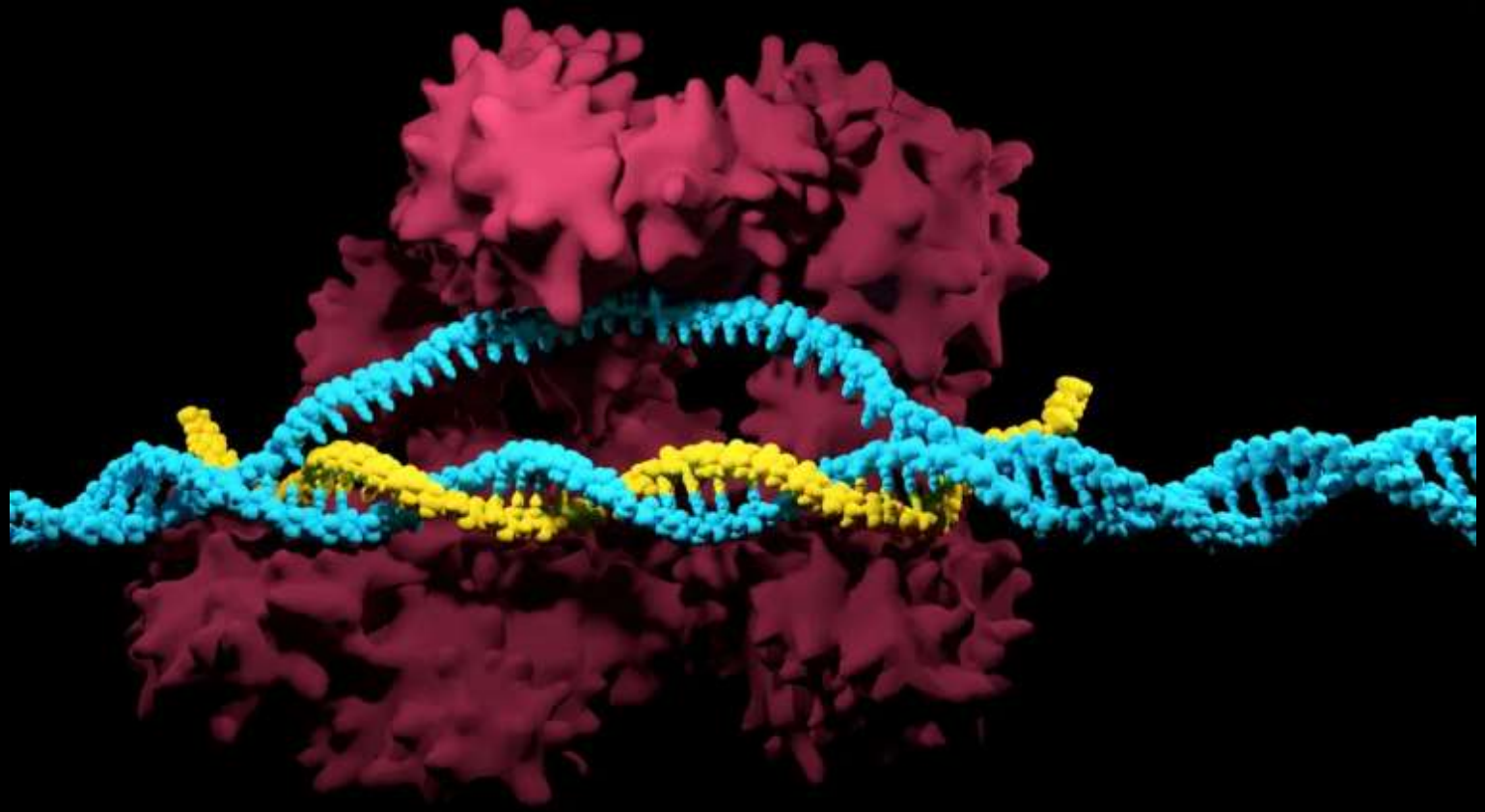


# ¿Cómo enseñar edición genética en las aulas?

**@LluisMontoliu**

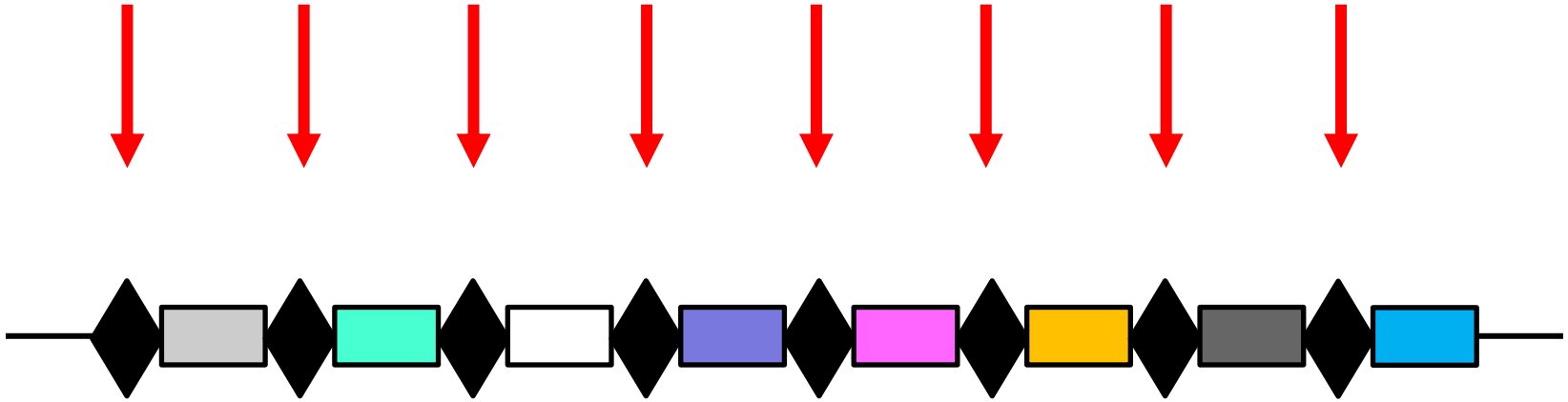
**CNB-CSIC y CIBERER-ISCIII, Madrid**


**Francisco Juan Martínez Mojica**


# Salinas de Santa Pola (Alicante)

# ADN

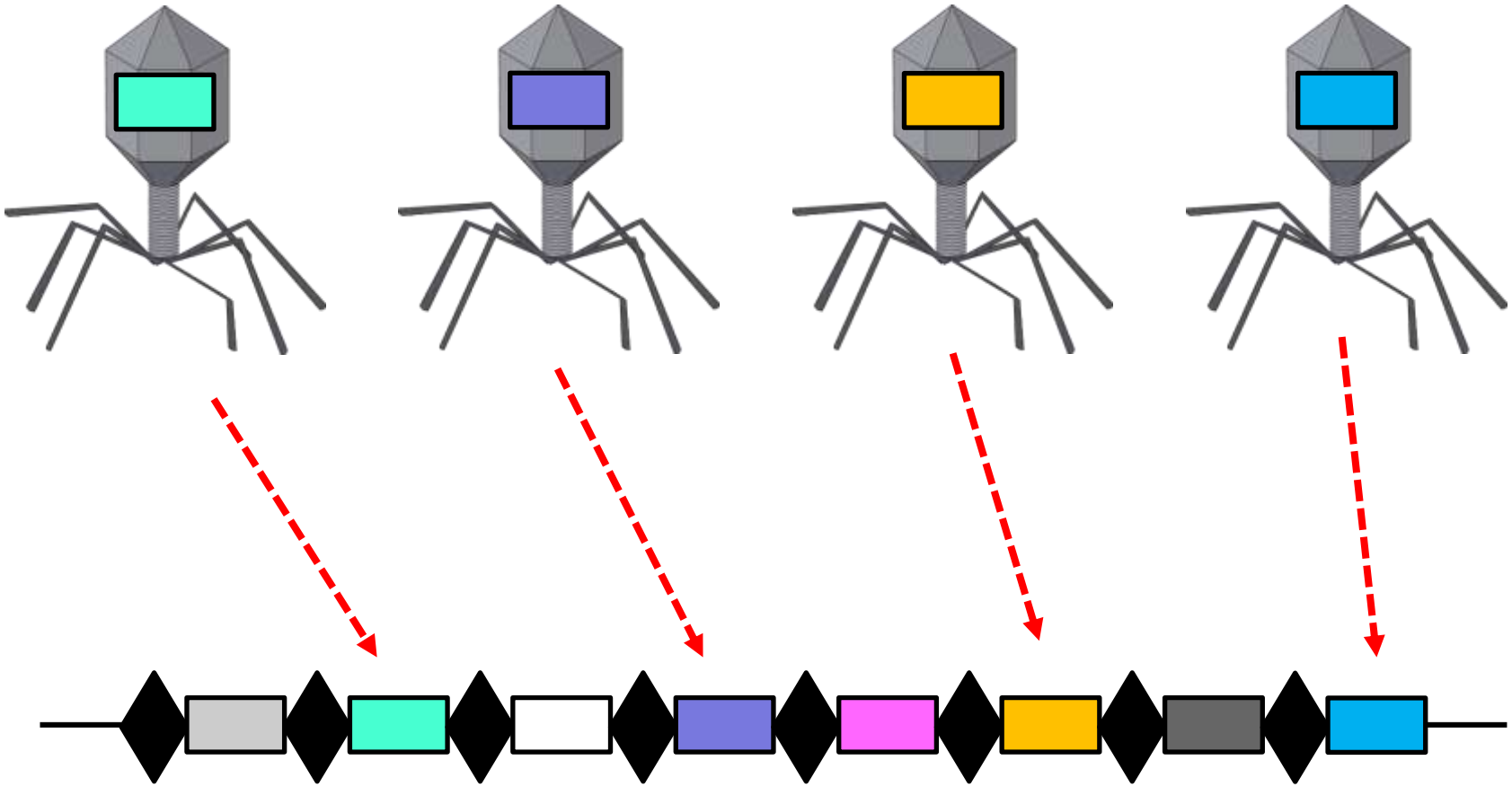
**2001 - 2002**



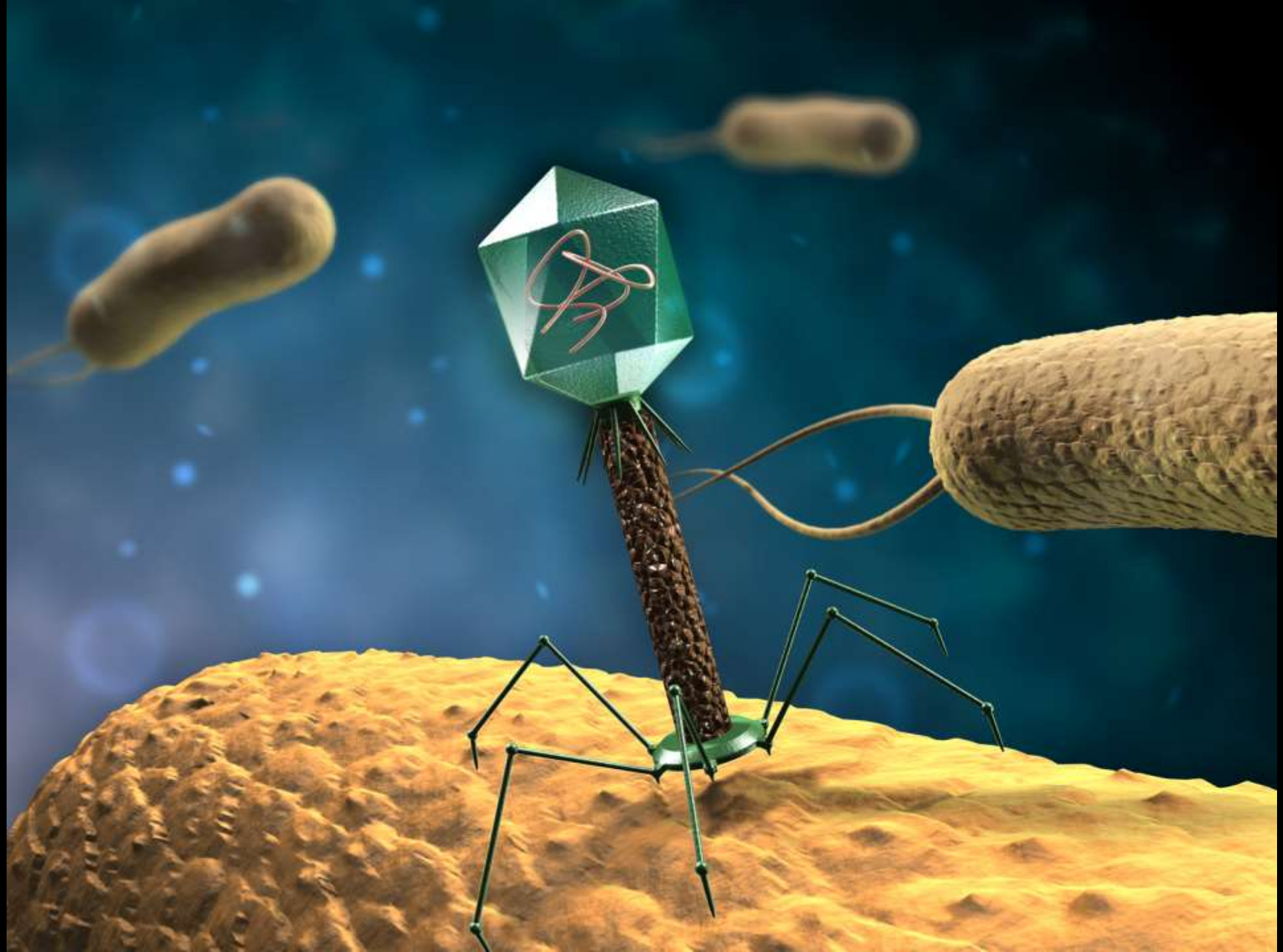
# C.R.I.S.P.R. (*crísper*)

*Clustered Regularly Interspaced Short Palindromic Repeats*

**Repeticiones cortas palindrómicas agrupadas y regularmente espaciadas**



2003 - 2005




SINC

1993

Francisco Mojica
University of Alicante, Spain

Rodolphe Barrangou
North Carolina State Univ, Raleigh, USA

Philippe Horvath
DuPont Nutrition and Health, France

Luciano Marraffini
The Rockefeller Univ, New York, USA

John van der Oost
Wageningen University, The Netherlands

Emmanuelle Charpentier
MPI for Infect. Biol., Berlin, Germany

Jennifer Doudna
Univ California Berkeley, CA, USA

Virginijus Siksnys
Vilnius University, Lithuania

Feng Zhang
BROAD-MIT, Cambridge, MA, USA

George Church
Harvard Med School, Boston, MA, USA

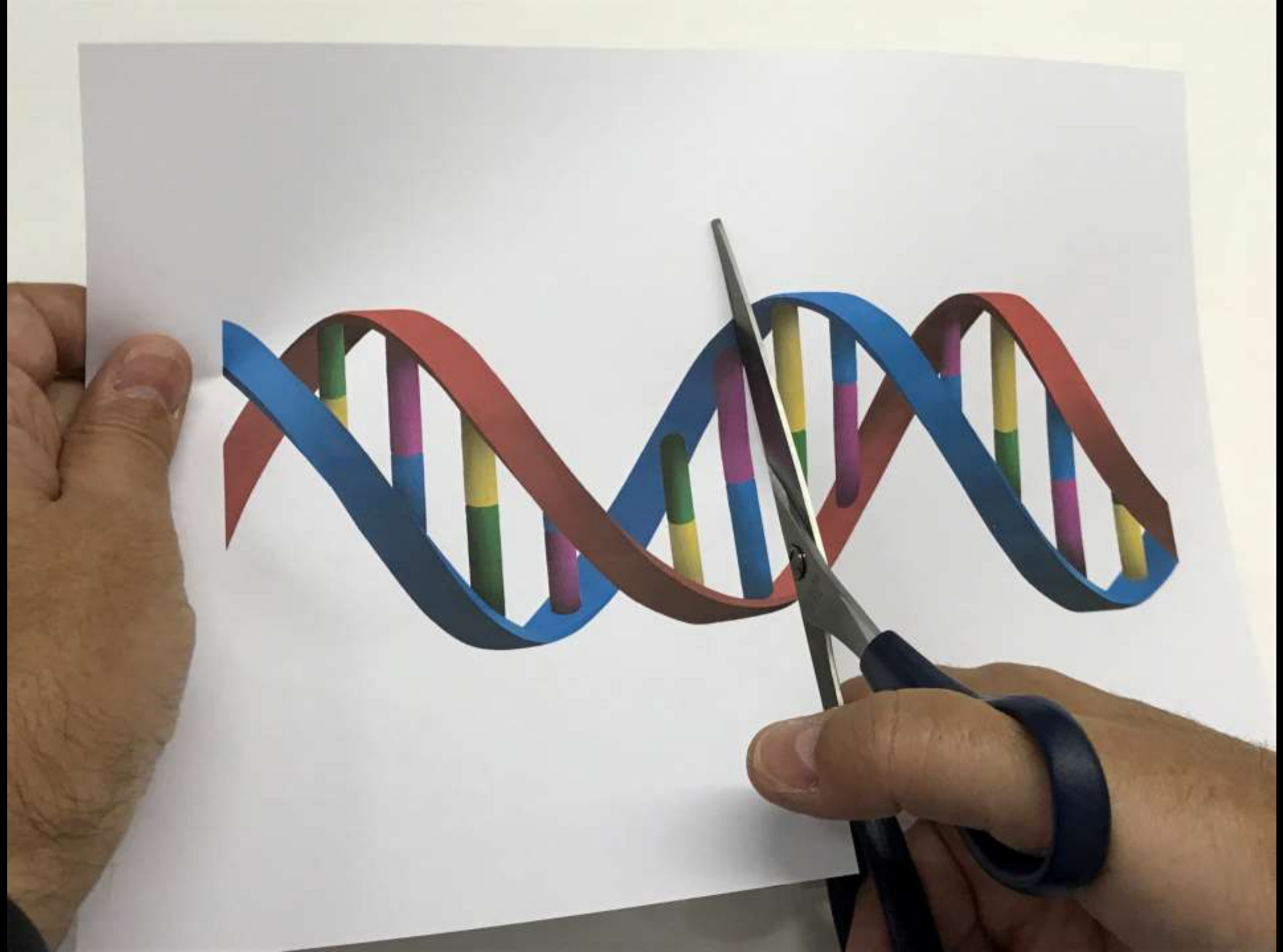
Rudolf Jaenisch
Whitehead Inst, Cambridge, MA, USA

J. Keith Joung
Mass Gen Hosp, Charlestown, MA, USA

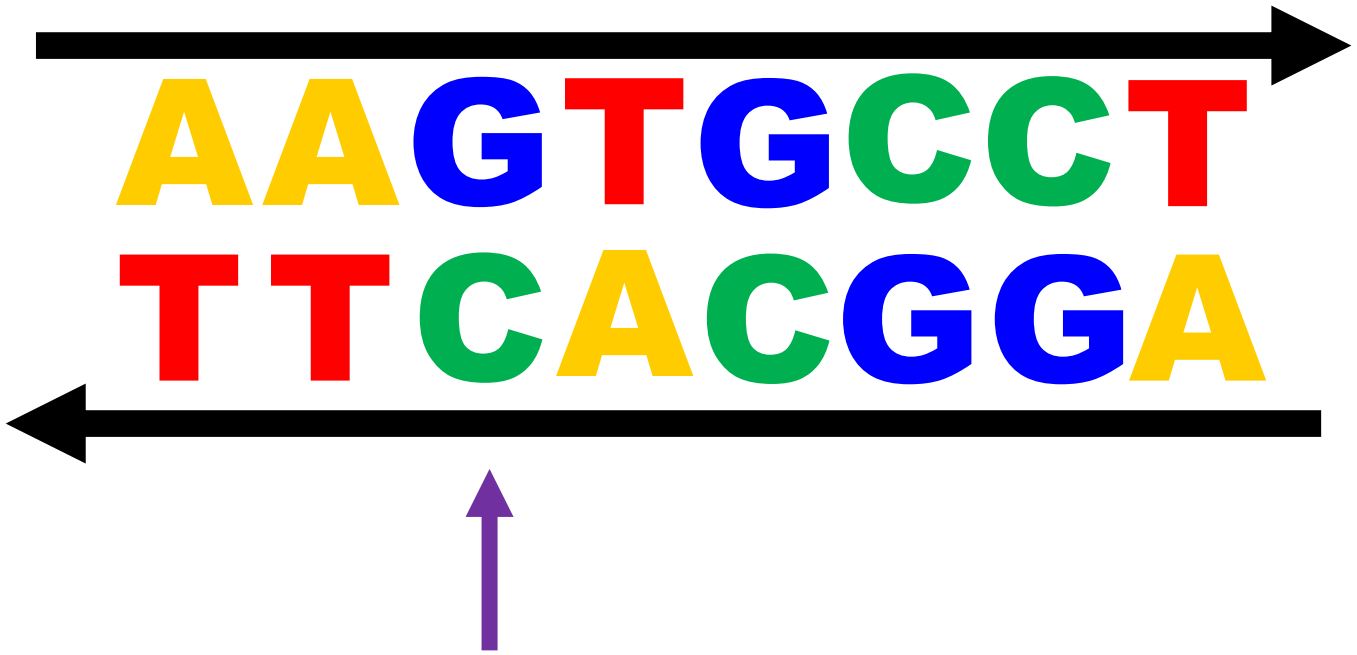
2013

David R. Liu
BROAD-MIT, USA

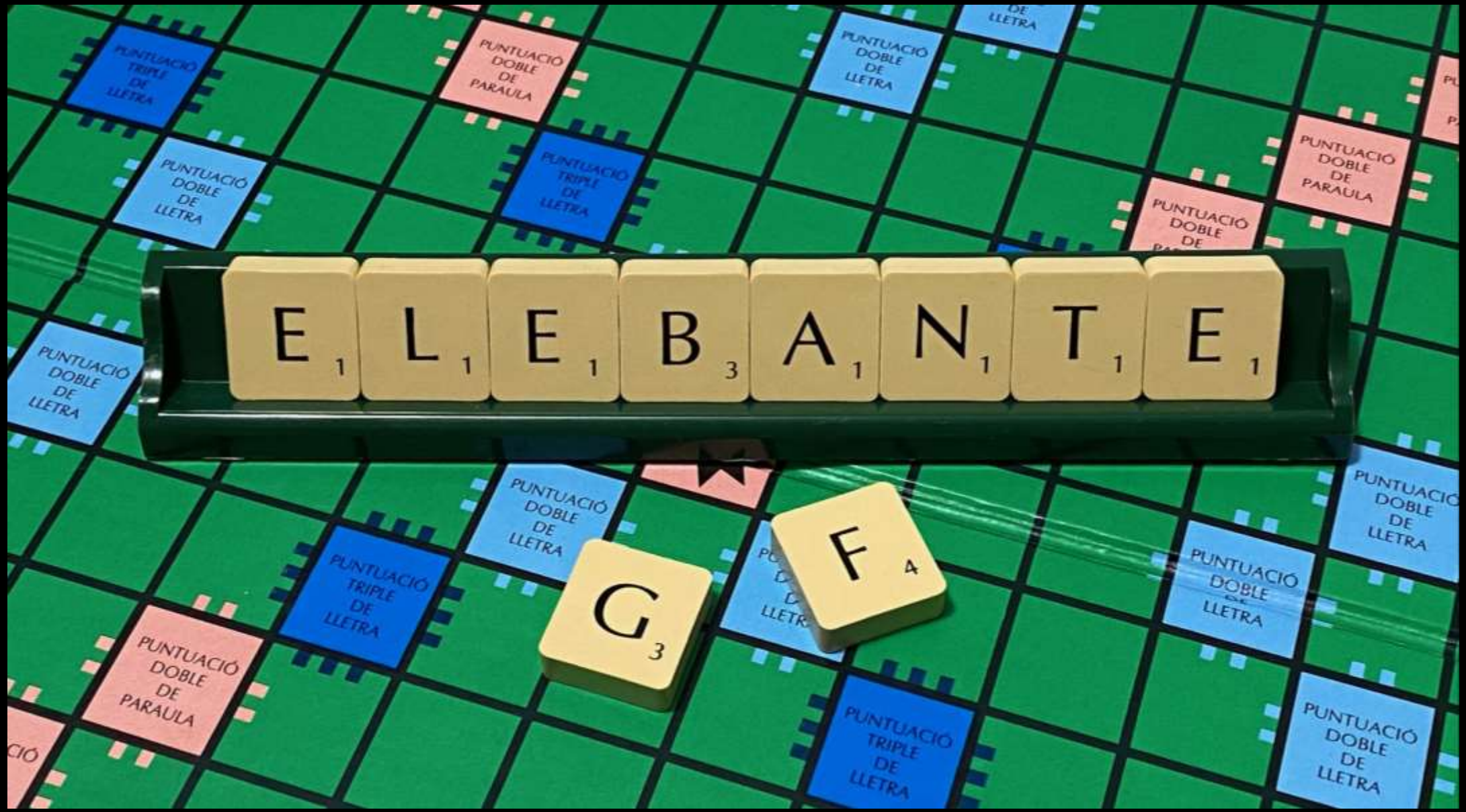
2016



# ADN



# ADN


ELEBANTE
G
F
PUNTUACIÓ DOBLE DE LLETRA
PUNTUACIÓ TRIPLE DE LLETRA
PUNTUACIÓ DOBLE DE PARAULA


E L E F A N T E
G
B


E L E G A N T E
F
B

# Burgos es una de las cumas de la humanidad

# Burgos es una de las cumas de la humanidad

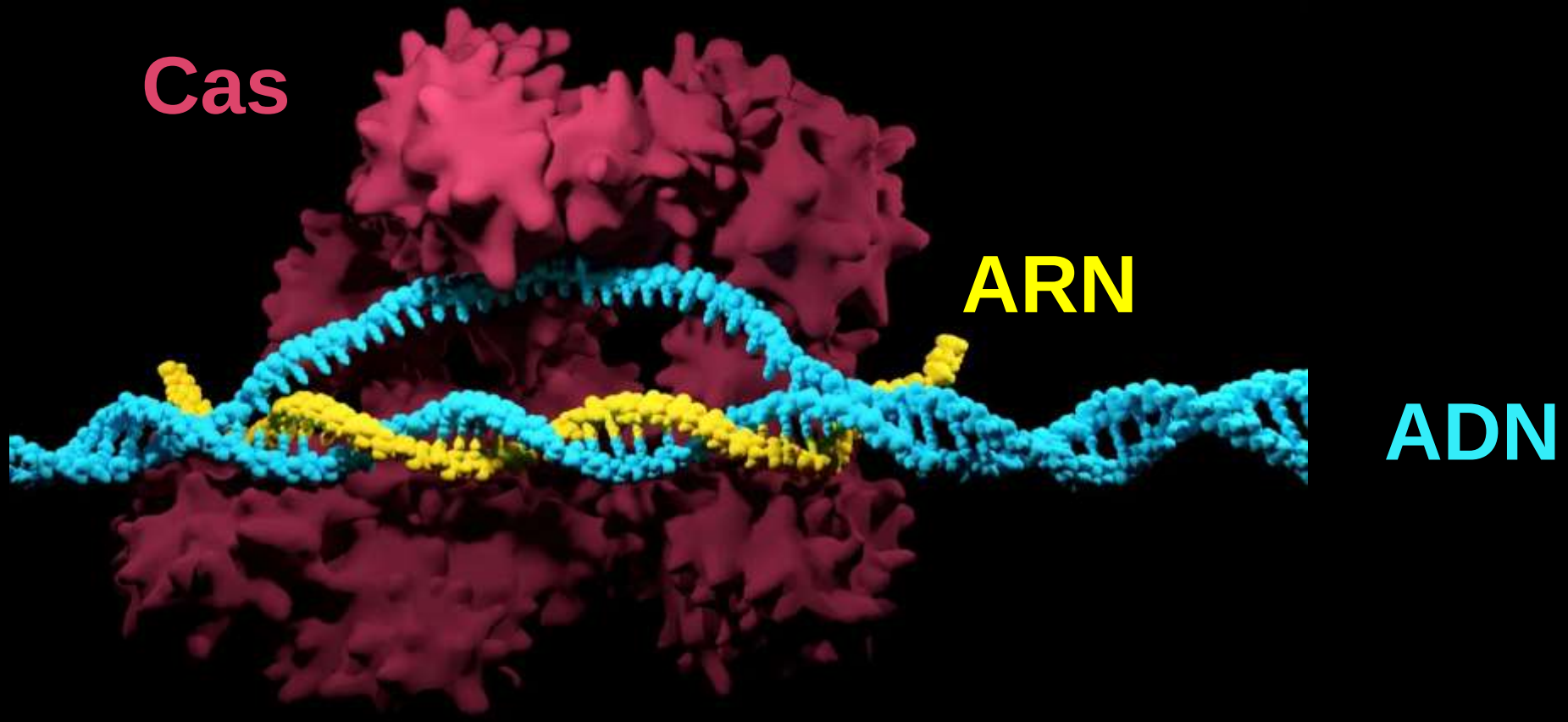
n

**Burgos es una de las cu-as de la humanidad**

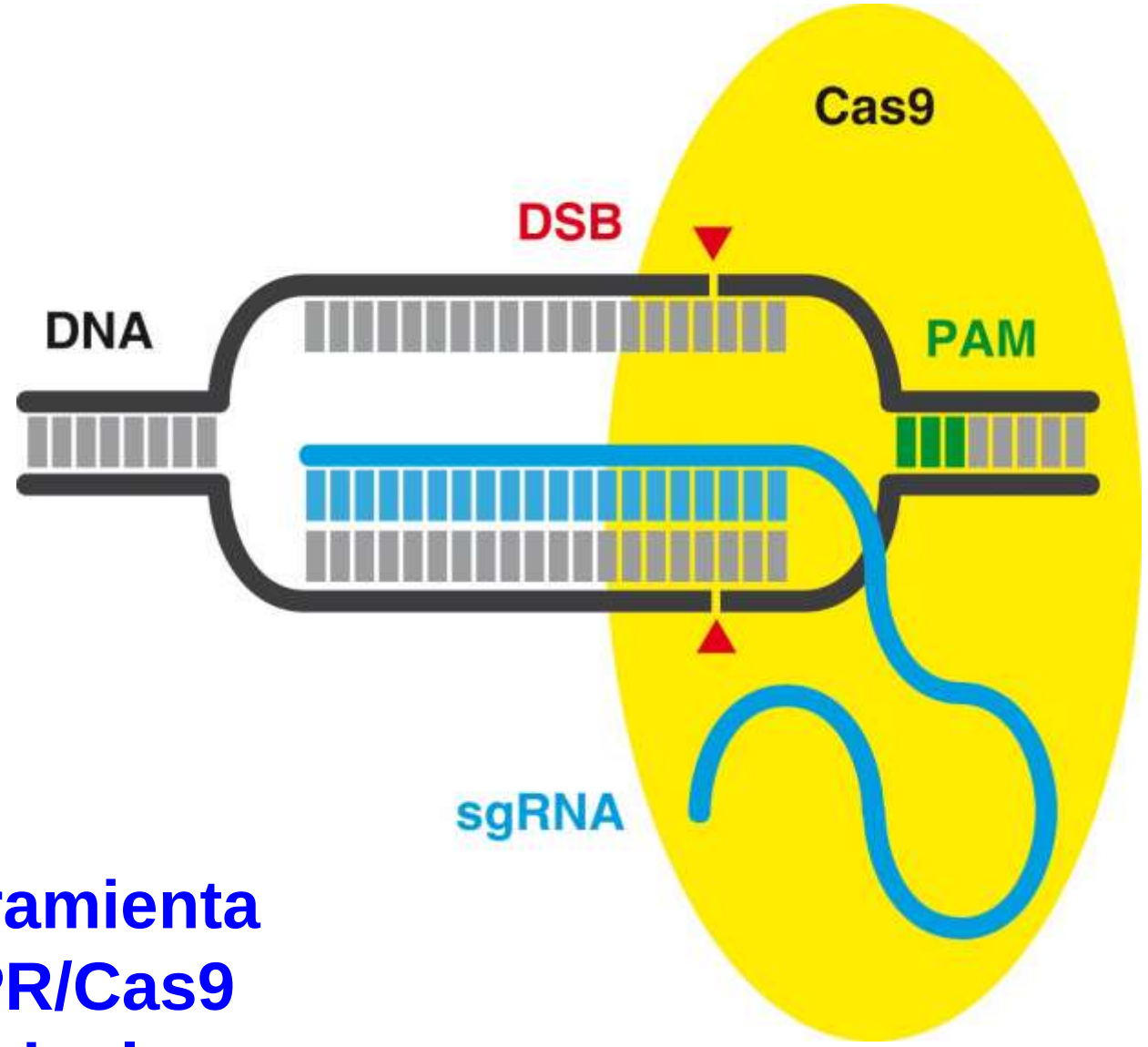
# Burgos es una de las cunas de la humanidad

**Burgos es una de las cunas de la humanidad**

# ¿Cómo funcionan las CRISPR?

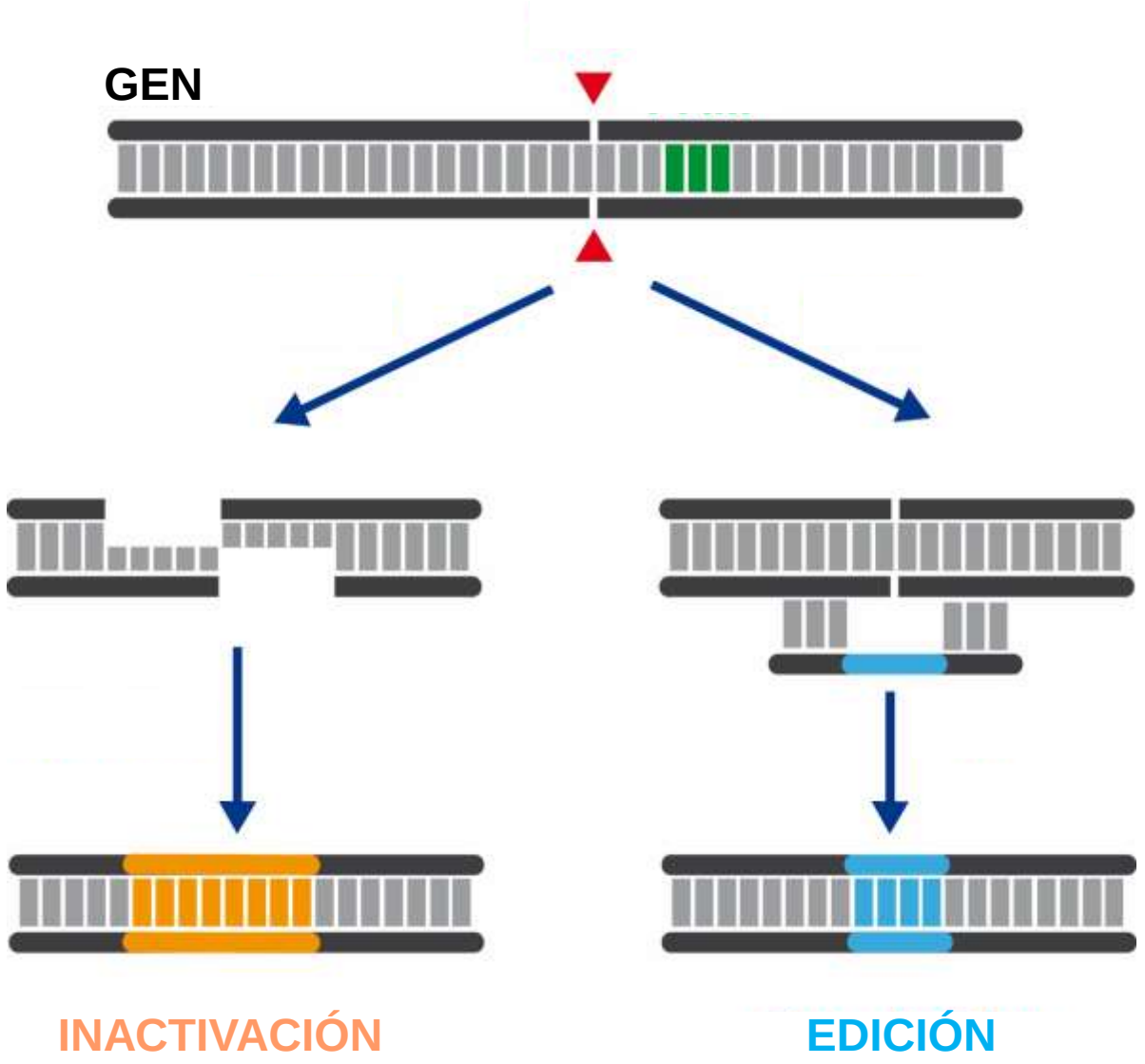


2012



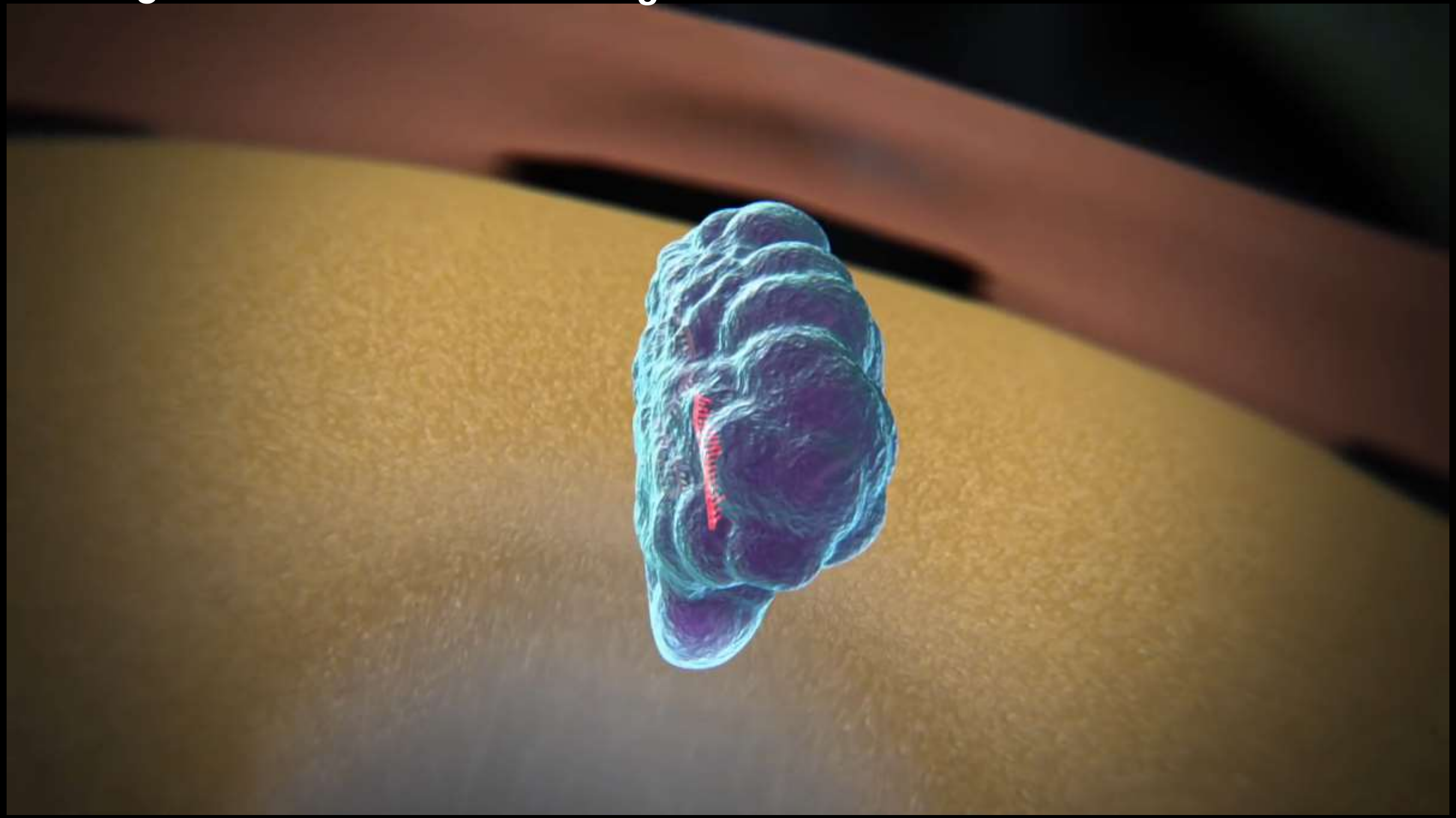

# La herramienta CRISPR/Cas9 original

# ¿Cómo funcionan las CRISPR?

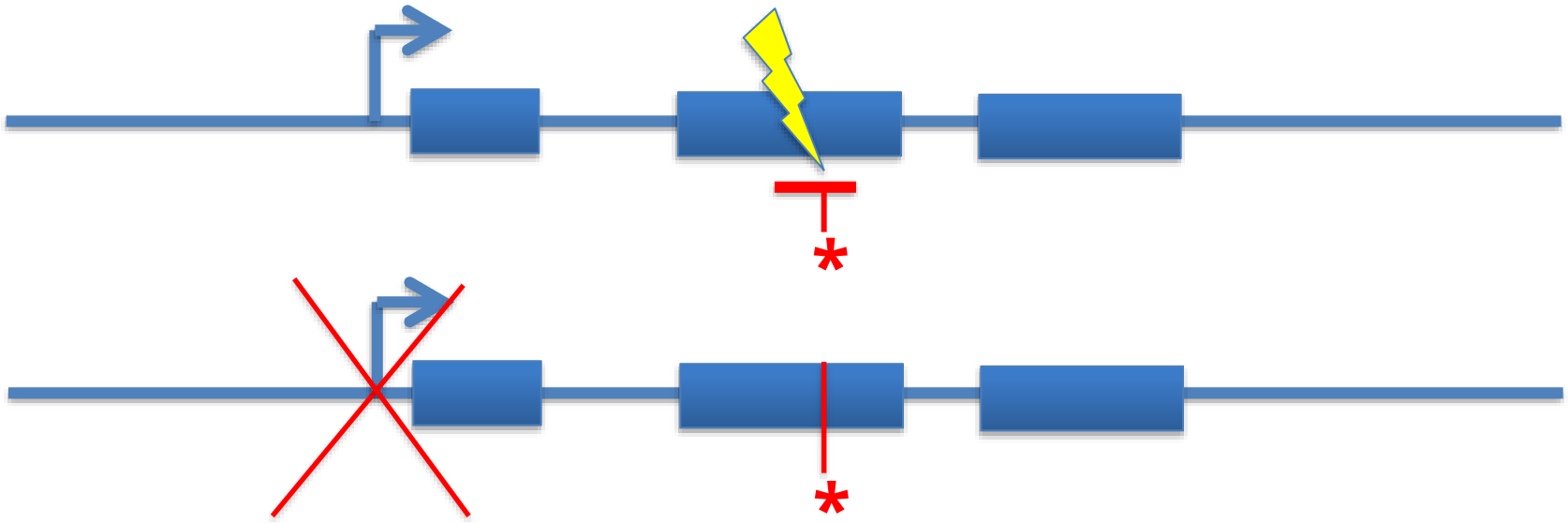


# ¿Cómo funciona la edición genética?

Movie by McGovern Institute for Brain Research at MIT



# Mutaciones puntuales



# Ratones CRISPR “avatar”

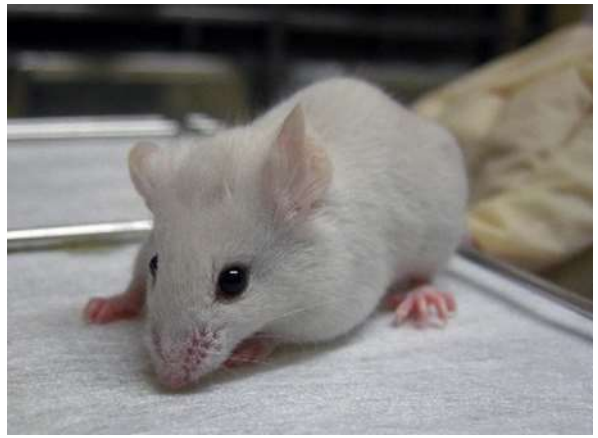
# Modelos animales de albinismo

**OCA1 (*TYR*) – OCA1A**

# OCA1 (*TYR*) – OCA1B

**OCA2 (*OCA2*)**

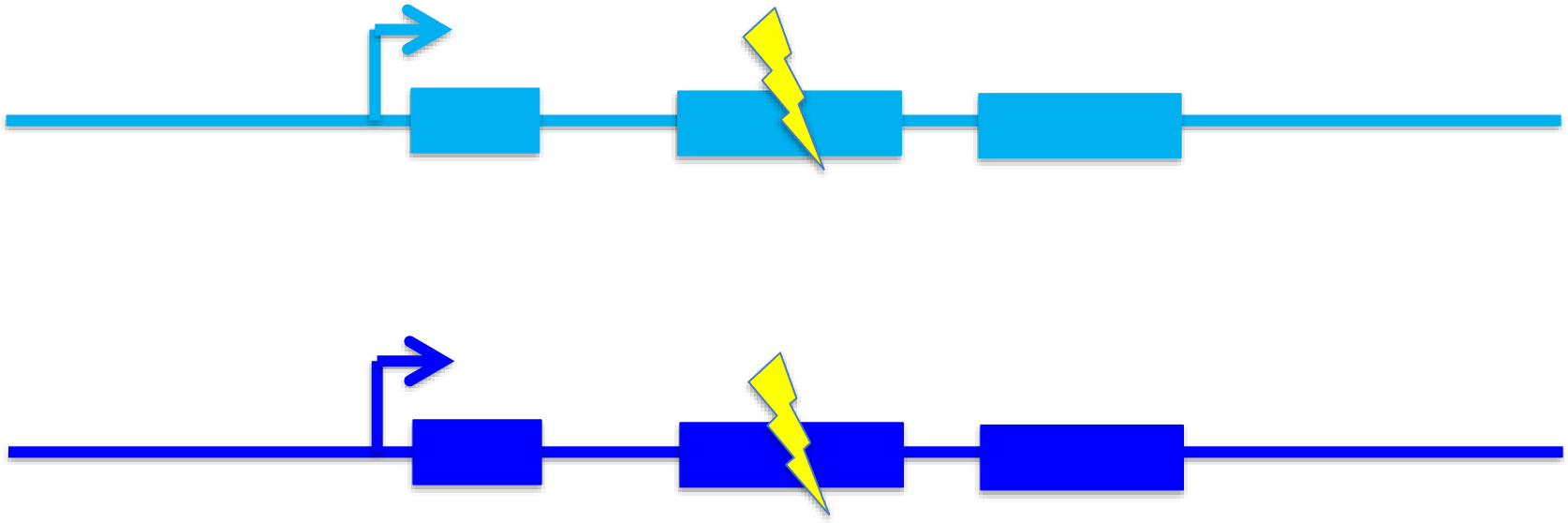
# OCA4 (*SLC45A2*)

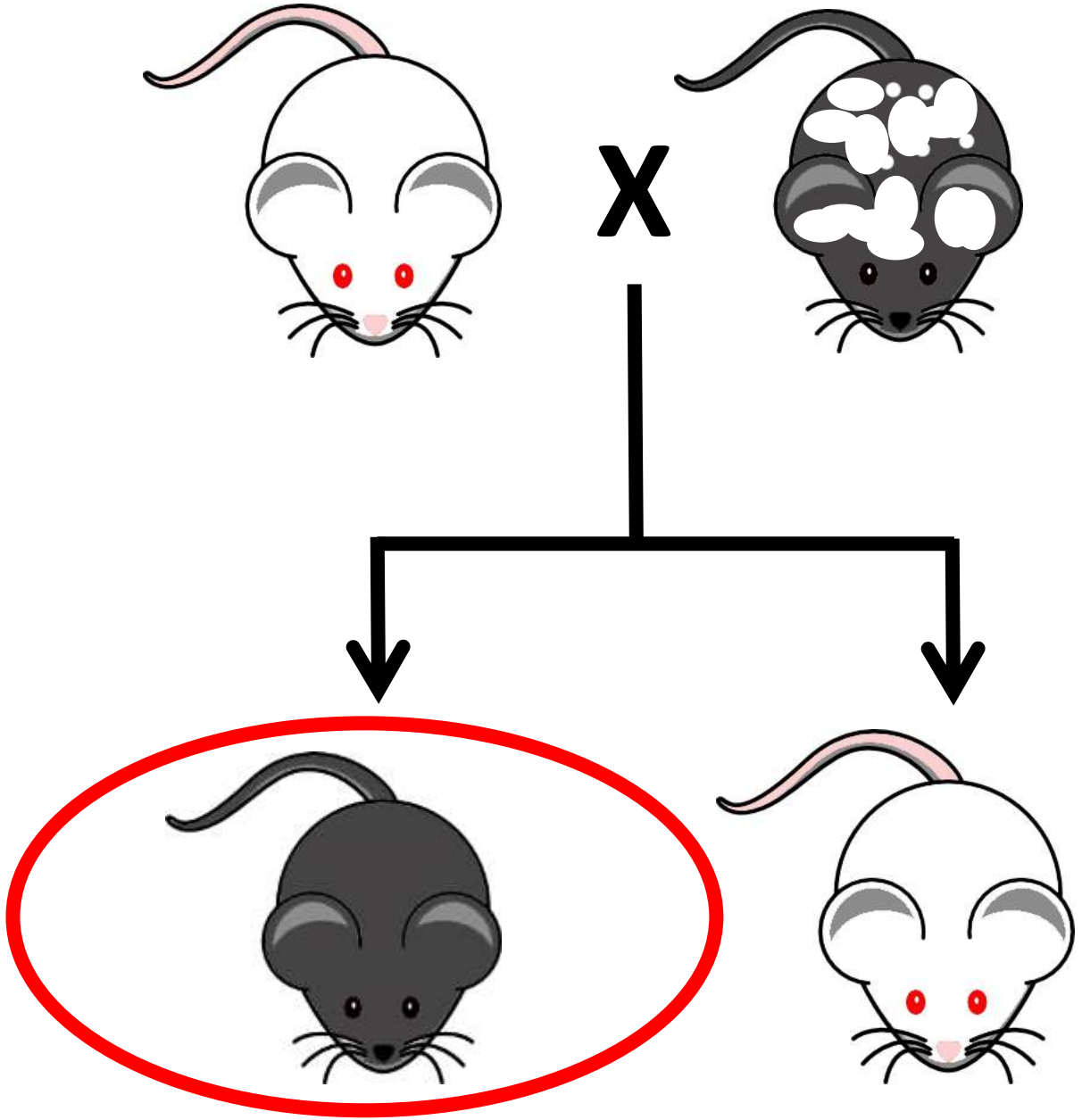
# Validación de tratamientos

# Limitaciones de las herramientas CRISPR de primera generación

# Inactivación de genes parecidos



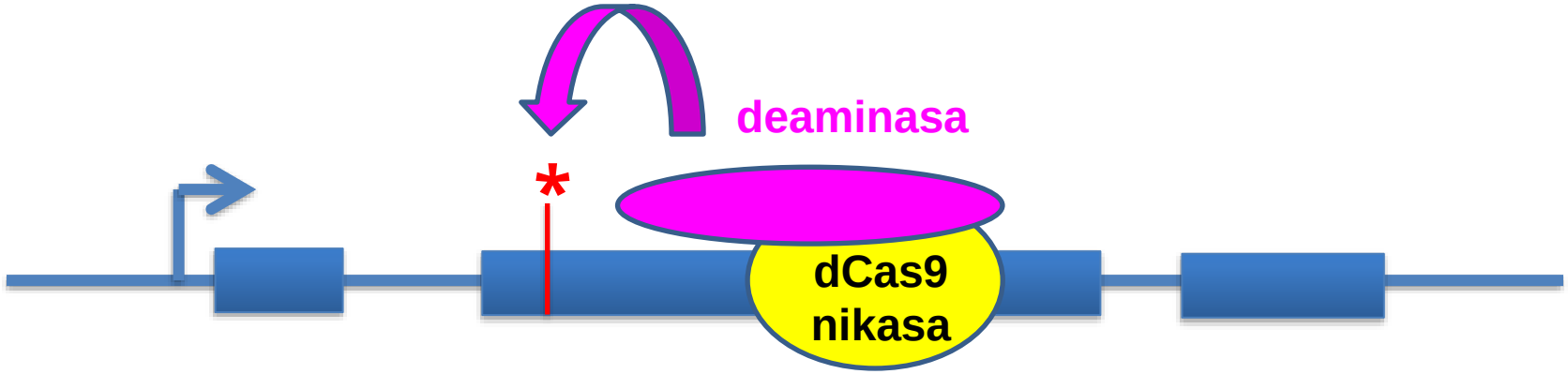
# Mosaicismo



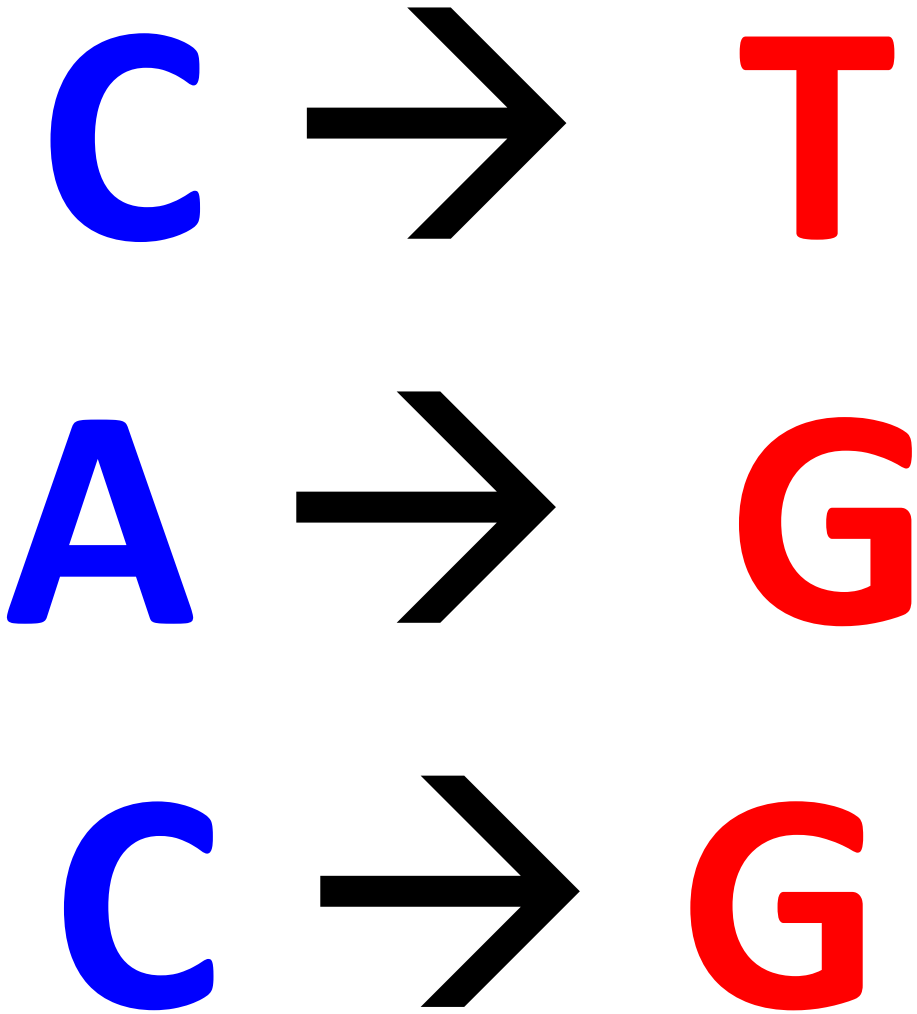
# Soluciones a las limitaciones de las herramientas CRISPR de primera generación


David R. Liu
BROAD Institute

# Editores de bases

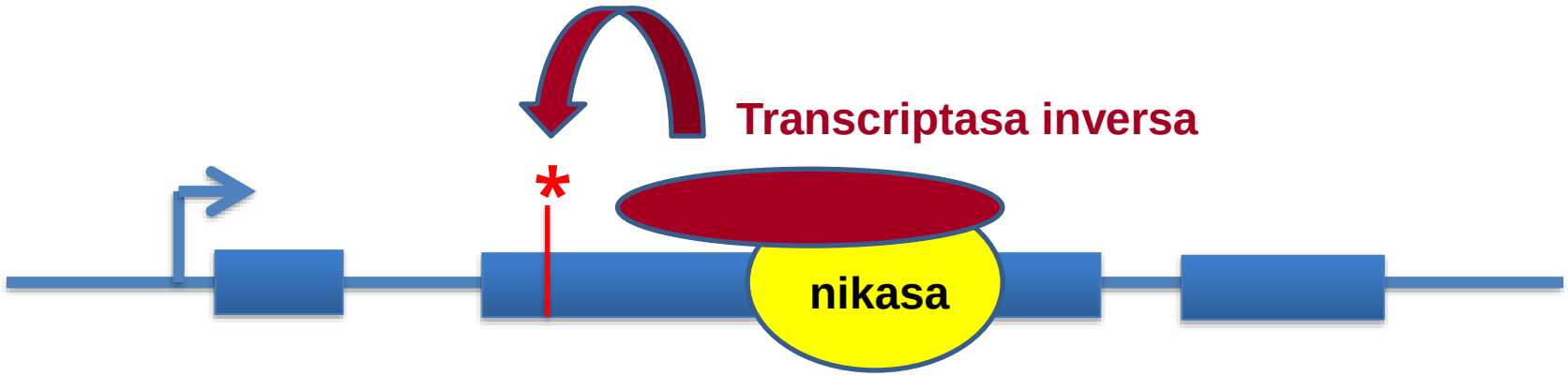



**Segunda generación de herramientas CRISPR**


C
T
A
G
C
G

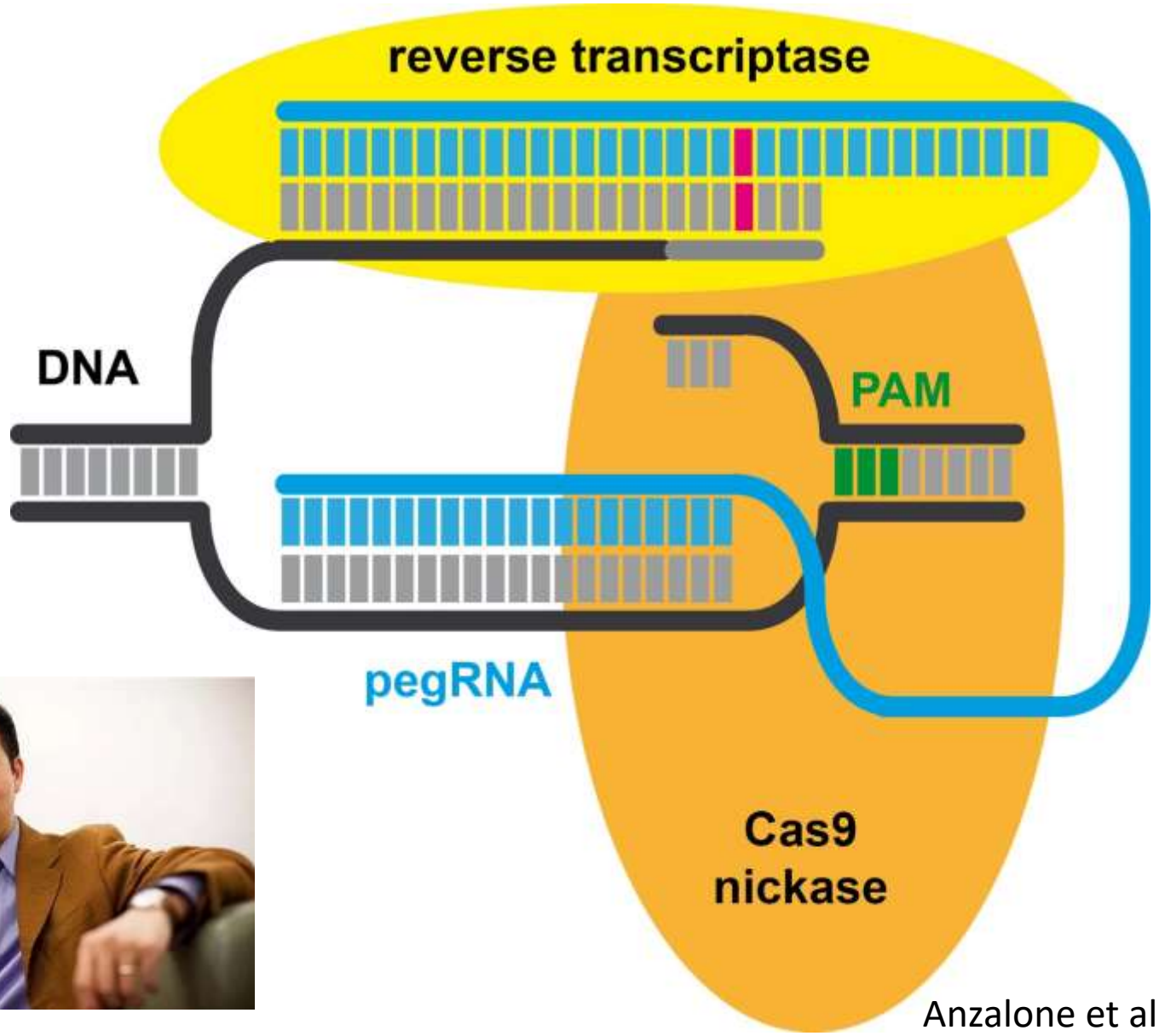

AGGATTAGAGCCGATAGCATACGATCAGTACGAT

# Édición de calidad




**Tercera generación de herramientas CRISPR**

# Edición de calidad




David Liu Lab

Anzalone et al. Nature 2019

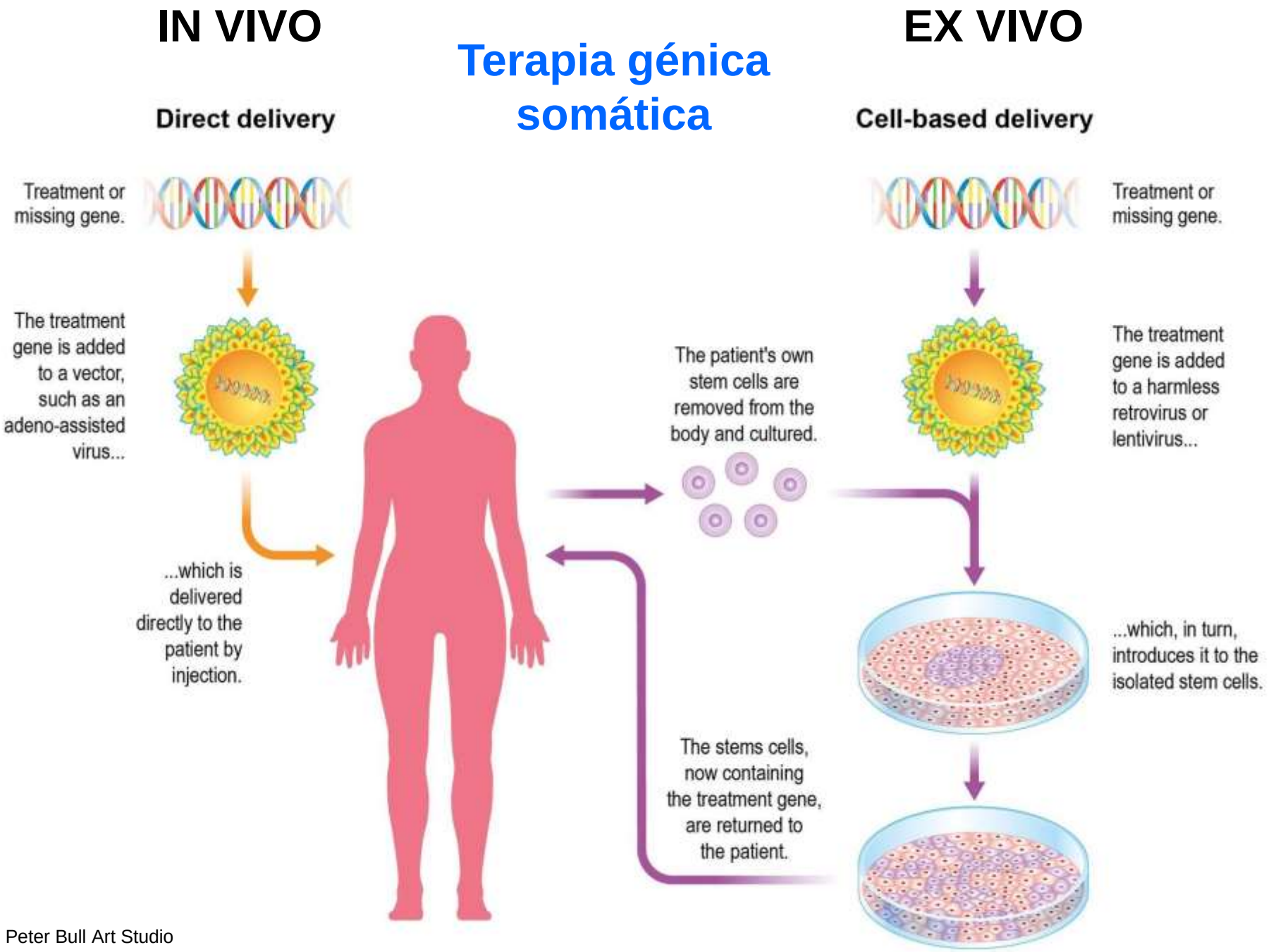
THE CONVERSATION
Rigor académico, oficio periodístico



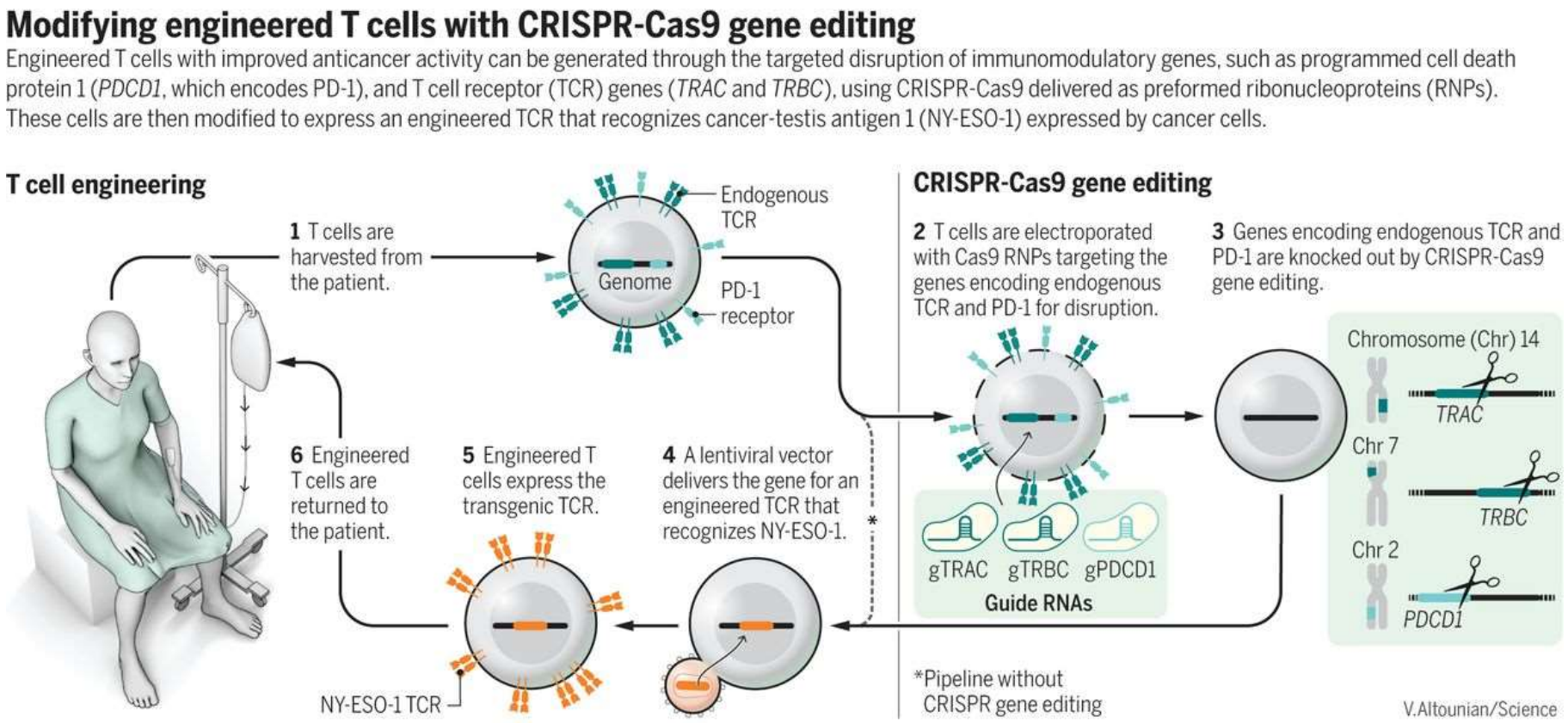
# La CRISPR más precisa hasta la fecha convierte la tijera genética en una navaja suiza

Publicado: 23 octubre 2019 22:30 CEST

# Terapia génica somática



# Inmunoterapia del cáncer con CRISPR




**6 Feb 2020**

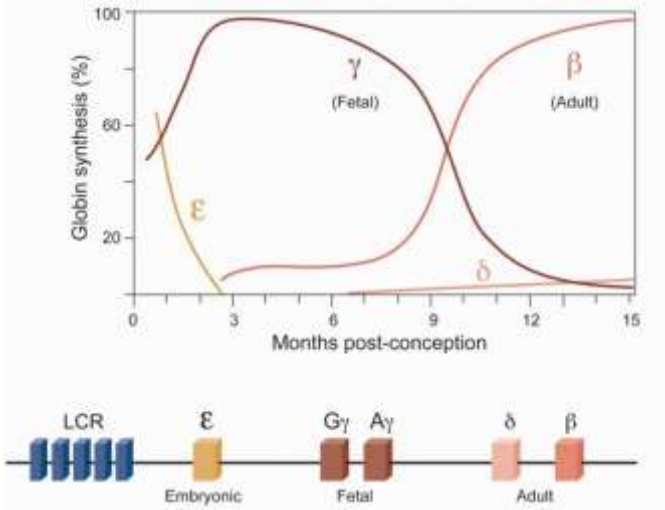
Hamilton & Doudna (Science, 2020)
Stadtmauer et al. (Carl June Lab, Science 2020)

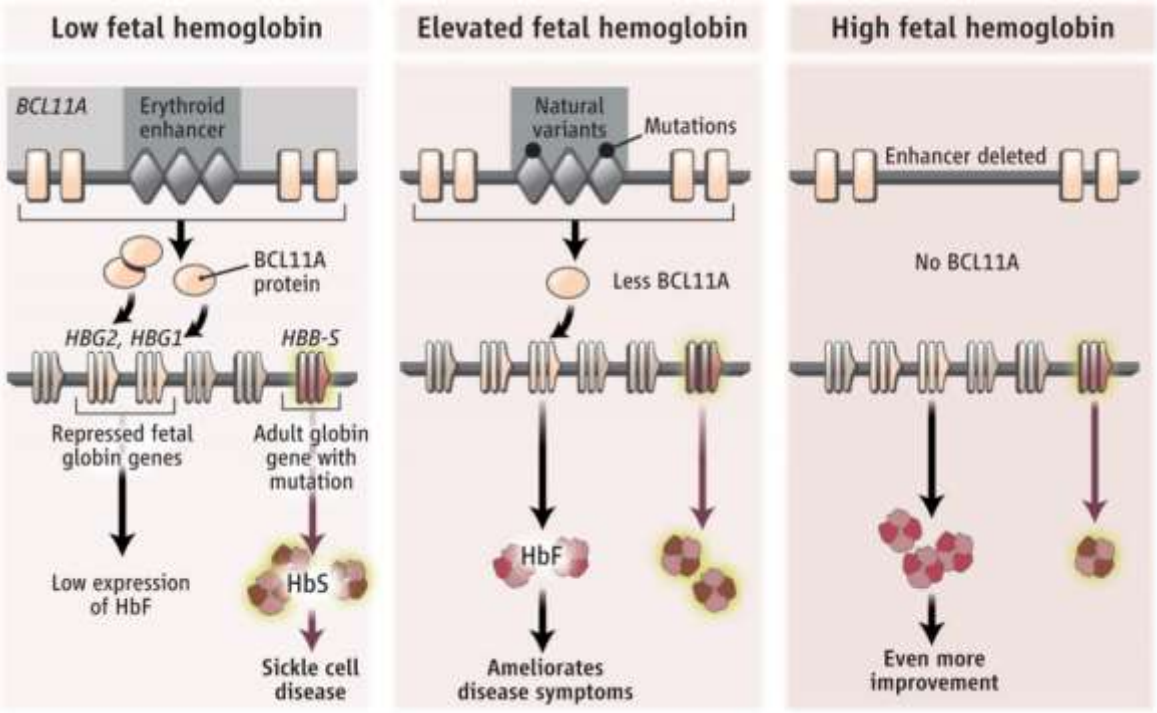
**Alyssa**

**Tratada su leucemia linfoblástica aguda de células T con editores de bases y con tecnología CAR-T**

**Mayo 2022**

# Curar la anemia falciforme con terapia génica CRISPR




Bauer Lab (Dana Farber-Boston Children's)

**Anemia falciforme**

**1:5.000/10.000**

CRISPR THERAPEUTICS

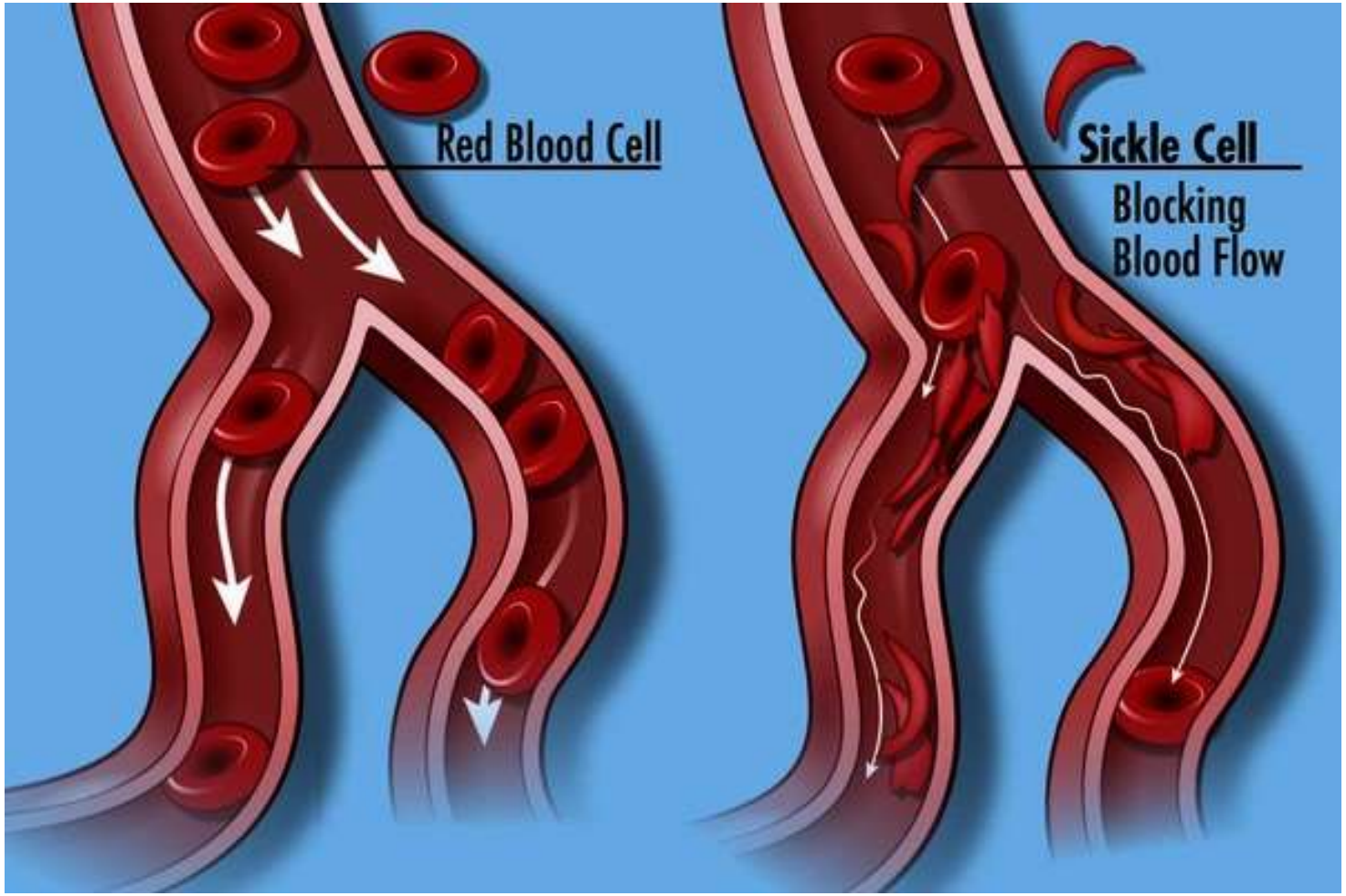
VERTEX

**Los primeros ensayos clínicos ex-vivo**
**Con CRISPR aprobado en Europa**

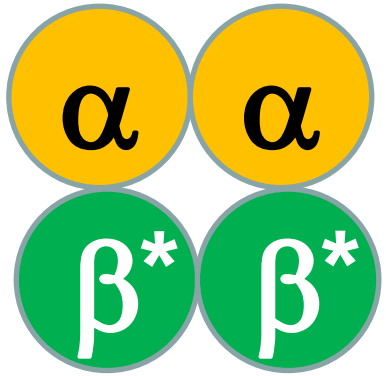
**November 2019**

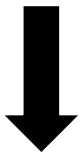
Glóbulo rojo normal

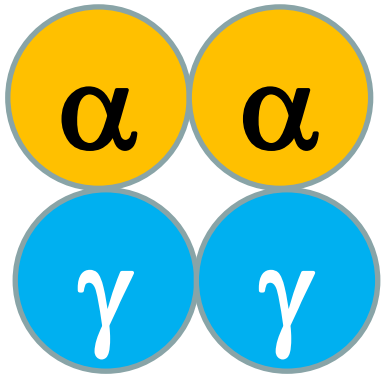
Glóbulo rojo en anemia falciforme




Hemoglobina en anemia falciforme



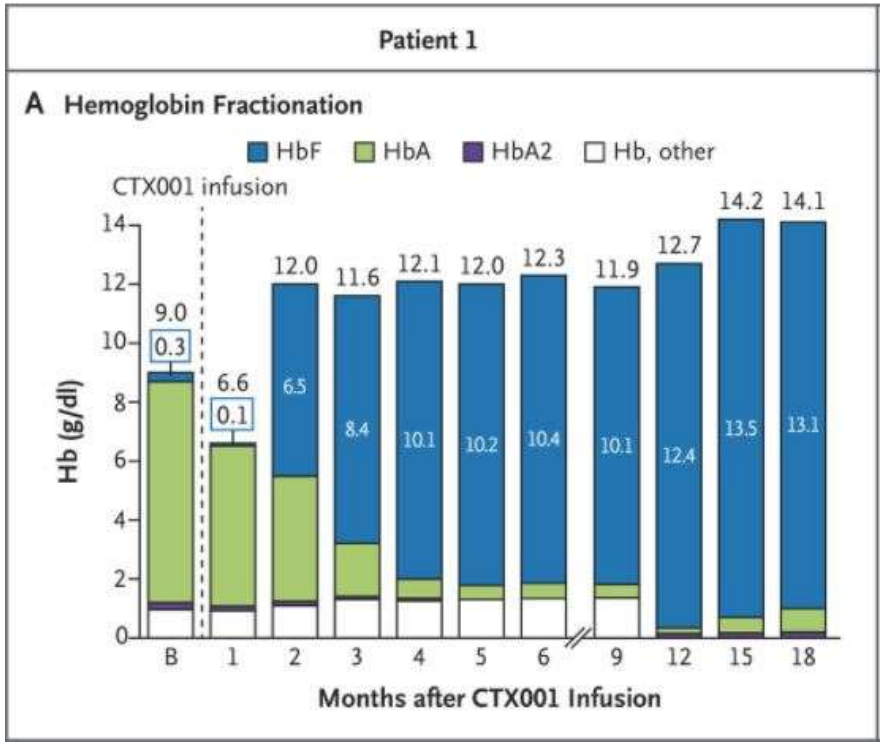






Hemoglobina curada con CRISPR

# CRISPR para tratar ANEMIA FALCIFORME

**Victoria Gray** (tratada 2 julio 2019)

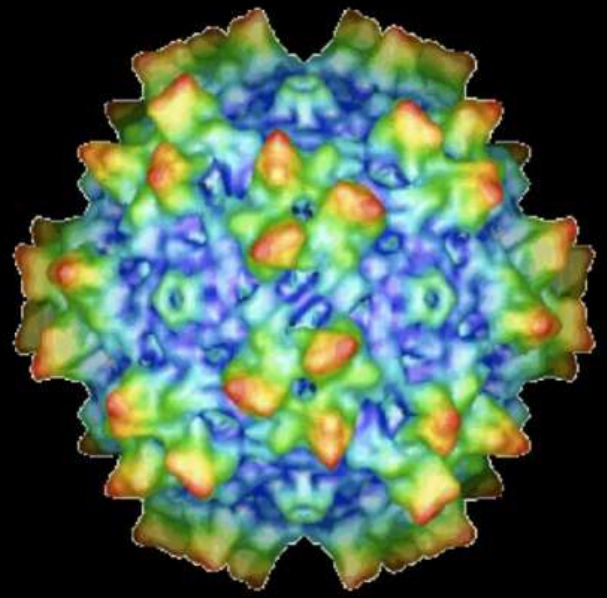



Frangoul *et al.* NEJM (5 December 2020)

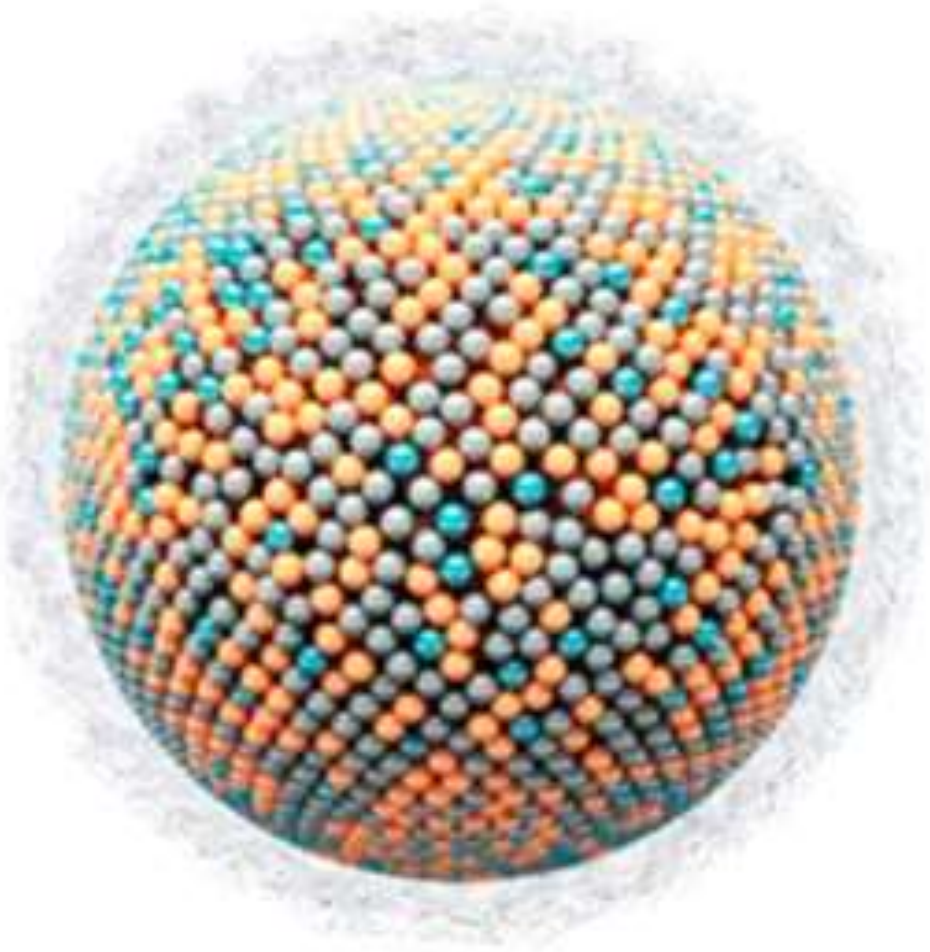
**Primer paciente curado en EE.UU. (2020)**

NPR

# La mayoría de ensayos clínicos CRISPR usan virus adenoasociados para administrarlas



AAV



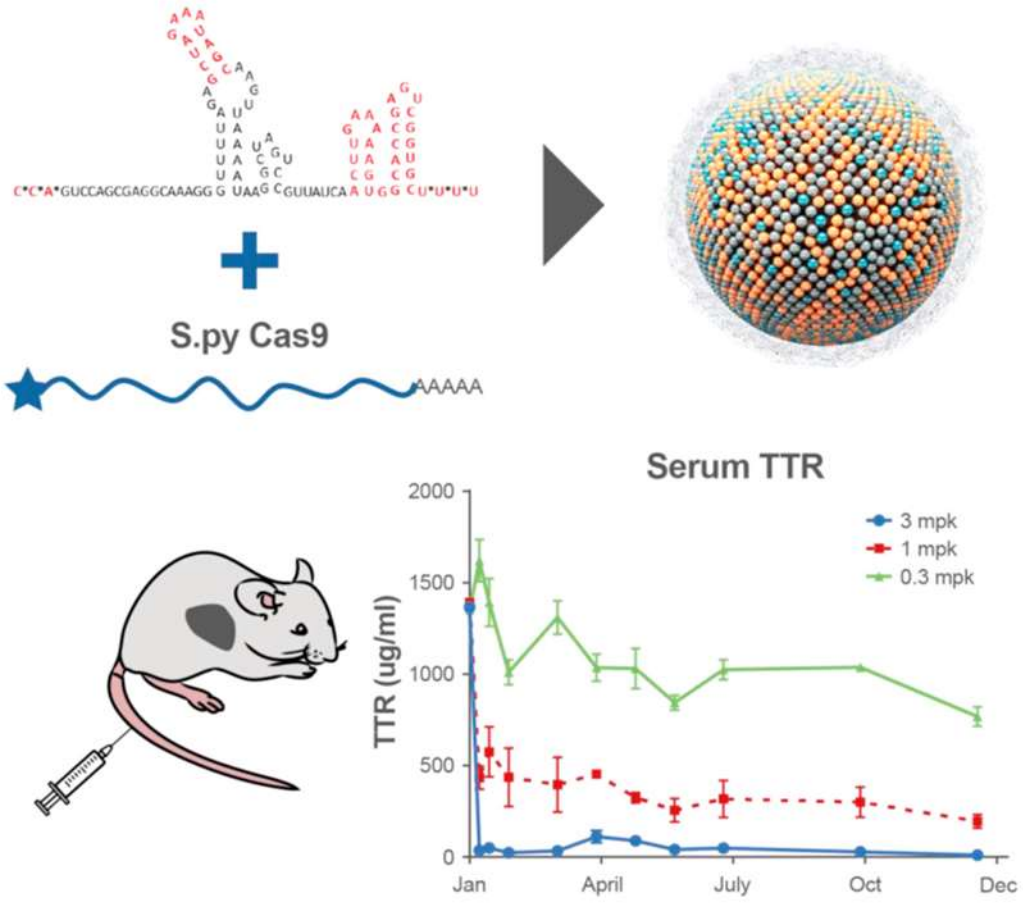
# Nanopartículas lipídicas


**Las primeras vacunas COVID-19 aprobadas**

| | | |
|---|---|---|
| Pfizer-BioNTech | FDA | 11 Dic 2020 |
| Moderna | FDA | 18 Dic 2020 |

# Amiloidosis congénita por transtiretina (ATTR)
# NANOTECNOLOGÍA - Nanopartículas

**1-9:1.000.000**

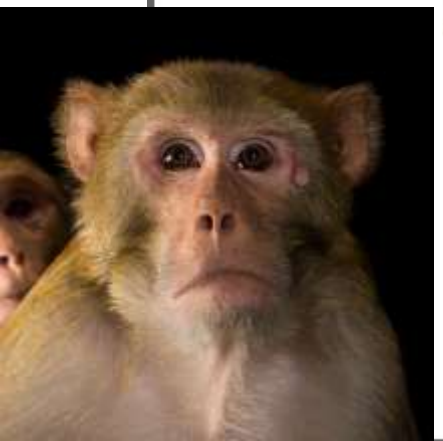


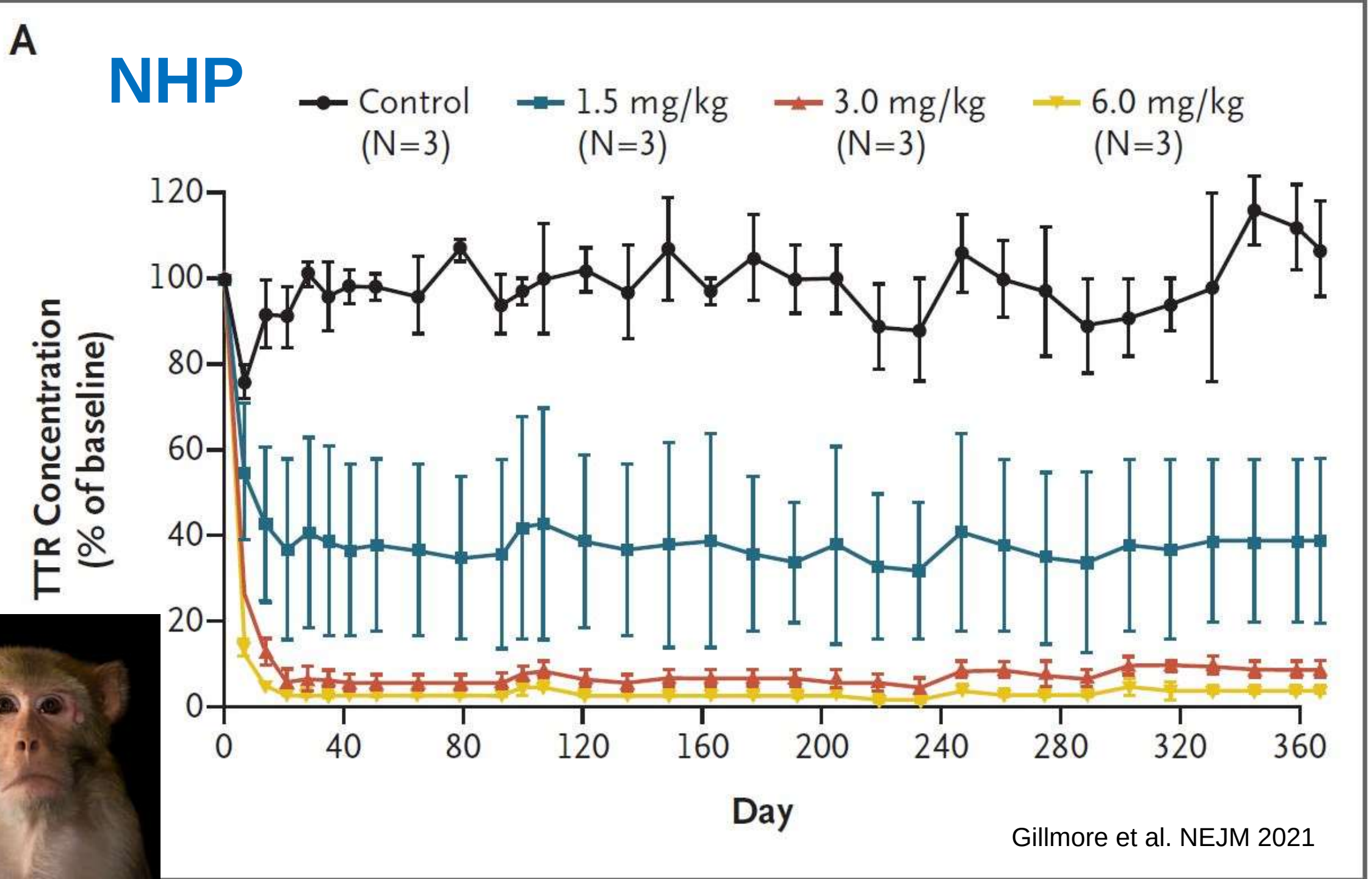


*Finn et al. Cell Reports* 2018 22, 2227-2235DOI: (10.1016/j.celrep.2018.02.014)


October 2020

# NHP



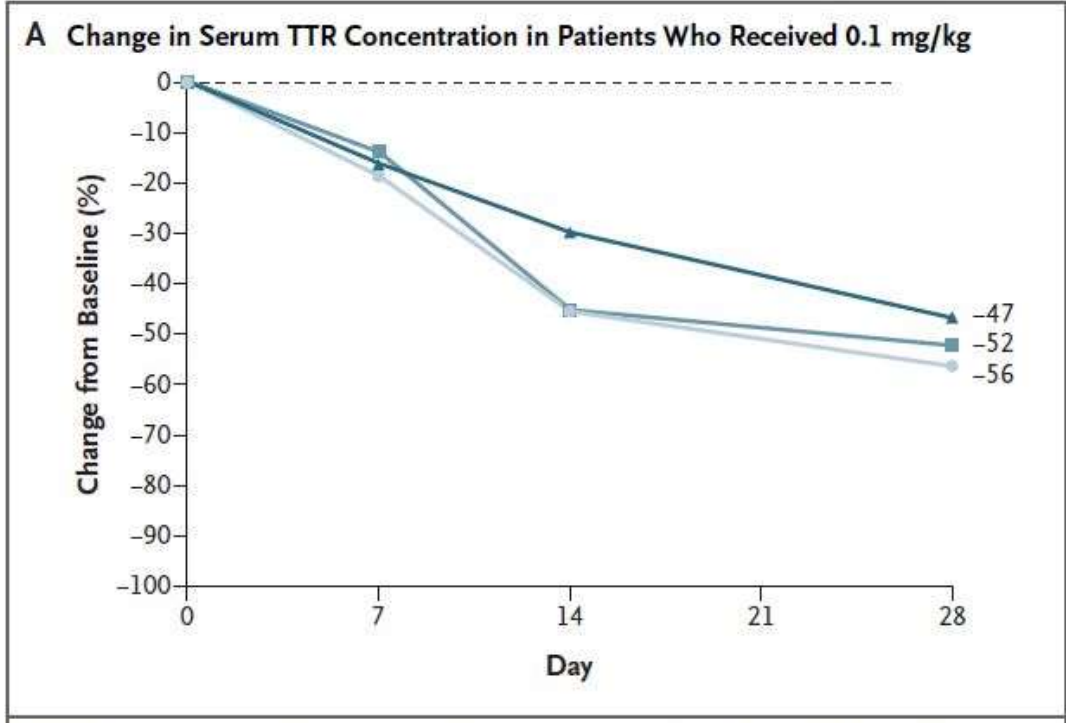


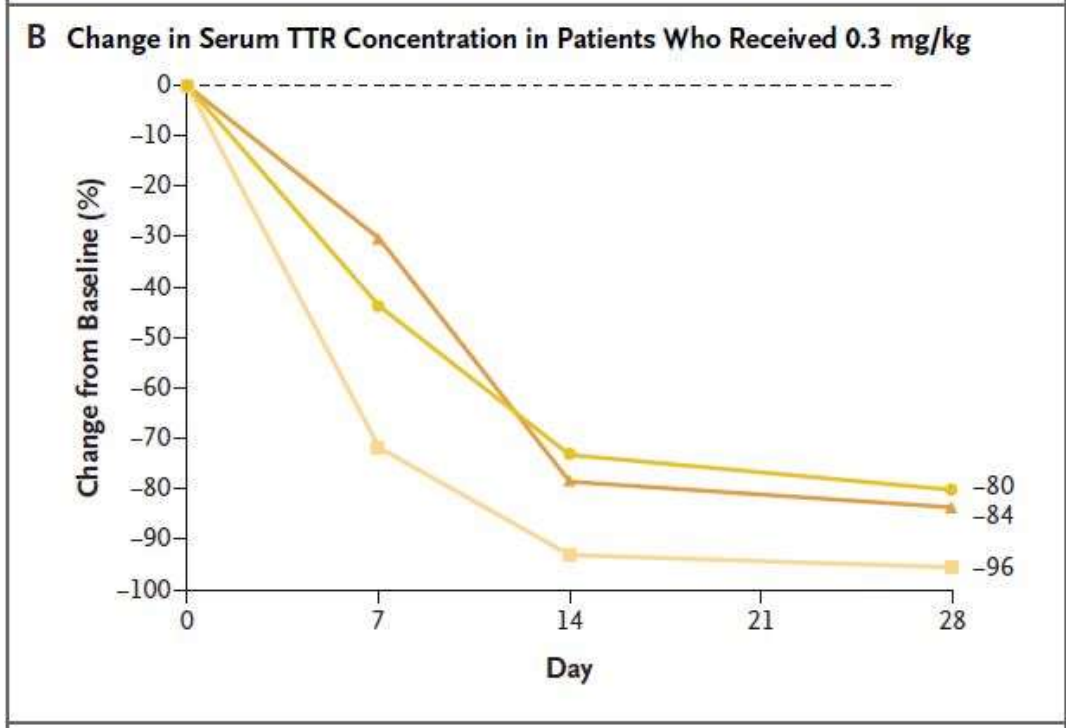

Gillmore et al. NEJM 2021

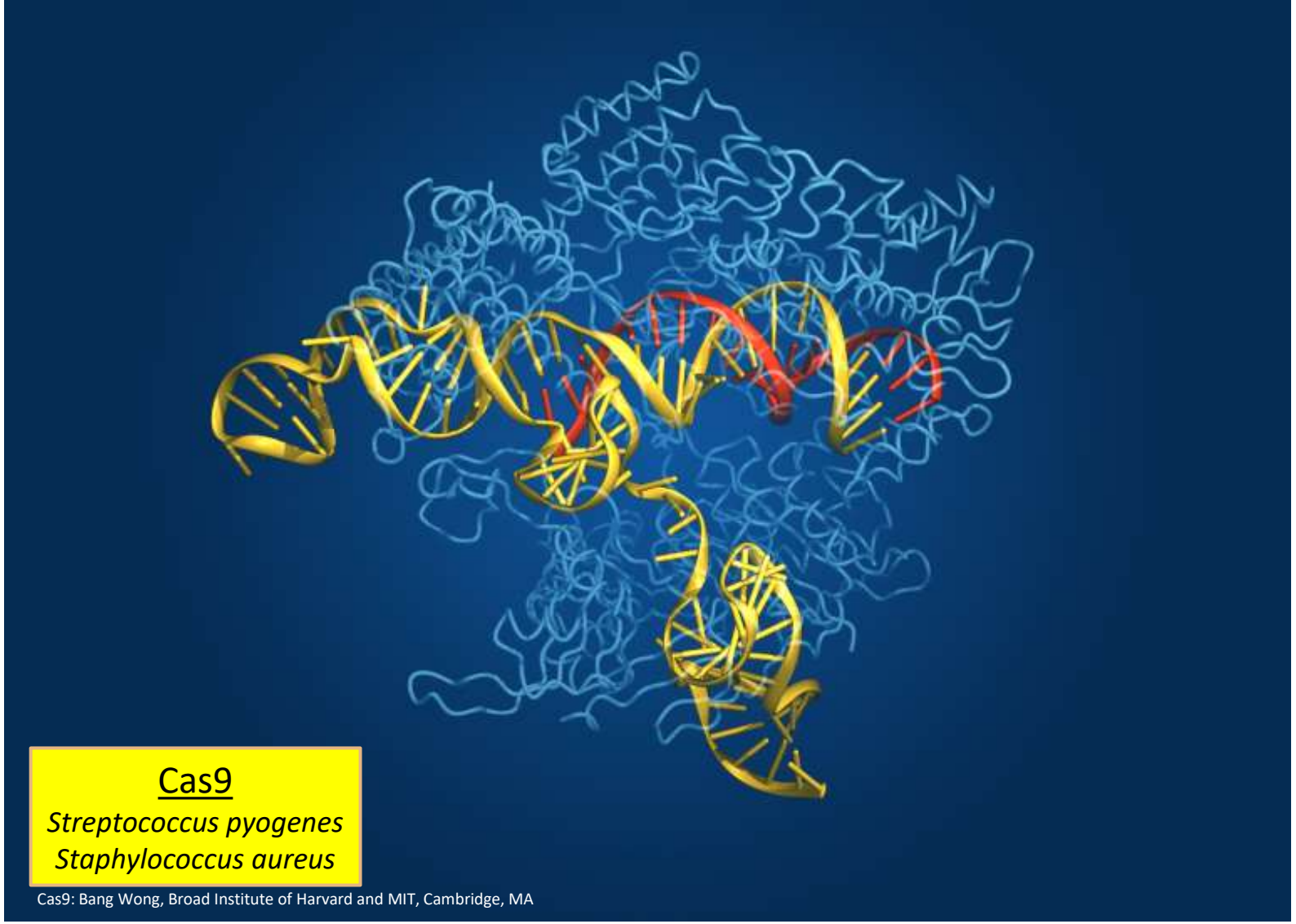
**Paddy Doherty**

## CRISPR para tratar la amiloidosis por transtiretina congénita

Gillmore et al. NEJM 2021





Cas9
*Streptococcus pyogenes*
*Staphylococcus aureus*

Cas9: Bang Wong, Broad Institute of Harvard and MIT, Cambridge, MA


Matthew Porteus **2019** ***Nature Med.***

# Las CRISPR actuales derivan de bacterias patógenas

Received: 6 February 2023 | Accepted: 12 April 2023

DOI: 10.1111/1751-7915.14266

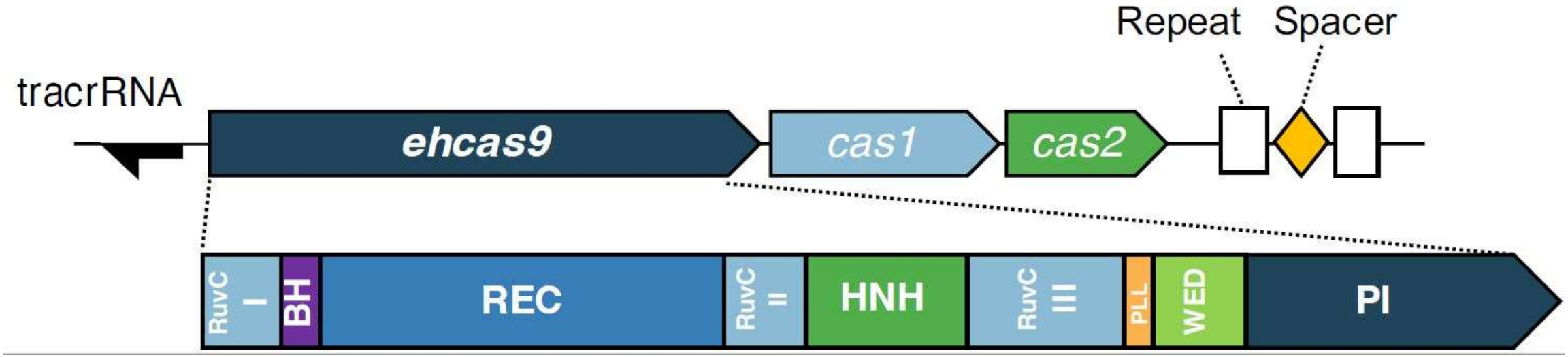
MICROBIAL BIOTECHNOLOGY Applied Microbiology International

RESEARCH ARTICLE

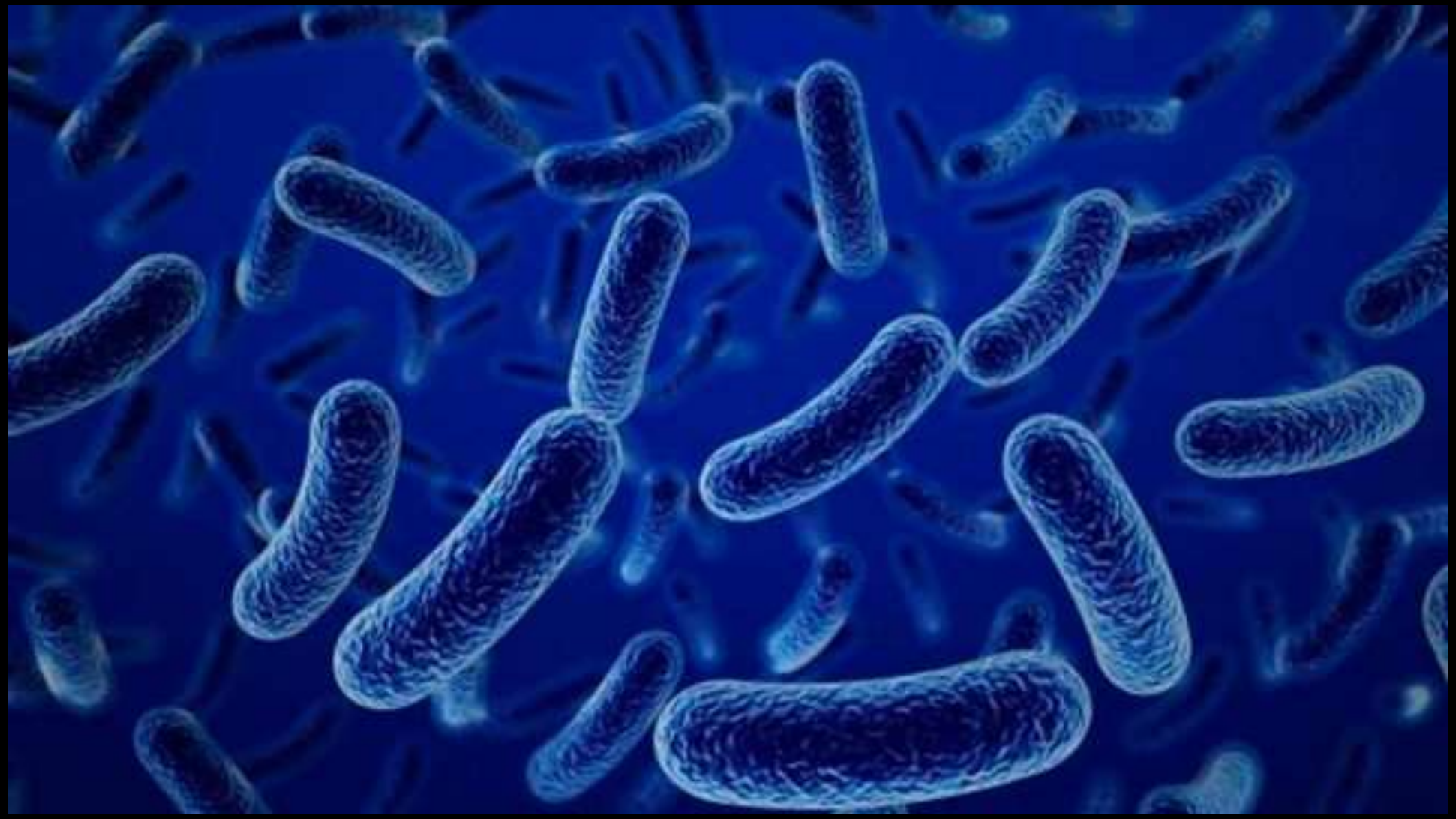
# Identification of the EH CRISPR-Cas9 system on a metagenome and its application to genome engineering

Belen Esquerra-Ruvira[1] | Ignacio Baquedano[1] | Raul Ruiz[1] |
Almudena Fernandez[2,3] | Lluis Montoliu[2,3] | Francisco J. M. Mojica[1,4]

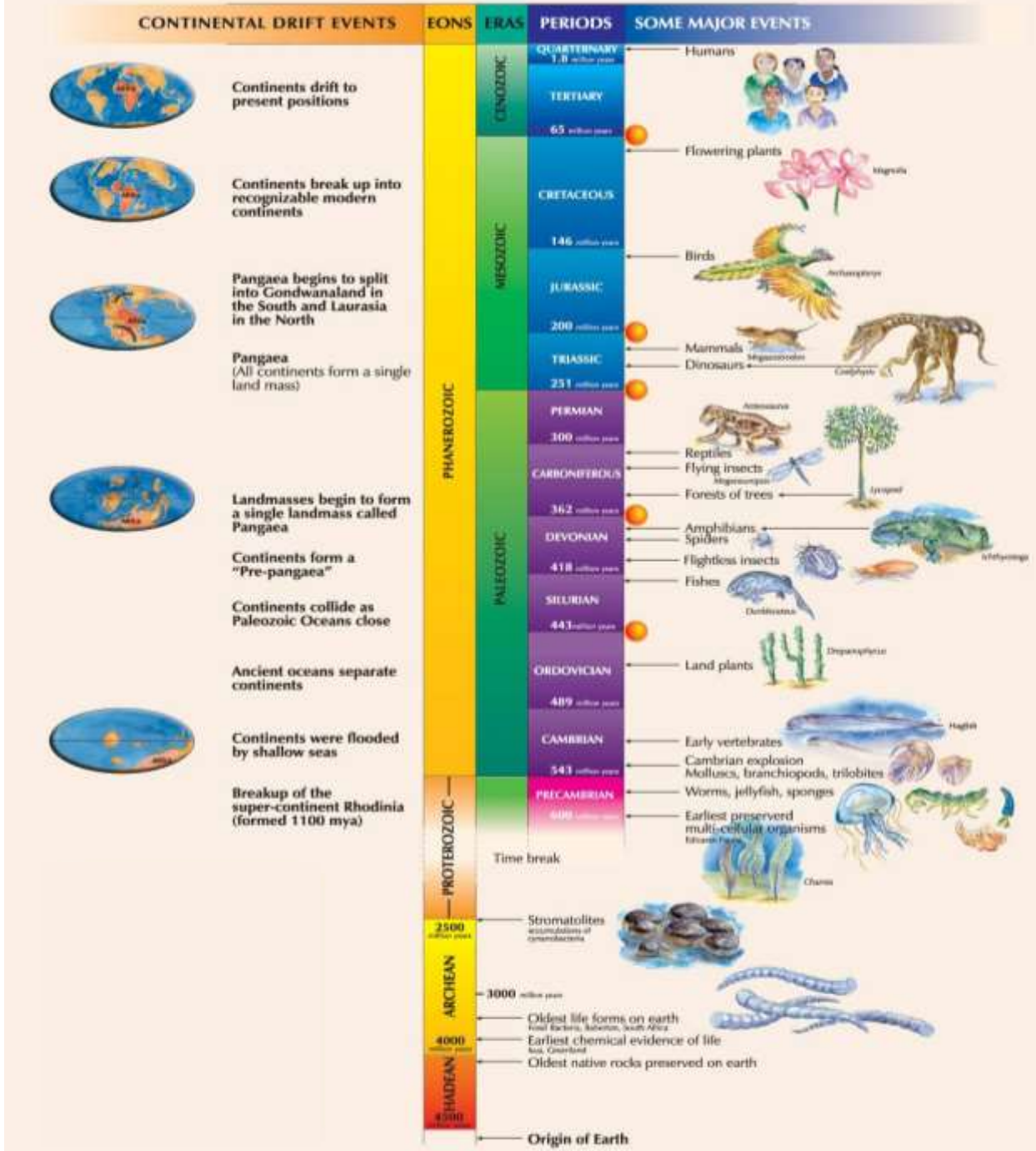
El Hondo, Elche




Esquerra-Ruvira et al. Microb. Biotech. 2023



# ¿Cómo serían las CRISPR-Cas ancestrales?




-37 PDCA

-65

-137 PCA

-150

-200 SCA

Resucitar Cas ancestrales

**Raúl Pérez- Jiménez Lab**
**CIC nanoGUNE**

-580

-1.000 BCA

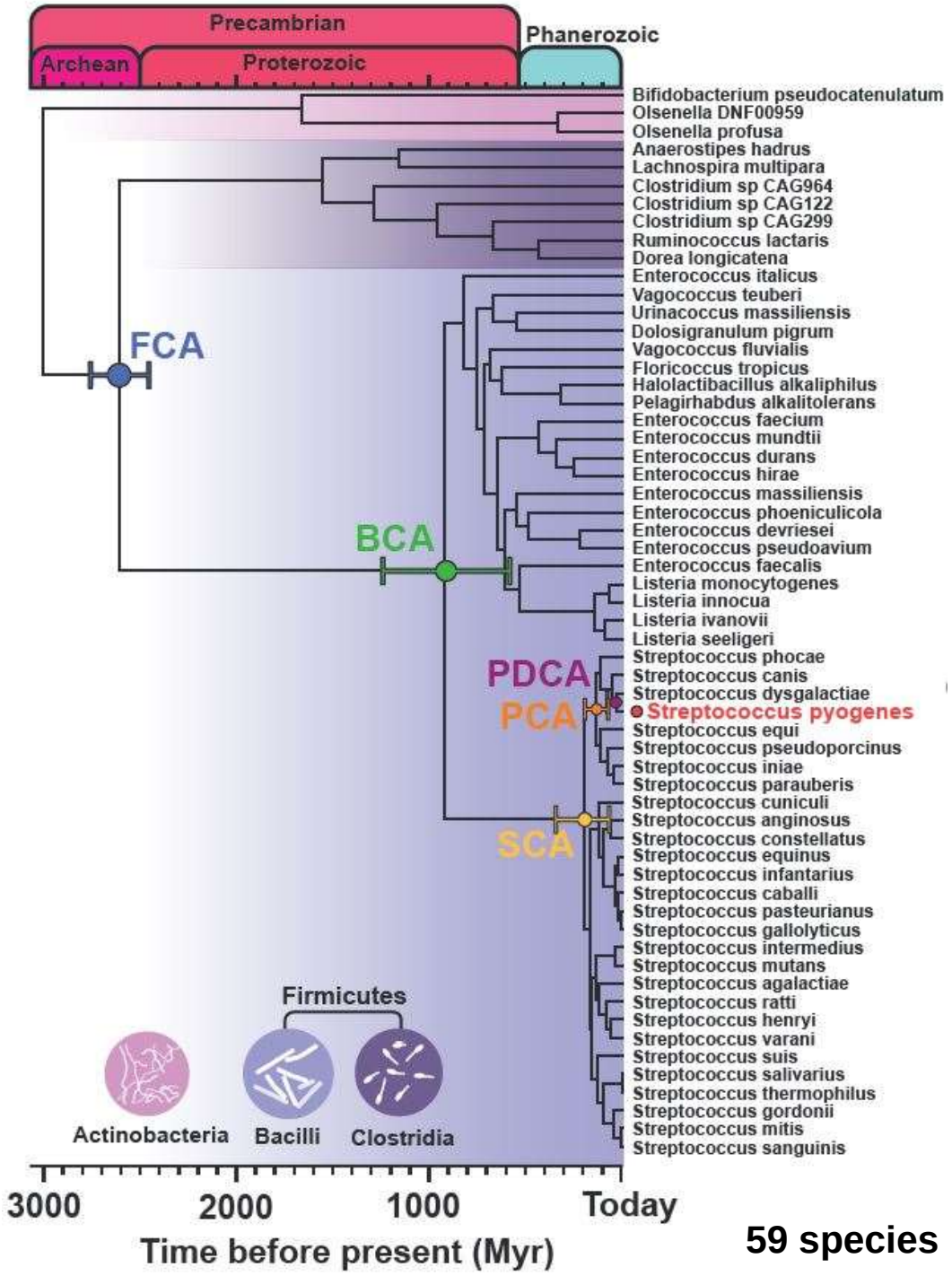
-1.850

-2.600 FCA

-3.500

-4.500

My




59 species

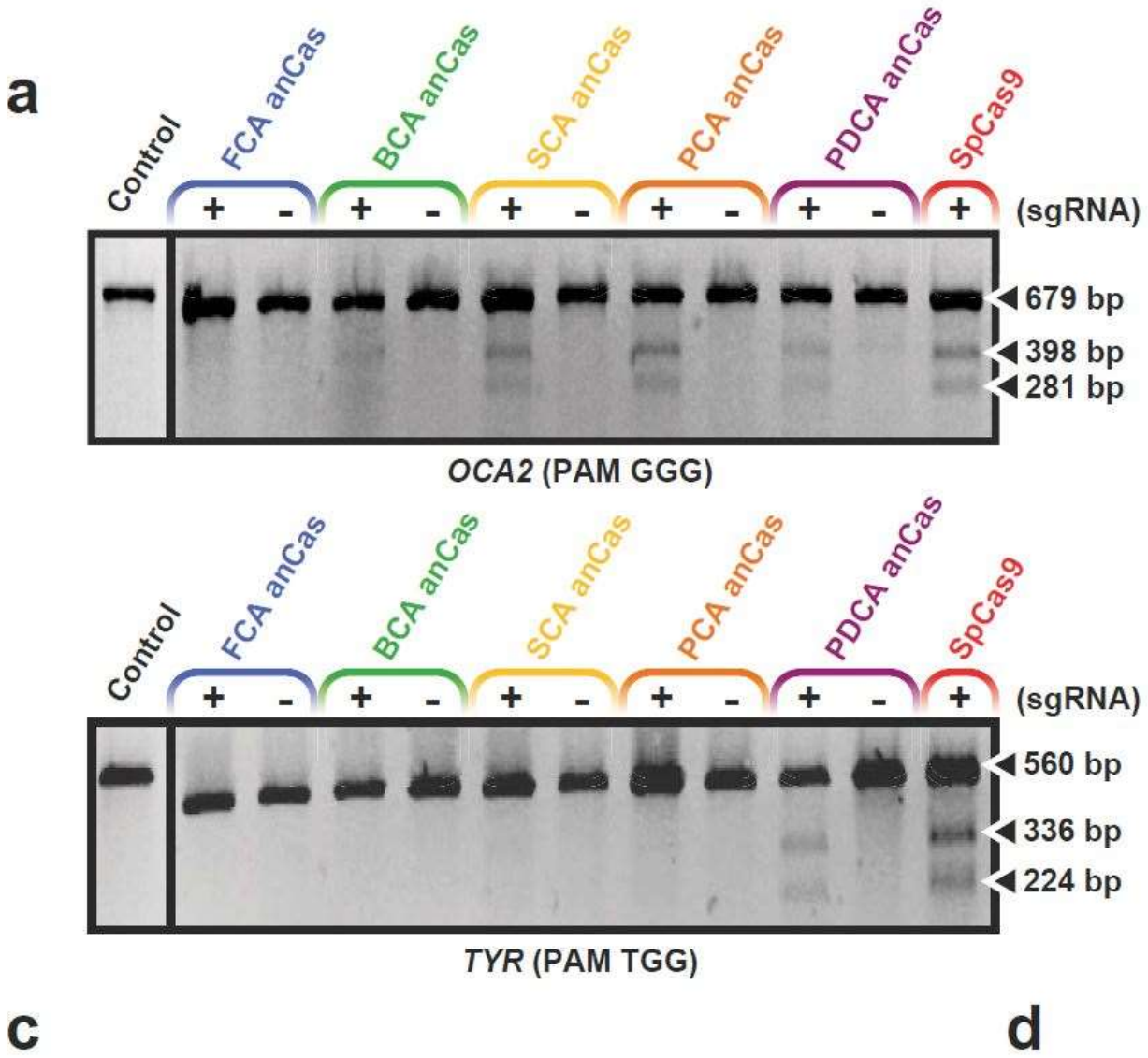
**Raúl Pérez-Jiménez Lab**
**CIC nanoGUNE**

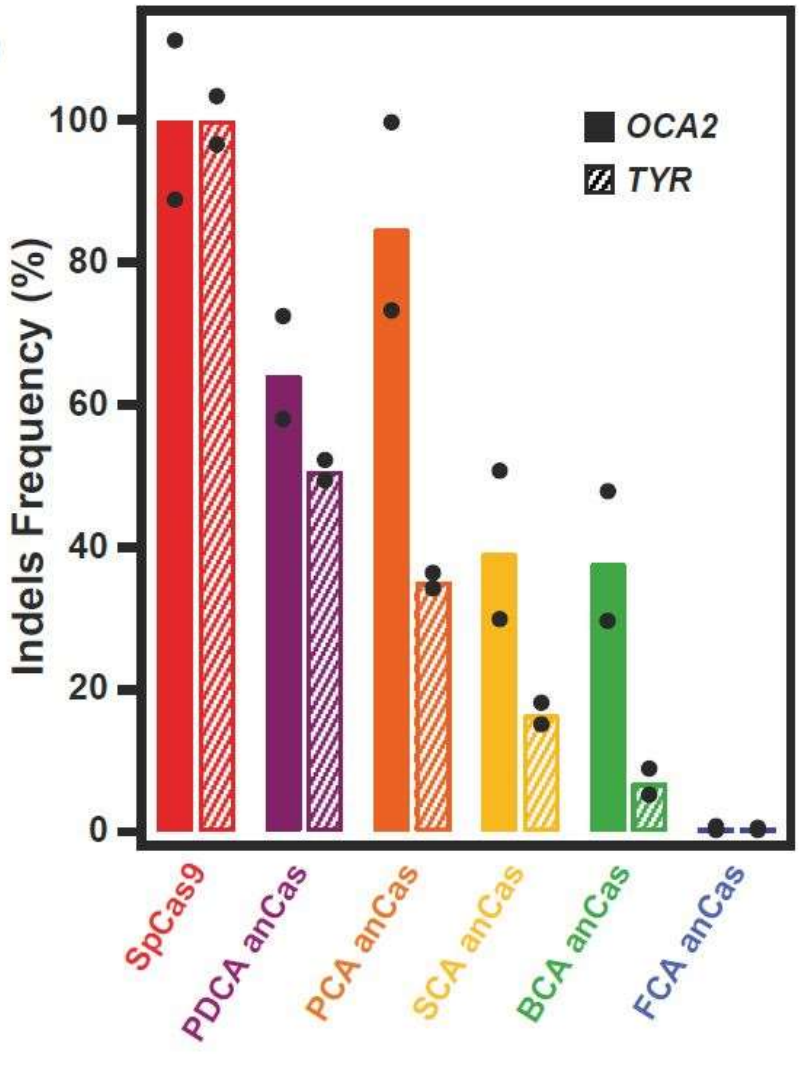
# Reconstrucción de secuencias Cas ancestrales mediante un procedimiento estadístico y superordenadores

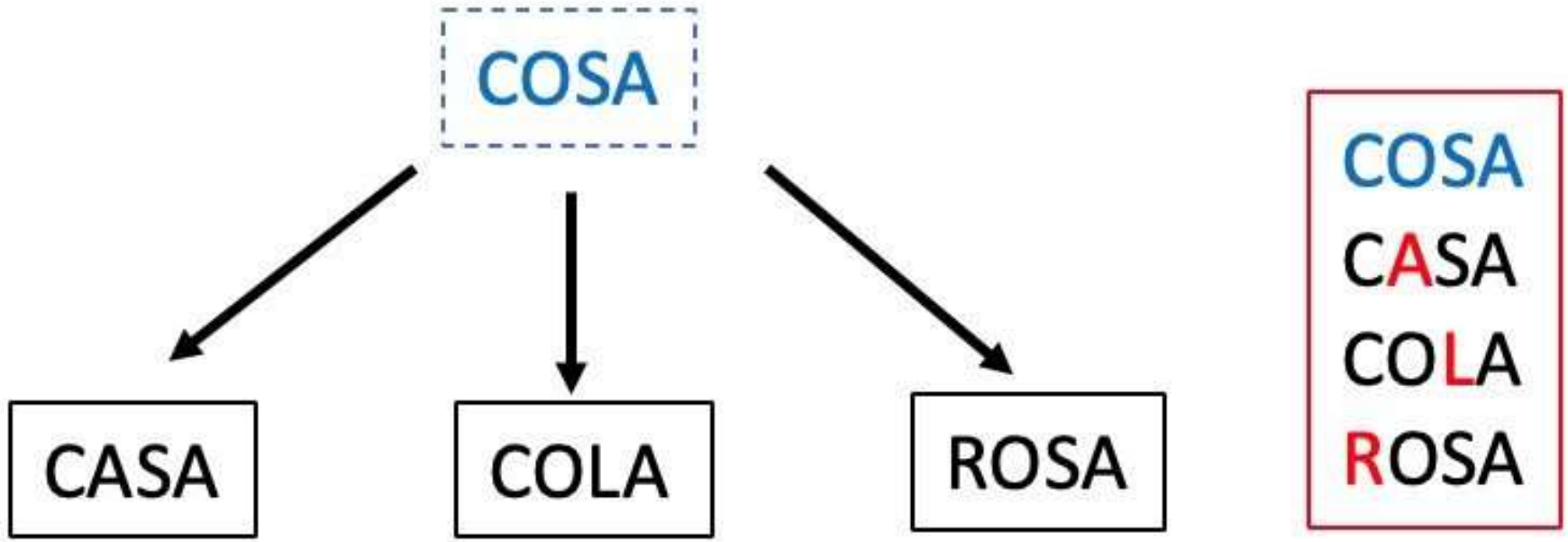
**53-96% similitud**

# Las Cas ancestrales funcionan como editores génicos en células humanas

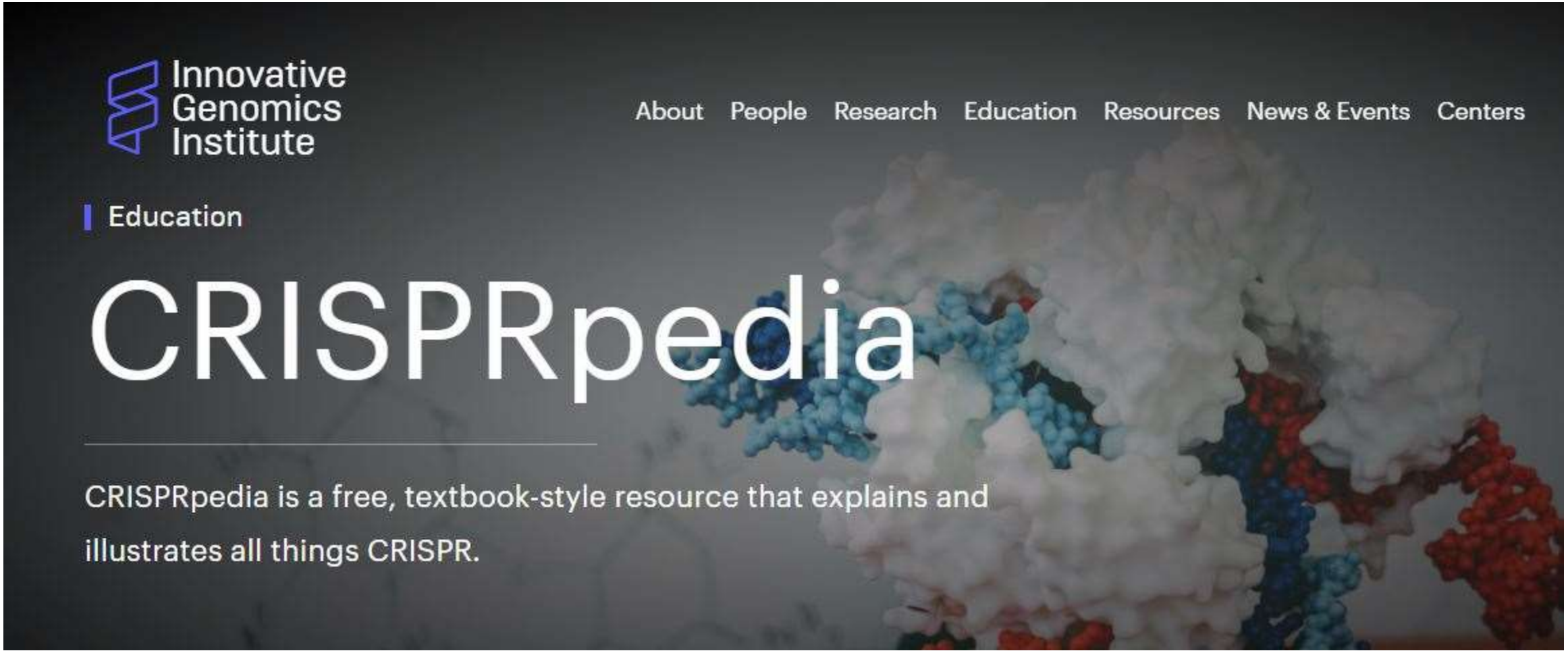
**Alonso-Lerma et al. 2023 Nature Microbiology**






Gemma Marfany (El Nacional, 2023)




**https://innovativegenomics.org/crisprpedia/**