

Serendipities of acquired immunity

Nobel Lecture

December 7, 2018

Tasuku Honjo

Kyoto University Institute for Advanced Study
and Graduate School of Medicine

My family (1955)



Cassini: Earth and Saturn

The Day the Earth Smiled



“

The telescopic view of Saturn fascinated me. I dreamed of becoming an astronomer.”

↑ Earth

Through the brilliance of Saturn's rings, Cassini caught a glimpse of a far-away planet and its moon. At a distance of just under 900 million miles, Earth shines bright among the many stars in the sky, distinguished by its bluish tint.

Saturn.

Actual photo taken on June 5, 2016

https://www.nasa.gov/mission_pages/cassini/multimedia/pia17171.html

Inspired by biography of Hideyo Noguchi (1876~1928)



Rockefeller Univ.

- Identified Syphilis spirochete as the cause of progressive paralysis
- Died in Ghana during pursuit of yellow fever pathogen

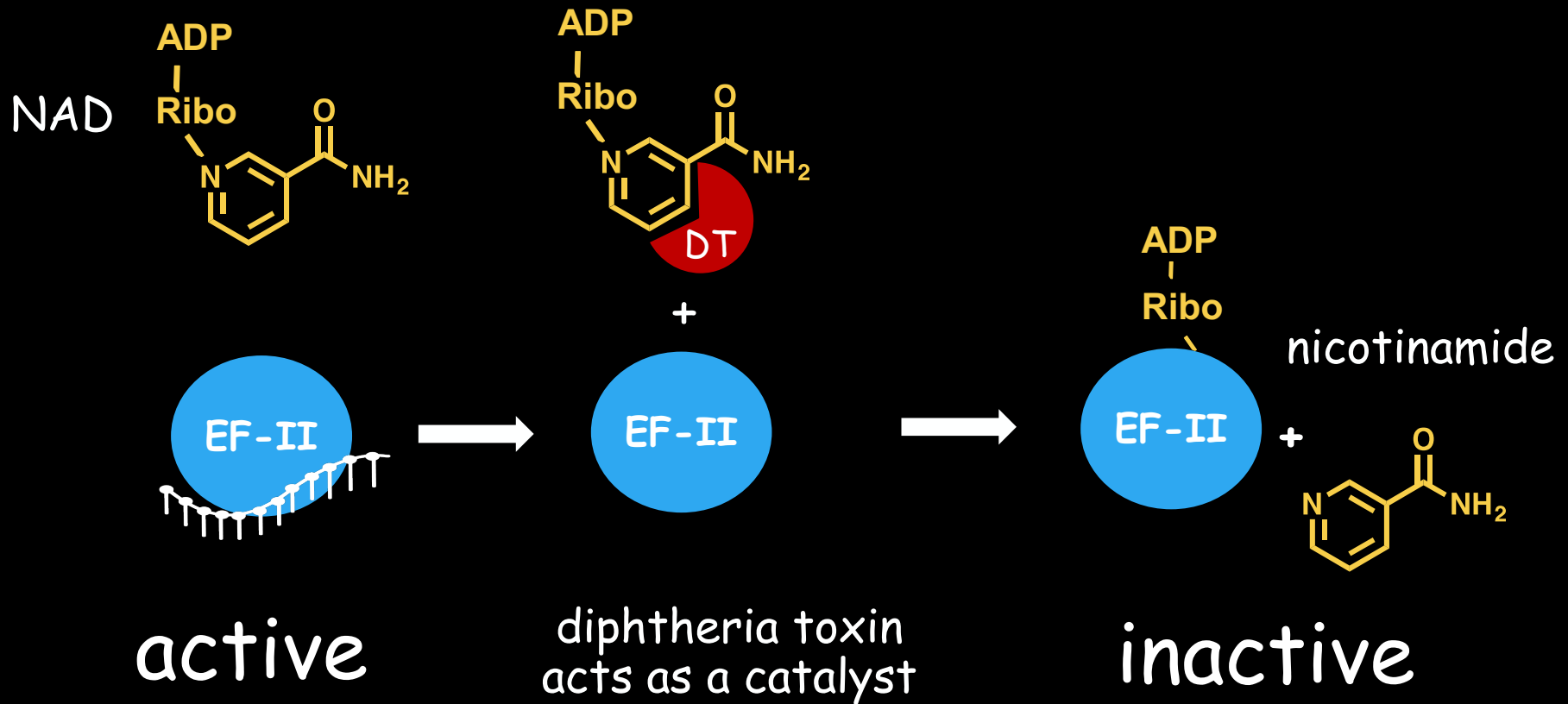
With Osamu Hayaishi



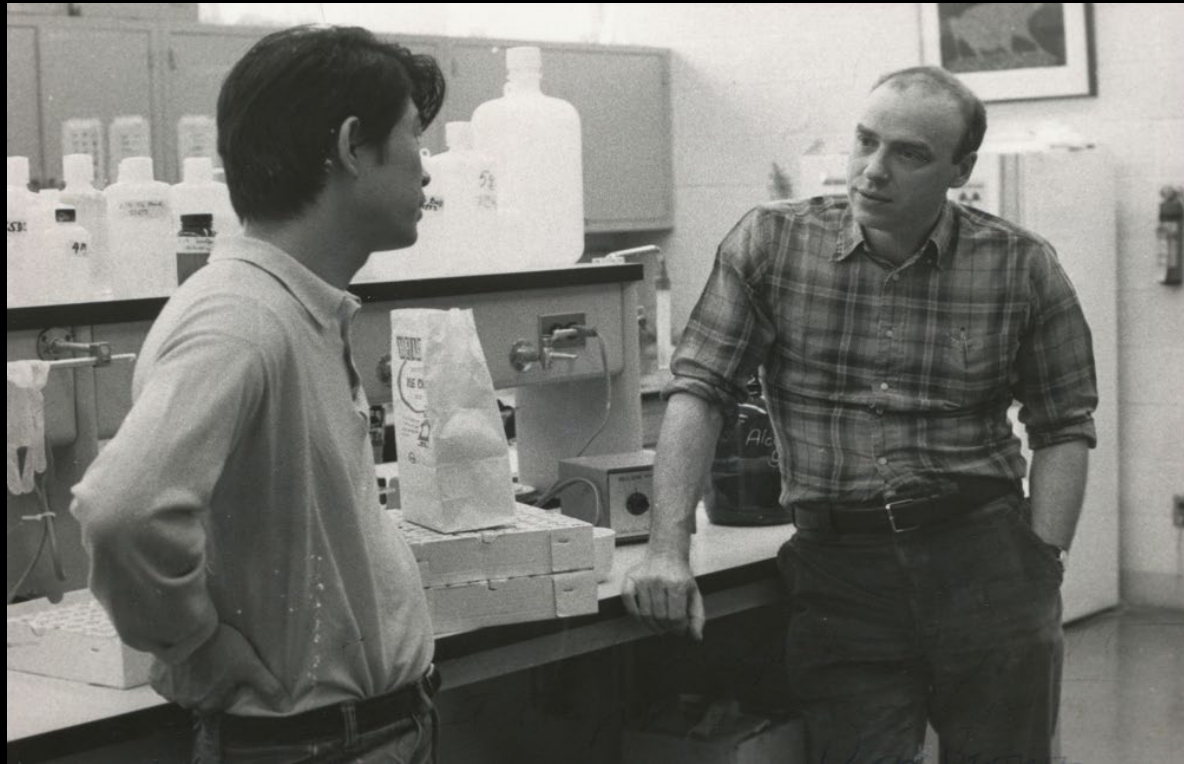
With Jacques Lucien Monod 1966



Diphtheria toxin inactivates protein synthesis factor by ADP-ribosylation



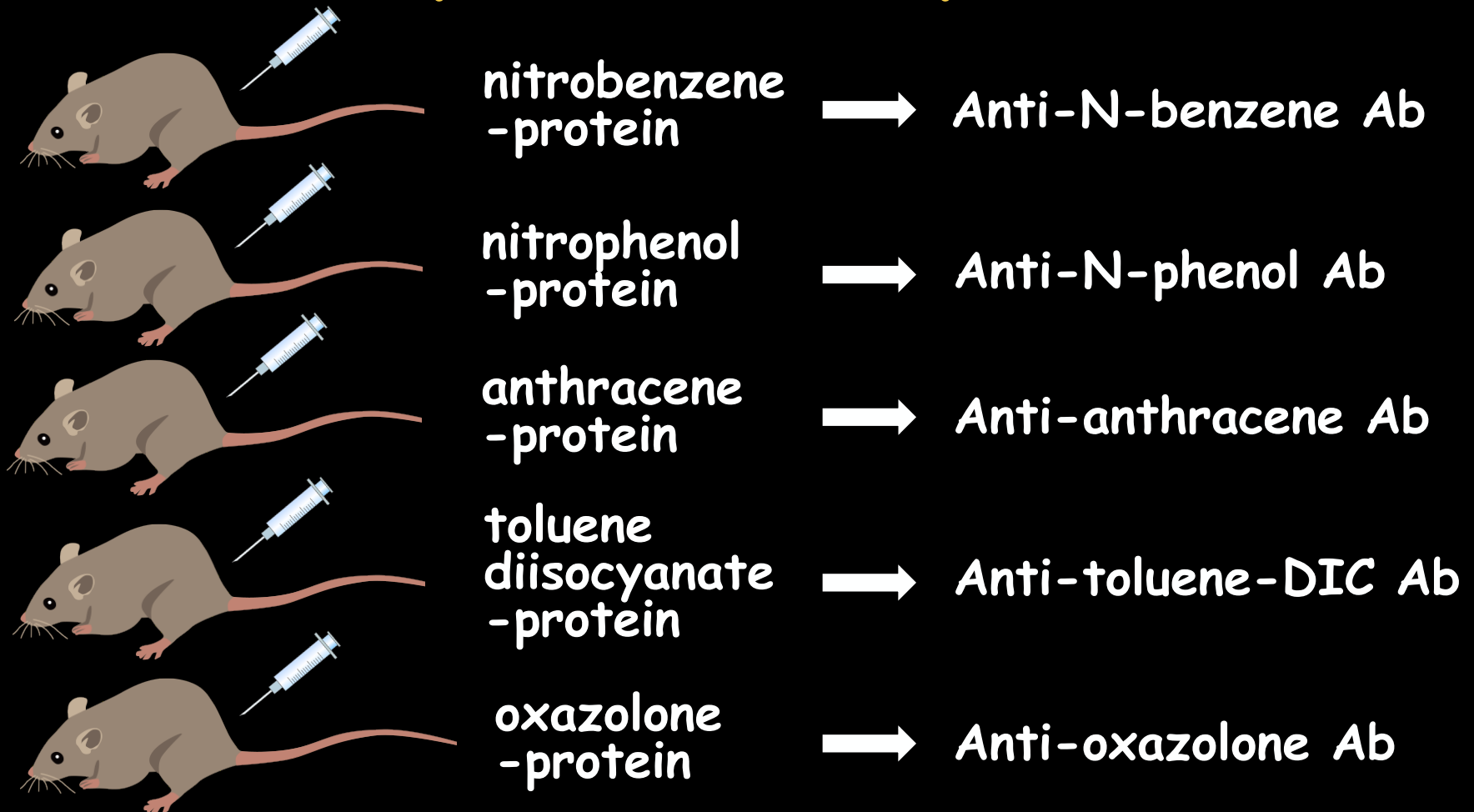
Donald Brown at Carnegie Institution in Baltimore 1971



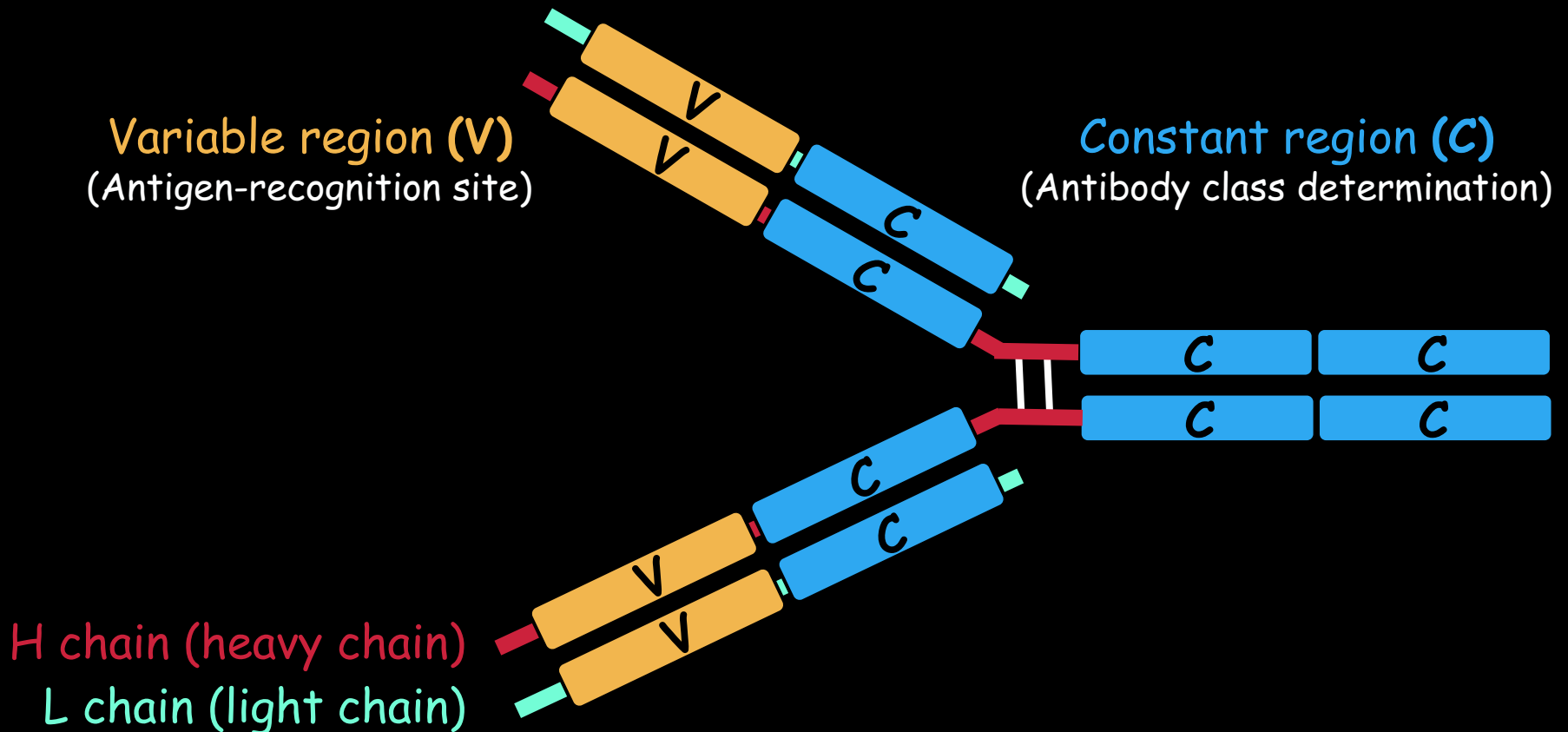
Mystery of immune response in 1950~1970

How can animals generate antibodies specific to an almost infinite number of antigens, including artificial chemicals?

Why can animals generate specific antibodies to almost all unexperienced compounds?



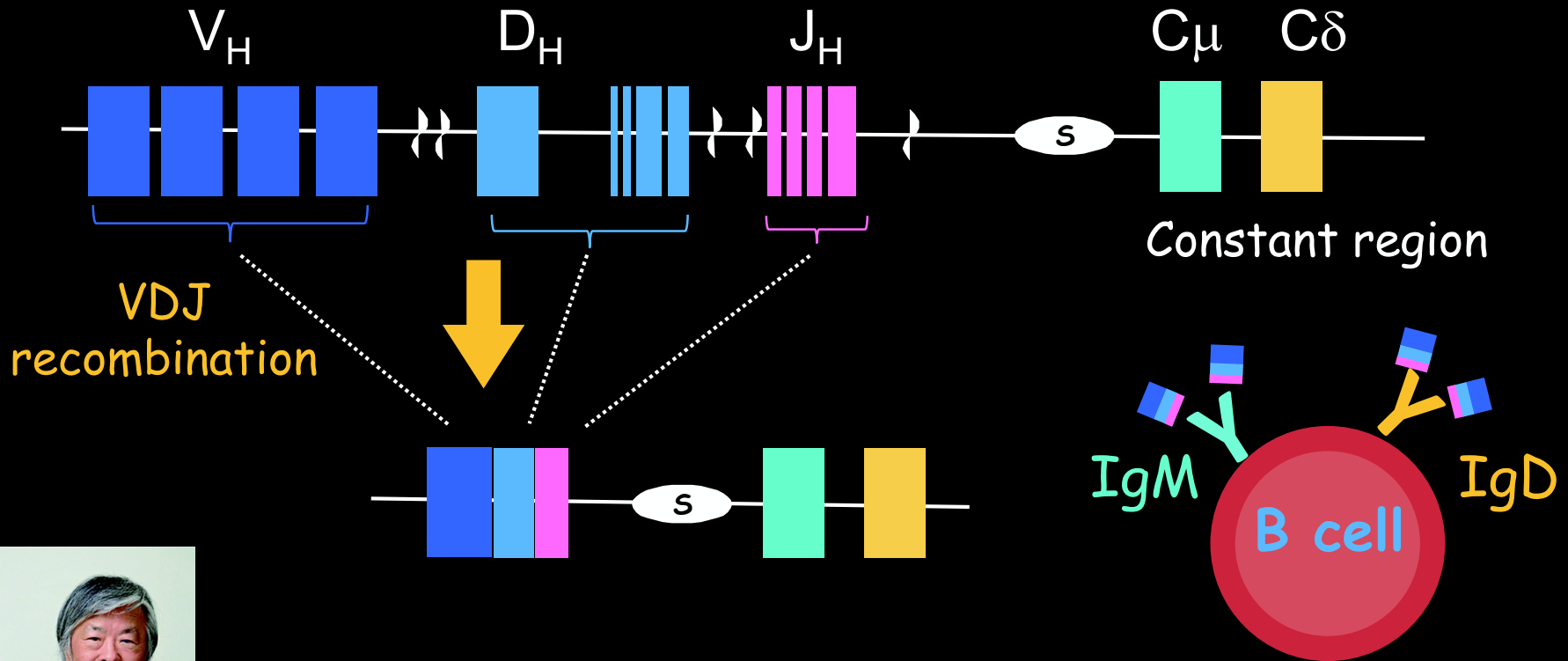
Structure of antibody identified by 1970



Philip Leder at NIH 1973



VDJ recombination generates V region repertoire during differentiation



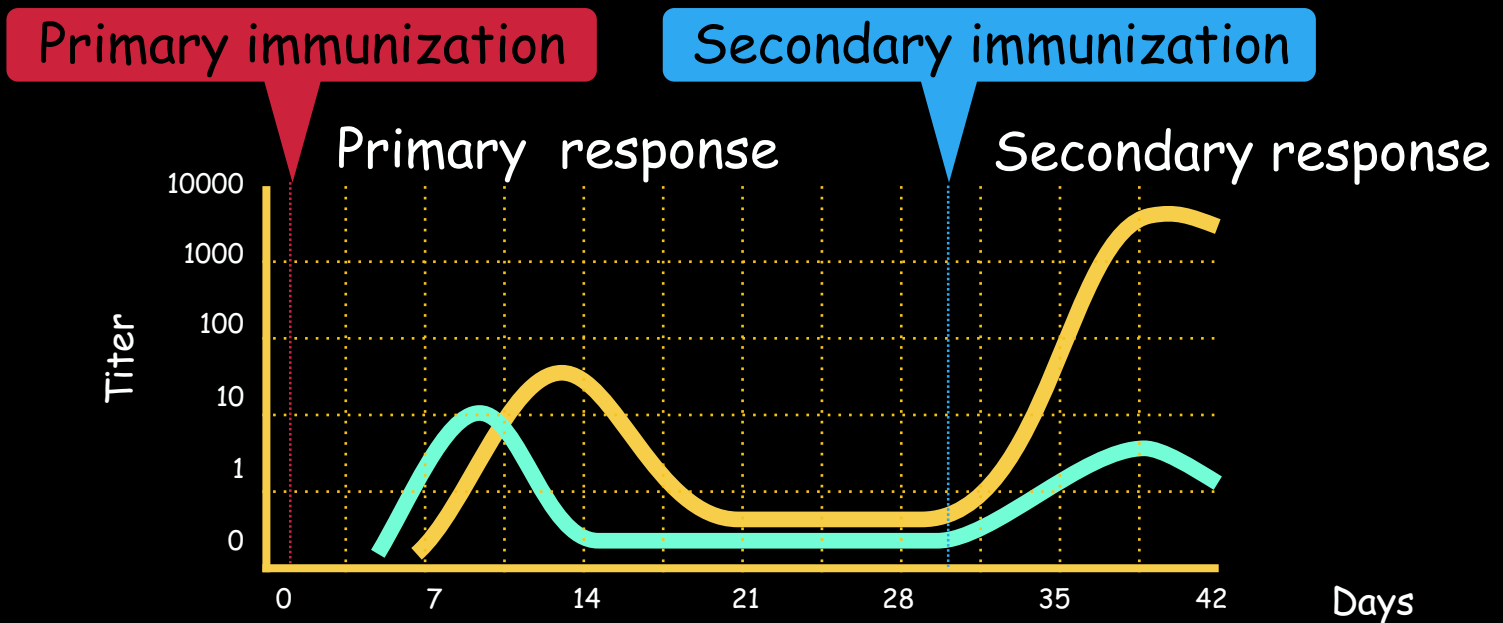
C. Brack et al., Cell (1978)

S. Tonegawa

University of Tokyo, Dept. of Nutrition (Professor Yoshinaga Mano) 1974

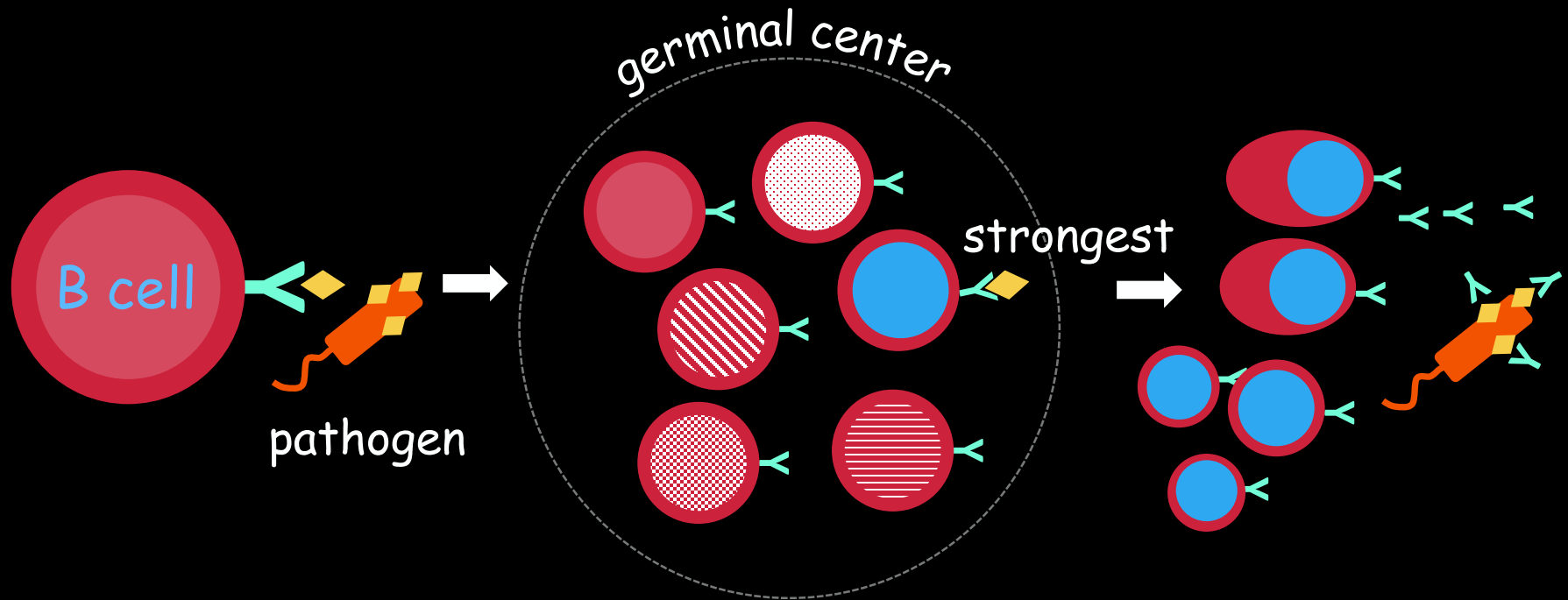


Antibody memory generation by vaccine (antigen) administration



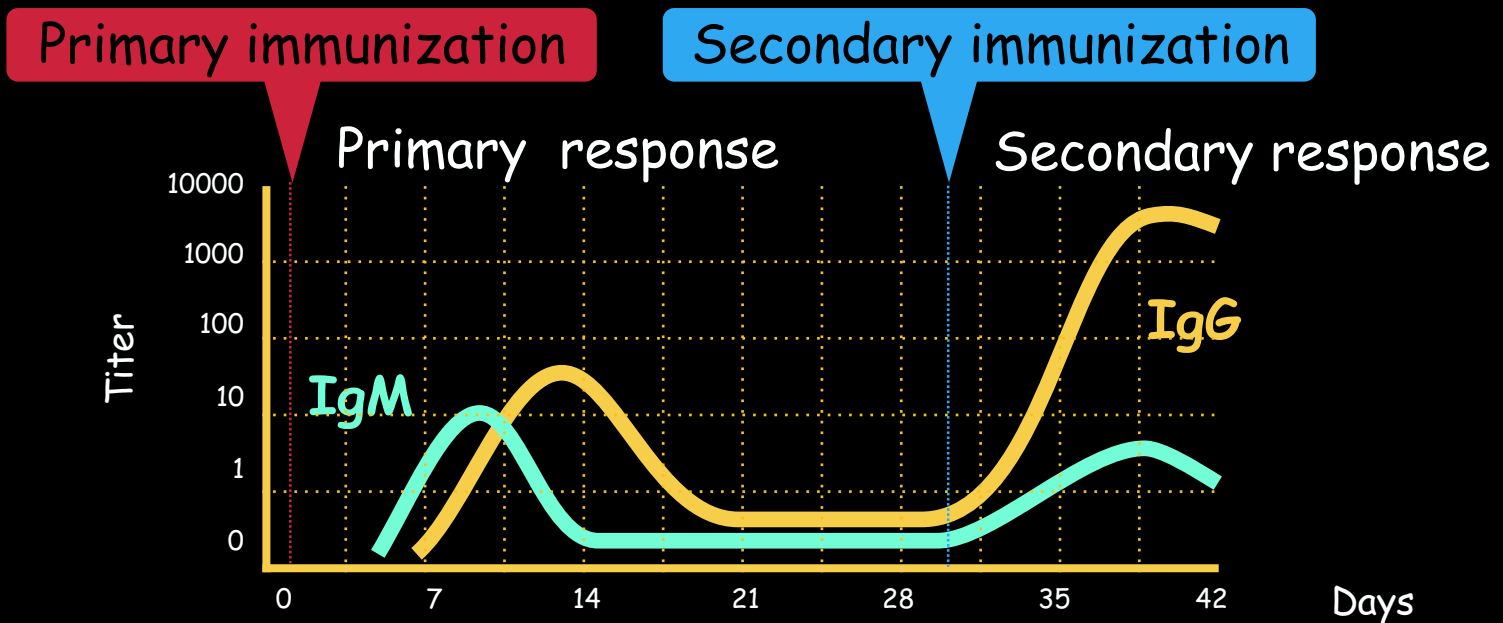
**Increase in antigen binding capability
(somatic hypermutation of variable region)**

Somatic hypermutation (SHM) mutates V region and only good antibodies are selected



Darwinian principle

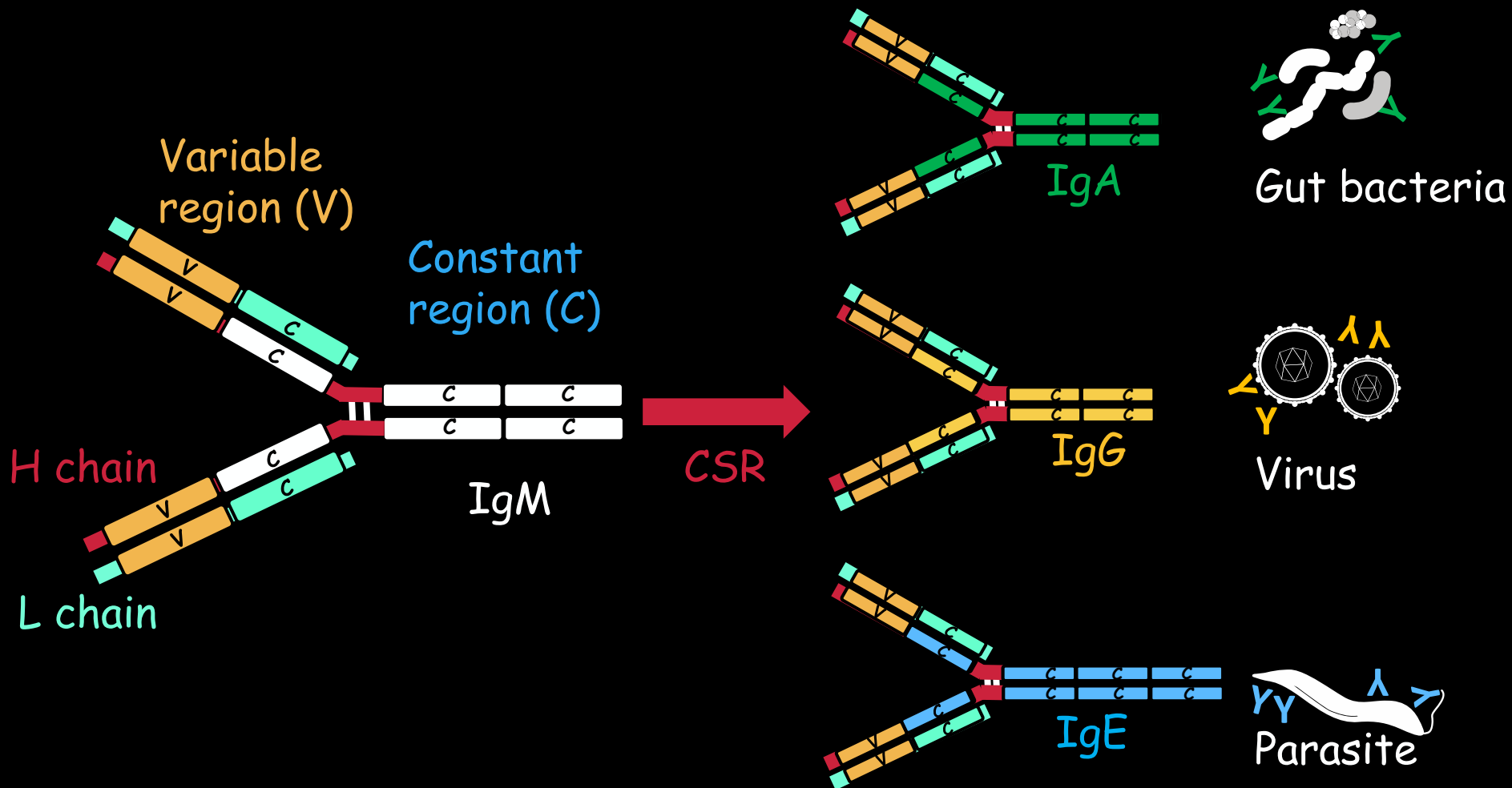
Antibody memory generation by vaccine (antigen) administration



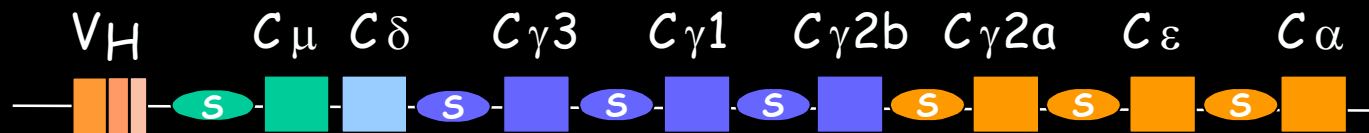
Increase in antigen binding capability
(somatic hypermutation of variable region)

? Increase in antigen processing ability
(class switch of constant region)

Class switching changes the H chain constant region and antibody function



Class switch recombination takes place by deletion of a large DNA segment



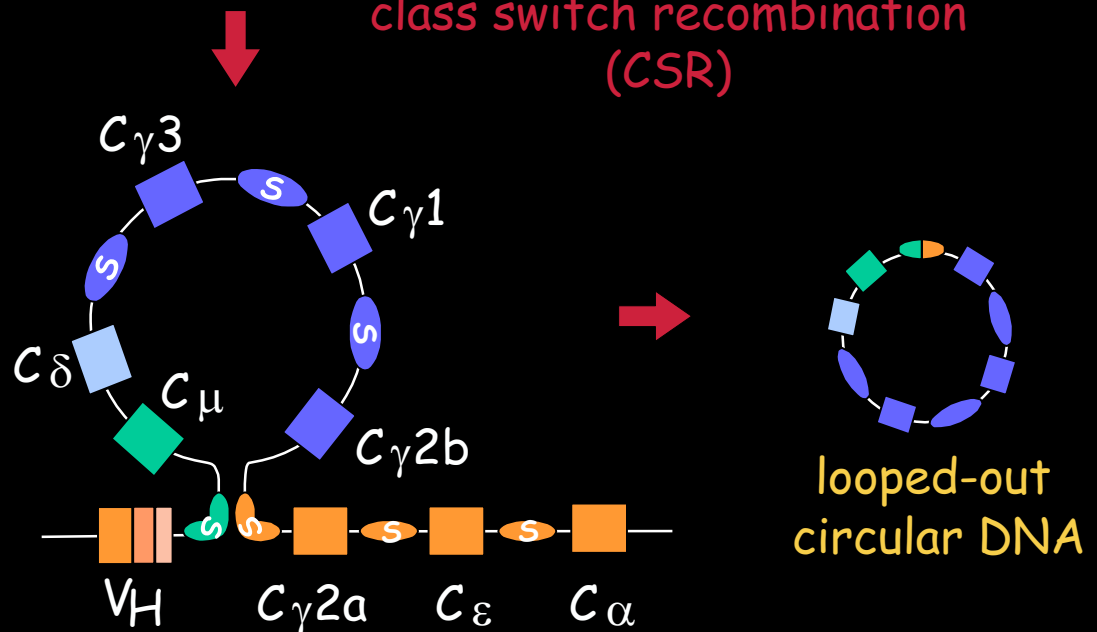
class switch recombination (CSR)



T. Kataoka



A. Shimizu



T. Honjo & T. Kataoka, PNAS (1978)

T. Kataoka *et al.*, PNAS (1980)

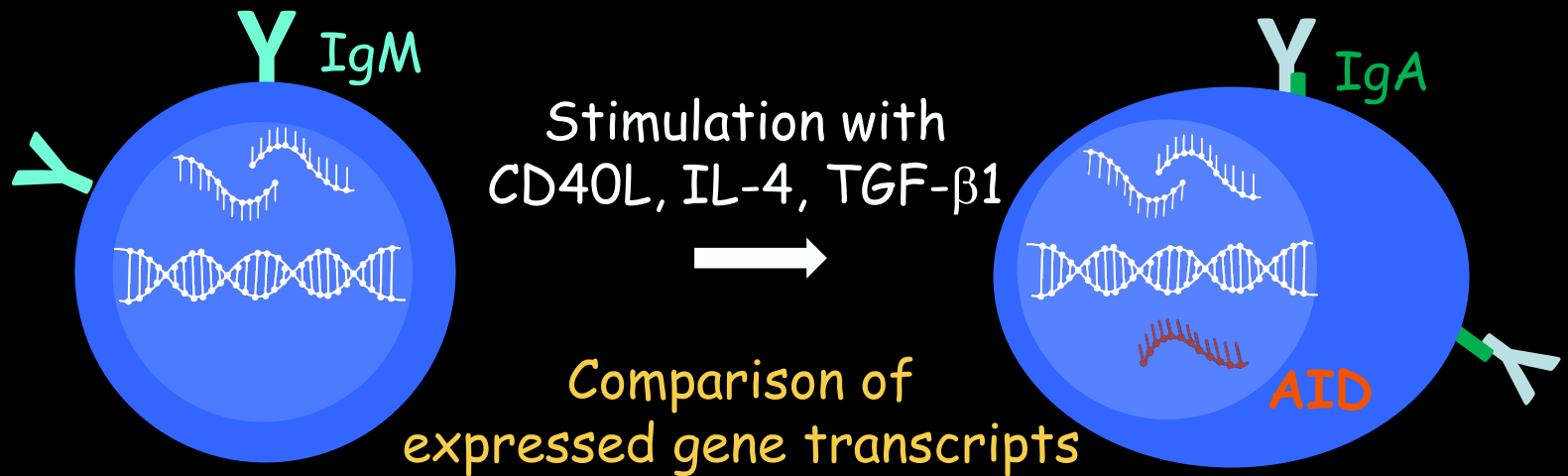
A. Shimizu *et al.*, Cell (1982)

The 55th Nobel Symposium
"Genetics of the Immune Response"
Saltsjobaden, Sweden, June 15 - 17, 1982



Matthias Wabl, Göran Möller (coorganizer) Leroy Hood

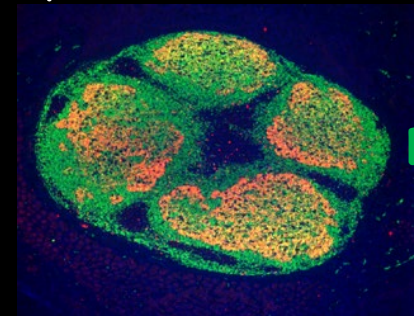
Discovery of AID by comparison of gene expression before and after CSR



Activation **I**nduced
cytidine **D**eaminase

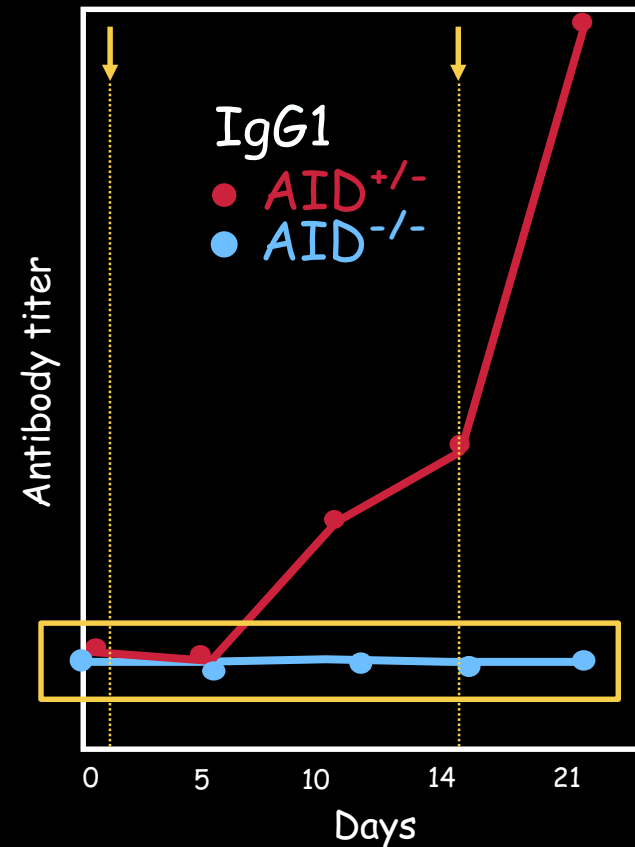
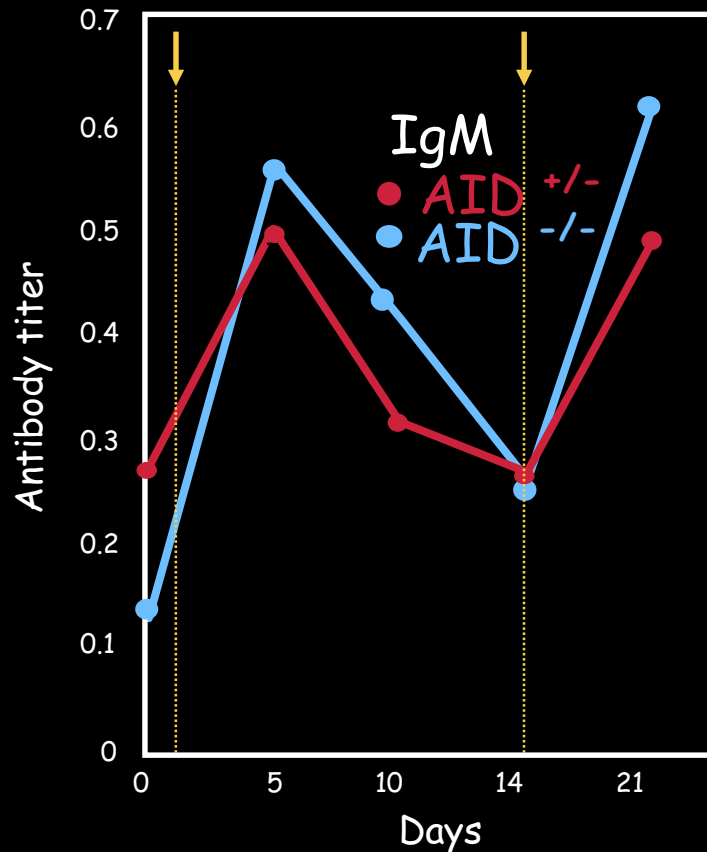


M. Muramatsu *et al.*, J.Biol. Chem. (1999)

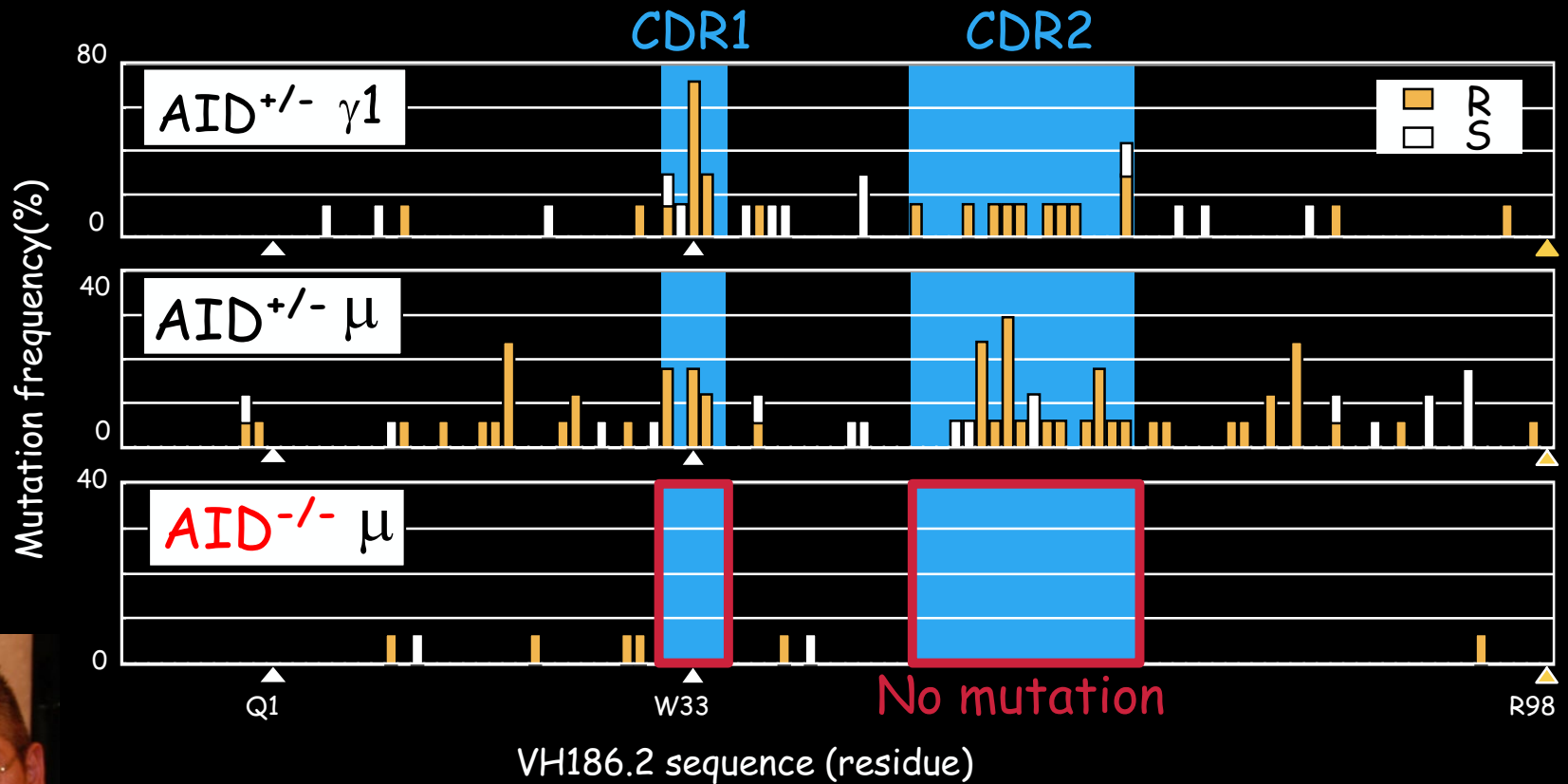


Expressed in germinal center

Defective IgG response to antigens (Sheep Red Blood Cell) in AID deficient mice



AID deficient mice fail to accumulate mutations



K. Kinoshita

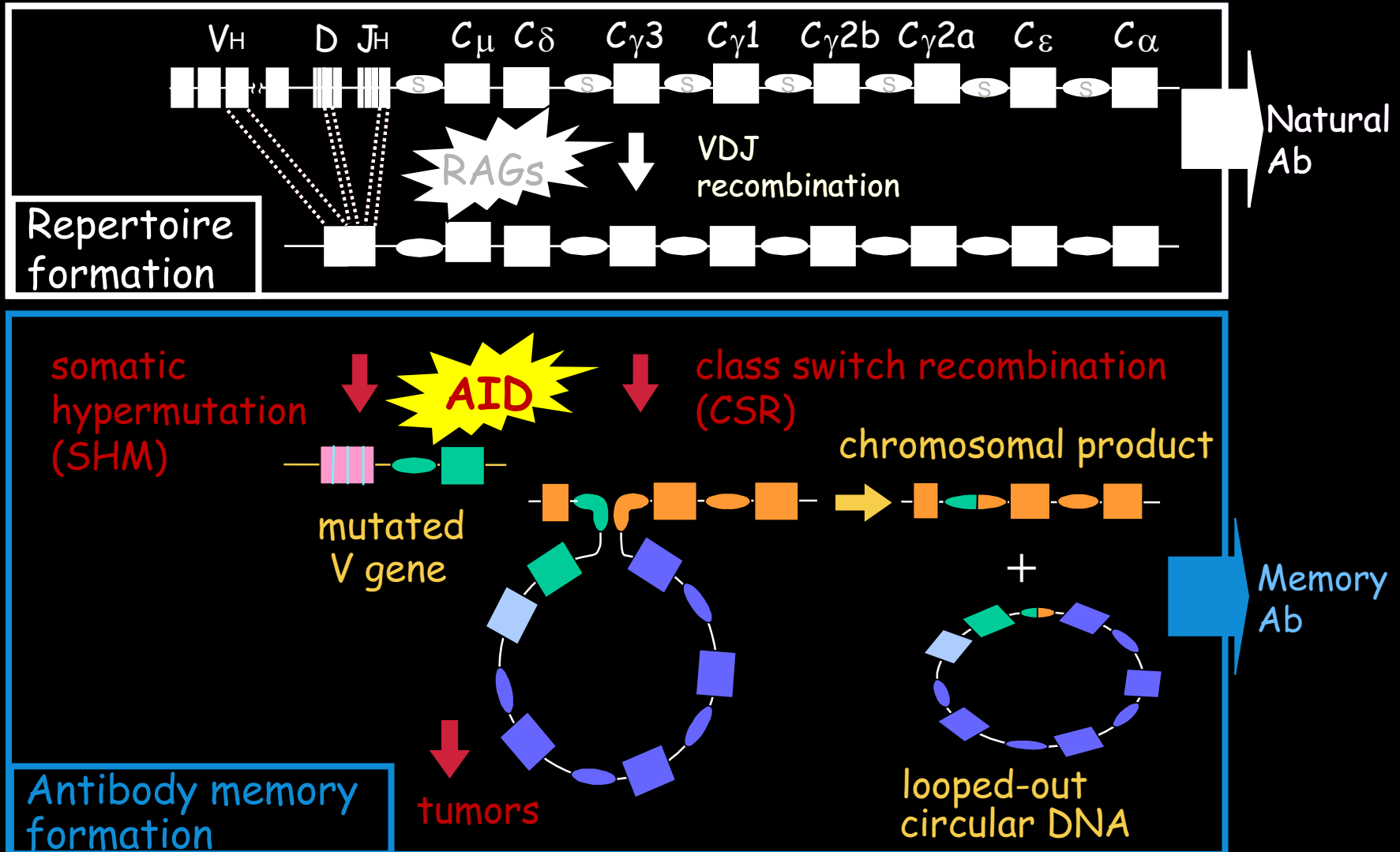
M. Muramatsu *et al.*, Cell (2000)

- AID deficiency in human is the cause of Hyper IgM Syndrome Type II: exactly the same phenotypes as mouse.

P. Revy et al., Cell (2000)

- Thus, AID is the enzyme that engraves antigen memory in the antibody gene, the mechanistic basis of vaccination.

AID engraves Ab memory in the genome for effective vaccination



Immune surveillance against cancer

Proposed by
Sir Frank Macfarlane Burnet (1970)

However, numerous attempts to develop immunotherapy were unsuccessful.

Cancer immunotherapy by boosting accelerators has not given convincing clinical outcomes

1. Cancer vaccine
2. *In vitro* activation of T lymphocytes
3. Cytokine treatment (IFN γ , IL-2, IL-12 etc)

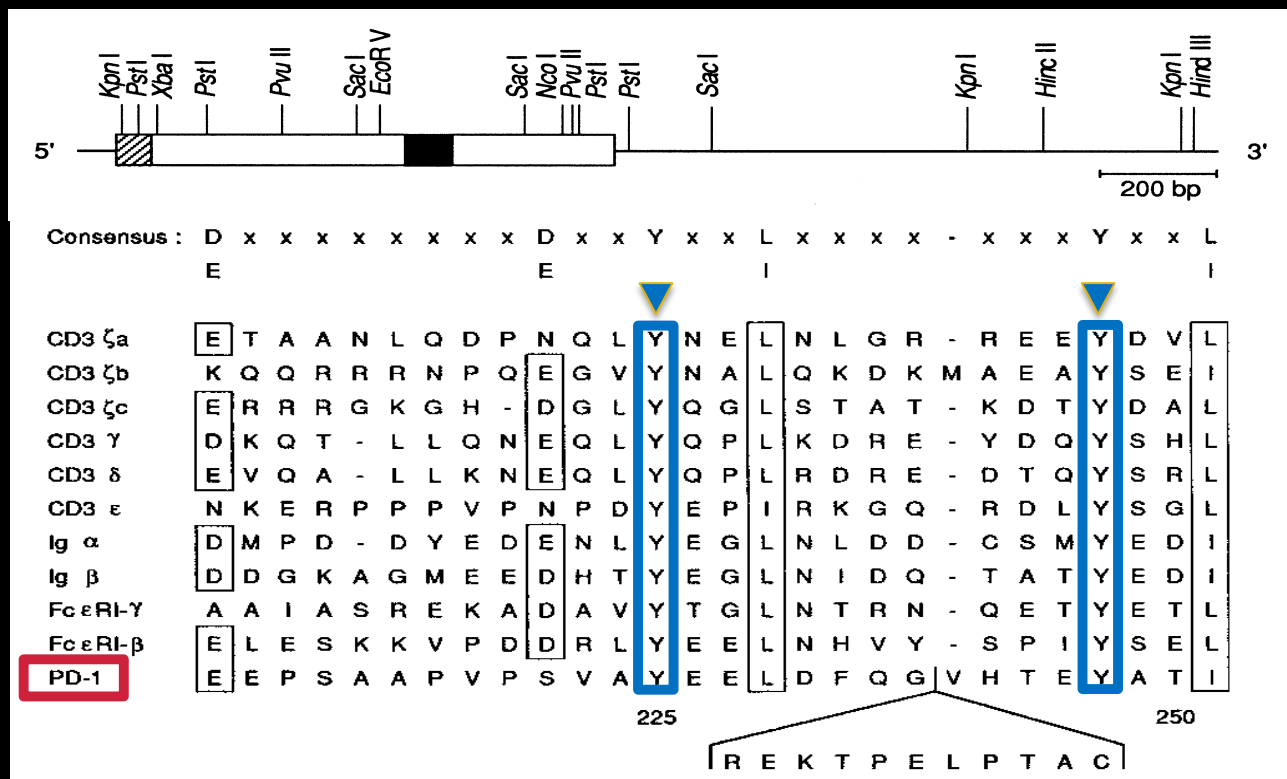
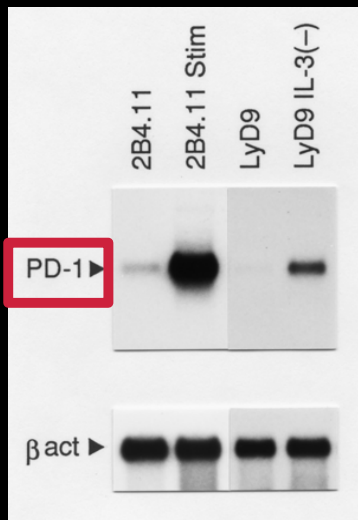
This was because no immune brake molecules were known before 1995

Brakes and accelerators control immune reactions like those in a car

action phase	drive	stop	action mode
parking [Activation]	ignition [CD28]	parking brake [CTLA4]	ON/OFF [Drastic]
driving [Attack]	accelerator [ICOS]	brake [PD-1]	~100k/h [Mild]

Discovery of PD-1 (programmed death-1) cDNA

Structure of cytoplasmic tail suggests PD-1 is a surface signaling molecule



Y. Ishida Y. Agata

Y. Ishida *et al.*, EMBO J. (1992)

A long journey to understanding the function of PD-1

- 1994 PD-1 knock out (KO) on mixed background mice, no phenotype change
- 1996 PD-1 KO on C57BL/6, no phenotype change for 6M

But over-response to antigen stimulation

- 1997 Nephritis and arthritis after 5M in PD-1 KO x lpr/lpr background
- 1998 Clear autoimmunity in PD-1 KO by 14M

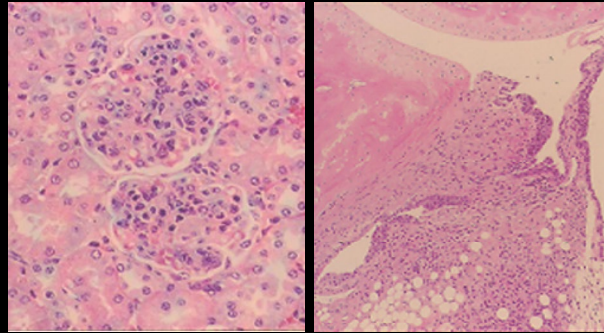
PD-1 is a negative regulator

C57BL/6

PD-1 KO (Knock Out)



Y. Nishimura
et al., Immunity (1999)



Nephritis Arthritis

BALB/c

WT

PD-1 KO



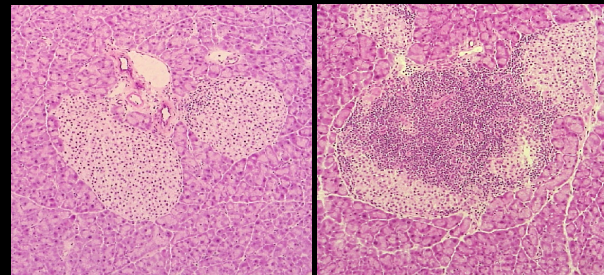
Dilated cardiomyopathy

NOD

NODxPD-1 KO



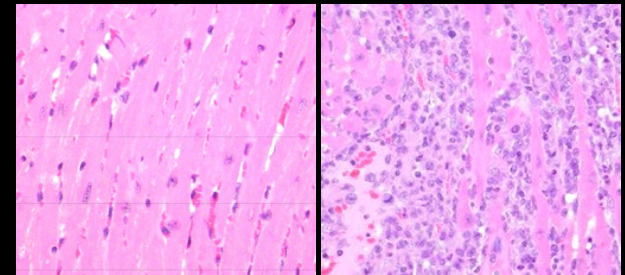
T. Okazaki *et al.*, Nat. Medicine (2003)
J. Wang *et al.*, Int. Immunol. (2010)



Diabetes

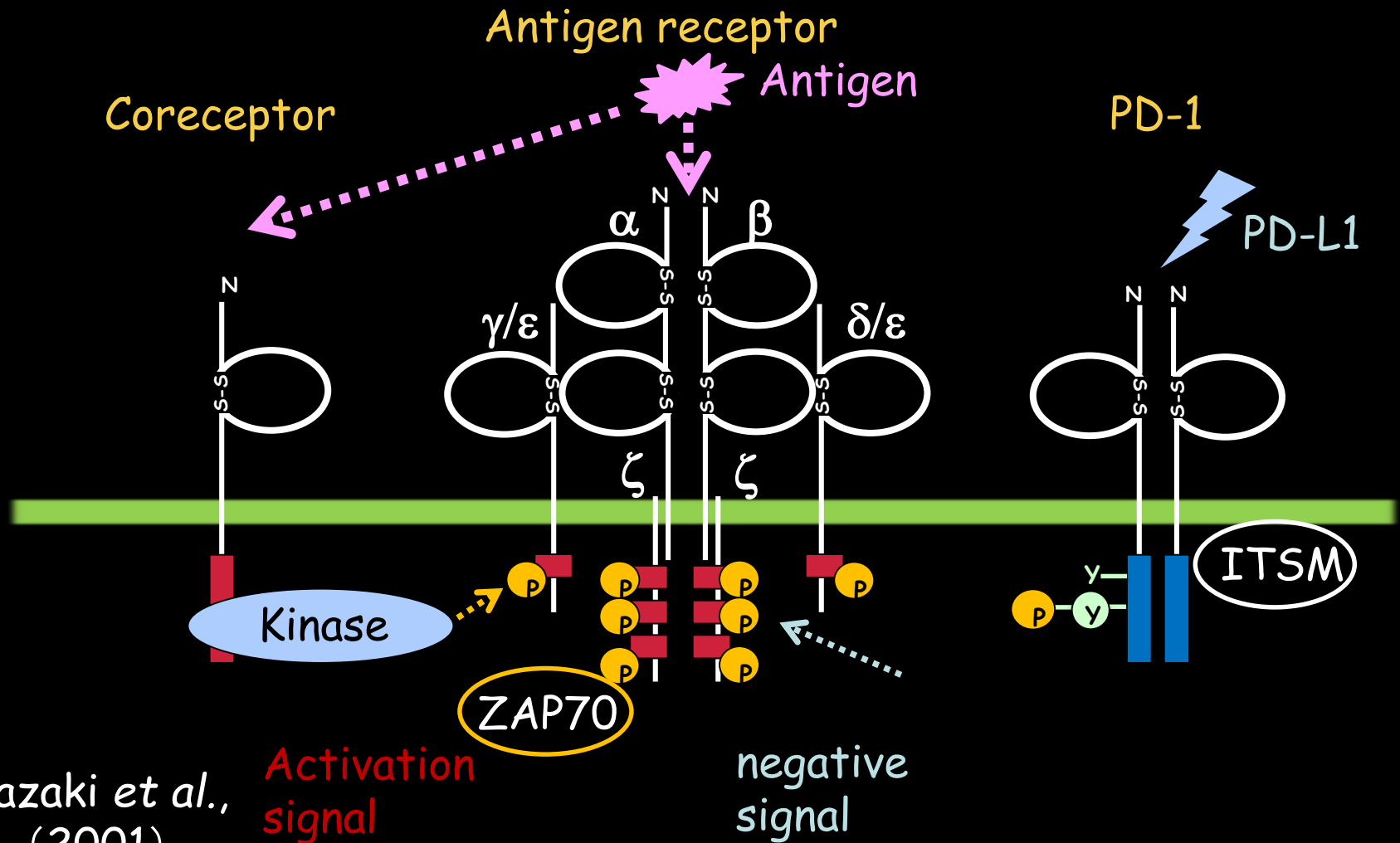
MRL

MRLxPD-1 KO

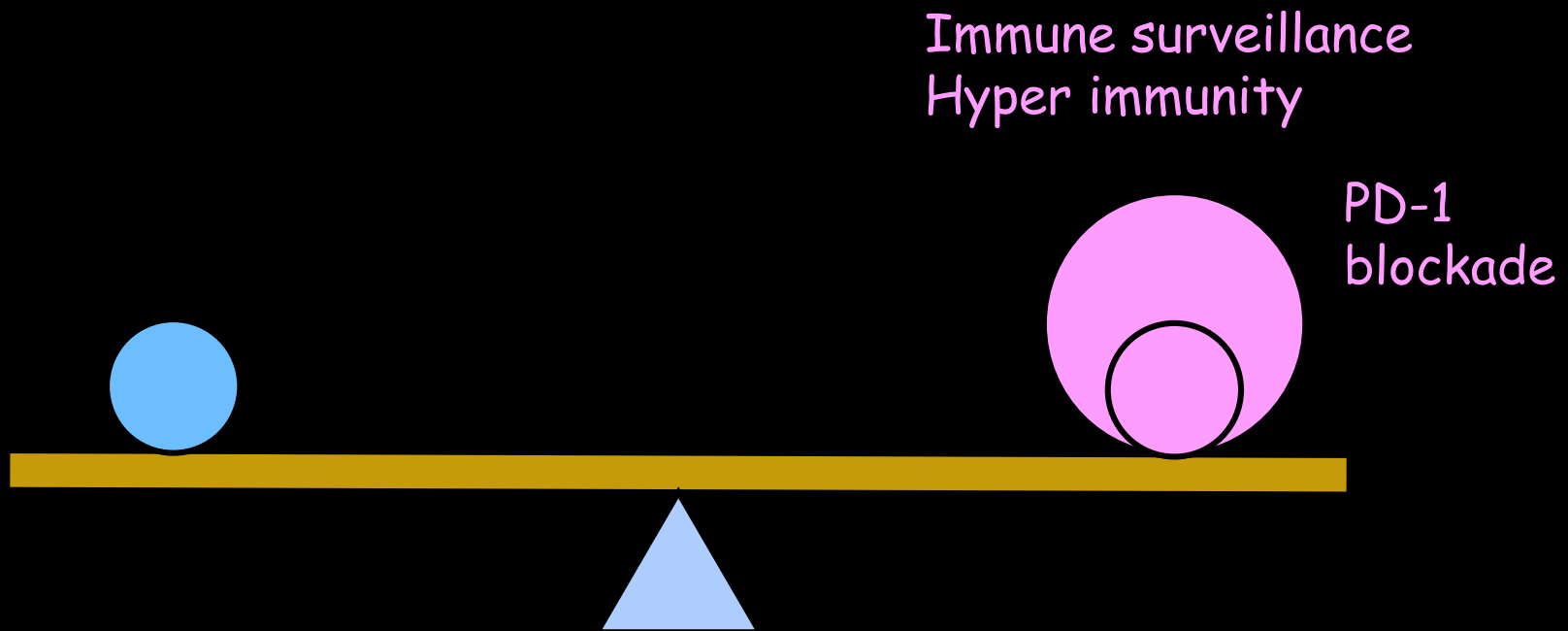


Myocarditis

Molecular mechanism of immune inhibition by PD-1 signaling



Balance between immune surveillance and immune tolerance

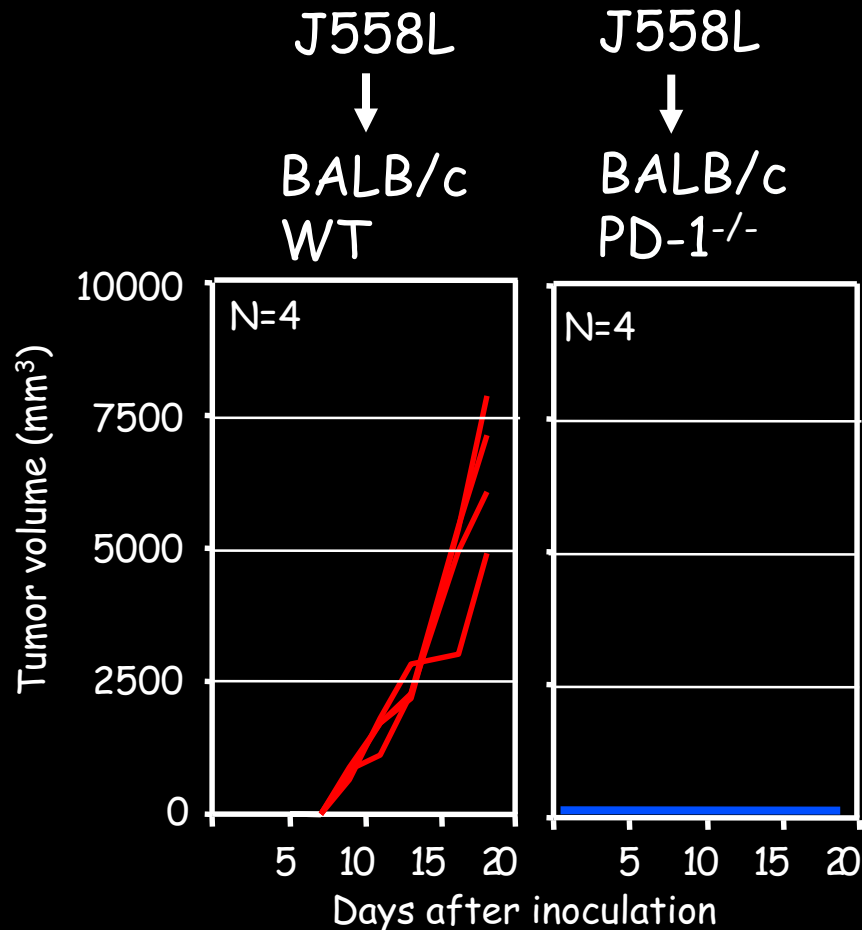


Immune surveillance
Hyper immunity

PD-1
blockade

Treatment of infectious
diseases and cancer
Risk of autoimmunity

Inhibition of tumorigenesis of myeloma (J558L) in PD-1^{-/-} mice



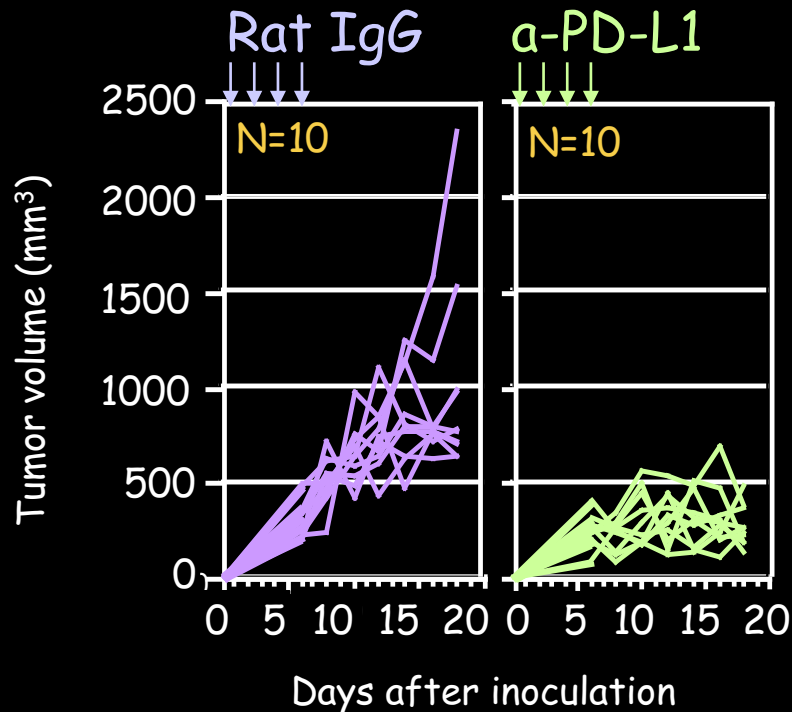
Inhibition of tumorigenesis of P815/PD-L1 by anti-PD-L1

P815/PD-L1 → DBA/2

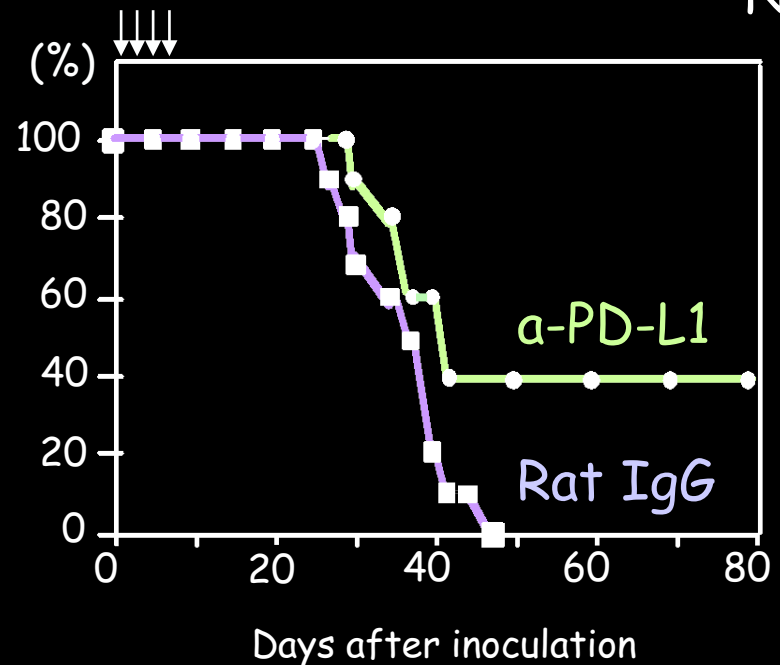


N. Minato

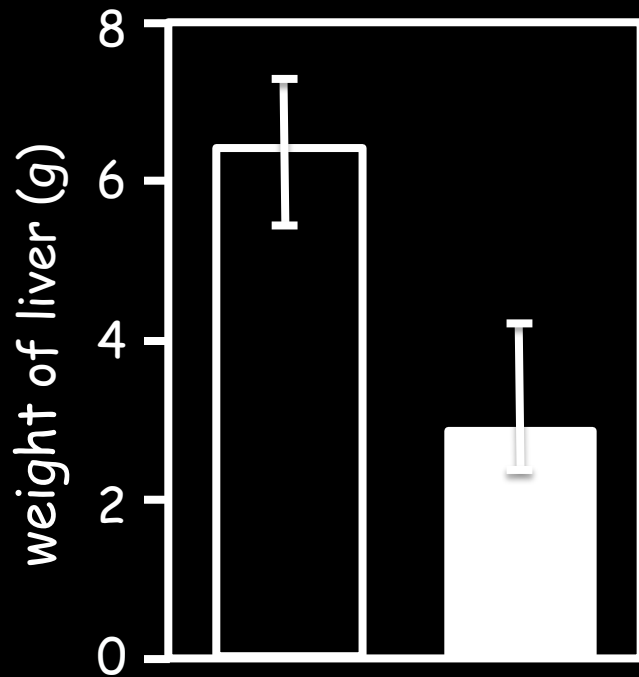
Tumor growth



Survival rate



PD-1 blockade inhibits metastasis of B16 melanoma (mouse model)



WT

Anti PD-1 Ab

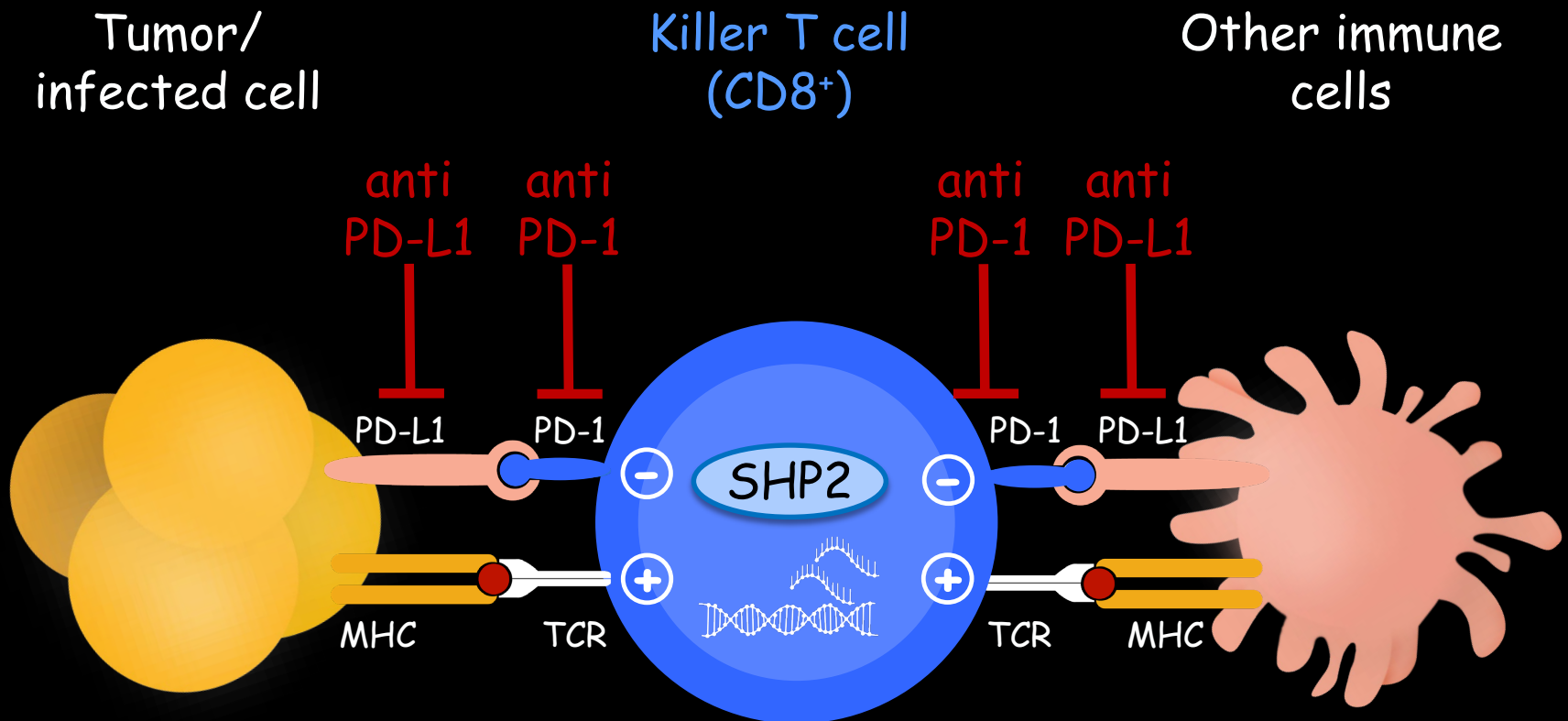
Spleen to liver



WT

Anti PD-1 Ab

PD-1 blockade by antibody against either PD-1 or PD-L1 can cure cancer



PD-L1 is expressed on various immune cells

Human anti-PD-1 antibody

Synthesized in mice containing human immunoglobulin gene by Medarex

Subclass: IgG4S228P

mutant IgG4 (S228P) stabilizes the protein and reduces ADCC

(antibody-dependent cell-mediated cytotoxicity)

KD = 2.6 nmol/L

Named Nivolumab

Approved as Investigation New Drug by FDA
(USA; Aug 1, 2006)

La Jolla Institute for Allergy & Immunology
Building Opening Symposium and 5th Ishizaka Lecture
September 14, 2006



Gary Koretzky, Tasuku Honjo, Anjana Rao, Rafi Ahmed, Jim Allison, Ralph Steinman

Clinical trials began in US (2006) and Japan (2008)

Summary of Phase I clinical trial

296 terminal stage patients recruited
Nivolumab treatment for two years

Complete or partial response rates

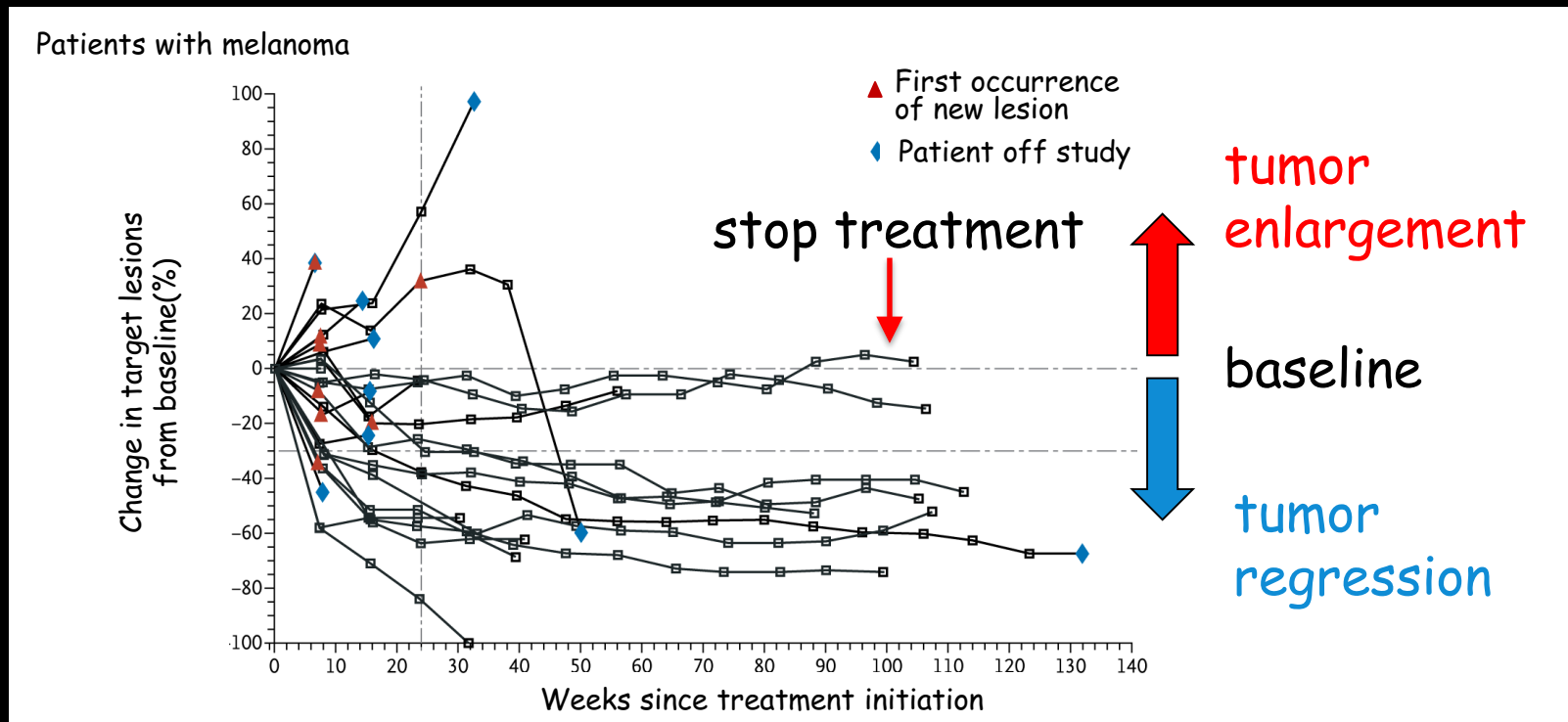
18% (76 patients) of non small cell lung cancer

28% (94 patients) of melanoma

27% (33 patients) of renal cell carcinoma

Durable response to PD-1 blockade

"Responses were durable; 20 of 31 responses lasted 1 year or more and some even after stopping therapy"



Phase II trial of anti-PD-1 antibody in patients with platinum-resistant ovarian cancer

Dose	total (n)	CR	PR	SD	PD	NE	RR	DCR
1 mg/kg	10	0	1	4	4	1	1/10 (10%)	5/10 (50%)
3 mg/kg	10	2	0	2	6	0	2/10 (20%)	4/10 (40%)
Total	20	2	1	6	10	1	3/20 (15%)	9/20 (45%)



Oct 21, 2011-Dec 7, 2014

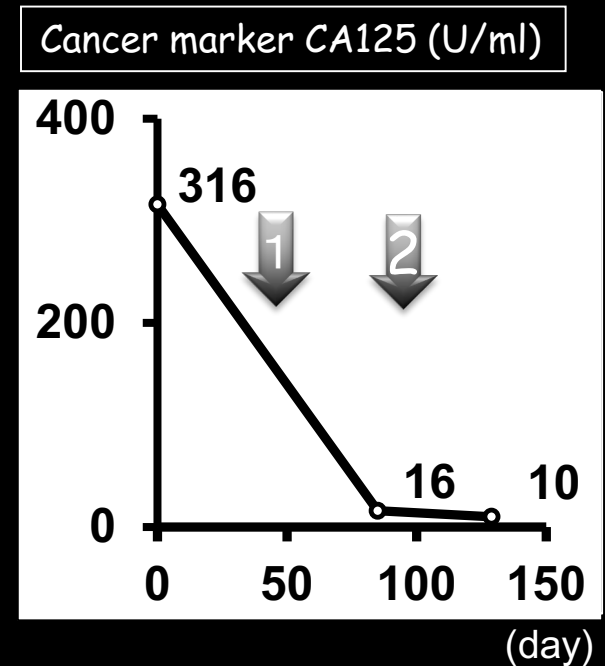
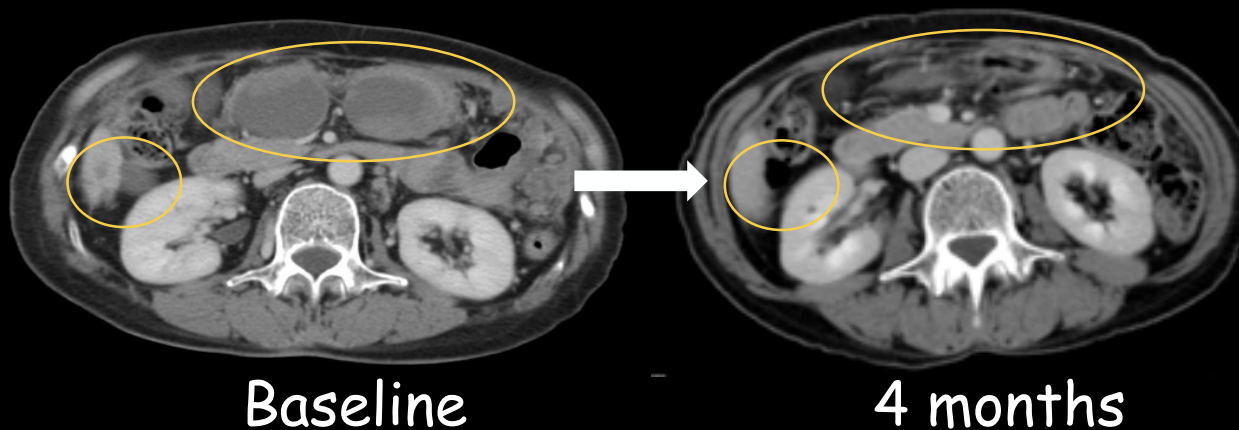
Tumor growth stopped in 40-50%
of terminal stage patients

J. Hamanishi *et al.*, J. Clin. Oncol. (2015)

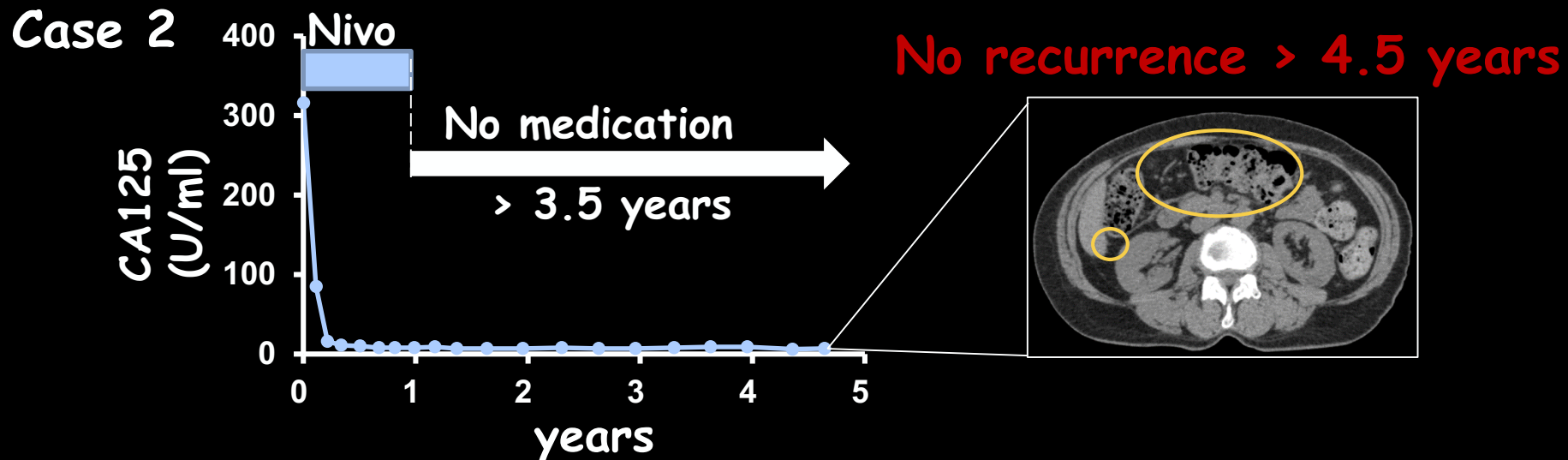
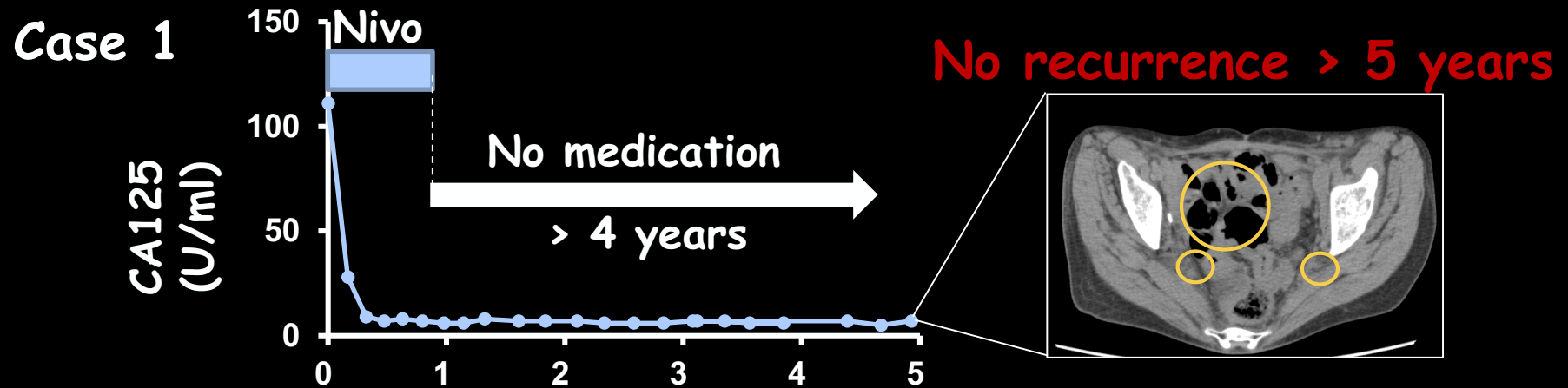
A responder with ovarian cancer (clear cell): Nivolumab 3mg/kg

History: 60 yr. Stage Ic with progressive disease after RSO, MMC/CPT11*3, SCH+BSO, CPT/CDDP*5, TC*2

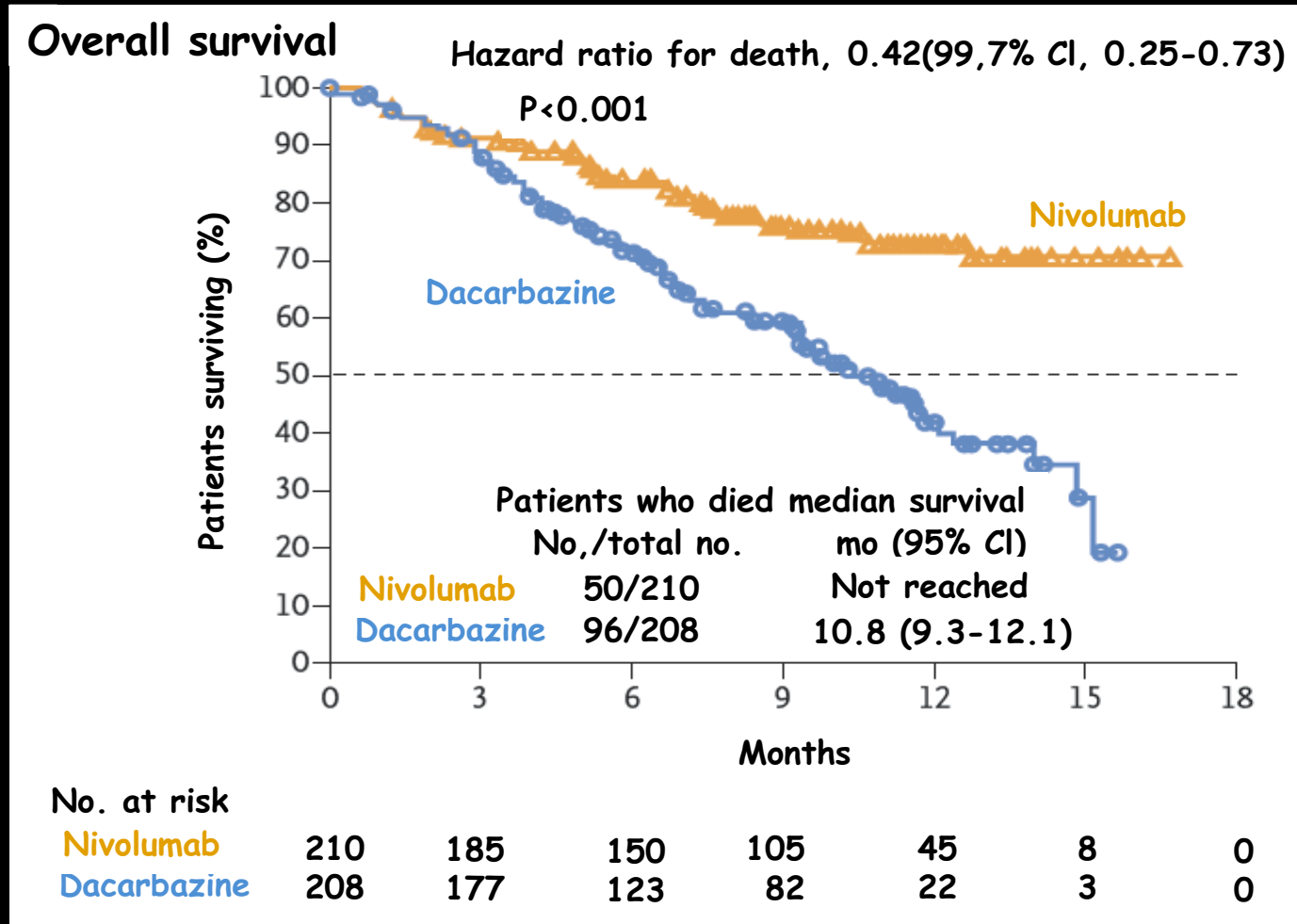
Peritoneal dissemination $\Rightarrow$ disappeared



Durable complete responses of ovarian cancer patients to Nivolumab



Randomized Study on Untreated Melanoma Patients with Nivolumab and Dacarbazine (chemotherapy)



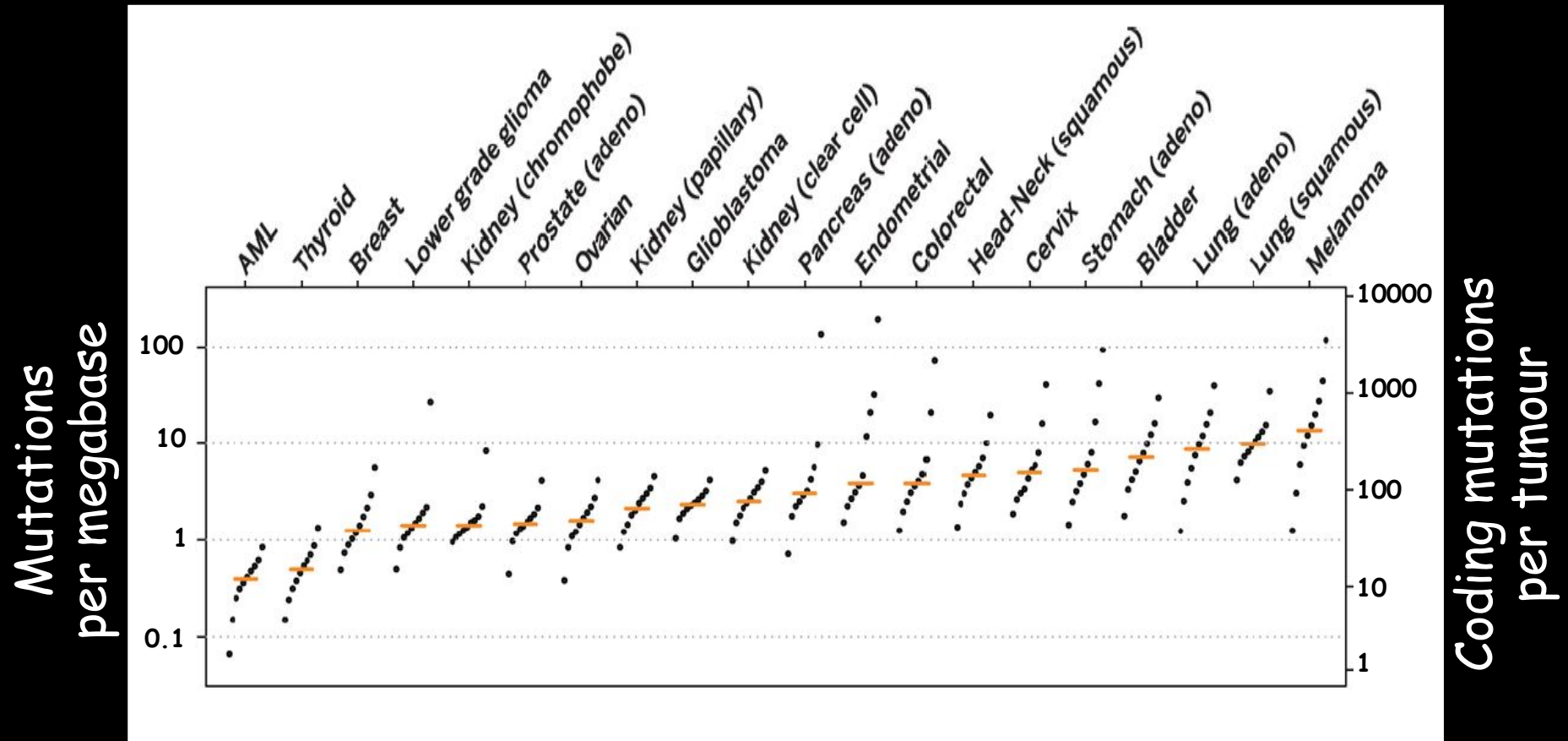
Cancers approved for PD-1 blockade therapy

- 2014 melanoma
- 2015 lung cancer
- 2016 renal cancer
Hodgkin's lymphoma
head and neck cancers
urothelial cancer
- 2017 colorectal cancer
gastric cancer
hepatocellular carcinoma
Merkel cancer
all highly mutated cancers
- 2018 cervical cancer
primary mediastinal large B-Cell lymphoma

Paradigm shift of cancer therapy by anti-PD-1 treatment

1. Less adverse effects because normal cells are unaffected
2. Effective for a wide range of tumors (more than 1000 clinical trials)
3. Durable effects to responders after stopping treatment

Cancer cells accumulate mutations



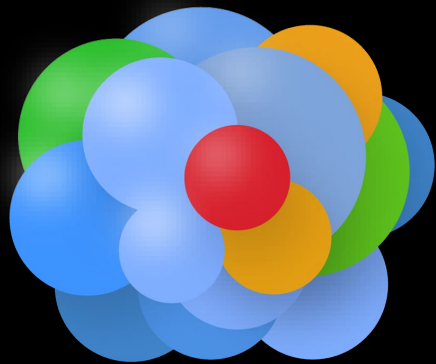
I. Martincorena *et al.*, Science (2015)

What we learned from huge cancer genome projects

1. Cancer cells accumulate a large number of mutations to express neo-antigens that can be recognized by the immune system as non-self. This is why cancer immunotherapy is effective.
2. Too many mutations to pinpoint the dominant mutations for targeted chemotherapy.

Continuous mutations generate resistant tumor cells

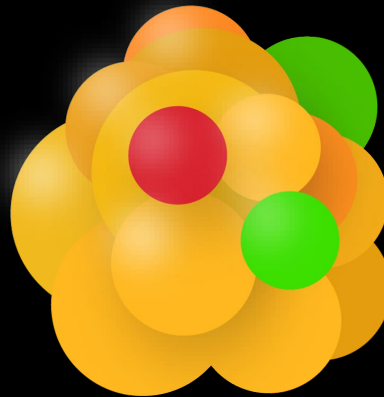
cancer
cells



drug A



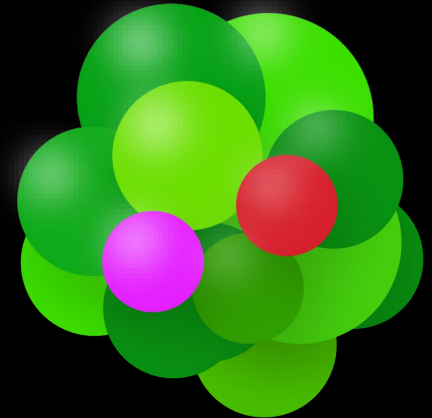
Selection
mutagenesis
Resistant cells grow



drug B



Selection
mutagenesis
Resistant cells grow



Lymphocytes can recognize many more mutants
& attack them

Current issues in PD-1 blockade therapy

Biomarkers for responders

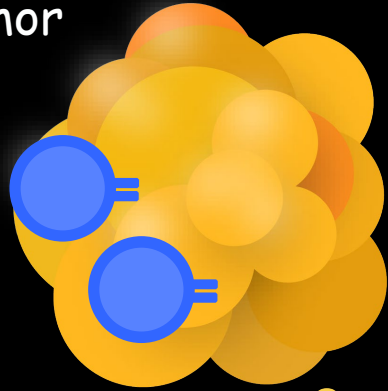
- High mutagenesis in tumors
- Potency of individual's immunity

Improvement of immunotherapy

- Accessibility of killer T cells to tumor sites
- Potentiation of killer T cell function

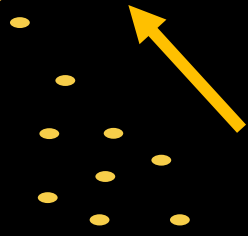
PD-1 blockade initiates killer T cell expansion in lymph nodes

Tumor

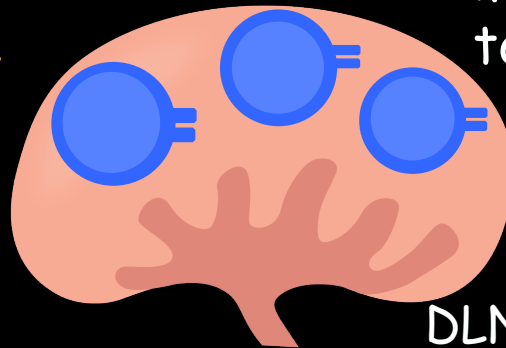


1. PD-1 blockade enhances killing within tumor which secretes chemokines

Chemokines attract killer T cell



2. PD-1 blockade enhances priming and induces chemokine receptor to help migration of new killer T cell towards tumor



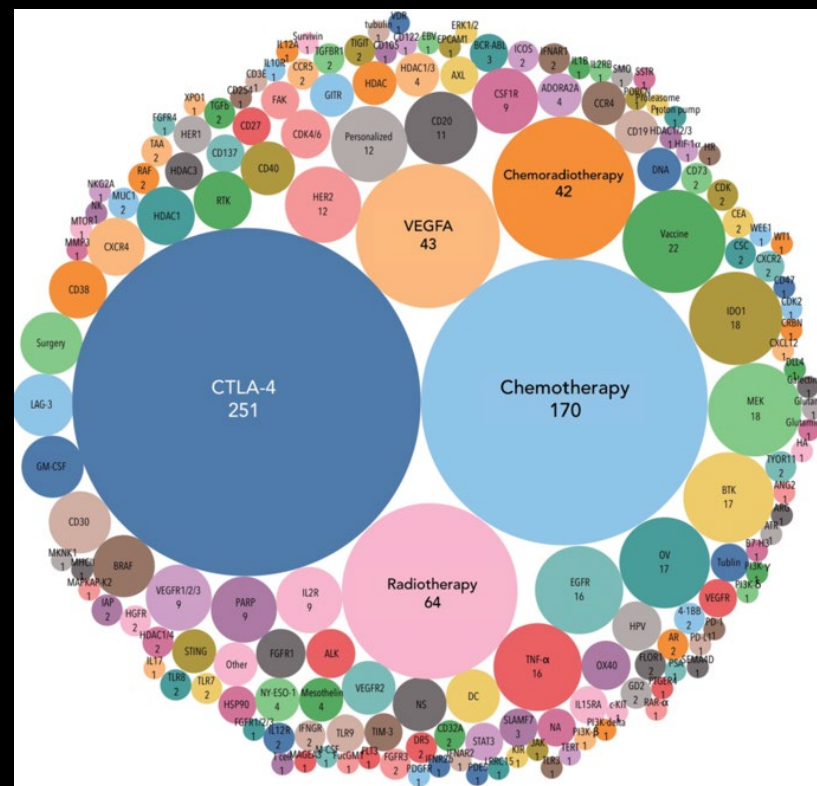
DLN
(draining lymph node)



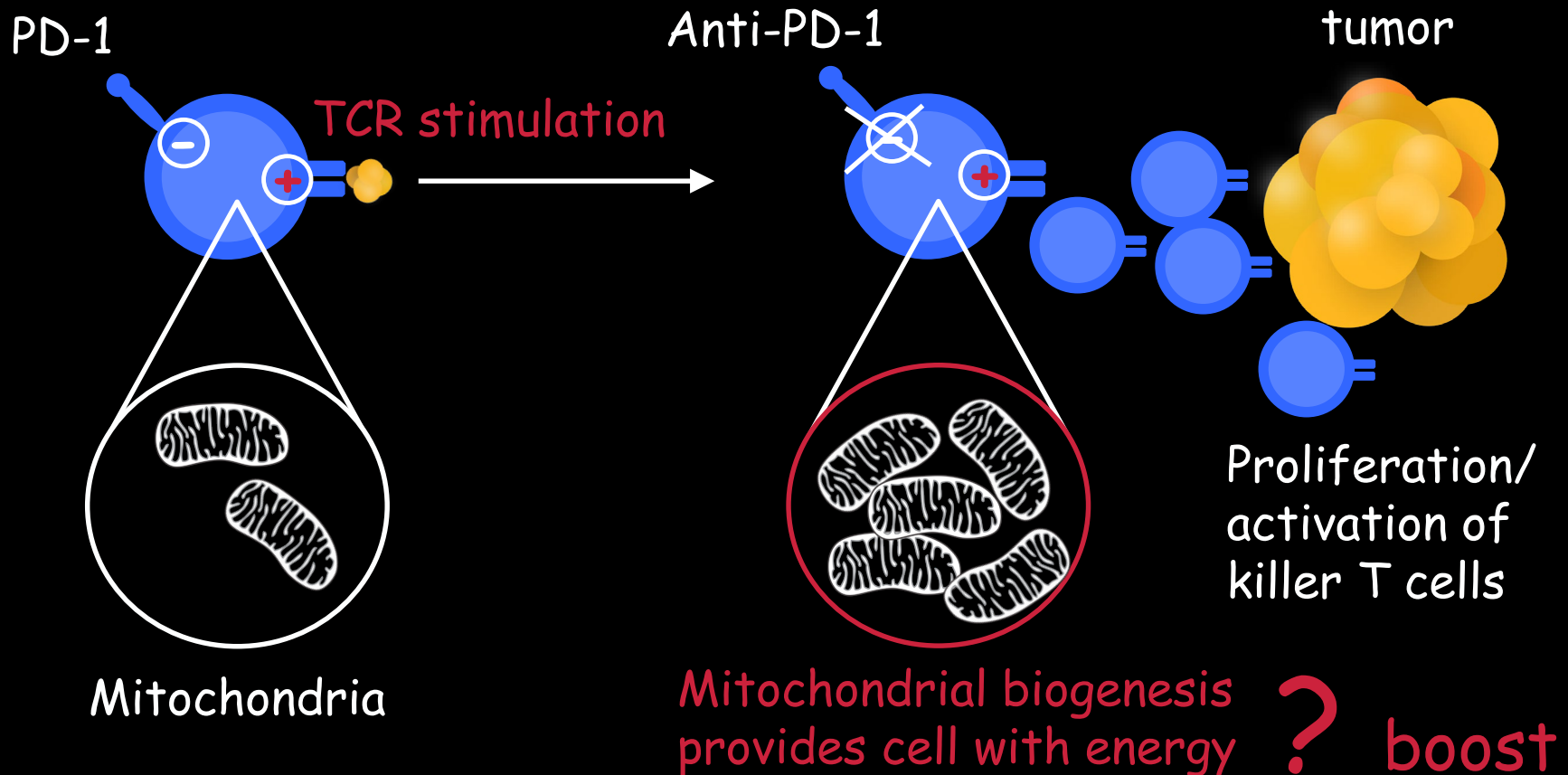
Cancer immunotherapy by PD-1-based combination studies underway in 2017

Numbers of PD-1 blockade trials using combinations with :

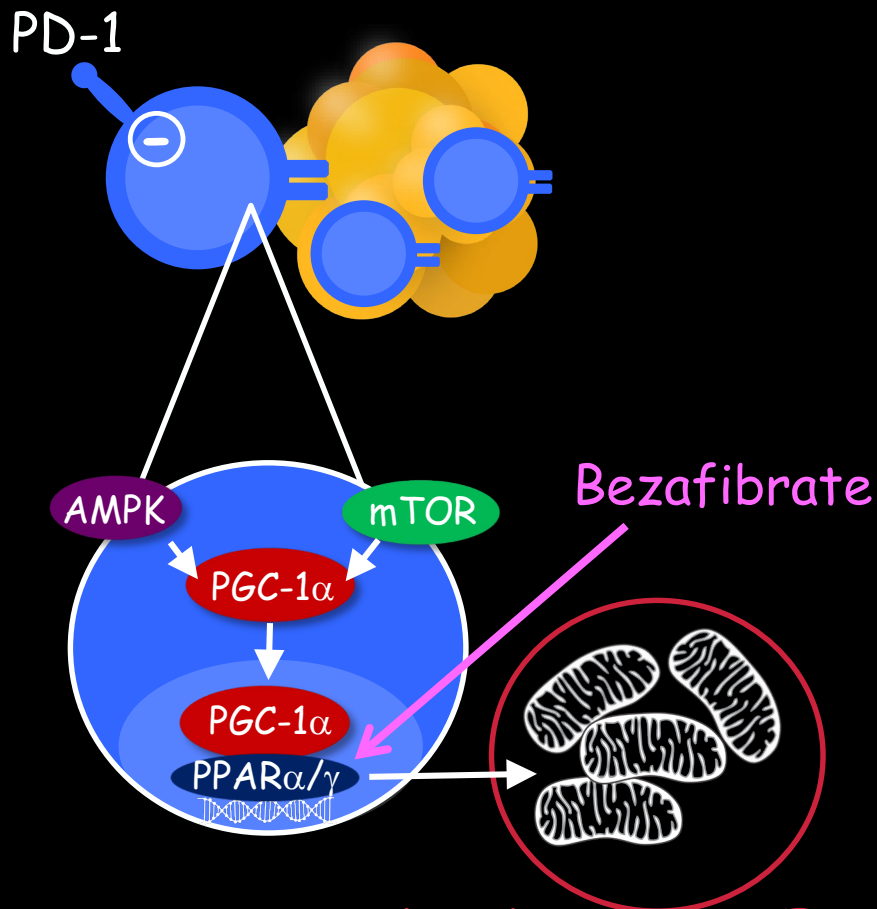
1. Anti-CTLA-4 agents: 251
2. Chemotherapies: 170
3. Radiotherapies: 64
4. Anti-VEGFA agents: 43
5. Chemoradiotherapy combos: 42



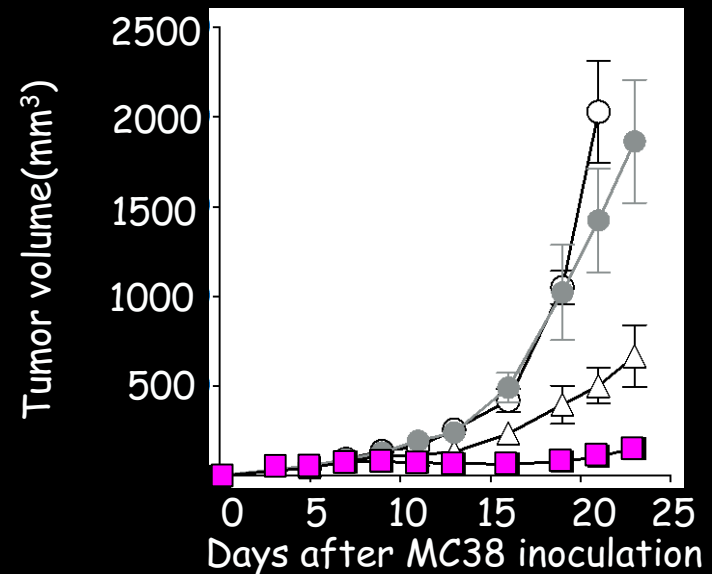
Requirement of mitochondrial activation for killer T cell activation and proliferation



Activation of PGC-1 α /PPAR complex improves the efficacy of PD-1 blockade



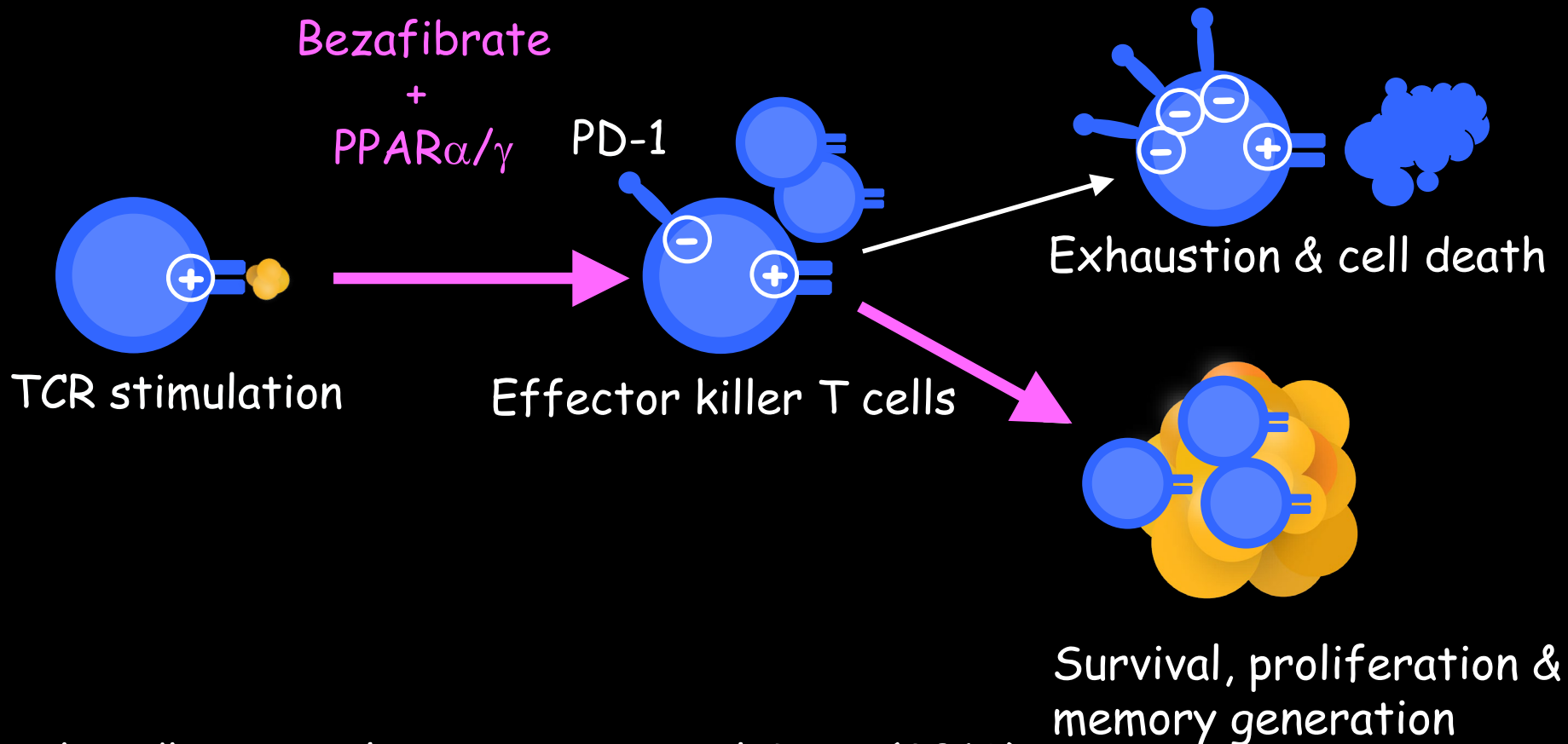
- Control
- ▲ Anti-PD-L1 mAb
- Anti-PD-L1 mAb + Bezafibrate
- Bezafibrate



Mitochondria mass + Energy boost

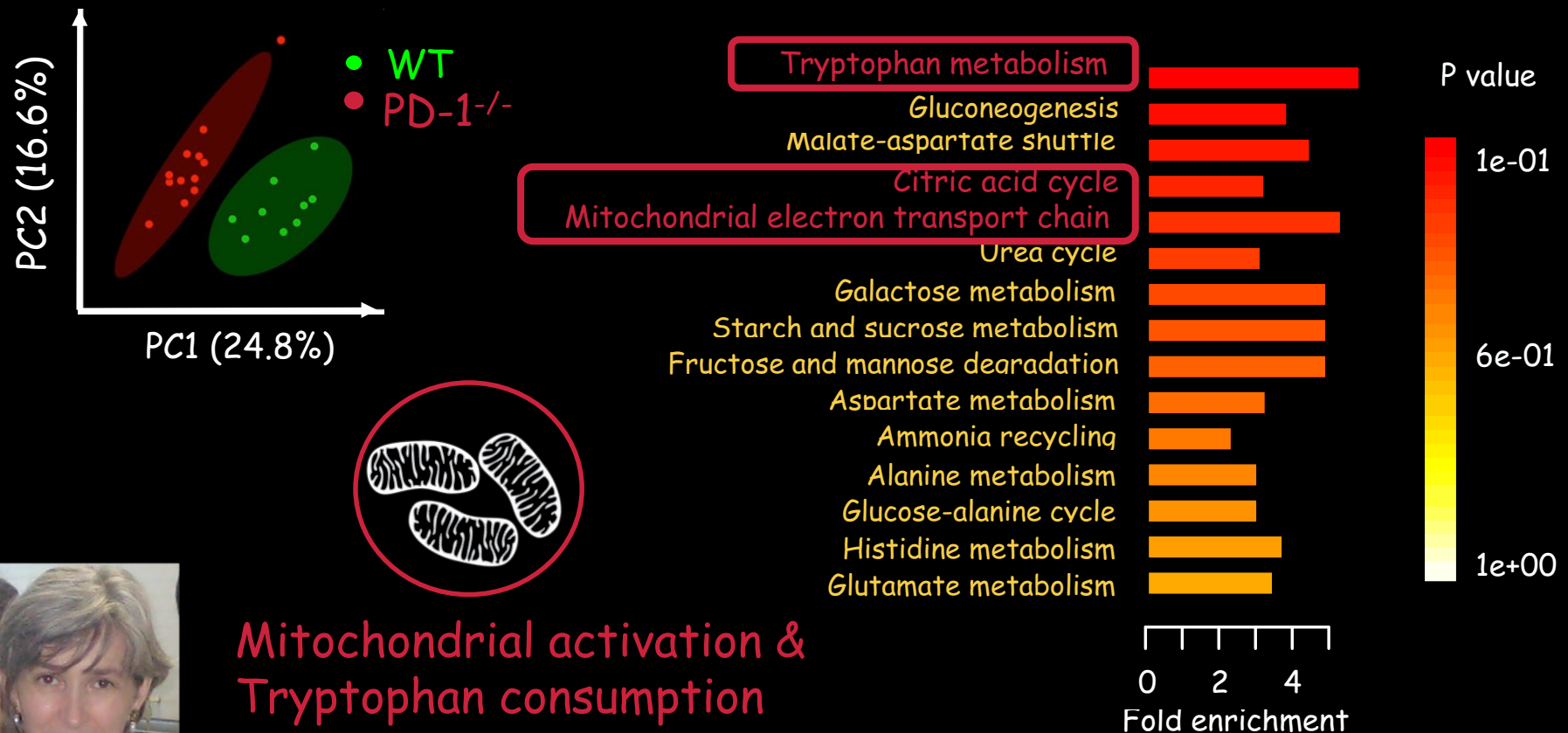
K. Chamoto et al., PNAS (2017)

Bezafibrate increases killer T cell proliferation and blocks cell death



P. Chowdhury *et al.*, Cancer Immunol. Res., (2018)

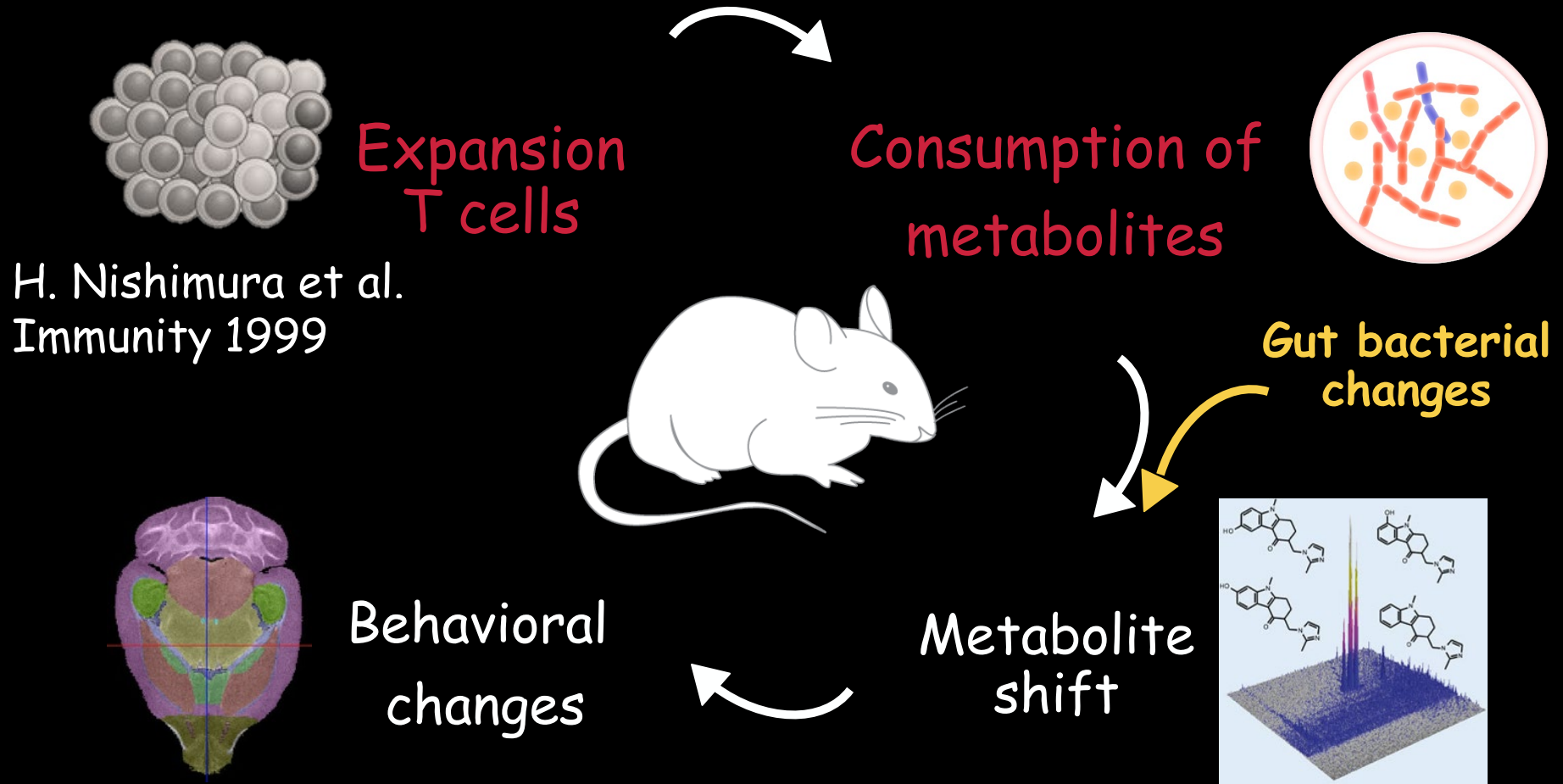
Hyperimmune activity can be read in blood biochemistry of PD-1^{-/-} mice



Sidonia Fagarasan

M. Miyajima, B. Zhang *et al.*, Nat. Immunol. (2017)

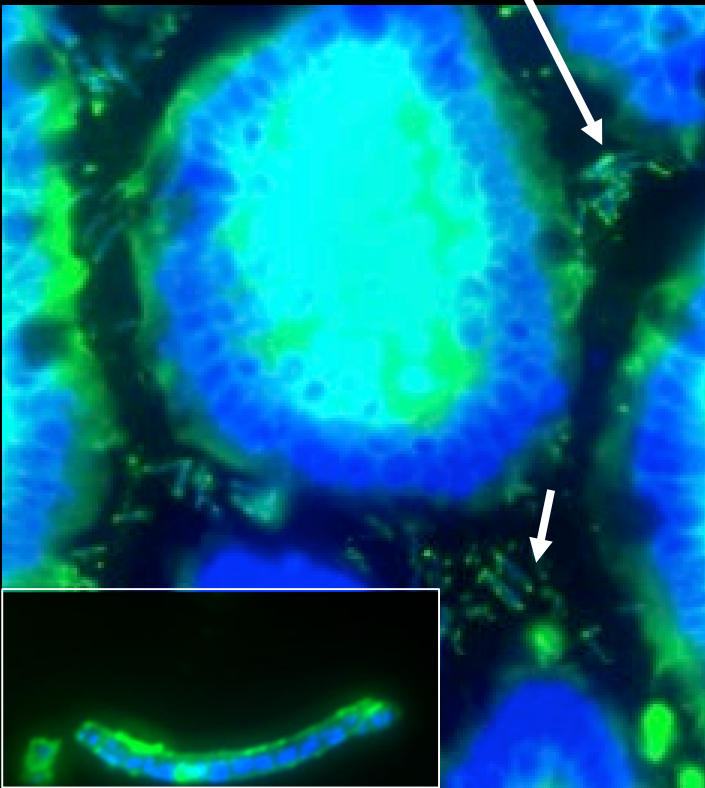
PD-1^{-/-} mice biology is very complex



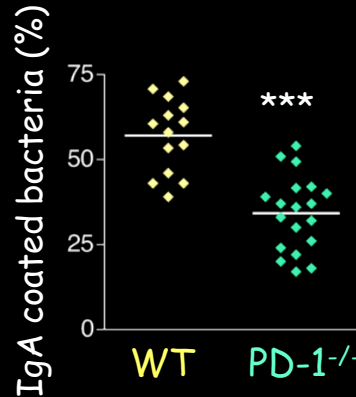
M. Miyajima, B. Zhang *et al.*, *Nat. Immunol.* (2017)

PD-1 selects IgA critical to microbiota regulation

IgA-coated bacteria in the gut



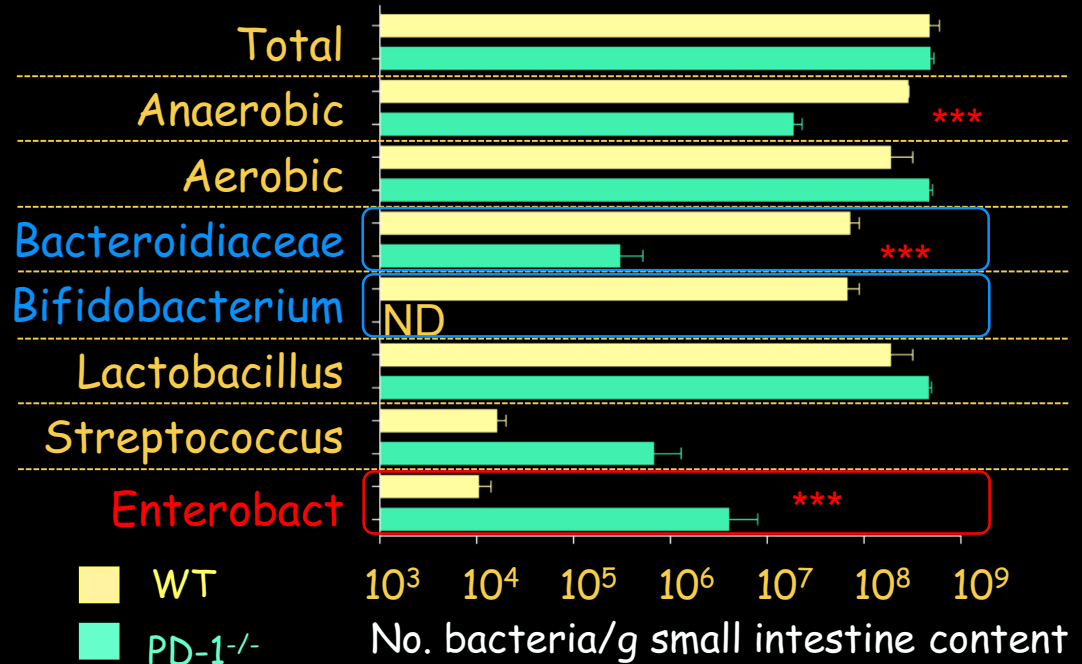
IgA DAPI



Less IgA-coating of bacteria in PD-1^{-/-} mice

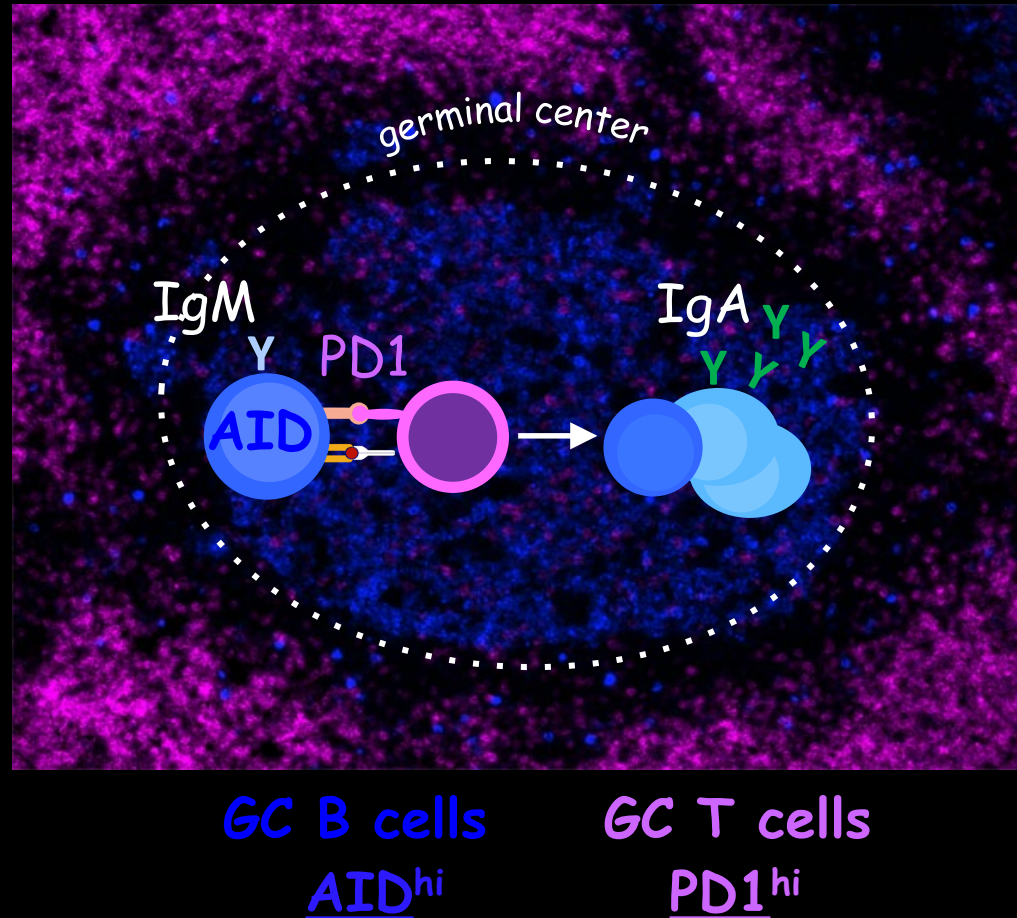


Bacterial dysbiosis

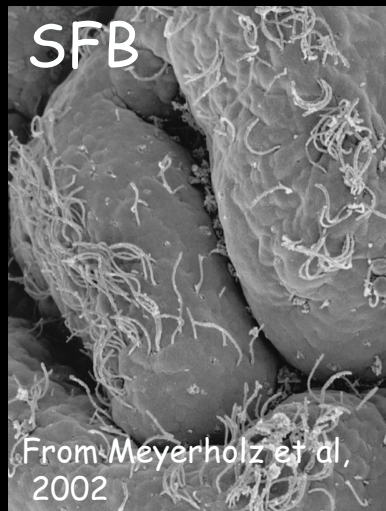
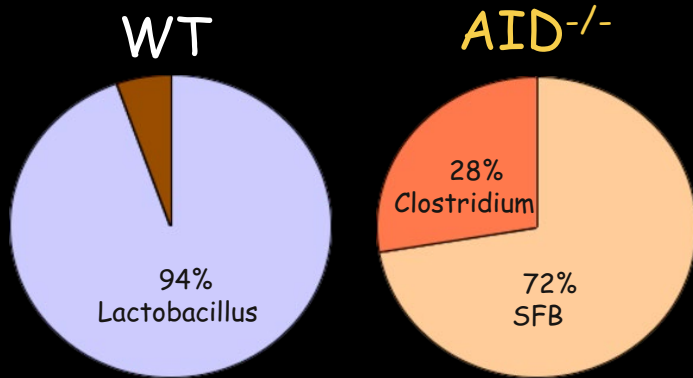


No. bacteria/g small intestine content

AID and PD-1 cooperate in germinal centers for high affinity IgA selection to maintain microbiome

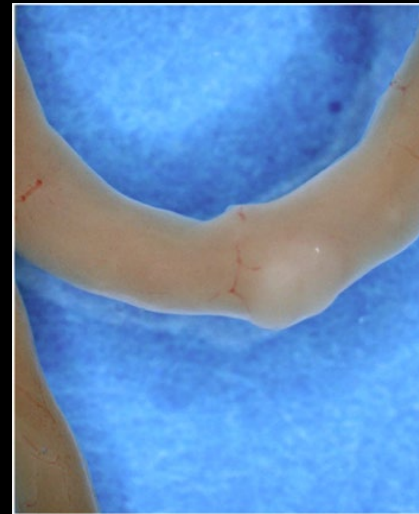


Critical role of AID for controlling microbiota & whole body immune homeostasis

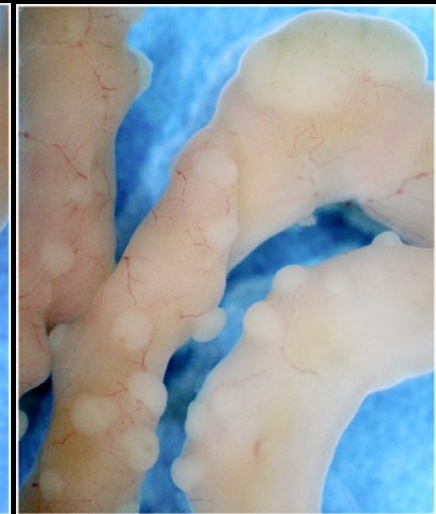


Mucosal immune activation

WT



AID^{-/-}



Systemic immune activation, spleen

WT



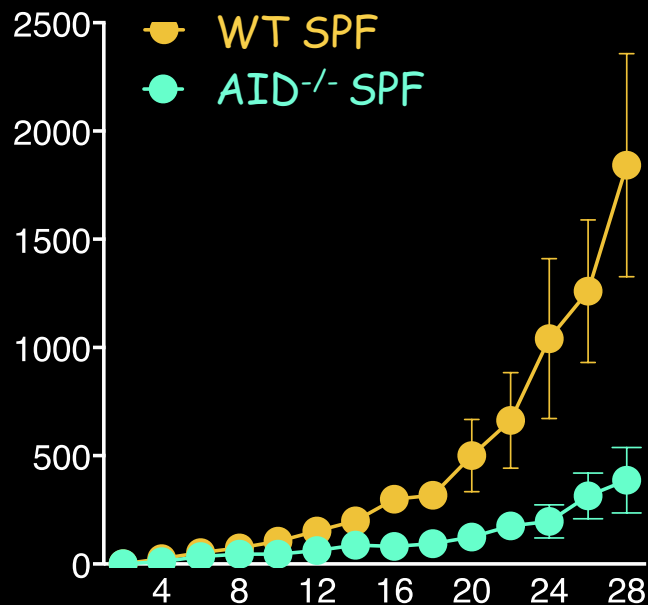
AID^{-/-}



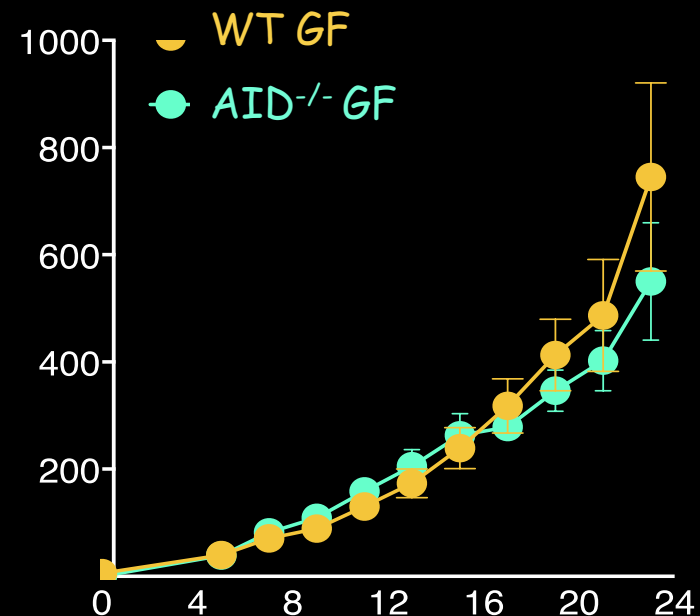
S. Fagarasan et al., Science (2002)
K. Suzuki et al., PNAS (2004)

Enhanced anti-tumor immunity in $AID^{-/-}$ mice depends on microbiota

Specific-pathogen free

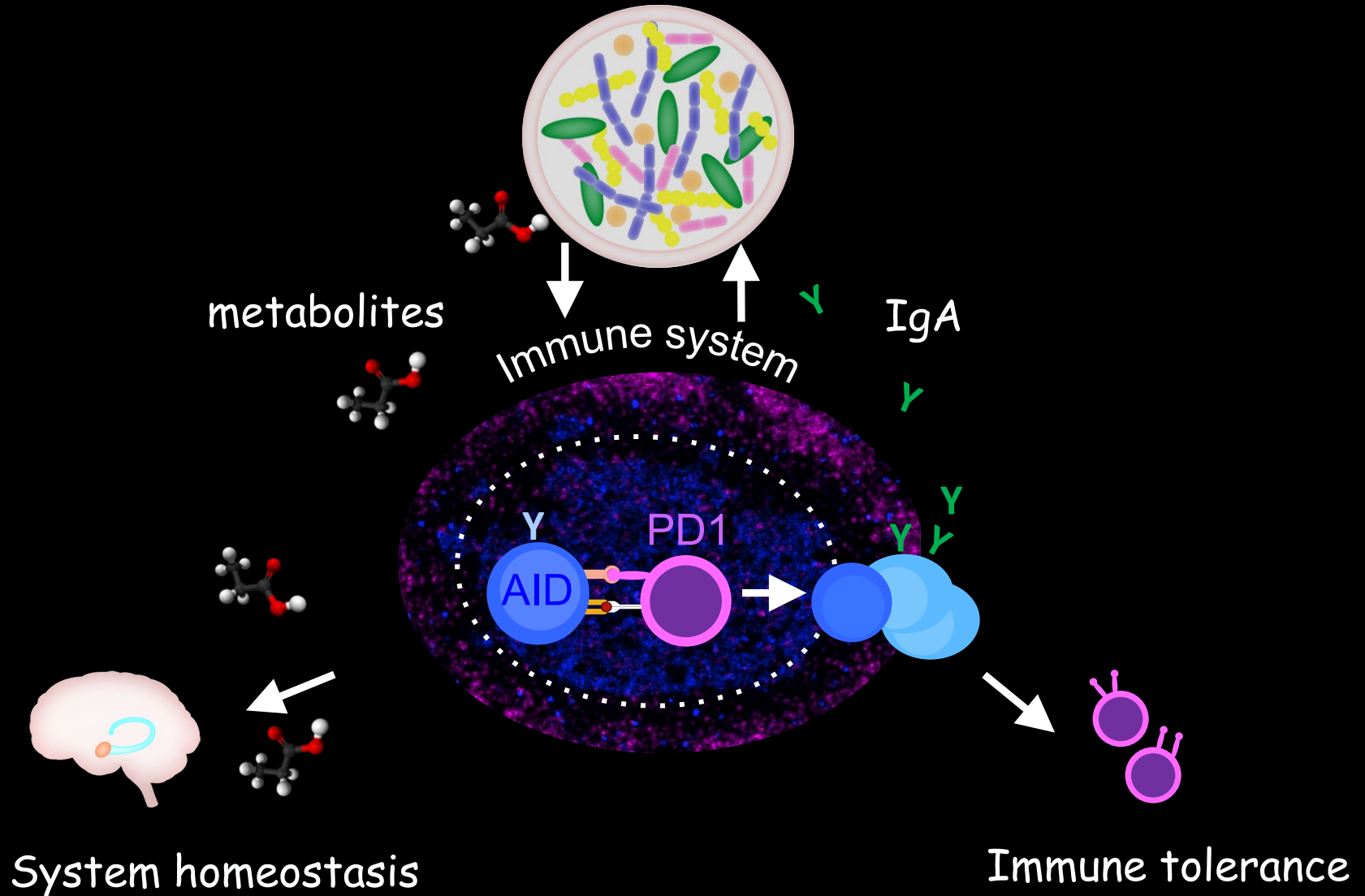


Germ free

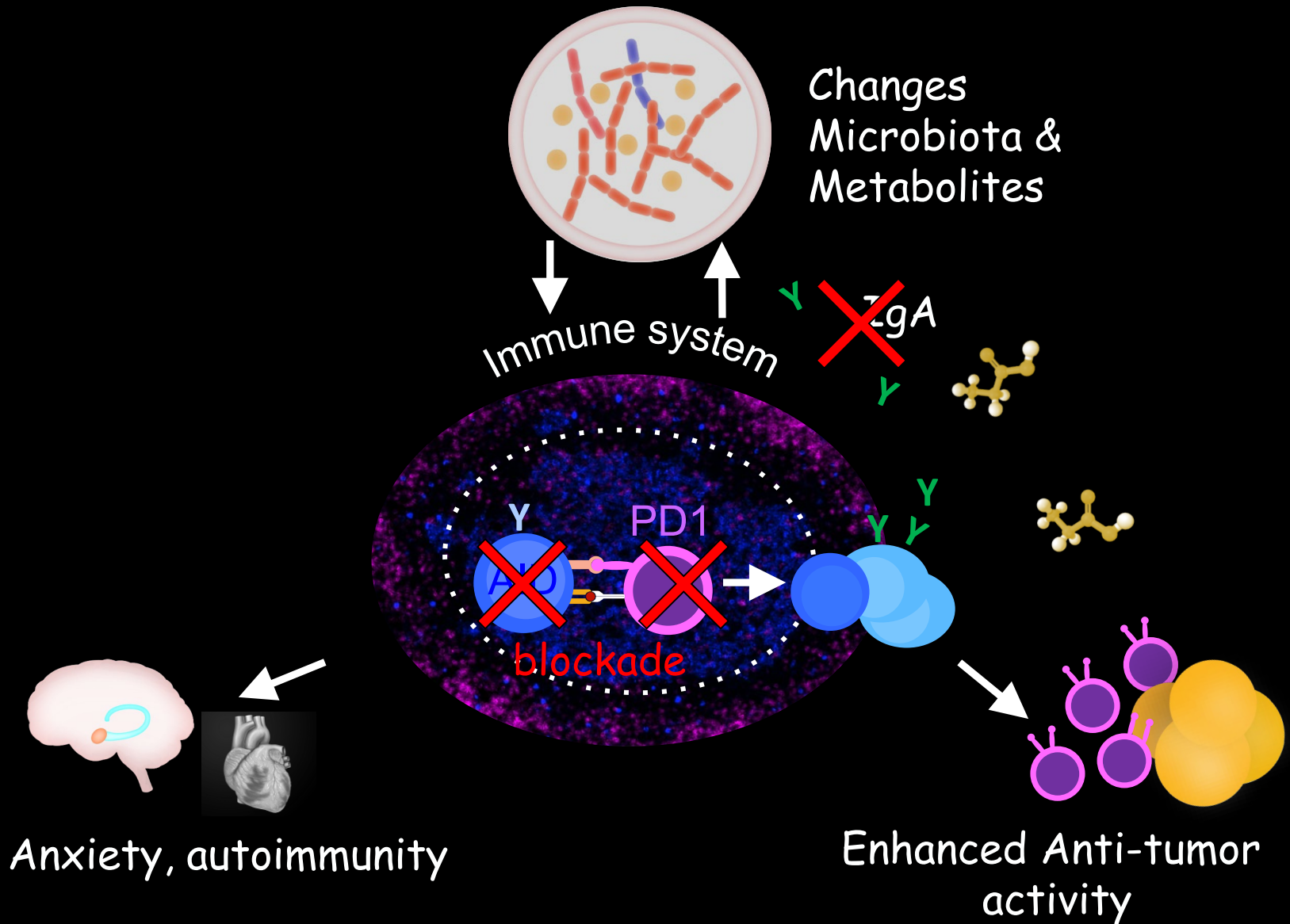


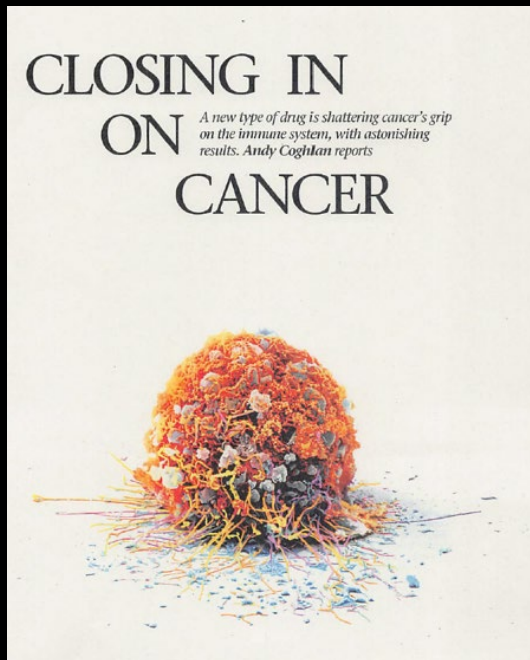
M. Akrami, R. Menzies, M. Miyajima, Y. Nakajima. unpublished data

Microbiome-immune system regulation



Microbiome-immune system regulation





CLOSING IN ON CANCER

Andy Coghlan
New Scientist, 5 March 2016

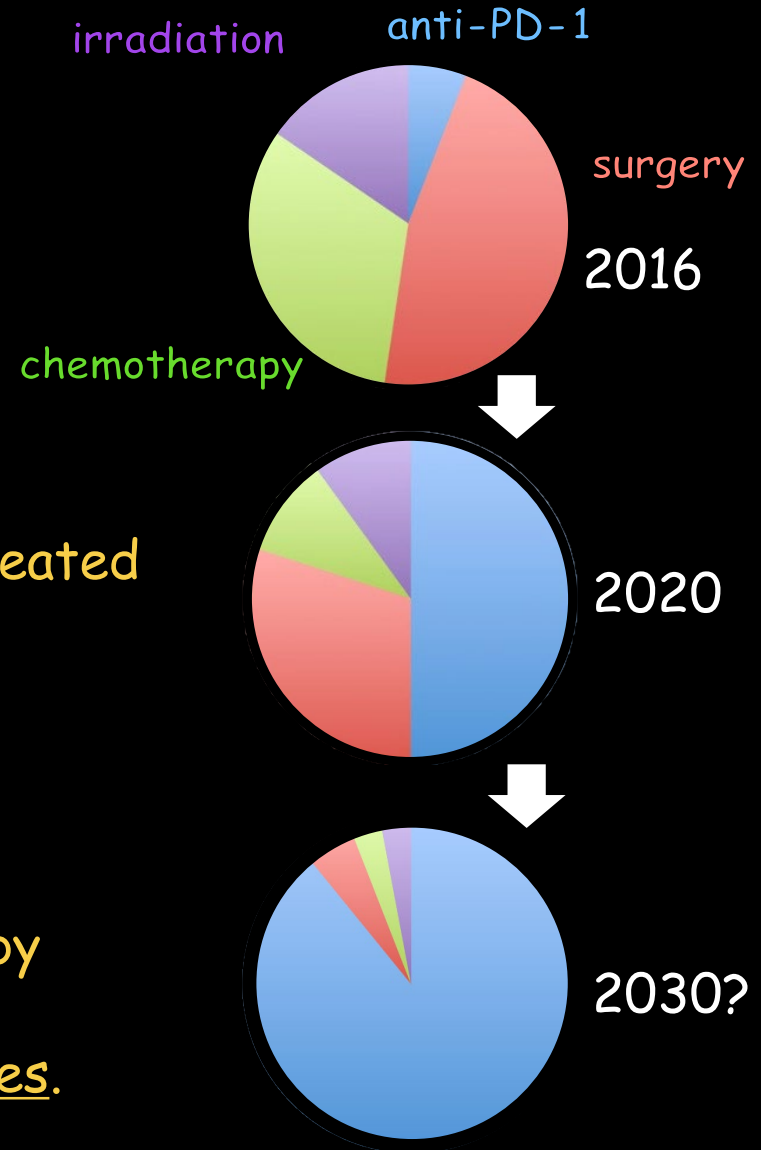
"We're at the point where we've discovered the **cancer equivalent of penicillin**" says Chen. Although penicillin itself couldn't cure all infections, it gave rise to a whole generation of antibiotics that changed medicine forever, consigning most previously fatal infections to history.

Future prospects in cancer therapy

1. Efficacy of PD-1 blockade therapy improved.

2. Many more cancers may be treated by immunotherapy.

3. Cancer may not completely disappear, but be controlled by immunotherapy. Cancer may become one of chronic diseases.

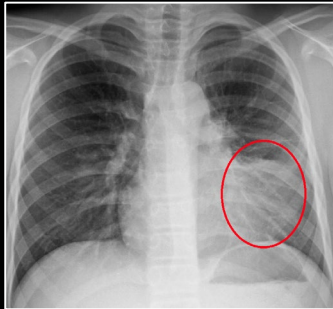


Enormous benefit by acquired immunity

20th century

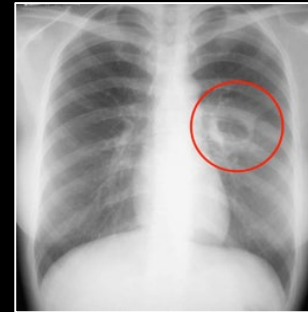
Eradication of infectious diseases
by vaccination and antibiotics

Pneumonia



Penicillin

Tuberculosis

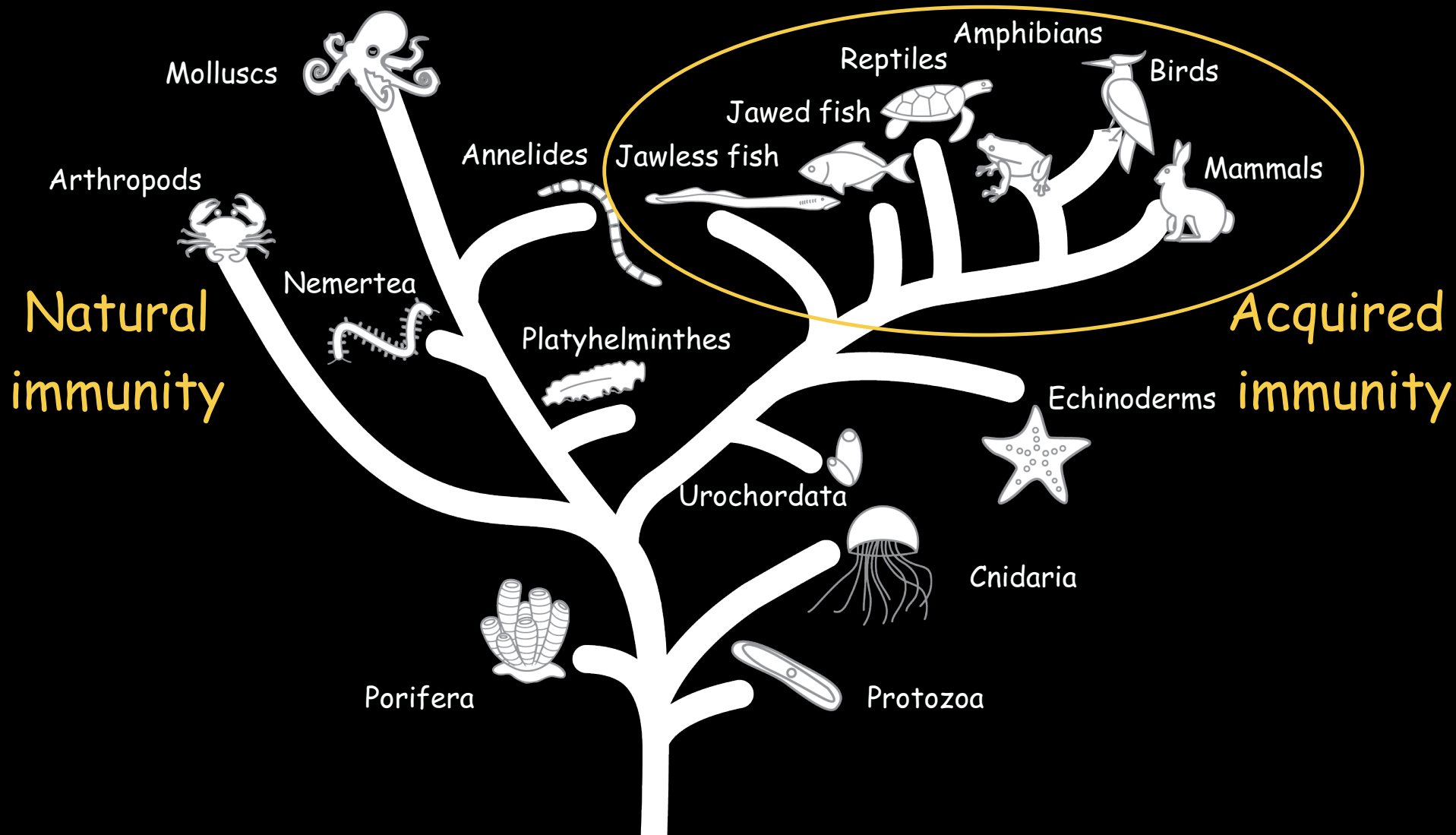


Streptomycin

21st century

Cancer may be controlled by
immunotherapy and its improvement
including microbiome manipulation

Acquired immunity evolved in vertebrates



Fortunate outcomes from evolution of acquired immunity

- Acquired immunity evolved in vertebrates as the **defense system** against pathogens. Consequently, the life span of vertebrates extended dramatically.
- Fortunately, cancer cells accumulate mutations and express **neo antigens**, which can also be recognized by acquired immunity.

Collaborators



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A. Durandy (Necker Hospital)
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Cancer immunotherapy by PD-1 blockade

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I. Konishi (Kyoto Univ.)

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NIHN



JSPS



AMED



NIBIOHN



ONO Pharmaceutical CO. LTD



Bristol-Myers Squibb



Tang Prize Foundation



Jane Coffin Child Memorial Fund



Thank you for your attention

