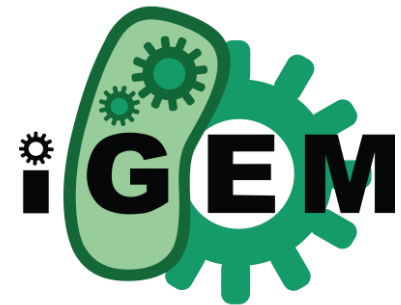


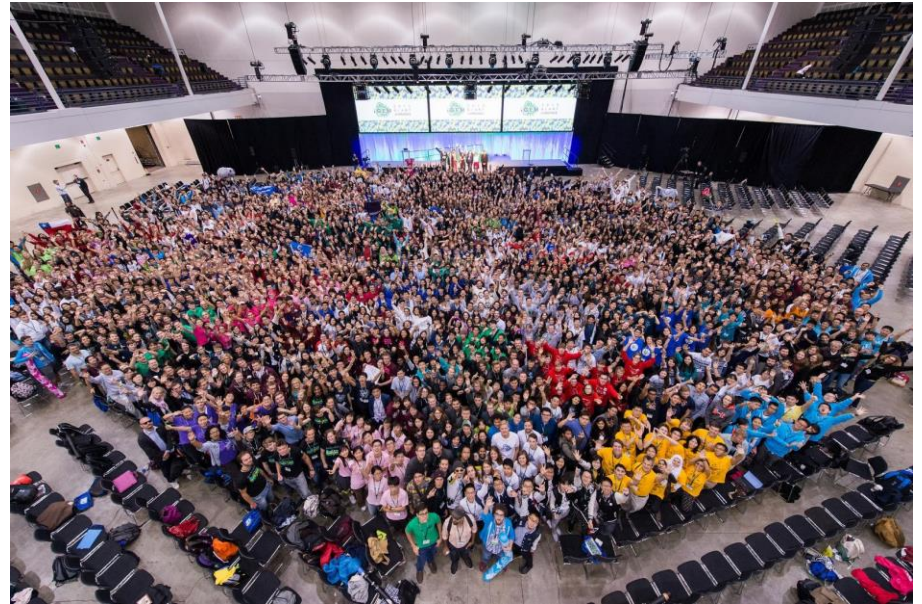
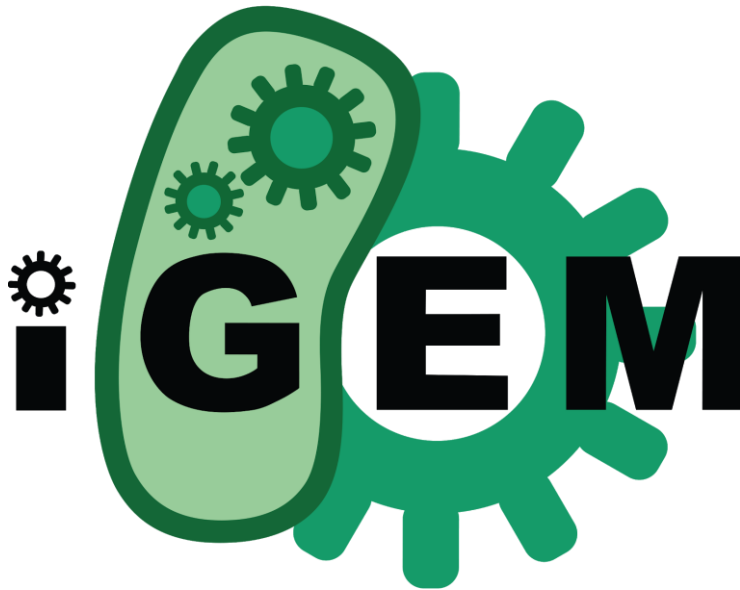
CRISPR-Cas System

Team NUS_Singapore-Sci
15th October 2018



What is iGEM

- The premiere synthetic biology competition for students all over the world.
- Present in October in **Boston, USA**



Our Team



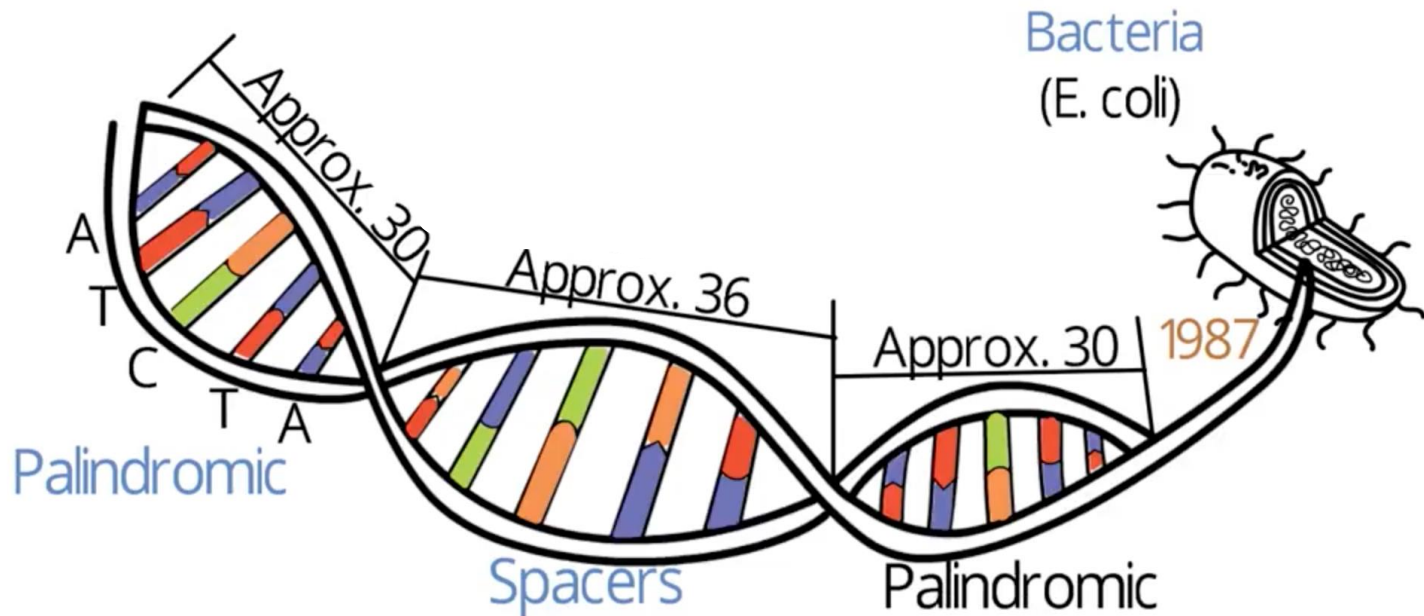
CONTENT

1. The discovery of CRISPR
2. Natural function of CRISPR
3. Application of CRISPR-Cas system
 - Gene editing and beyond
 - Case Study
 - Nucleic acid detection
 - RNA editing
4. Ethical Issues on gene editing

Discovery of CRISPR

1987

Japanese scientist discovered palindromic sequences separated by spacer in bacteria E.Coli
They did not know the function



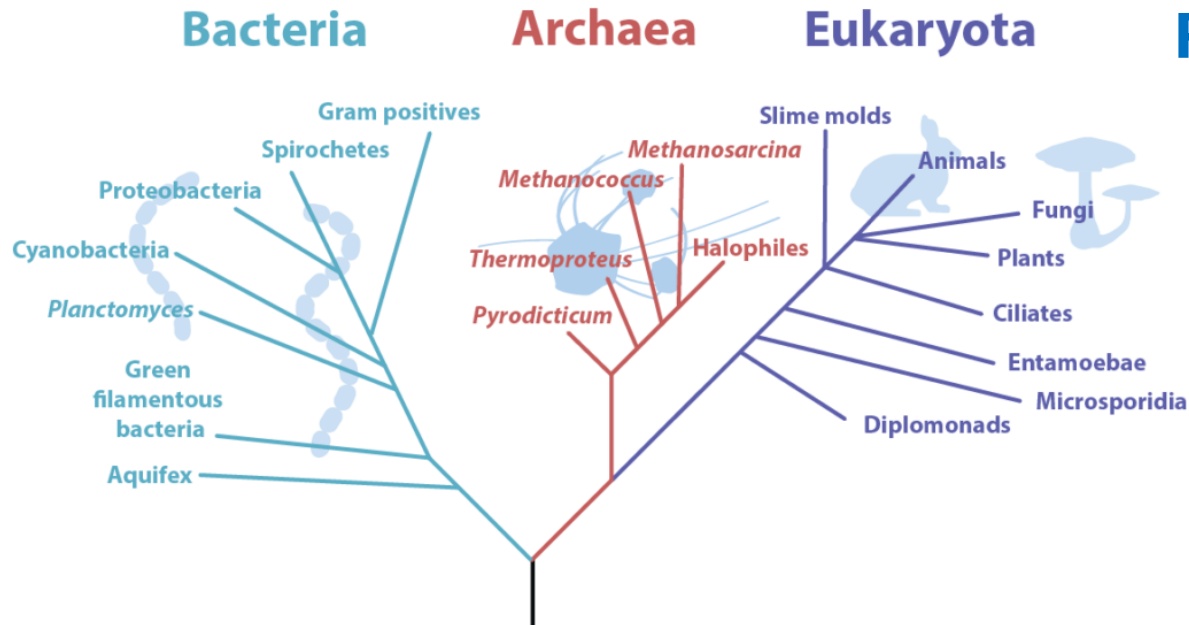
Discovery of CRISPR

1990

Fransisco Mojica discovered same palindromic sequences separated by spacer in Archaea

If both **Bacteria** and **Archaea** have..
must be something **important!**

Named
Clustered
Regular
Interspaced
Short
Palindromic
Repeats



Discovery of CRISPR

1990: Fransico Mojica

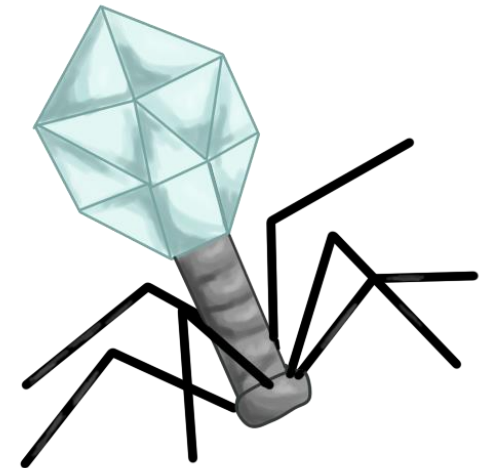
Why?
**Protect bacteria against
virus**



Archaea spacer
sequence



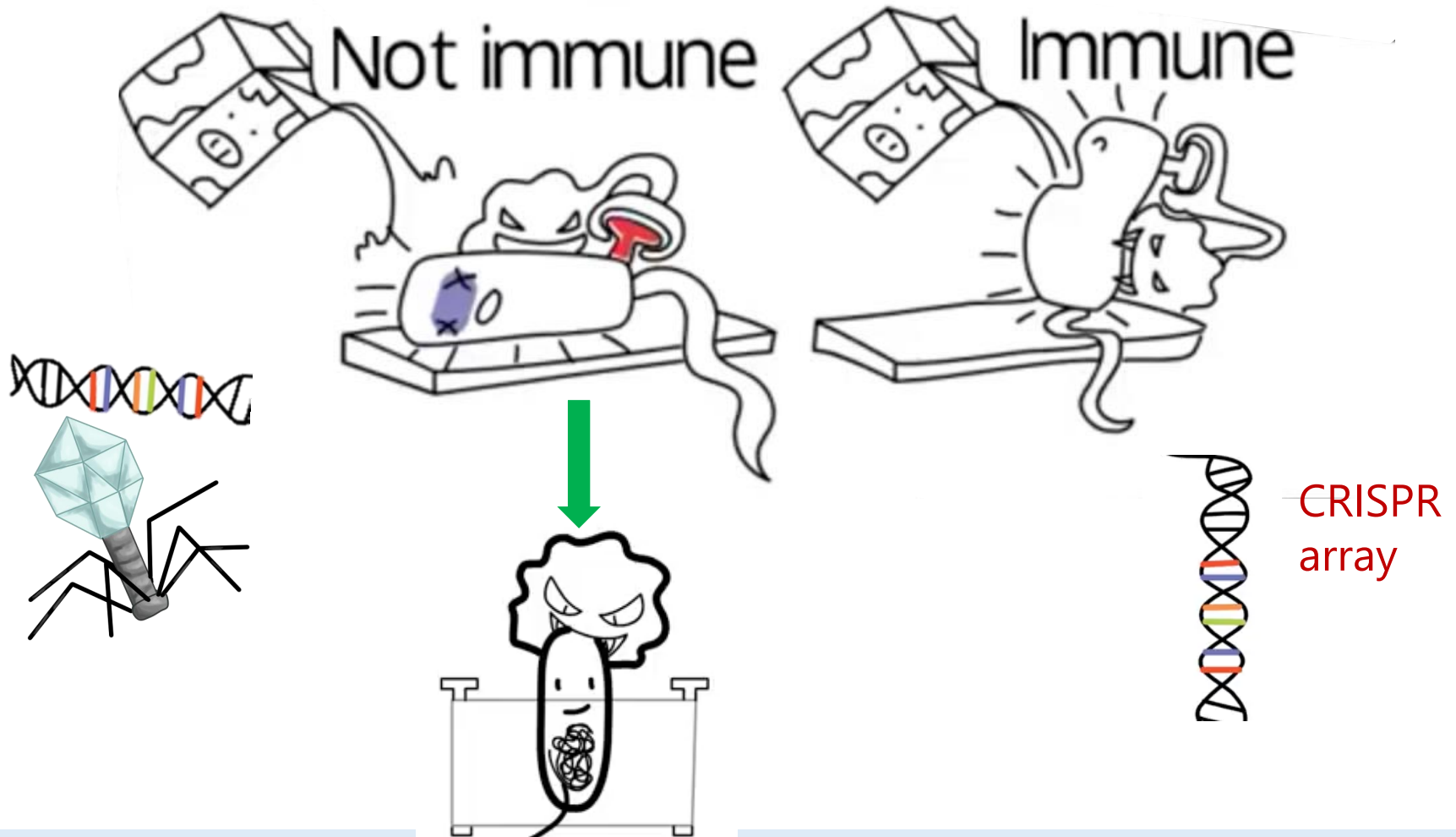
BLAST: search for
similar DNA
sequence in other
organism



Virus

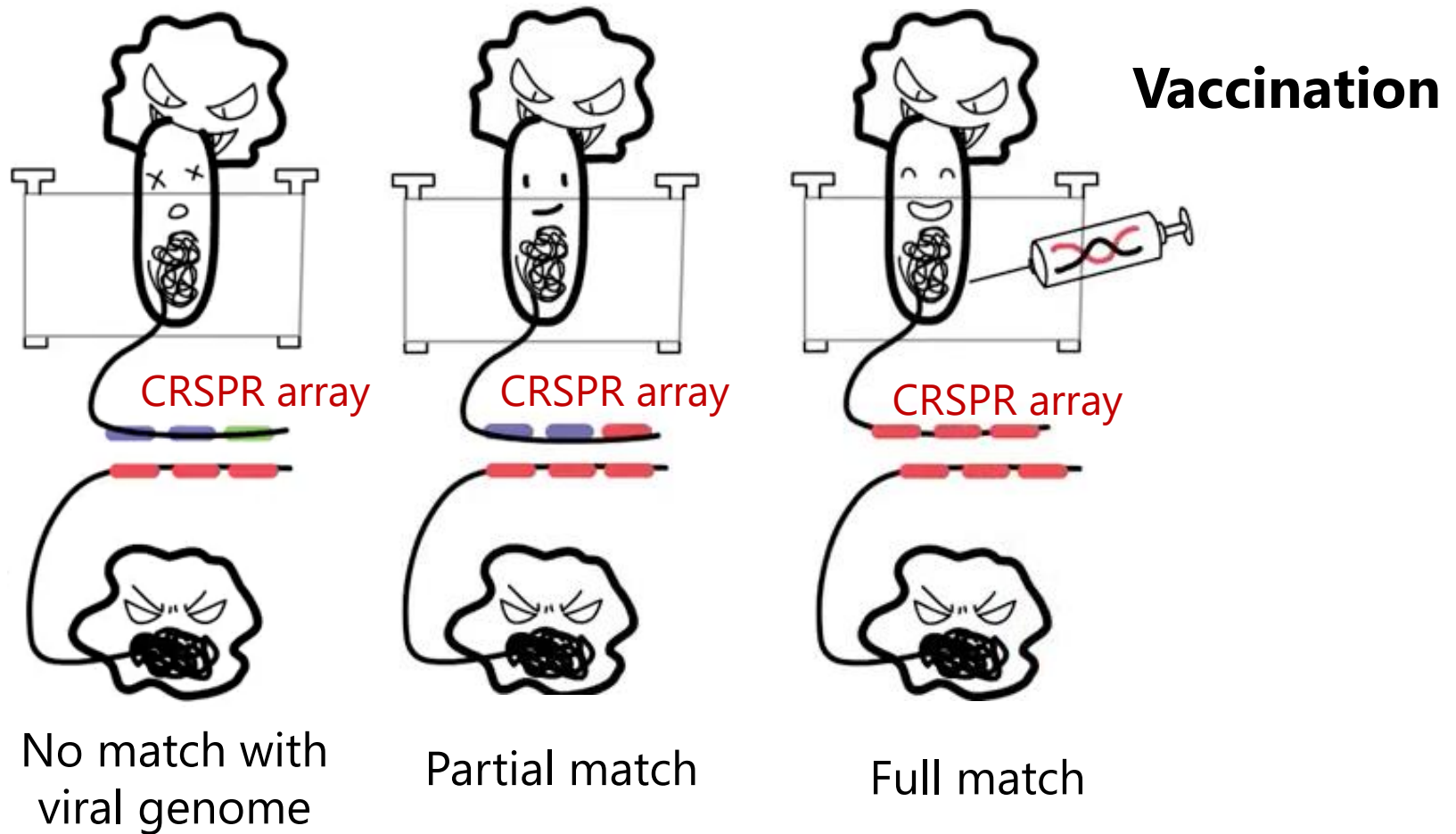
Discovery of CRISPR

2006: Discovery in yogurt factory

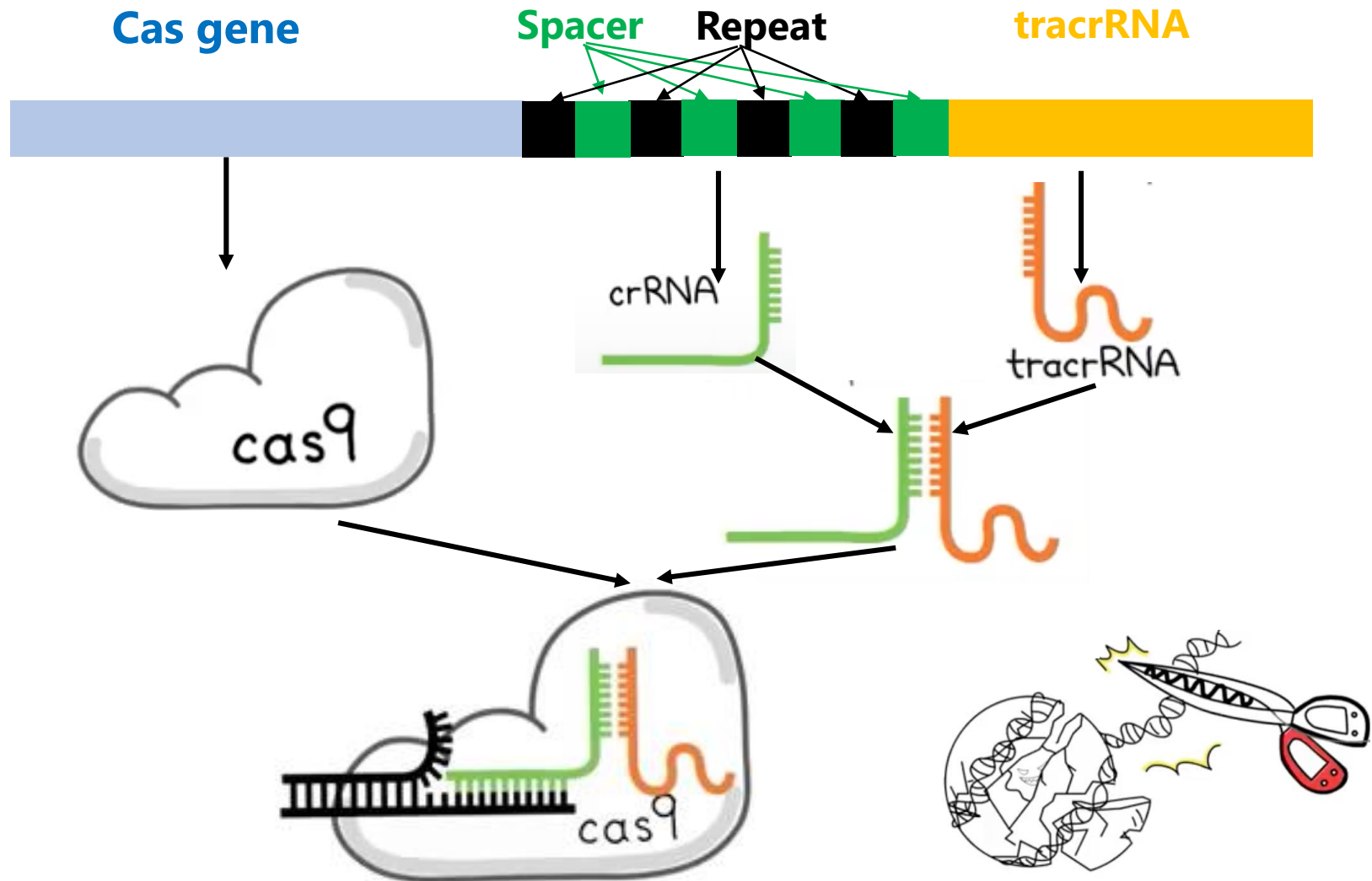


Discovery of CRISPR

2006: Discovery in yogurt factory



Discovery of CRISPR: Full CRISPR system



How does CRISPR help bacteria to defend against viral infection

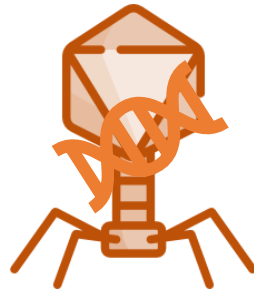
- **C**lustered **R**egularly **I**nterspaced **S**hort **P**alindromic **R**epeats



 Short palindromic repeats

How does CRISPR help bacteria to defend against viral infection

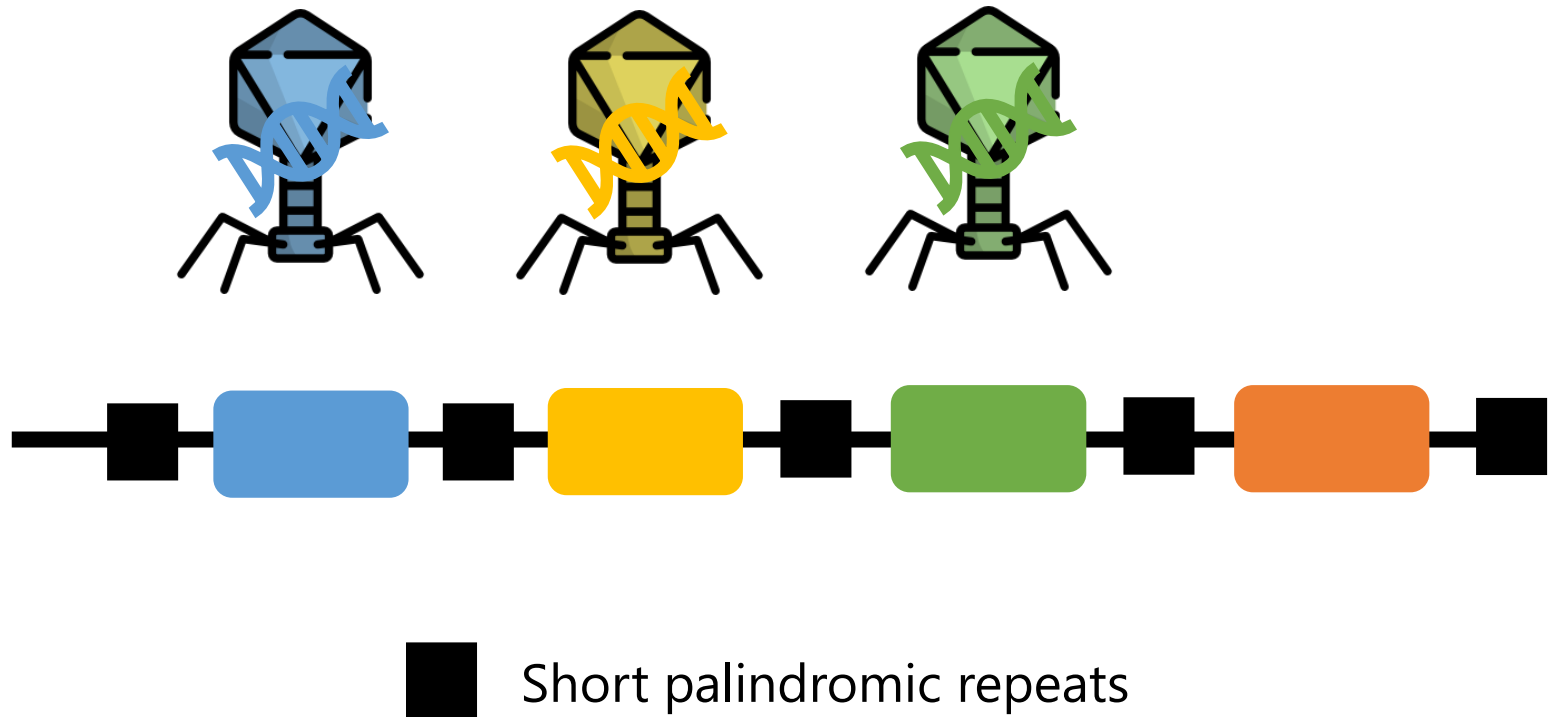
- **C**lustered **R**egularly **I**nterspaced **S**hort **P**alindromic **R**epeats



Short palindromic repeats

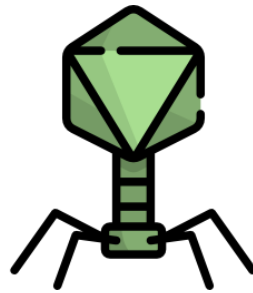
How does CRISPR help bacteria to defend against viral infection

- **C**lustered **R**egularly **I**nterspaced **S**hort **P**alindromic **R**epeats



How does CRISPR help bacteria to defend against viral infection

- **C**lustered **R**egularly **I**nterspaced **S**hort **P**alindromic **R**epeats



Short palindromic repeats

**But... how does it defend
against the viral infection?**

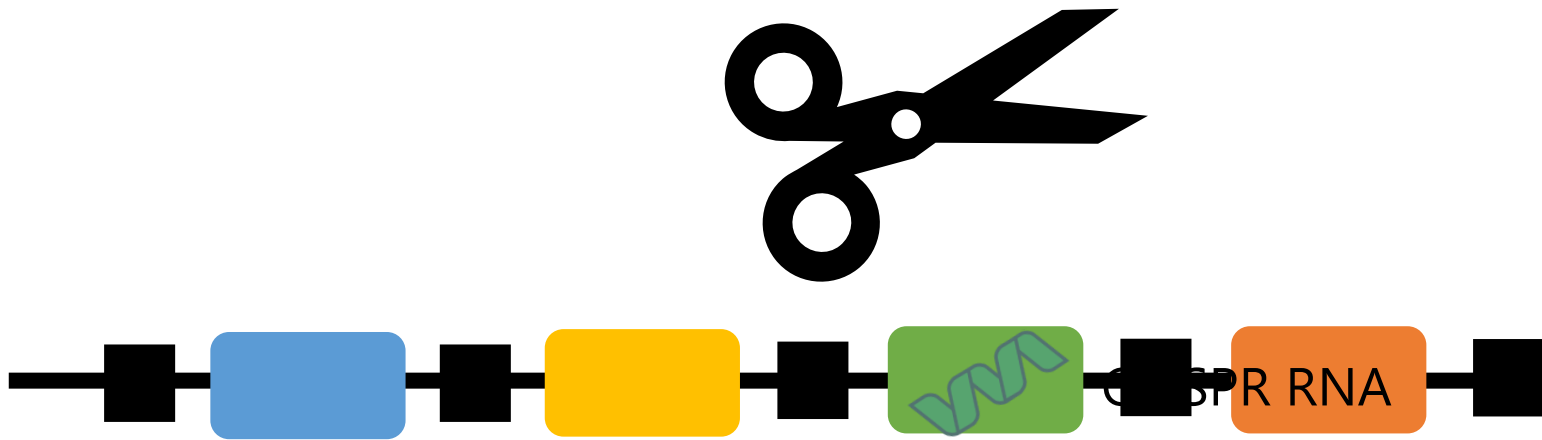
Introducing...



Cas9

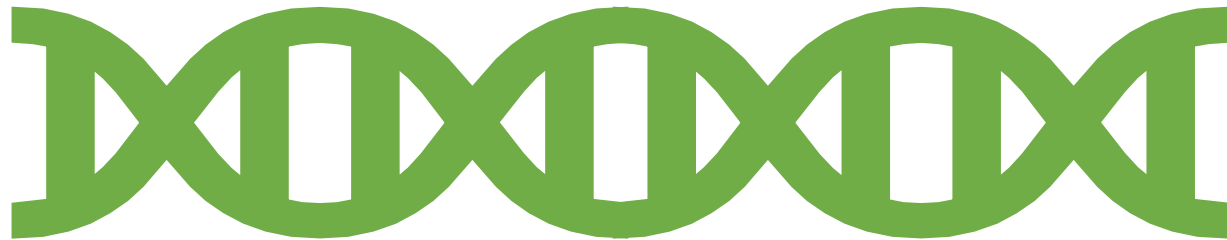
Cas9

- **CRISPR-associated protein 9**
- **RNA guided** DNA endonuclease enzyme




Cas9

- **CRISPR-associated protein 9**
- RNA guided **DNA endonuclease** enzyme



Cas9 acts as molecular scissors to cleave DNA strands

A black pair of scissors is shown cutting through a green DNA double helix. The scissors are positioned diagonally, with the blades meeting at the center of the DNA molecule, symbolizing the cleavage action of the Cas9 enzyme.

What is dCas9?

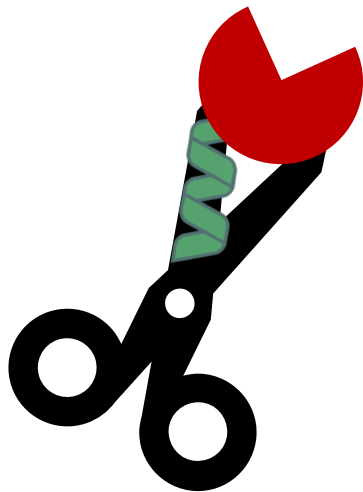
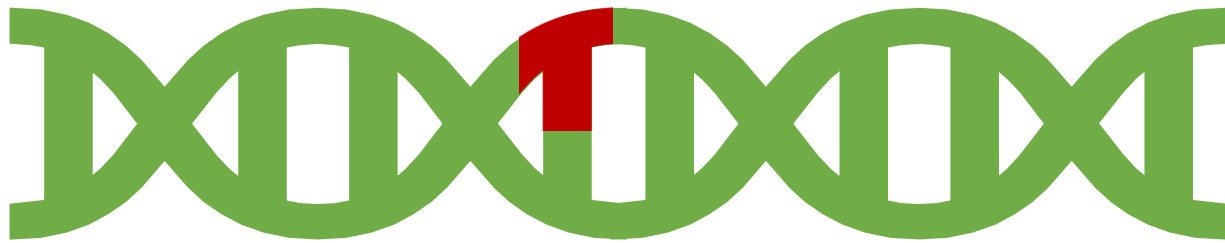
dCas9



What about dCas?

[CRISPR-Cas genome editing and Beyond?](#) (1:37)

Changing CRISPR/dCas for Base Editing



Applications of CRISPR/Cas System and its Potential

Genetic Engineering Will Change Everything Forever –
CRISPR (6:35-7:53)

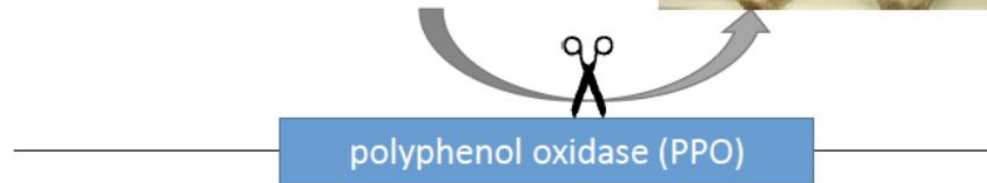
Gene Editing Misconceptions and Ethical Concerns

CRISPR not consider as GMO?

NATURE | NEWS



Gene-edited CRISPR mushroom escapes US regulation



- Polyphenol oxidase (PPO) causes browning of mushrooms during storage
- CRISPR to introduce mutations to 1 out of 6 *PPO* genes
- 30% reduced activity
- Prolonged storage time

Waltz, Nature 2016



Gene-edited CRISPR mushroom escapes US regulation



- US Department of Agriculture (USDA) will not regulate a CRISPR modified mushroom (*"No foreign DNA present"*)
- Cultivated and sold without passing through the agency's regulatory process
- First CRISPR-edited organism to be approved

Kahoot time!

**Please take out your phones!
go to kahoot.it**

Common misconceptions

#1 MISCONCEPTION :

Gene functions do not affect each other and changes are predictable

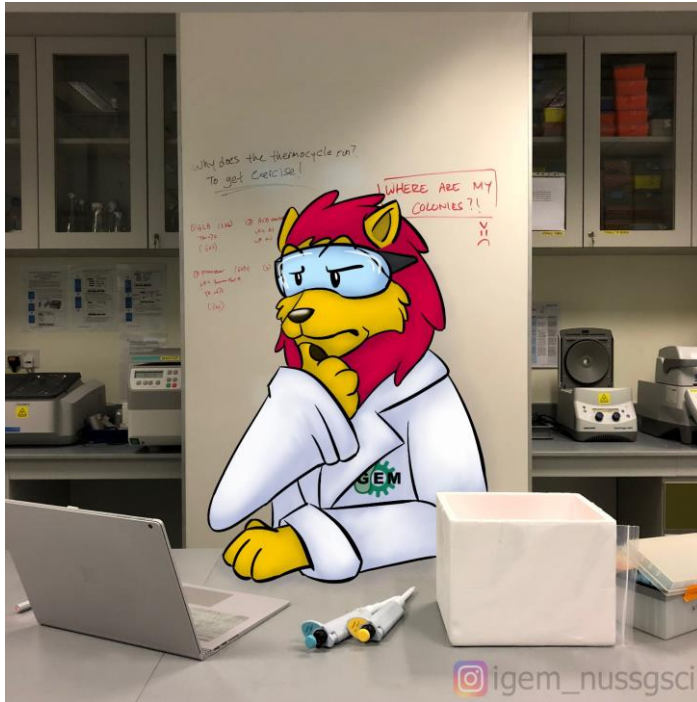
#2 MISCONCEPTION :

Gene editing can make changes to all DNA in the cell.

#3 MISCONCEPTION :

DNA bases in a gene cannot be changed or altered in our body.

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