Timms, Andrew Wakeham, Arda Shahinian, Kelvin P. Lee, Craig B. Thompson, Henrik Griesser, Tak W. Mak⁽¹⁾

Science 10 Nov 1995:
Vol. 270, Issue 5238, pp. 985-988
DOI: 10.1126/science.270.5238.985

Angiogenesis Inhibitors

Genetic Validation

Endostatin, Angiostatin ●

VEGF Inhibitors ●

Biological Understanding

"All the News That's Fit to Print"

The New York Times

VOL. CXLVII . . . No. 51,146

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NEW YORK, MONDAY, MAY 3, 1996

It missed the greatest New York metropolitan area.

Late Edition
New York: Today, Variable clouds and rain, a low shower. High 74. Tonight, cloudy, fog. Low 54. Tomorrow, cloudy, late rain. High 70. Yesterday, high 65, low 55. Details, page 31.

\$2.50

A Cautious Awe G greets Drugs That Eradicate Tumors in Mice

By GINA KOLATA

Within a year, if all goes well, the first cancer patient will be injected with two new drugs that can eradicate any type of cancer, with no obvious side effects and no drug resistance — in mice.

Some cancer researchers say the drugs are the most exciting treatment that they have ever seen. But then they temper their enthusiasm with caution, noting that the history of cancer treatments is full of high expectations followed by dashed hopes when drugs with remarkable effects in animals are tested in people.

Still, the National Cancer Institute has made the drugs its top priority, said Dr. Richard D. Klausner, the director. Dr. Klausner called them "the most

HOPE IN THE LAB
A special report.

cancer researcher at the Harvard Medical School, was wary. "We are all driven by hope," Dr. Grossman said. "But a sober scientist waits for the data." And until the drugs are given to humans, he said, the crucial data simply do not exist.

So far, the drugs are the only ones ever tested that can seemingly eradicate all tumors in mice, even gigantic ones, equivalent to a two-pound growth in a person. The best that other cancer drugs have done is slow the growth of these large tumors.

Dr. Klausner called them "the most exciting treatment that they have ever seen. But then they temper their enthusiasm with caution, noting that the history of cancer treatments is full of high expectations followed by dashed hopes when drugs with remarkable effects in animals are tested in people."

"I am putting nothing on higher priority than getting this into clinical trials," Dr. Klausner said. The mouse studies are "remarkable and wonderful," he said, and "very compelling." But he pointed out that the studies were in mice and so, when it comes to humans, he said he wanted to emphasize "the ifs."

The new drugs, angiostatin and endostatin, work by interfering with the blood vessels' tendency

2 Drugs Eradicate Tumors in Mice

Continued From Page 1

to grow. They kill off the cells that sprout new blood vessels in the tumor, and then the tumor starves and dies. The drugs are injected into the tumor, and the tumor shrinks and dies. The drugs are injected into the tumor, and the tumor shrinks and dies. The drugs are injected into the tumor, and the tumor shrinks and dies.

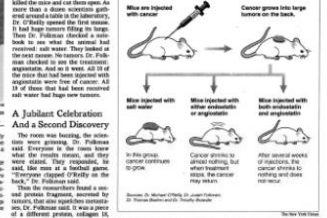
Dr. Julius Folkman is credited with the cancer-killing discovery. He found that tumors need blood vessels to grow. He found that tumors need blood vessels to grow. He found that tumors need blood vessels to grow.

Dr. Folkman said that the drugs are the most exciting treatment that they have ever seen. But then they temper their enthusiasm with caution, noting that the history of cancer treatments is full of high expectations followed by dashed hopes when drugs with remarkable effects in animals are tested in people.

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Hope for a Breakthrough

Drugs called angiostatin and endostatin could naturally in small quantities in the human body and work by interfering with the blood vessels that tumors need to survive and grow. When tested together in mice, the drugs made tumors disappear and not return.



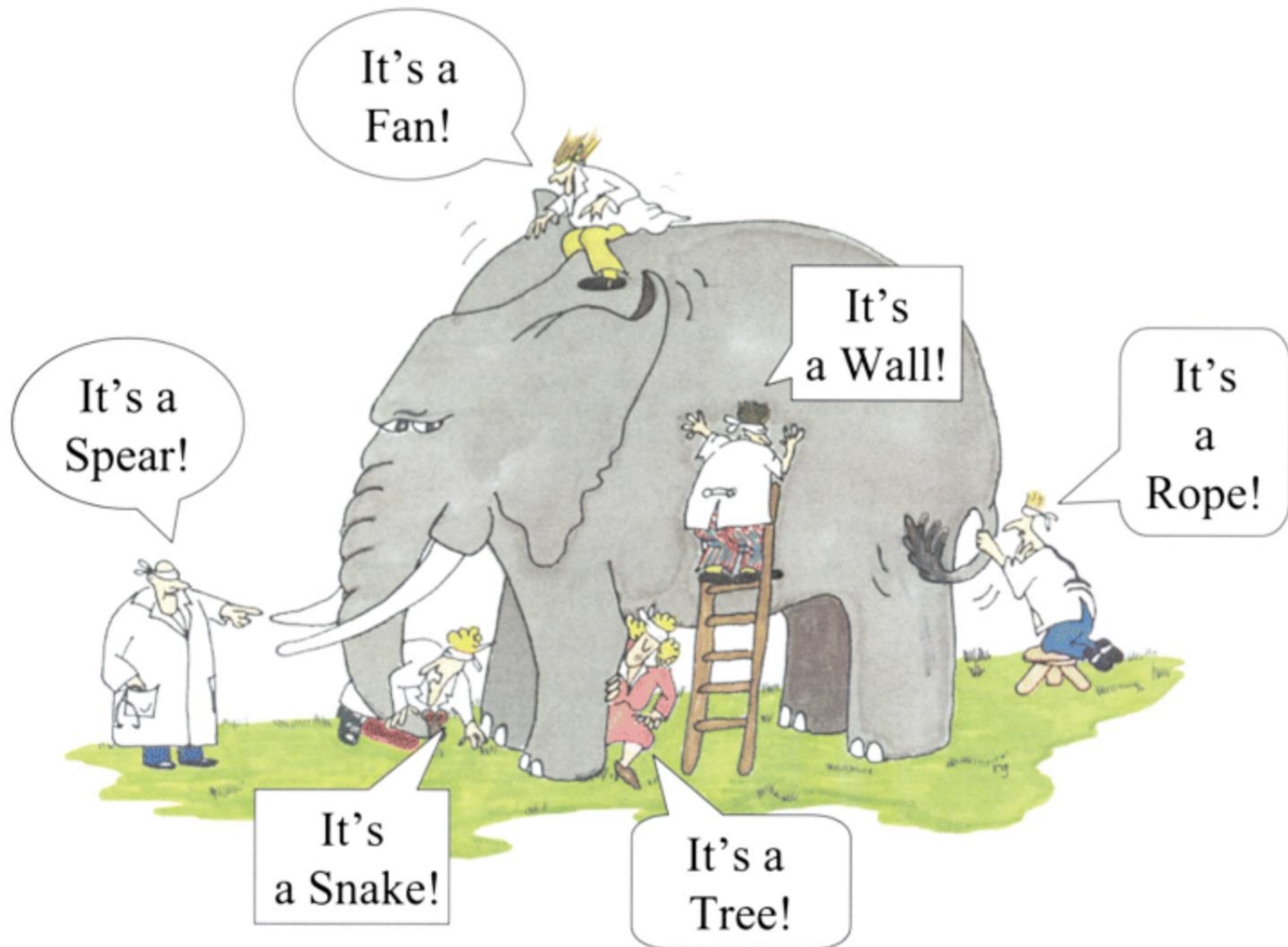
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Genetics Establishes Causality

Δ Target Gene $\rightarrow$ Δ Phenotype

Studying Cancer Without Genetics on Your Side



Genetic Validation Can Take Multiple Forms

- **Germline Human Variants/Mutations**
- **Recurrent Somatic Mutations**
- **(Properly Controlled) Somatic Knockout/Knockdown Studies (e.g. with CRISPR/Cas9, RNA Interference)**
- **Mouse Knockout Studies**

Drugging the “Undruggable”

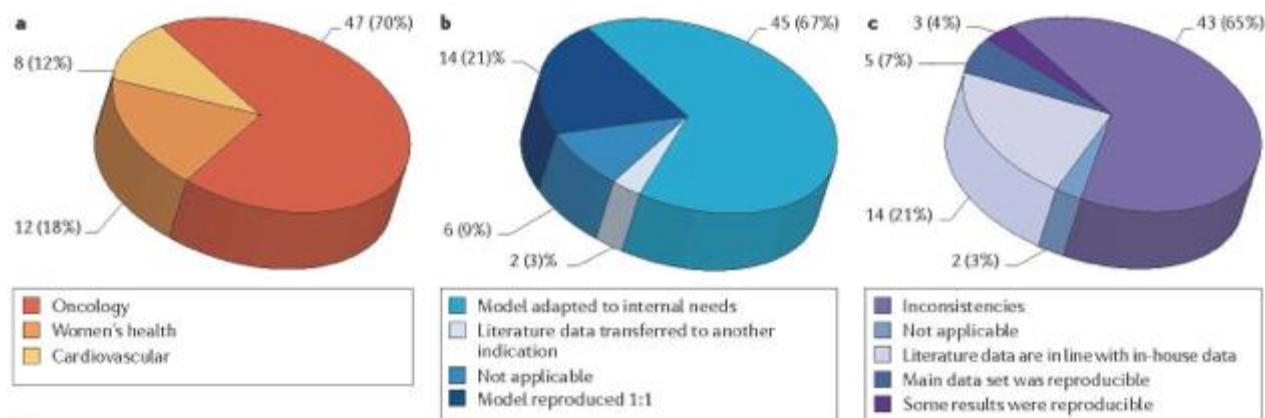
- **Target Critical Downstream Effectors (Epistasis)**
- **Look for Allosteric Inhibitors**
- **Exploit Synthetic Lethal Interactions**
- **Search for Degraders**

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- Mouse Knockout Studies

Believe it or not: how much can we rely on published data on potential drug targets?

Florian Prinz, Thomas Schlange and Khusru Asadullah

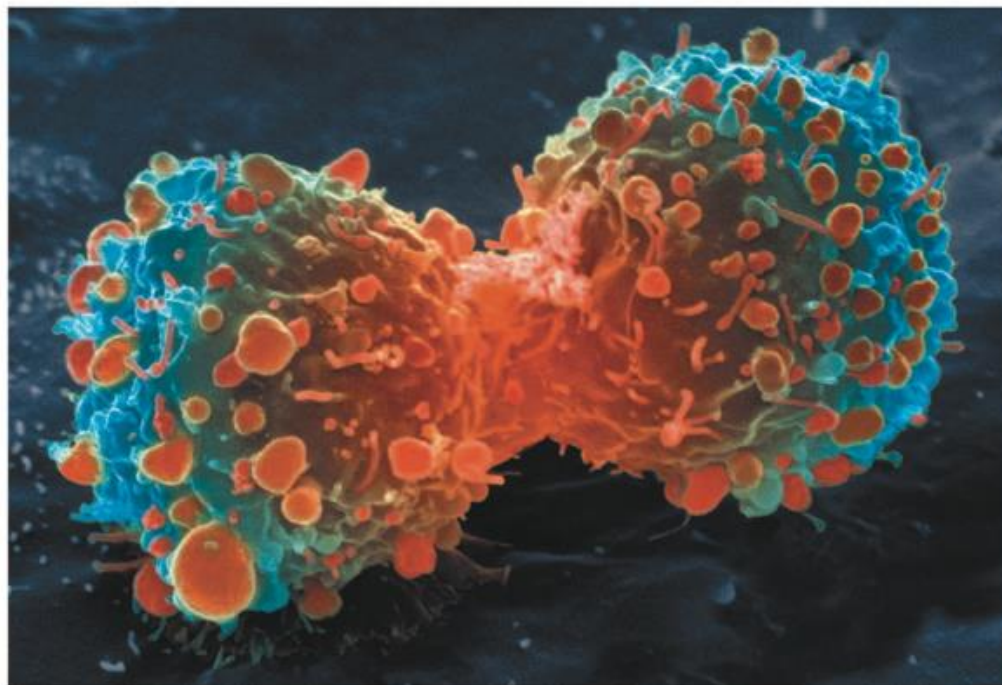


d

	Model reproduced 1:1	Model adapted to internal needs (cell line, assays)	Literature data transferred to another indication	Not applicable
In-house data in line with published results	1 (7%)	12 (86%)	0	1 (7%)
Inconsistencies that led to project termination	11 (26%)	25 (60%)	2 (5%)	4 (9%)

COMMENT

S. SCHMIDTKE/SPR



Many landmark findings in preclinical oncology research are not reproducible, in part because of inadequate cell lines and animal models.

Raise standards for preclinical cancer research

C. Glenn Begley and Lee M. Ellis propose how methods, publications and incentives must change if patients are to benefit.

PERSPECTIVES

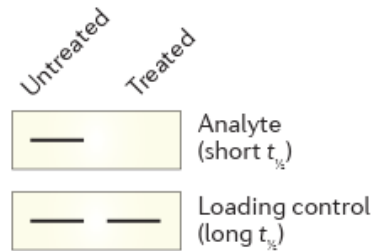
OPINION

Common pitfalls in preclinical cancer target validation

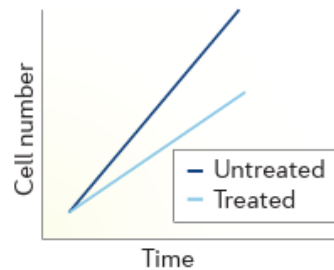
William G. Kaelin Jr

Common "Down" Assays in Cancer Biology

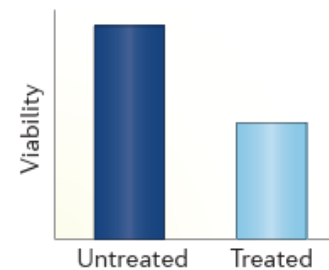
a Pharmacodynamics



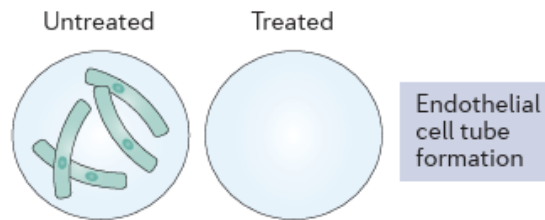
b Proliferation



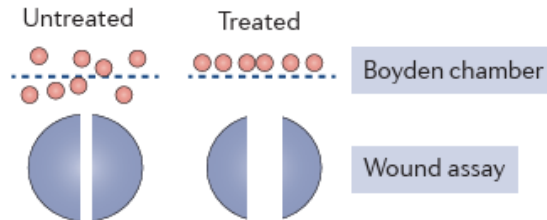
c Apoptosis



d Angiogenesis



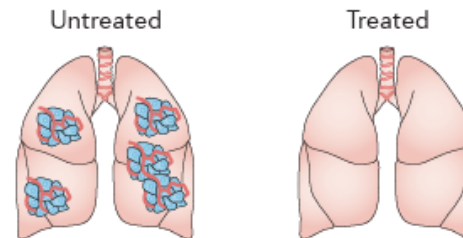
e Invasion



f Tumour formation



g Metastasis



The Off-Target Problem

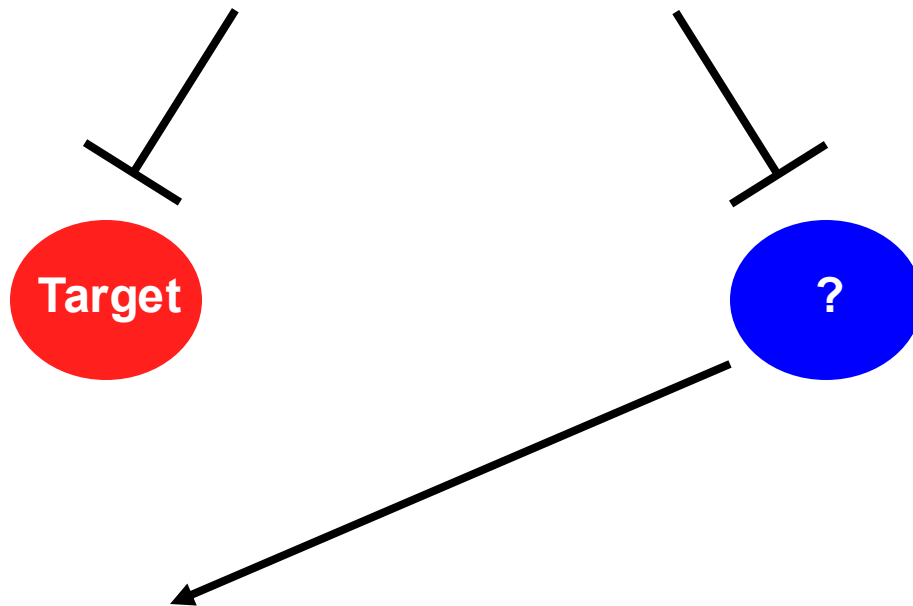
Perturbant (e.g. Drug, siRNA)



Perturbant Phenotype

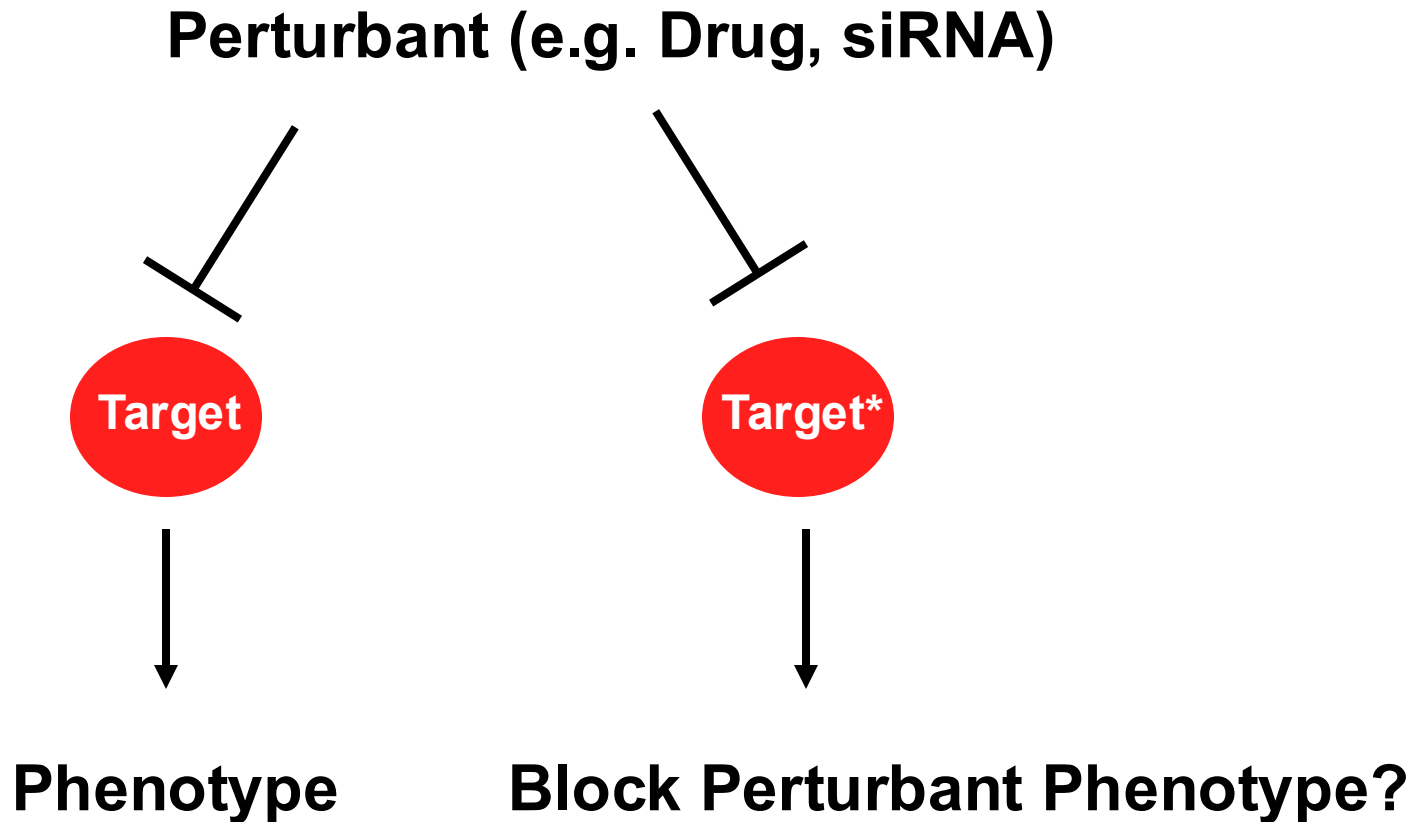
The Off-Target Problem

Perturbant (e.g. Drug, siRNA)



Perturbant Phenotype

Rescue with Perturbant-Resistant Target



LETTER

doi:10.1038/nature19795

On-target efficacy of a HIF-2 α antagonist in preclinical kidney cancer models

Hyejin Cho¹, Xinlin Du², James P. Rizzi², Ella Liberzon¹, Abhishek A. Chakraborty¹, Wenhua Gao¹, Ingrid Carvo^{1,3}, Sabina Signoretti^{1,3}, Richard K. Bruick⁴, John A. Josey², Eli M. Wallace² & William G. Kaelin Jr^{1,5}

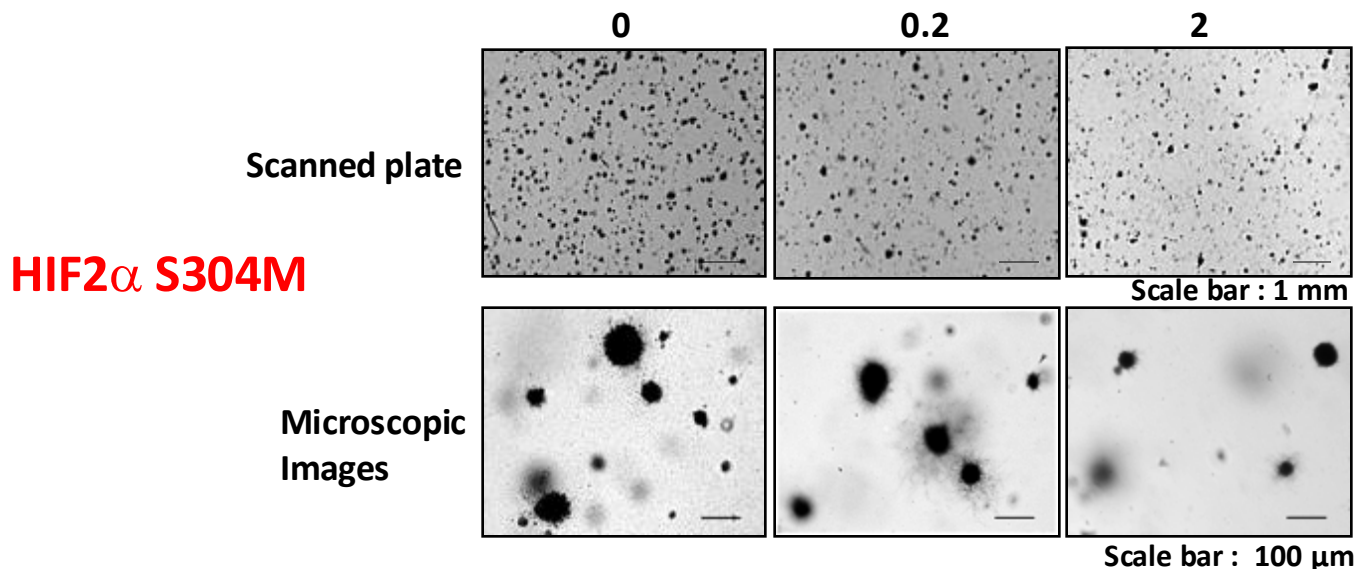
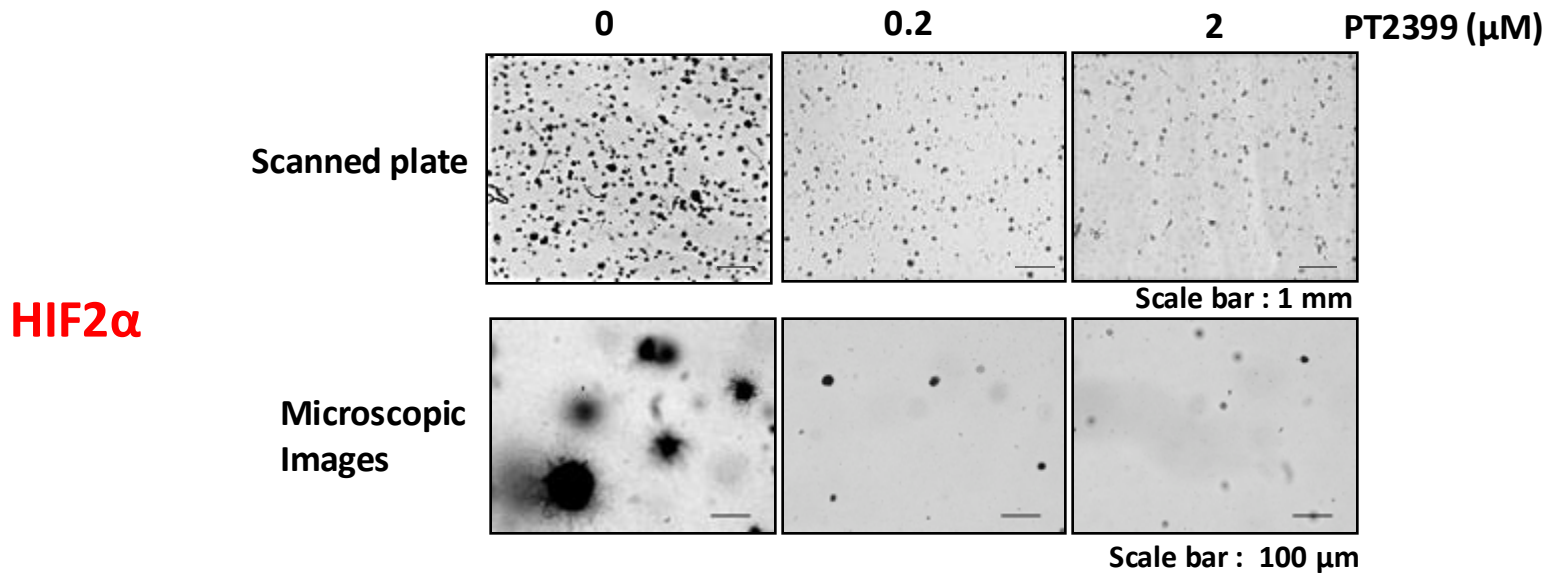
LETTER

doi:10.1038/nature19796

Targeting renal cell carcinoma with a HIF-2 antagonist

Wenfang Chen^{1,2,3*}, Haley Hill^{1,2*}, Alana Christie^{1*}, Min Soo Kim^{1,4*}, Eboni Holloman^{1,2}, Andrea Pavia-Jimenez^{1,2}, Farrah Homayoun^{1,2}, Yuanqing Ma^{1,2}, Nirav Patel^{1,2}, Paul Yell⁵, Guiyang Hao⁶, Qurratulain Yousuf^{1,2}, Allison Joyce^{1,2}, Ivan Pedrosa^{1,6}, Heather Geiger⁷, He Zhang^{1,4}, Jenny Chang¹, Kevin H. Gardner^{8,9,10}, Richard K. Bruick^{1,11}, Catherine Reeves⁷, Tae Hyun Hwang^{1,4}, Kevin Courtney^{1,2}, Eugene Frenkel^{1,2}, Xiankai Sun^{1,6}, Naseem Zojwalla¹², Tai Wong¹², James P. Rizzi¹², Eli M. Wallace¹², John A. Josey¹², Yang Xie^{1,4}, Xian-Jin Xie^{1,4}, Payal Kapur^{1,13}, Renée M. McKay^{1,2} & James Brugarolas^{1,2}

Inhibition of VHL-Defective Renal Cell Carcinoma by PT2399: Soft Agar Assays



Kidney Cancer Models

- **Cancer Cell Lines in 2D Cultures**
- **Cancer Cell Lines in Soft Agar**
- **Subcutaneous Cell Line Xenografts**
- **Orthotopic Cell Line Xenografts**
- **Patient-Derived Xenografts**
- **Genetically-Engineered Mouse Models**

Choosing Models

- **The Suitability of a Model DEPENDS ON THE QUESTION(S) BEING ASKED**
- **Corroborating Lines of Evidence are a Scientist's Best Friend***

Genetic Validation of CTLA-4

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Science 10 Nov 1995:
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DOI: 10.1126/science.270.5238.985

Challenges Making VHL-/- ccRCC GEMS

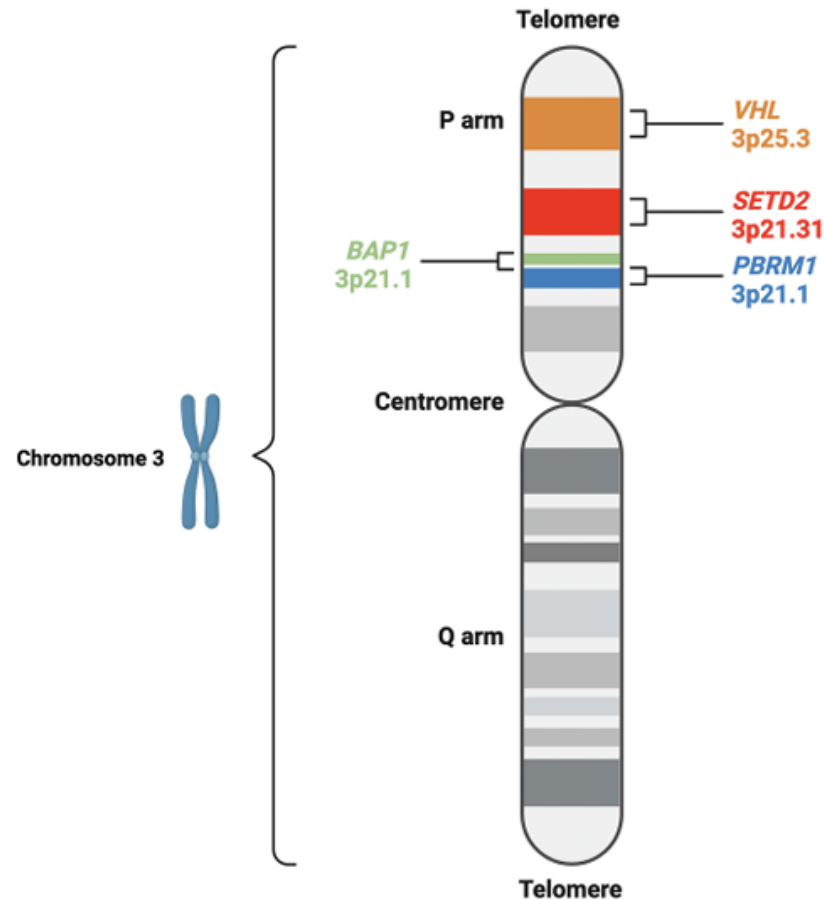
Germline Cancer Gene Mutations: Humans and Mice Might be “Wired” Differently with Respect to Tissue Tropism

Humans

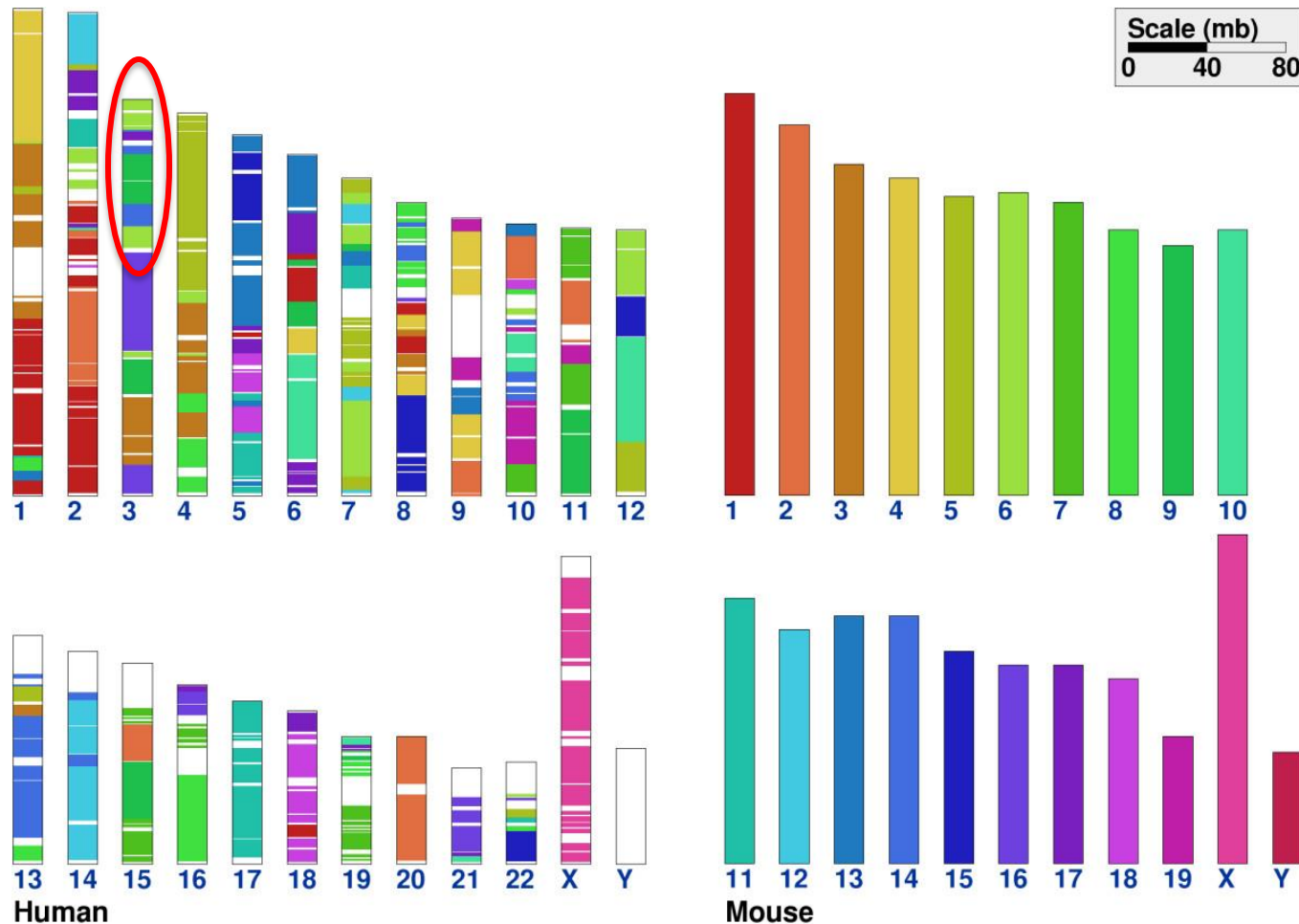
Mice

RB1	Retinoblastoma, Sarcoma	Pituitary and Thyroid NE Tumors
TP53	Breast Ca, Sarcoma, etc.	Lymphoma
APC	Large Bowel Cancer	Small Bowel Cancer
BRCA1	Breast Ca, Ovarian Ca, etc.	None
VHL	Kidney Ca, etc.	None

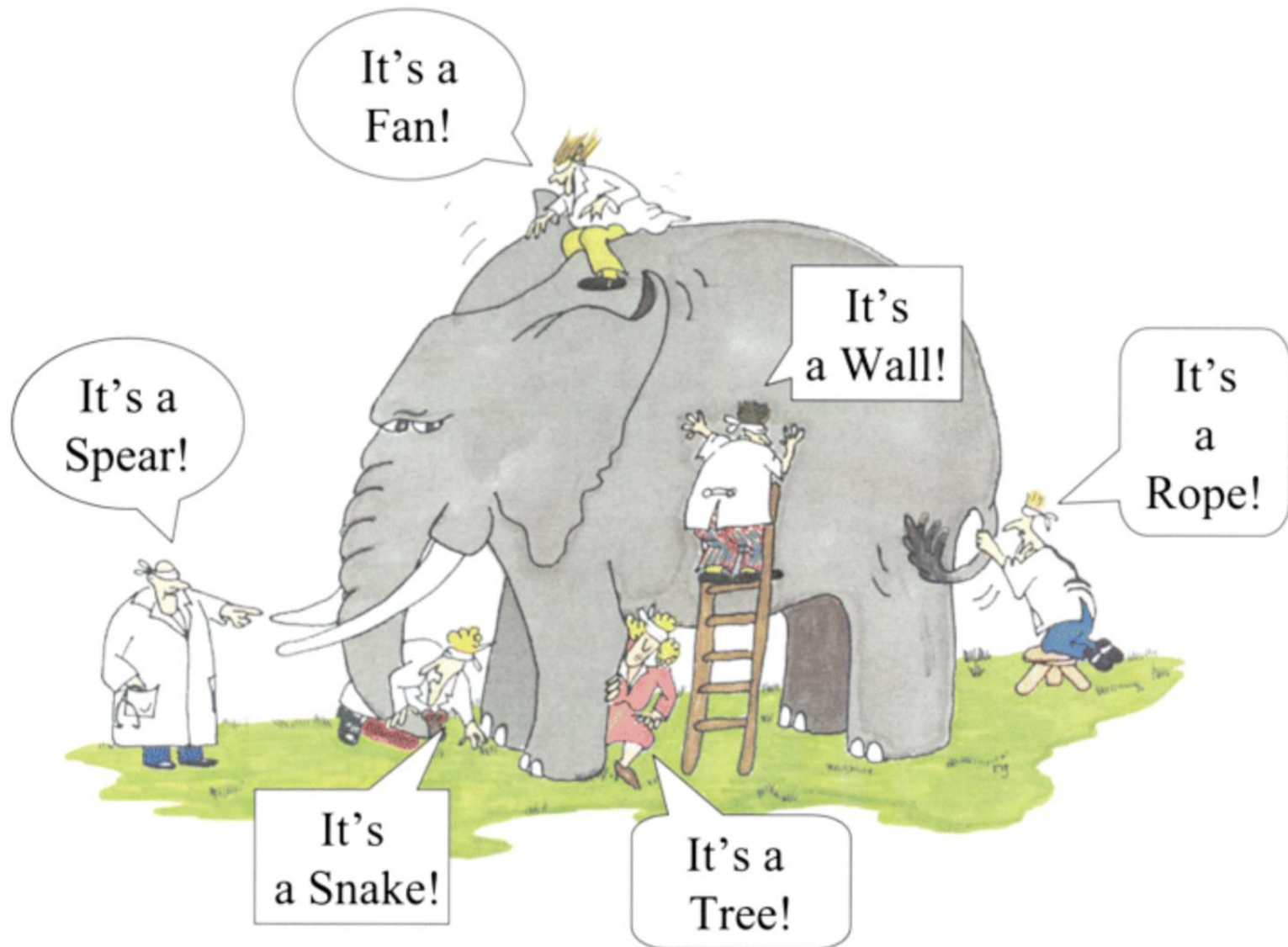
Chromosome 3p Loss Affects Multiple Kidney Cancer Suppressors



Lack of Synteny Between Humans and Mice

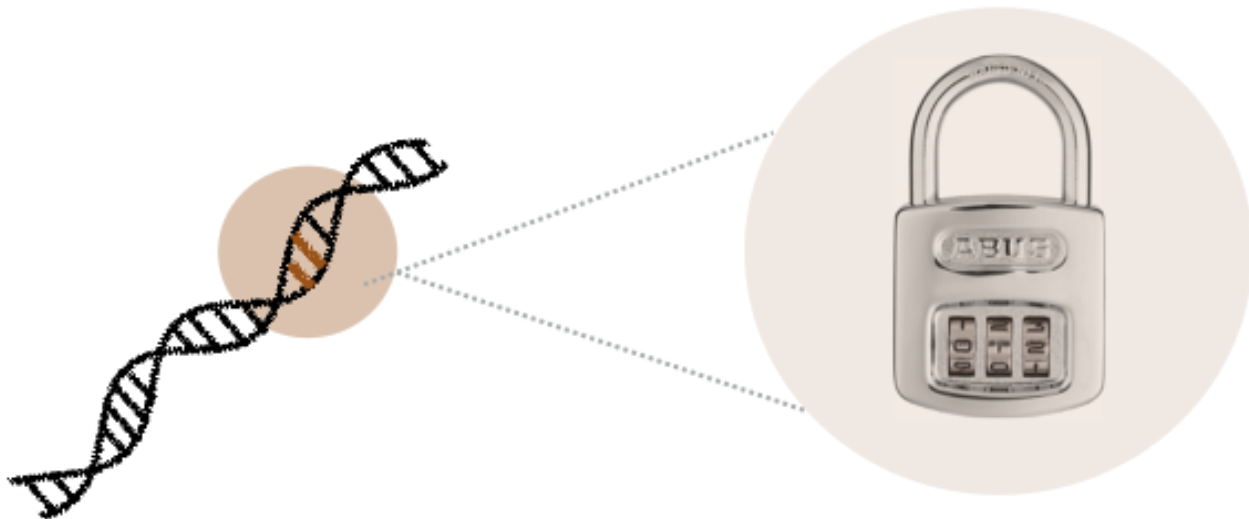


Studying Cancer Without Genetics on Your Side



The Multiple Mutation Problem

“Cancer Cells harbor too many mutations- we will never be able to fix them all” (countless naysayers)

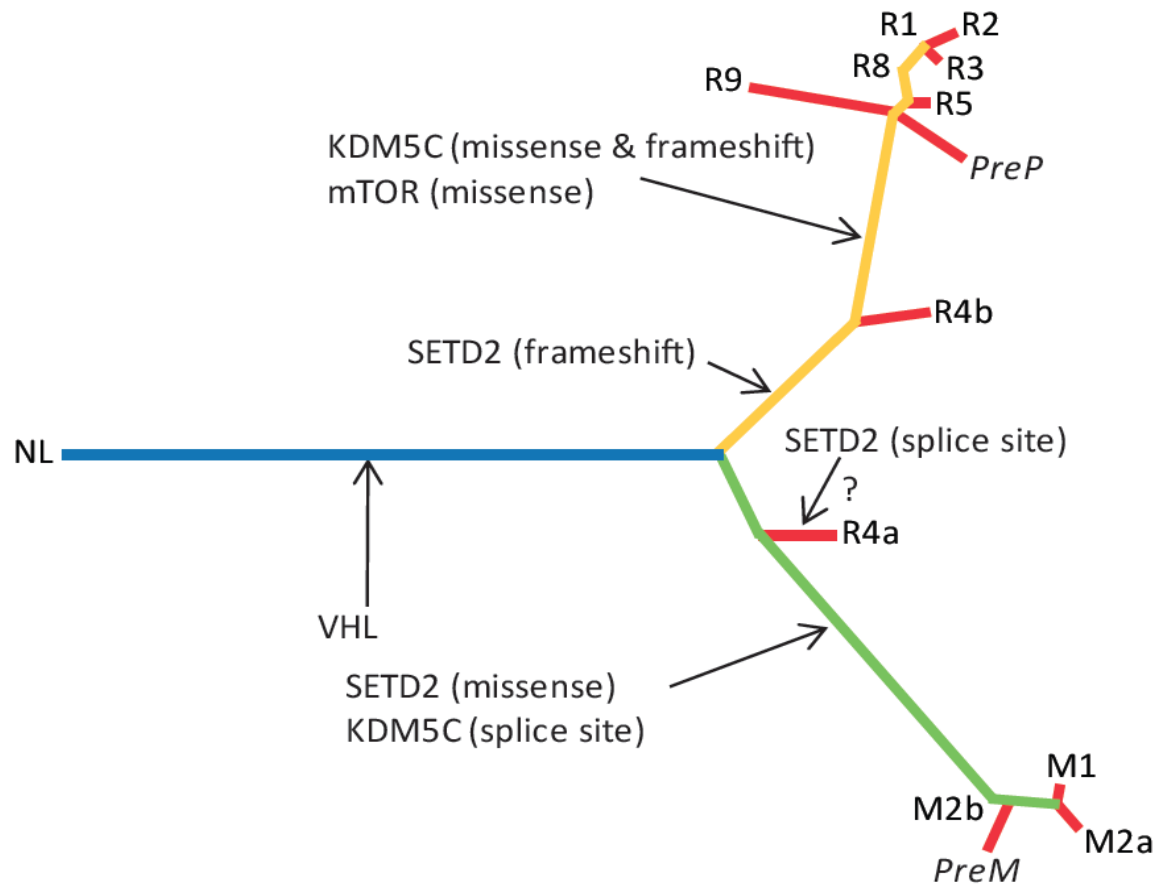


Genetic Interdependence

Hypothesis: Some Late Mutations (Driver or Passenger) in a Tumor are Only Tolerated Because of the Mutations that Preceded Them

If True, Correcting Early Mutations should Selectively Kill Tumor Cells

Truncal vs Branch Mutations During Tumor Evolution: One ccRCC Example



Successfully Targeting Early Mutations

- **Her2/Neu –Breast Cancer**
- **EGFR – Lung Cancer**
- **c-KIT - GIST**
- **B-Raf - Melanoma**
- **ALK – Lung Cancer**
- **ROS- Lung Cancer**
- **Etc.**

Clinical Response and Resistance to RAF Inhibition in Melanoma



October, 2009



January, 2010

Solution 1: Combination Chemotherapy

1 cm³ Tumor = 10⁹ Cells

Tumor Burden in Most Cancer Patients = 10¹⁰-10¹² Cells

Assume 1/1,000,000 Cells Resistant to Any One Drug

Then:

Chance of Cure with Single Agent is Approximately ZERO

However:

**Probability of Any One Cell Being Resistant to 3 Non-Cross
Resistant Drugs is $10^{-6} \times 10^{-6} \times 10^{-6} = 10^{-18}$**



A curative combination cancer therapy achieves high fractional cell killing through low cross-resistance and drug additivity

Adam C Palmer^{1†}, Christopher Chidley^{1†}, Peter K Sorger^{1,2*}

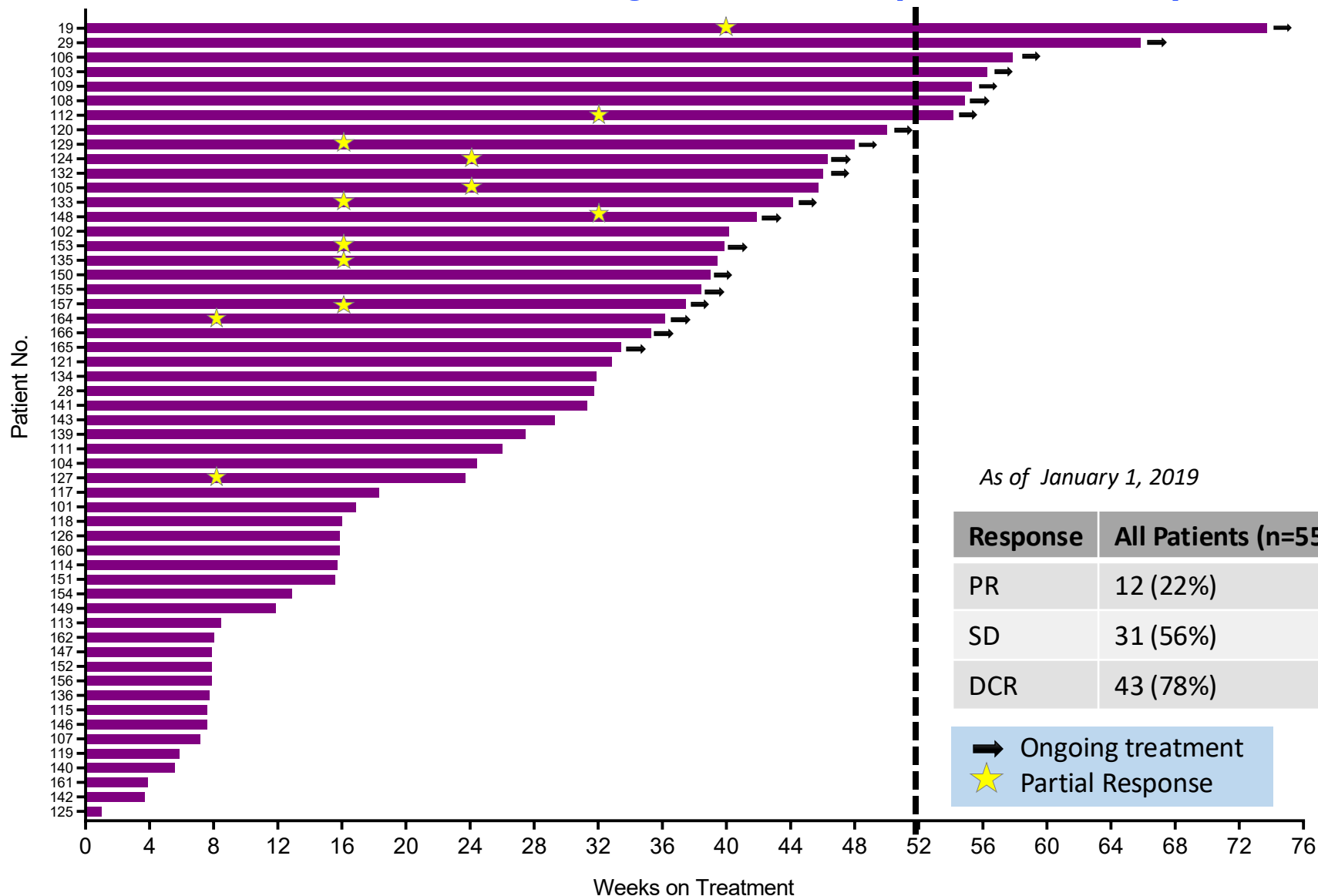
¹Laboratory of Systems Pharmacology, Harvard Medical School, Boston, United States; ²Department of Systems Biology, Harvard Medical School, Boston, United States

Getting to Combinations

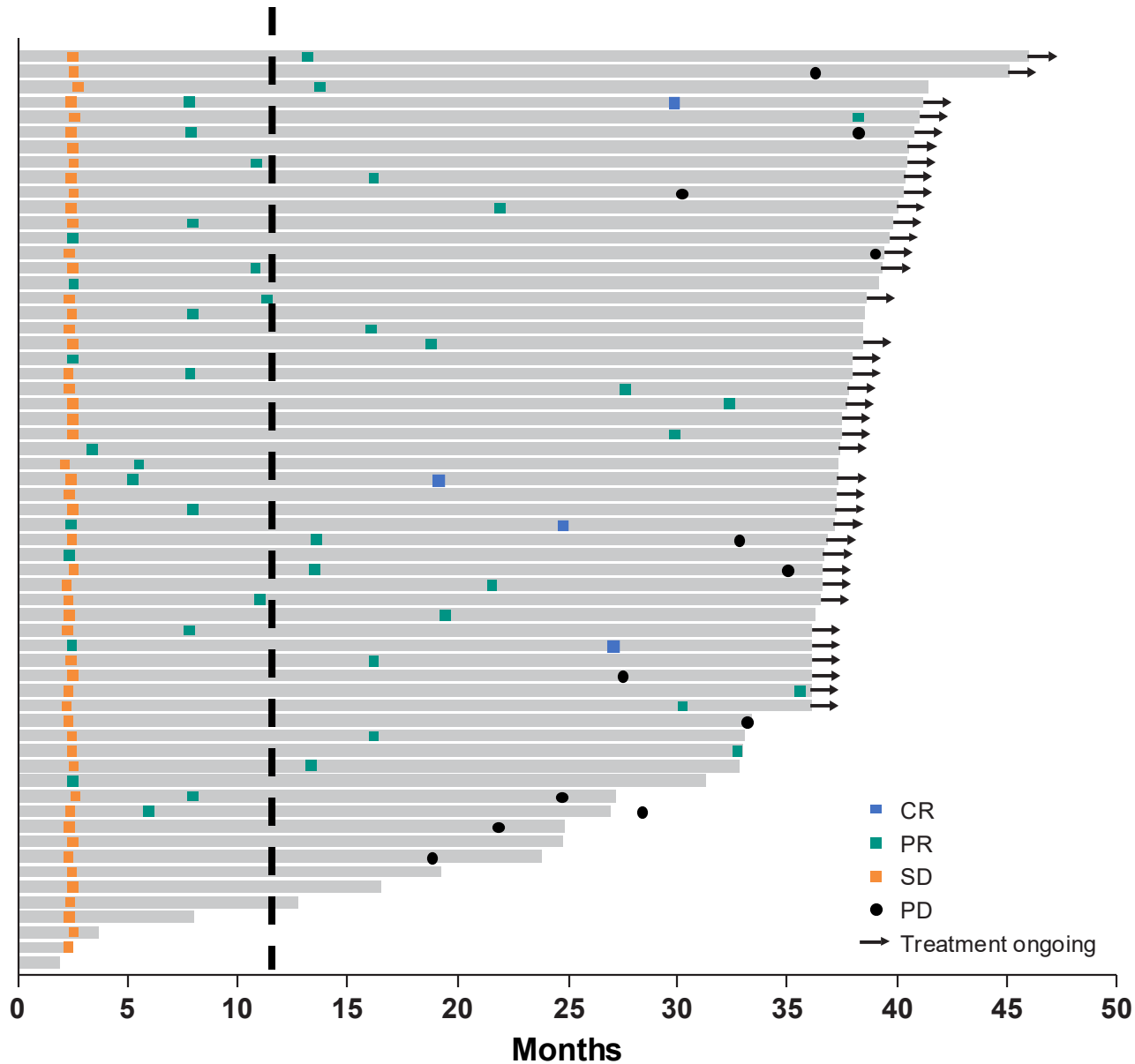
- **Potency and Specificity Will Matter (!)**
 - The best drug for the eventual combination might not be the best drug as monotherapy
- **Need to Get Out of Rotating Single Agent Mindset**
- **Beware of Using Drugs at Single Agent MTD**
- **Need to Think Creatively About Clinical Trial Designs**

Solution 2. Treat Earlier!

HIF2 Inhibitor Belzutifan in Patients with Advanced Kidney Cancer (Phase 1/2)

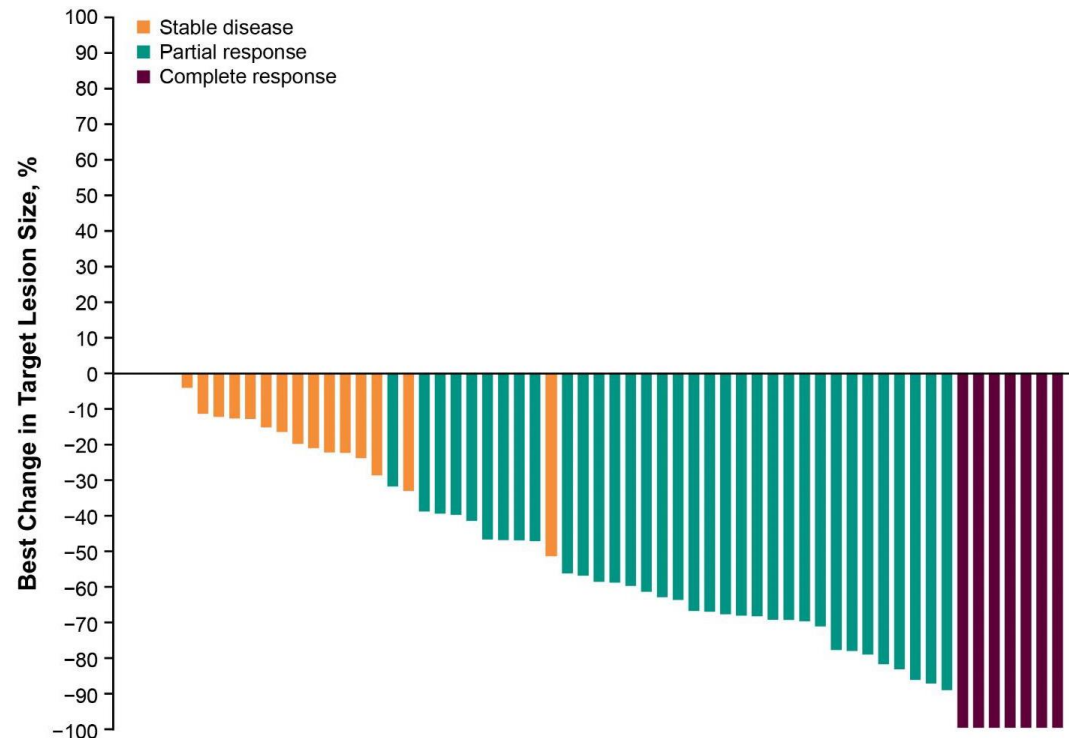


Treatment of VHL Disease Patients with Belzutifan



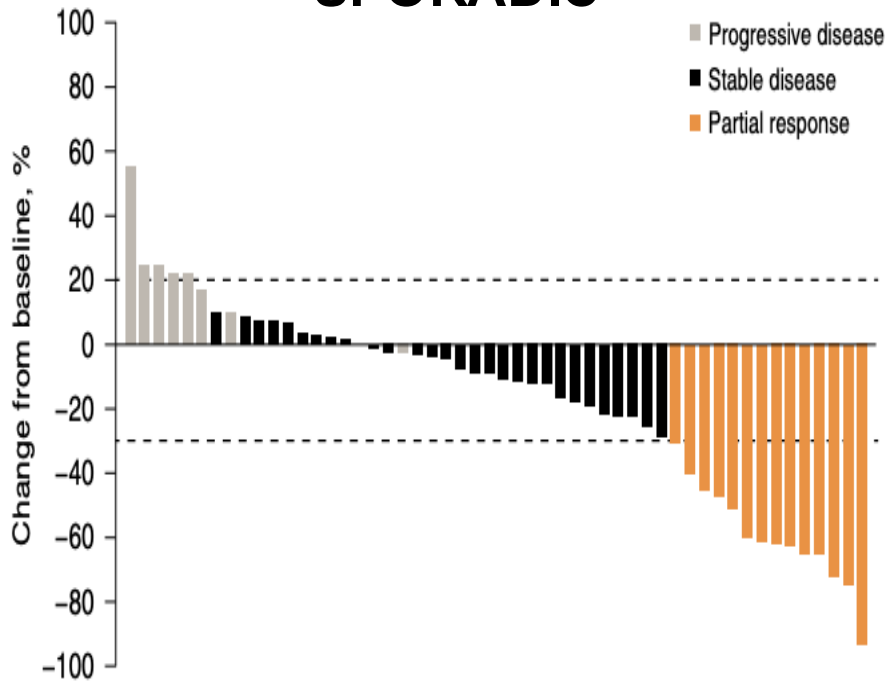
Treatment of VHL Disease–Associated RCC with Belzutifan

	RCC N = 61
ORR, % (95% CI)	67 (54-79)
Best response n (%)	
CR	7 (11)
PR	34 (56)
SD	19 (31)
PD	0
NE ^a	1 (2)



Treatment of ccRCC with Belzutifan

SPORADIC



Choueiri et al Nat Med 2021

VHL DISEASE



Srinivasan et al Lancet Oncology 2025

