

# Immunity against SARS-CoV-2

## VACCINES candidates

### Lecture 7

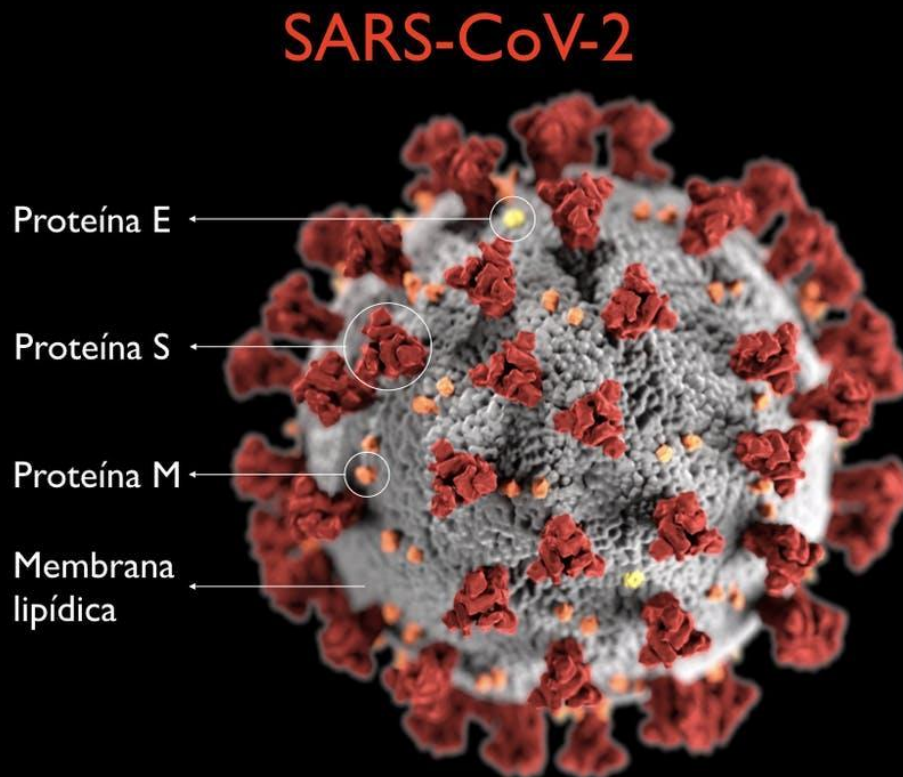
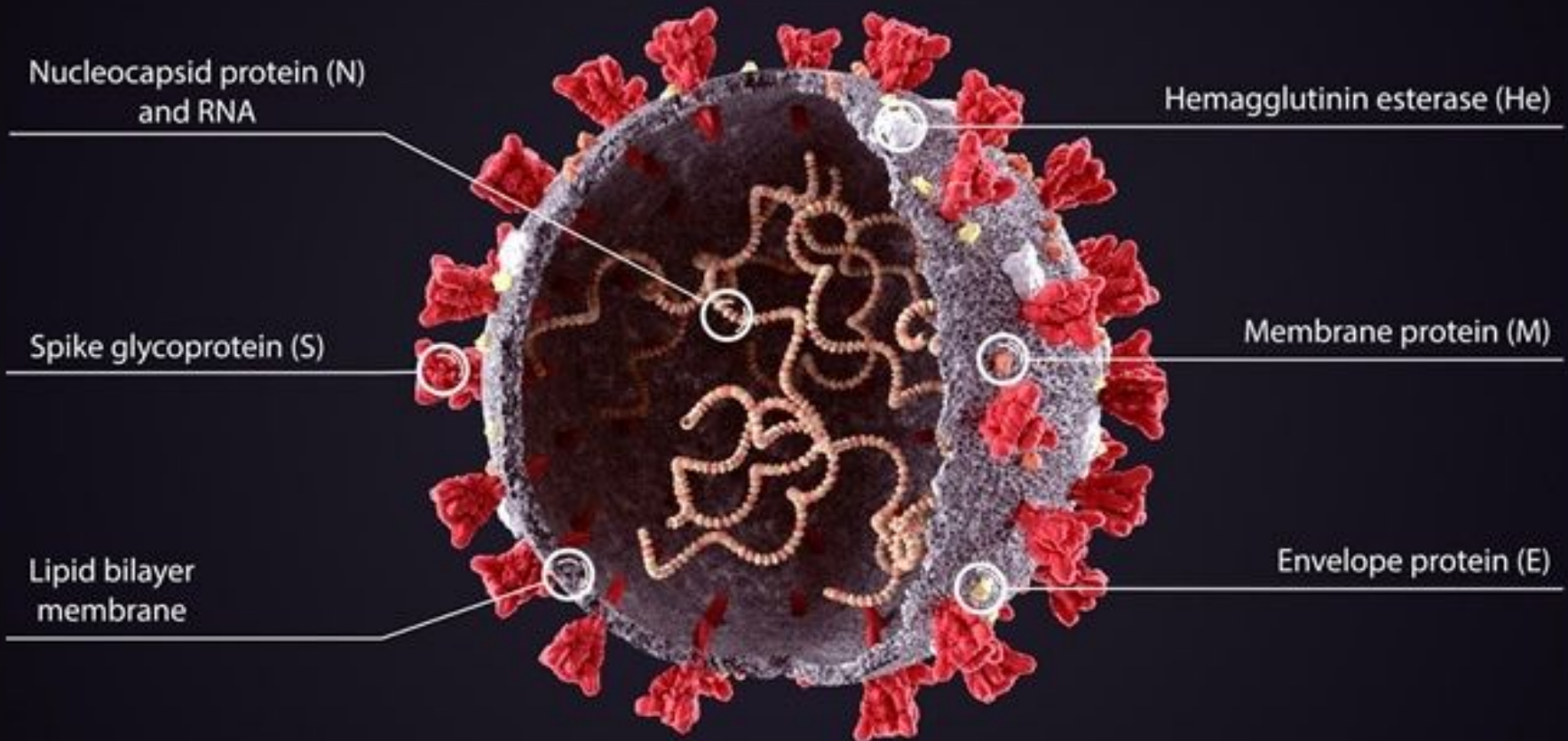


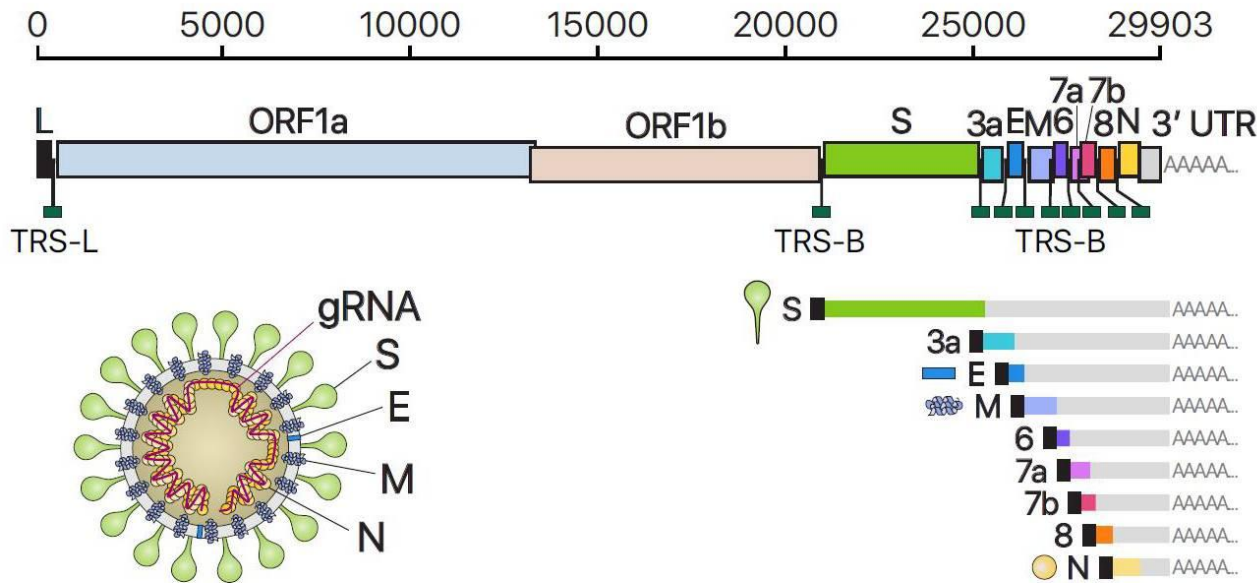
Ilustración adaptada de *Centers for Disease Control and Prevention, USA*

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2020

# SARS-CoV-2 structure (Type IV according to Baltimore classification)



# SARS-CoV-2 genome



Genome: single stranded RNA messenger 29.9kb long, encoding 13 ORFs.

Coronavirus genomes have the longest RNA virus genome known..

The proteins are expressed by two ways: primary translation of polyprotein that initiates the infection, and after some replication, subgenomic mRNA expression which produces all structural proteins

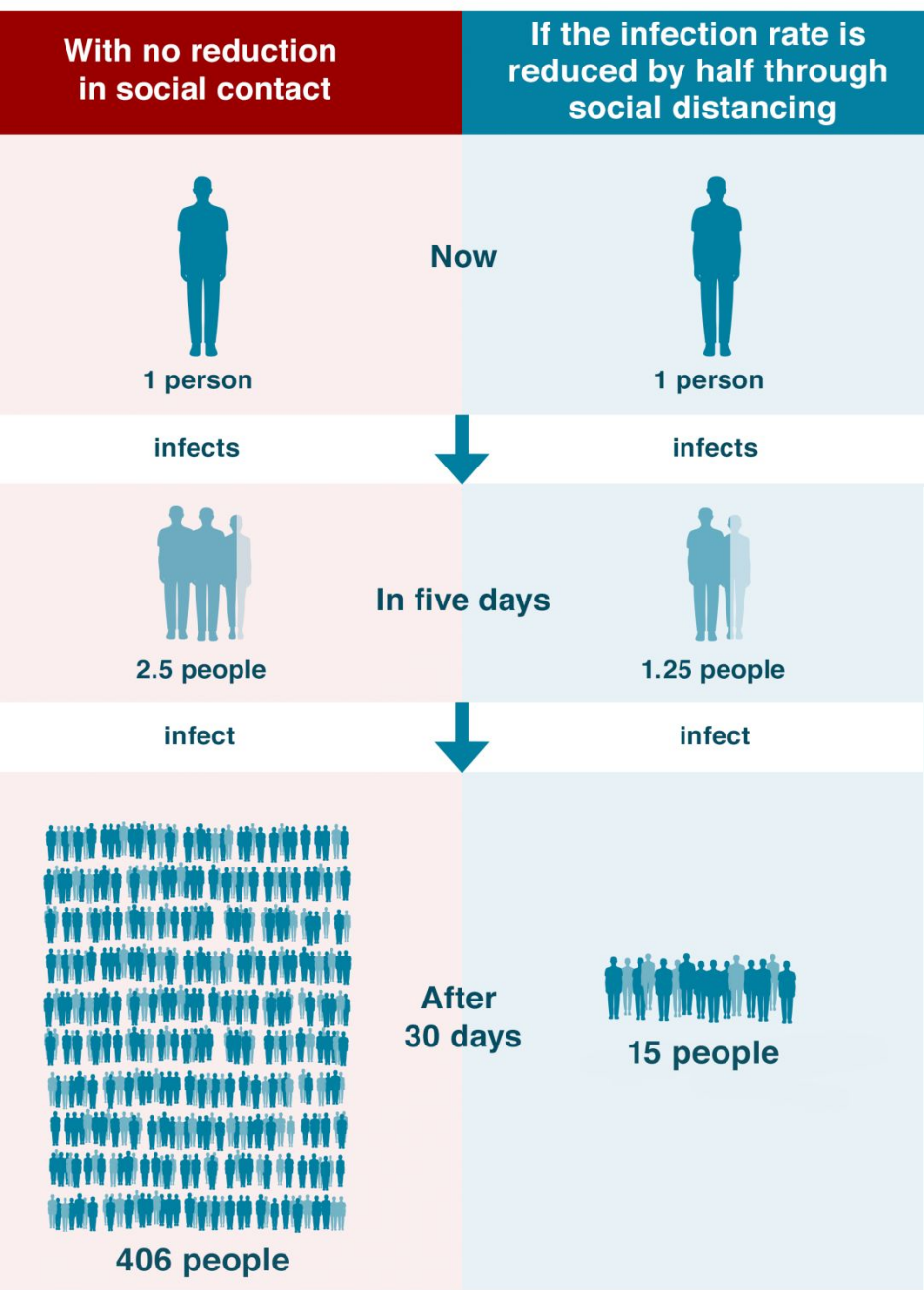
# **SARS-CoV-2 causes an infectious causes COVID-19**

- Most people infected with the COVID-19 virus will experience mild to moderate respiratory illness and recover without requiring special treatment. Older people, some children and those with underlying medical problems like cardiovascular disease, diabetes, chronic respiratory disease, and cancer are more likely to develop serious illness.

# There are basically three ways to stop the Covid-19 disease

1. Extraordinary restrictions on free movement and assembly, as well as aggressive testing, to interrupt its transmission entirely;
2. Just wait until enough people get infected and develop NATURAL ACQUIRED ACTIVE immunity (herd immunity);
3. A vaccine that could protect everyone by developing ARTIFICIAL ACQUIRED ACTIVE immunity (also herd immunity)

# Why everyone should be social distancing



# 1. Restrictions measures interrupt transmission entirely (immunity is not developed)

## CORONAVIRUS PREVENTION





2. **Get infected** - another way for a herd immunity, aside from vaccines. Some die, and the rest develop antibodies and/or cell-mediated immunity.


There have been two killer coronaviruses before:

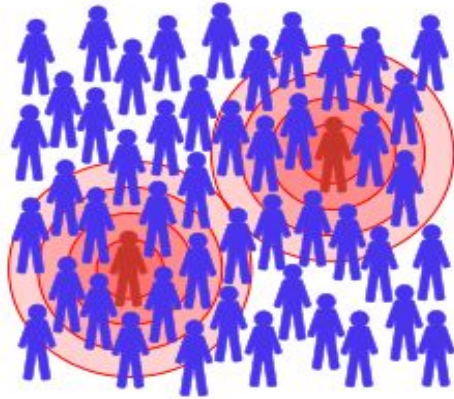
- **SARS-CoV** infected only 8,000 people, killing 774 (about 10%), and was contained in 7½ months.
- **MERS** has never stopped but is rare. Since arising in 2012 it's infected 2,519 people, killing 35% of them (866 deaths so far).

A novel **SARS-CoV-2** by now infects more than 5,6 mln people, kills more than 351,000 of them, and is not going to stop.

 = not immunized, but still healthy

 = immunized and healthy

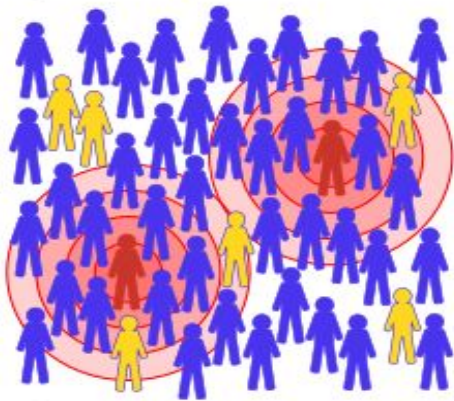
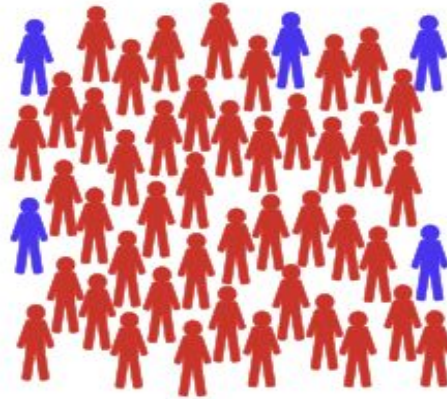
 = not immunized, sick, and contagious



No one is immunized.



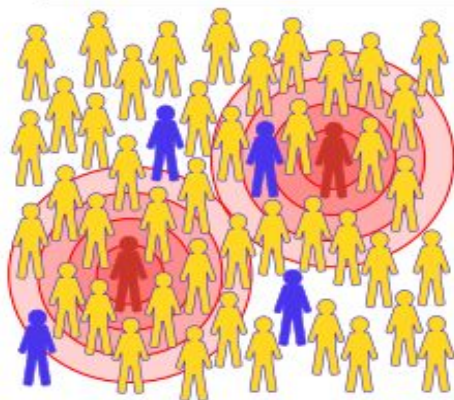
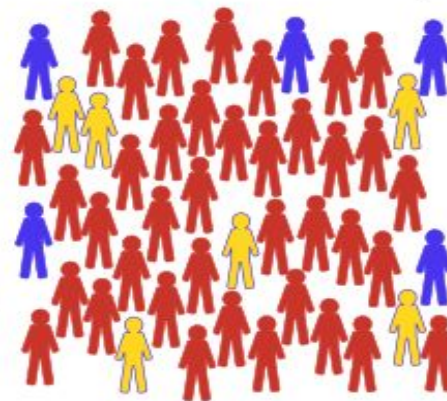
Contagious disease spreads through the population.



Some of the population gets immunized.



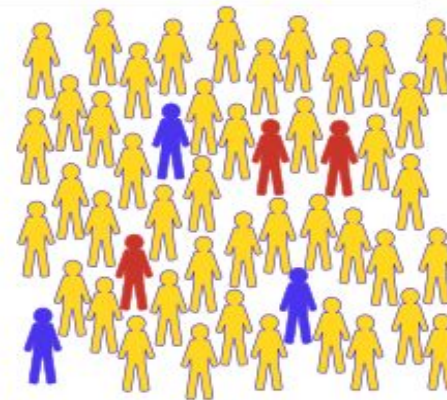
Contagious disease spreads through some of the population



Most of the population gets immunized.



Spread of contagious disease is contained.



**Herd immunity** is an epidemiological concept that describes the state where a population is sufficiently immune to a disease that the infection will not spread within that group.



## 2. How many people should be infected to develop herd immunity?

- For mumps, 92 percent of the population should be immune for the disease to stop spreading entirely. This is what's known as the herd immunity threshold.
- COVID-19 is less infectious than mumps, with the proportion of people who need to be infected is lower but still high, sitting at around 70 percent of the entire population.

And what happens if 70 percent of an entire population gets sick, and due to fatality rate around 0,5-1% , how many of them will die?

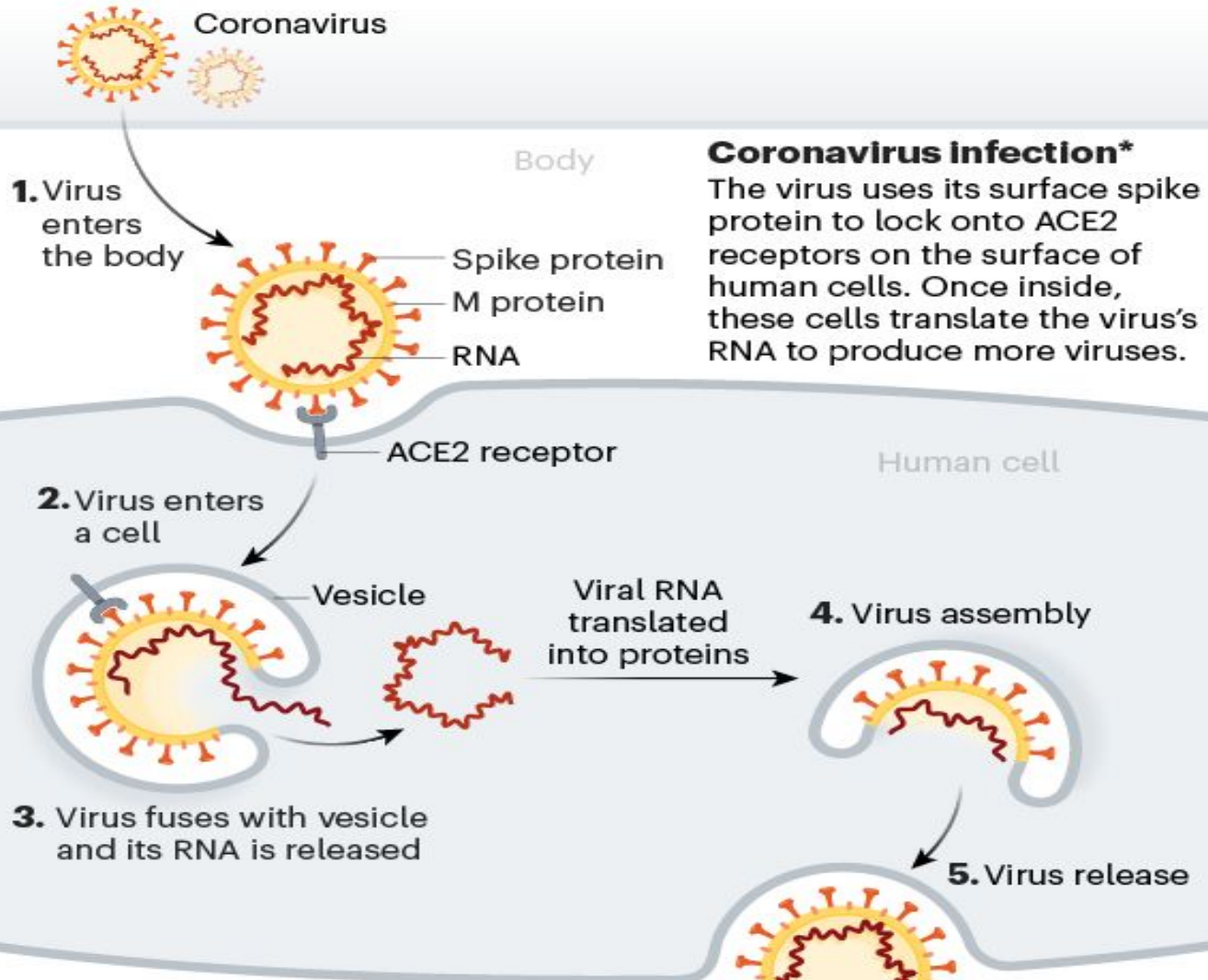
It is a catastrophic outcome, and is a nonsense, but not preventive measure.

- .

# 3. Vaccination-the only way for the herd immunity.

## VACCINE BASICS: HOW WE DEVELOP IMMUNITY

The body's adaptive immune system can learn to recognize new, invading pathogens, such as the coronavirus SARS-CoV-2.



How we develop immunity against virus

3. Virus fuses with vesicle and its RNA is released

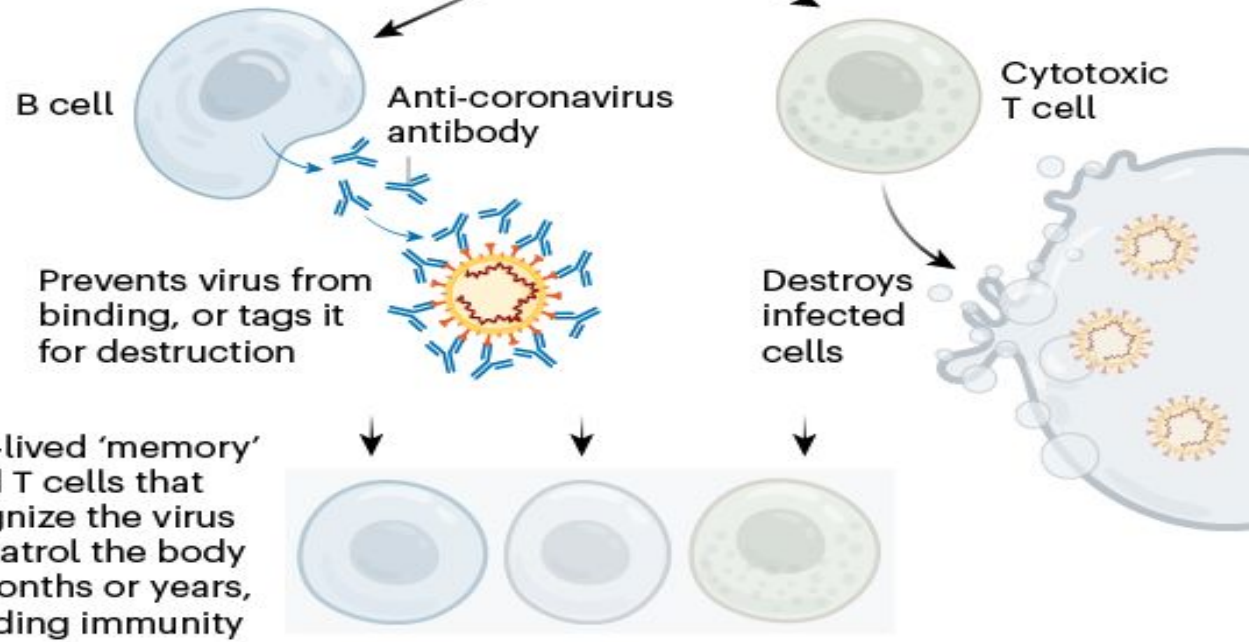
5. Virus release

### Immune response\*

Specialized 'antigen presenting cells' (APCs) engulf the virus and display portions of it to activate T-helper cells.

T-helper cells enable other immune responses:  
B cells make antibodies that can block the virus from infecting cells, as well as mark the virus for destruction.  
Cytotoxic T cells identify and destroy virus-infected cells.

# How we develop immunity against virus

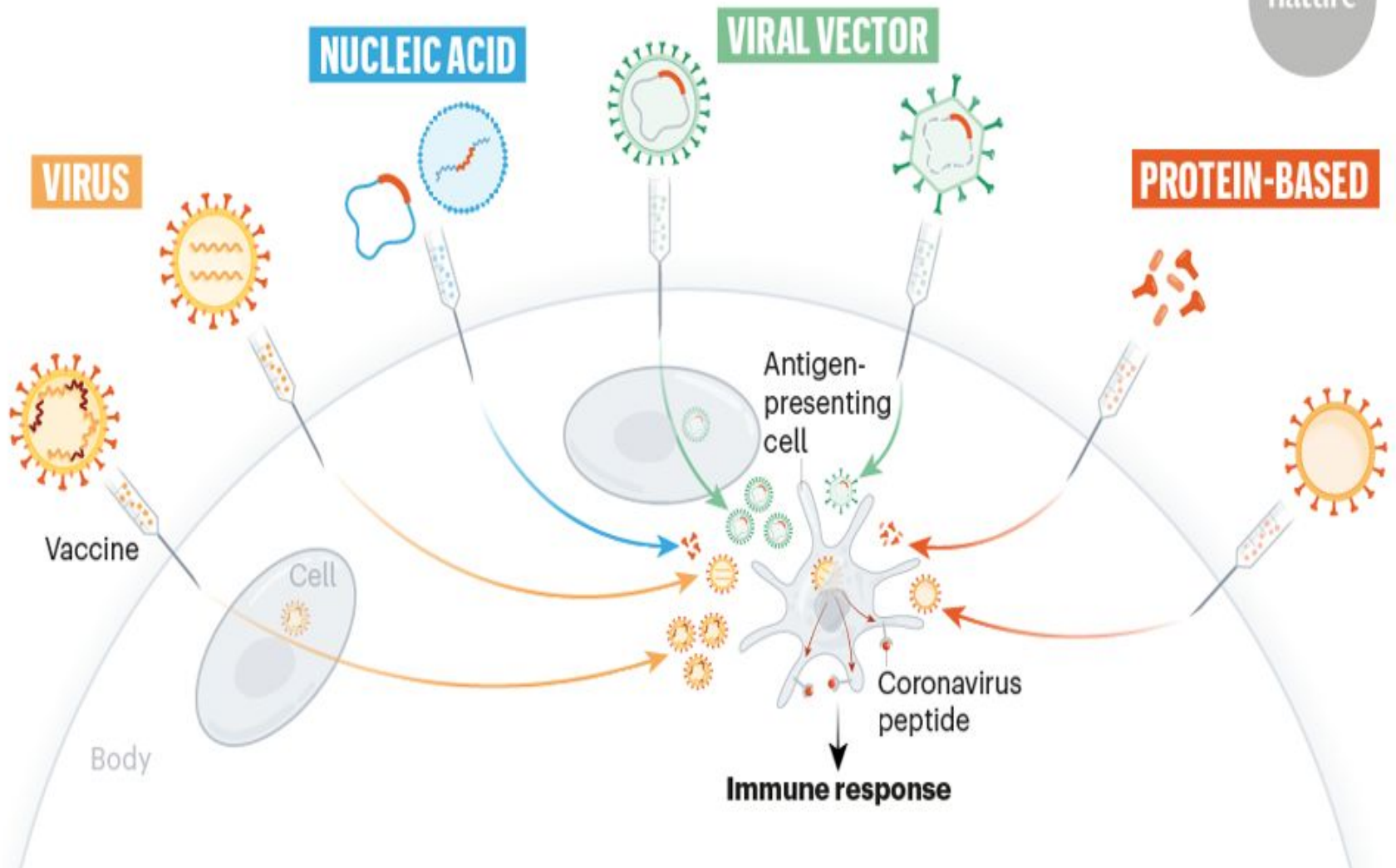


Long-lived 'memory' B and T cells that recognize the virus can patrol the body for months or years, providing immunity

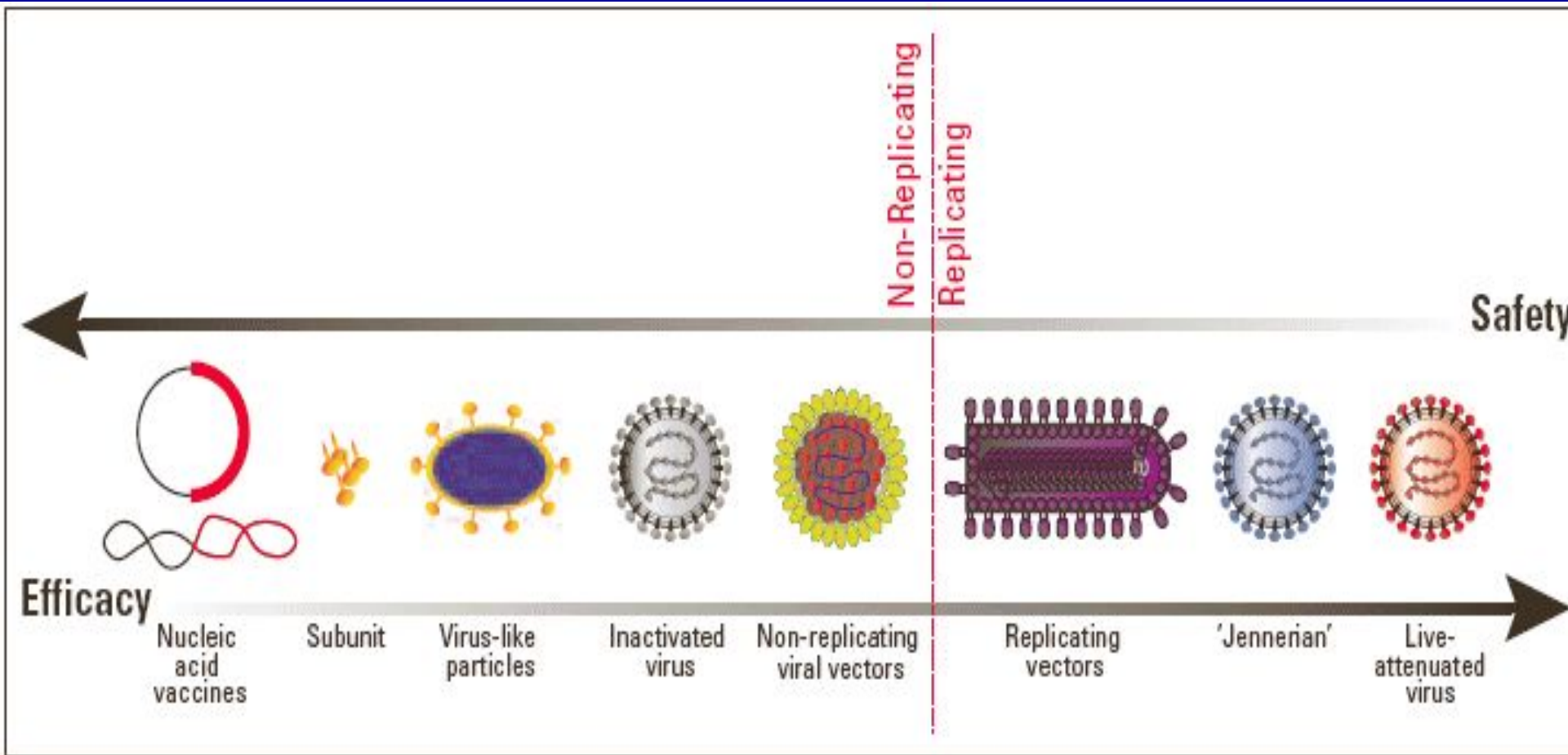
\*Simplified

# CORONAVIRUS VACCINE CANDIDATES

nature



# Important features of vaccines: Safety and efficiency



# AN ARRAY OF VACCINES

## Virus

Inactivated

Weakened

## Viral vector

Replicating

Non-replicating

## Nucleic acid

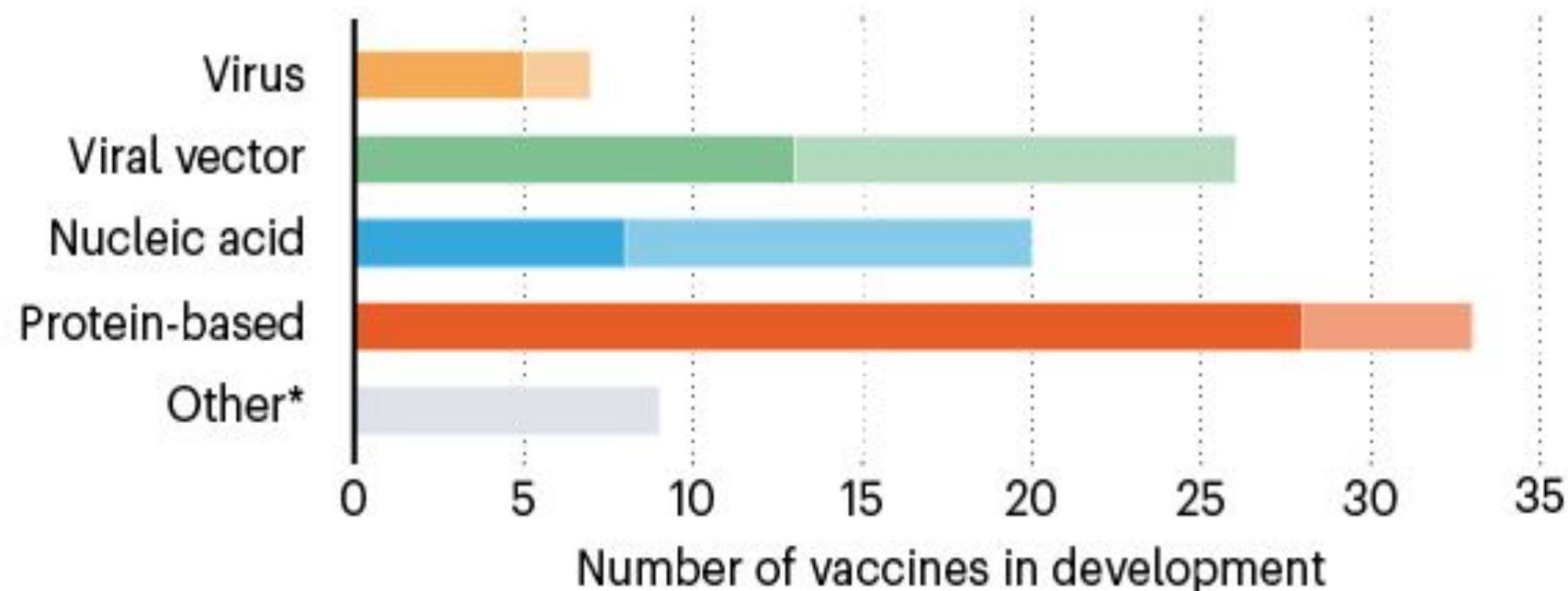
DNA

RNA

## Protein-based

Protein subunit

Virus-like particles

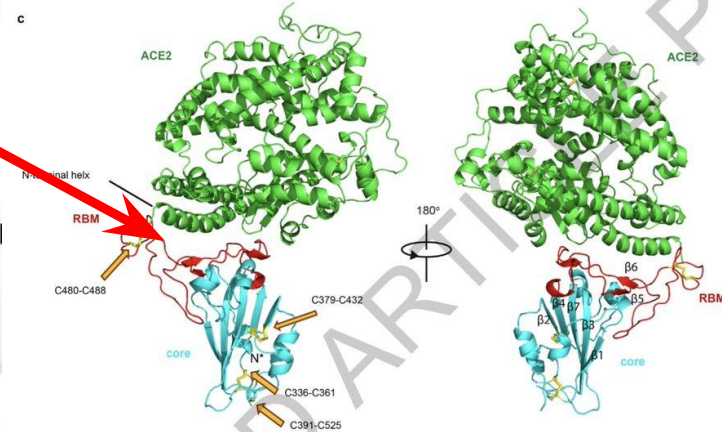
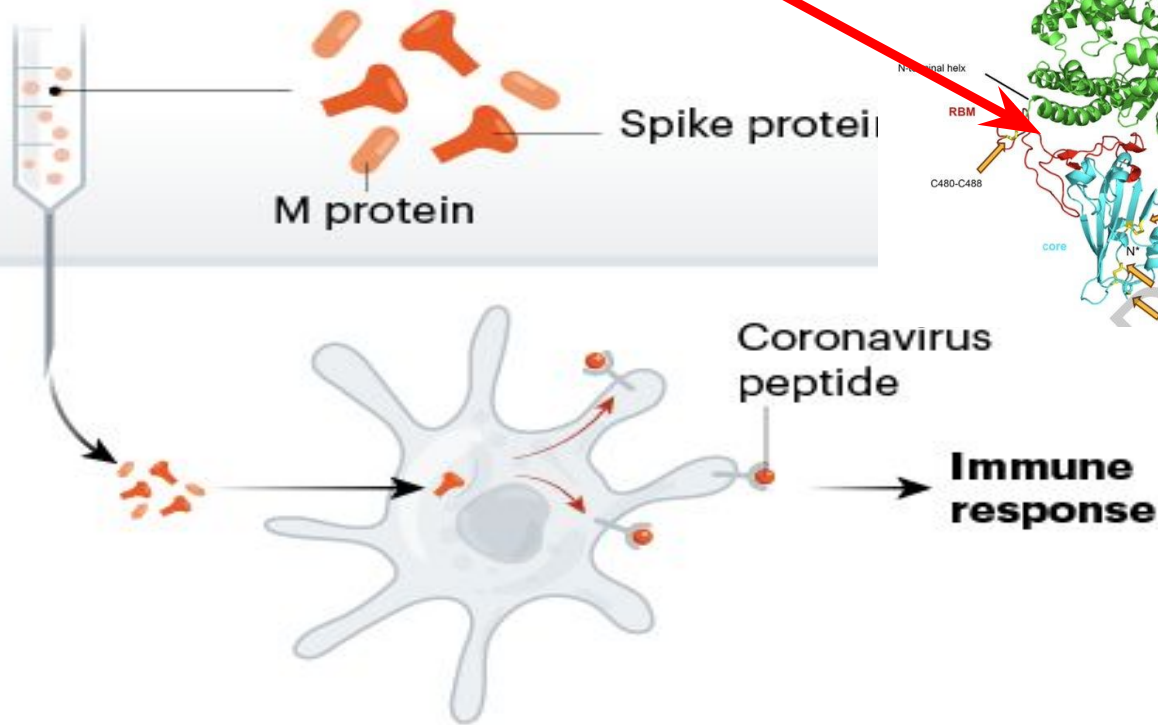


\* Other efforts include testing whether existing vaccines against poliovirus or tuberculosis could help to fight SARS-CoV-2 by eliciting a general immune response (rather than specific adaptive immunity), or whether certain immune cells could be genetically modified to target the virus.

# PROTEIN-BASED VACCINES

## Protein subunits

Twenty-eight teams are working on vaccines with viral protein subunits — most are focusing on the virus's spike protein or a key part of it called the receptor binding domain. Similar vaccines against the SARS virus protected monkeys against infection but haven't been tested in people. To work, these vaccines might require adjuvants — immune-stimulating molecules delivered alongside the vaccine — as well as multiple doses.



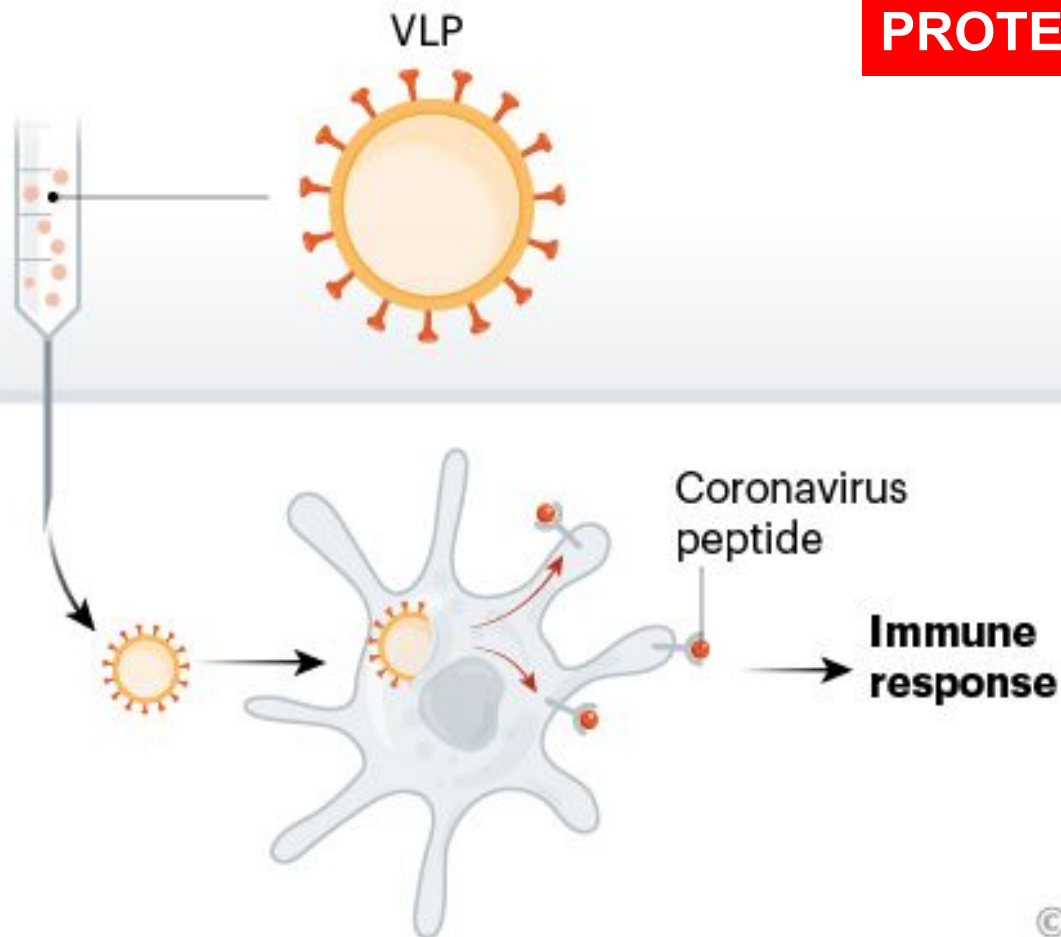
## Virus-like particles

Essentially, a hollow virus-like particle is a protein structure that mimics the infectious

## Virus-like particles

Empty virus shells mimic the coronavirus structure, but aren't infectious because they lack genetic material. Five teams are working on 'virus-like particle' (VLP) vaccines, which can trigger a strong immune response, but can be difficult to manufacture.

**PROTEIN-BASED VACCINES**





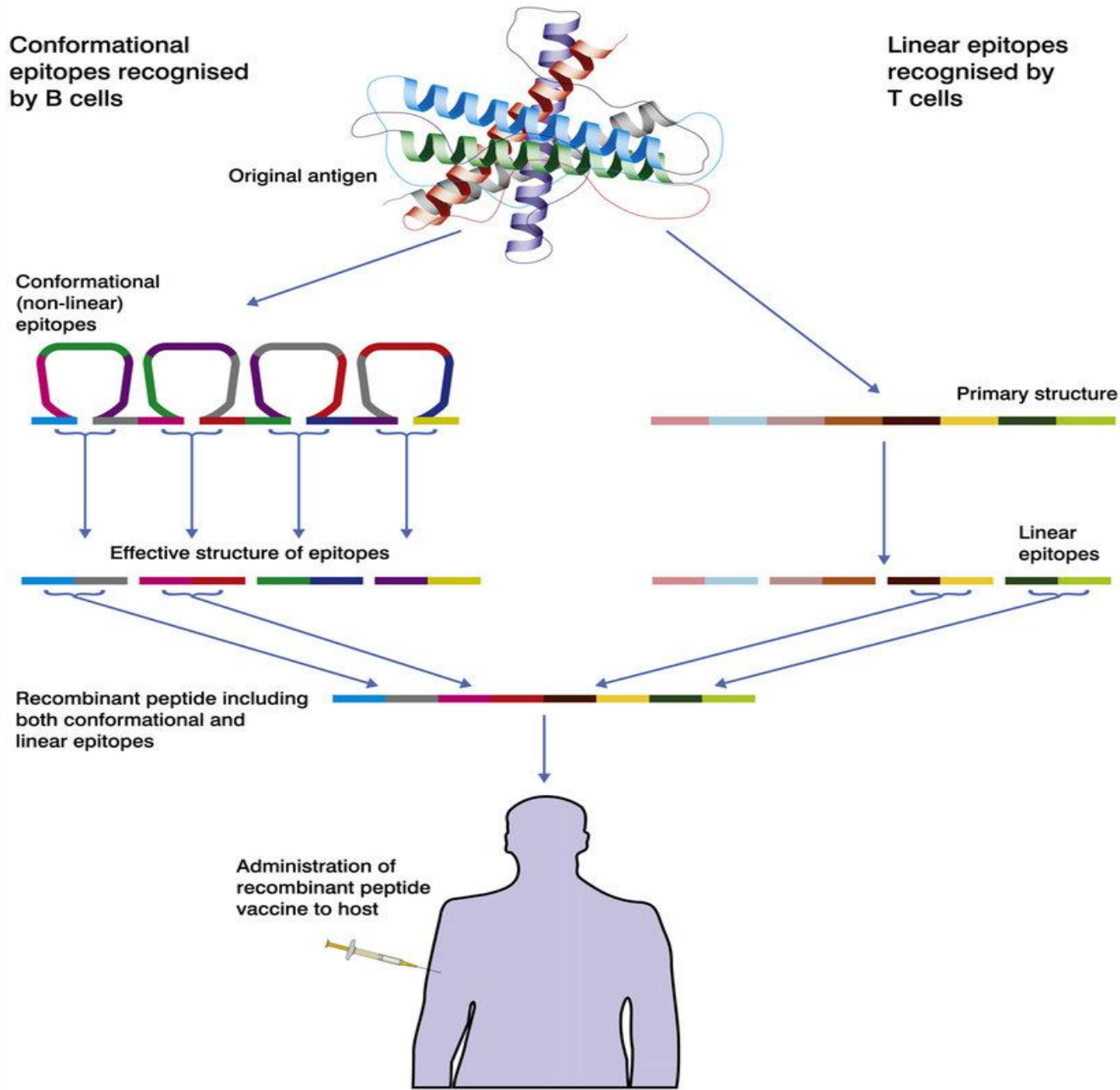
# Subunit vaccines for viruses

They can be developed after identification of the microbial components, that elicit a protective immune response—protective antigens (S for SARS).

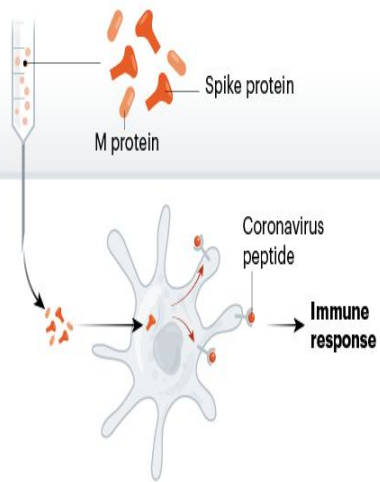
Immunogenic component may be isolated from viruses:

- (1) by biochemical means (*chemical vaccines*) or
- (2) by genetic engineering (*recombinant vaccines*) involving the expression of cloned viral genes in bacteria or eukariotic cell.

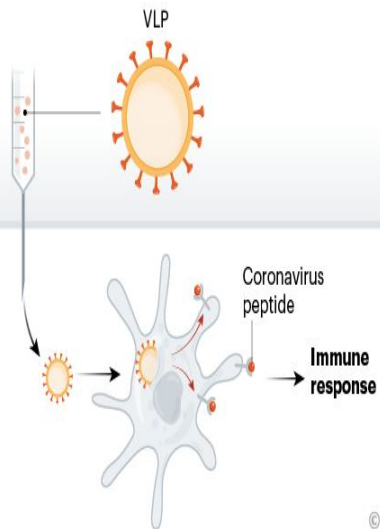
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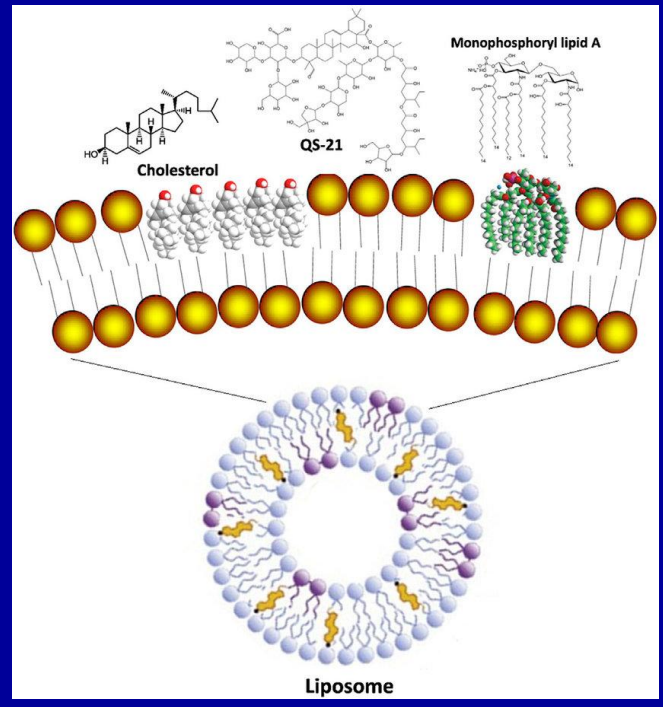
**Virus-like particles**  
Empty virus shells mimic the coronavirus structure, but aren't infectious because they lack genetic material. Five teams are working on 'virus-like particle' (VLP) vaccines, which can trigger a strong immune response, but can be difficult to manufacture.



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# PROTEIN-BASED VACCINES disadvantages

## Subunit VNs are poor immunogens and need to be administered with **adjuvants** or inside small lipid membrane vesicles - **liposomes**.

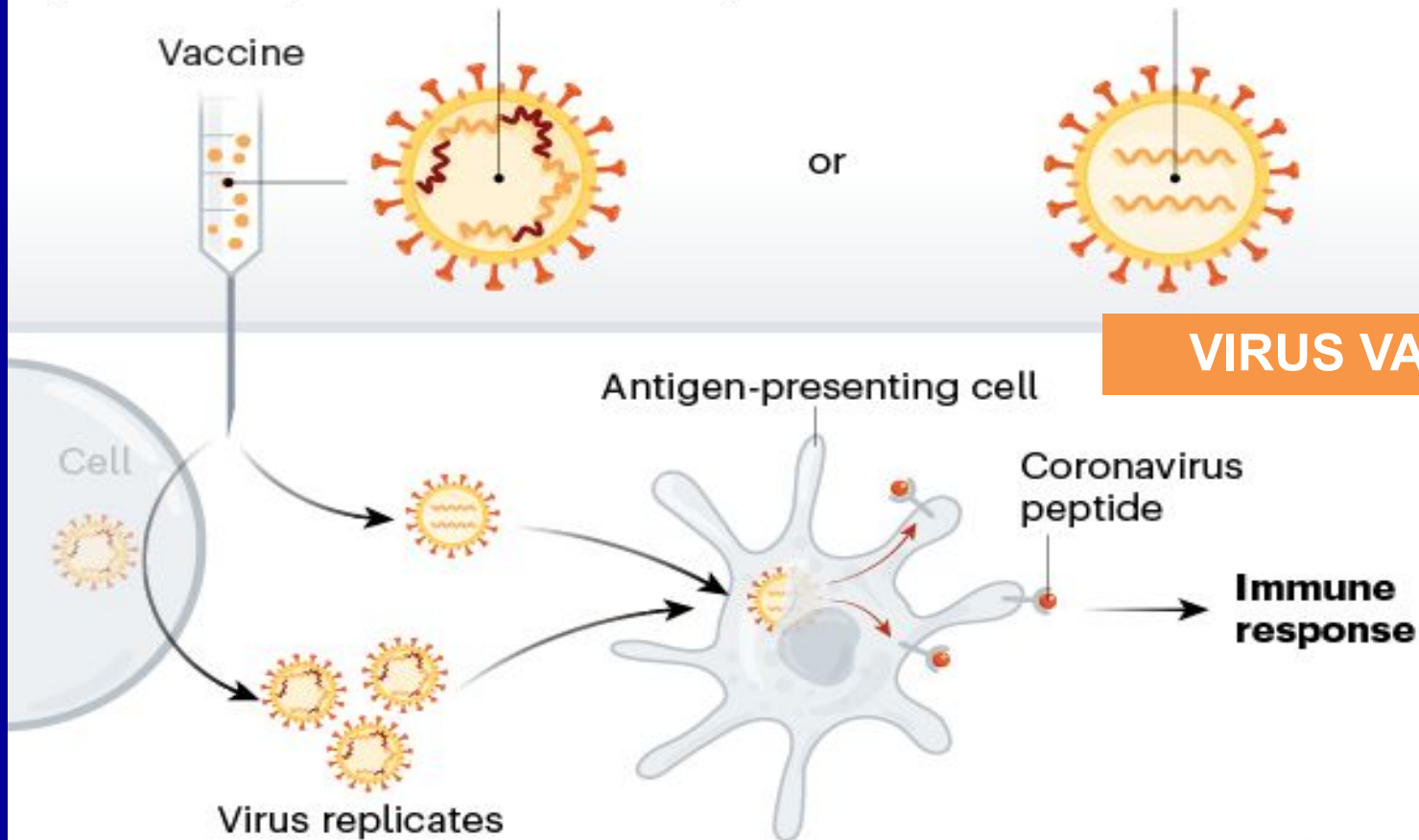


## Weakened virus

A virus is conventionally weakened for a vaccine by being passed through animal or human cells until it picks up mutations that make it less able to cause disease. Codagenix in Farmingdale, New York, is working with the Serum Institute of India, a vaccine manufacturer in Pune, to weaken SARS-CoV-2 by altering its genetic code so that viral proteins are produced less efficiently.

## Inactivated virus

In these vaccines, the virus is rendered uninfecious using chemicals, such as formaldehyde, or heat. Making them, however, requires starting with large quantities of infectious virus.



## VIRUS VACCINES

# Live vaccines: **The advantages**

(1) The **immunity is long live**, and mimics the normal immune responses.

(2) When vaccine is administered orally, **SIgA** is secreted in the **gut** and **oropharynx to protect the mucous**.

This **prevents** the establishing of **carrier state** and **facilitates** the **eradication** of the **virus** from the community.

(3) Live vaccines are administered in **low doses**. Basically **one single administration** is enough for protection because **organisms multiply** in a body.

# Live vaccines: **The disadvantages**

- (1) they may cause disease in immunosuppressed individuals;
- (2) the vaccine may revert to virulent form;
- (3) the COVID – 19 vaccine may cause the effect of the virus (cytokine “storm”)

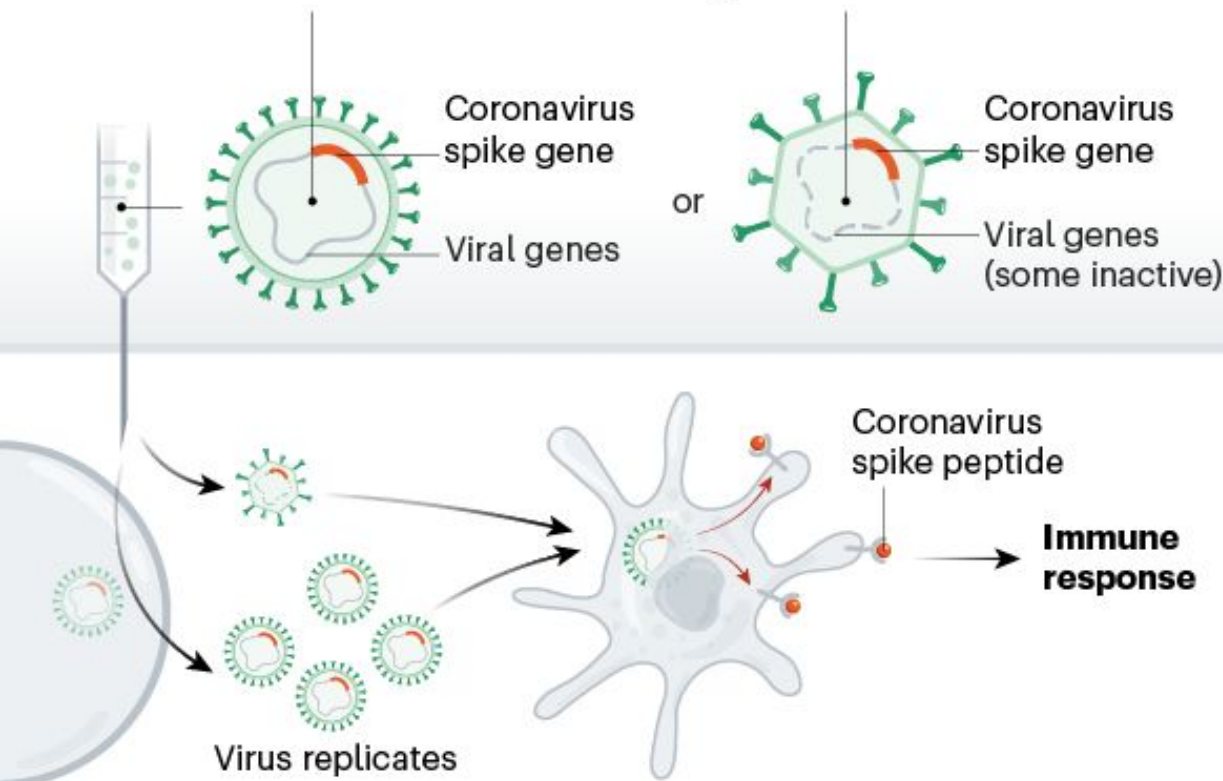
# VIRAL-VECTOR VACCINES

## Replicating viral vector (such as weakened measles)

The newly approved Ebola vaccine is an example of a viral-vector vaccine that replicates within cells. Such vaccines tend to be safe and provoke a strong immune response. Existing immunity to the vector could blunt the vaccine's effectiveness, however.

## Non-replicating viral vector (such as adenovirus)

No licensed vaccines use this method, but they have a long history in gene therapy. Booster shots can be needed to induce long-lasting immunity. US-based drug giant Johnson & Johnson is working on this approach.



Platforms for vector vaccines

Disadvantage:  
Anti-vector  
immunity

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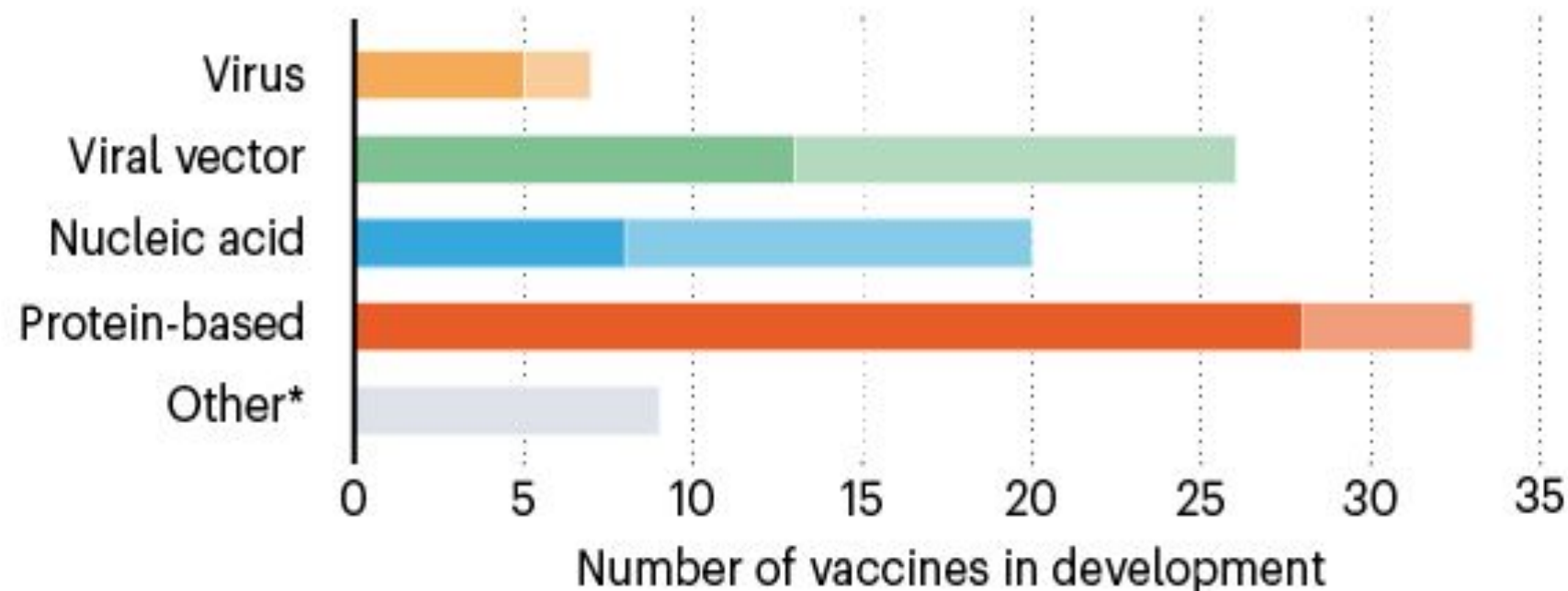
DNA

RNA

## Protein-based

Protein subunit

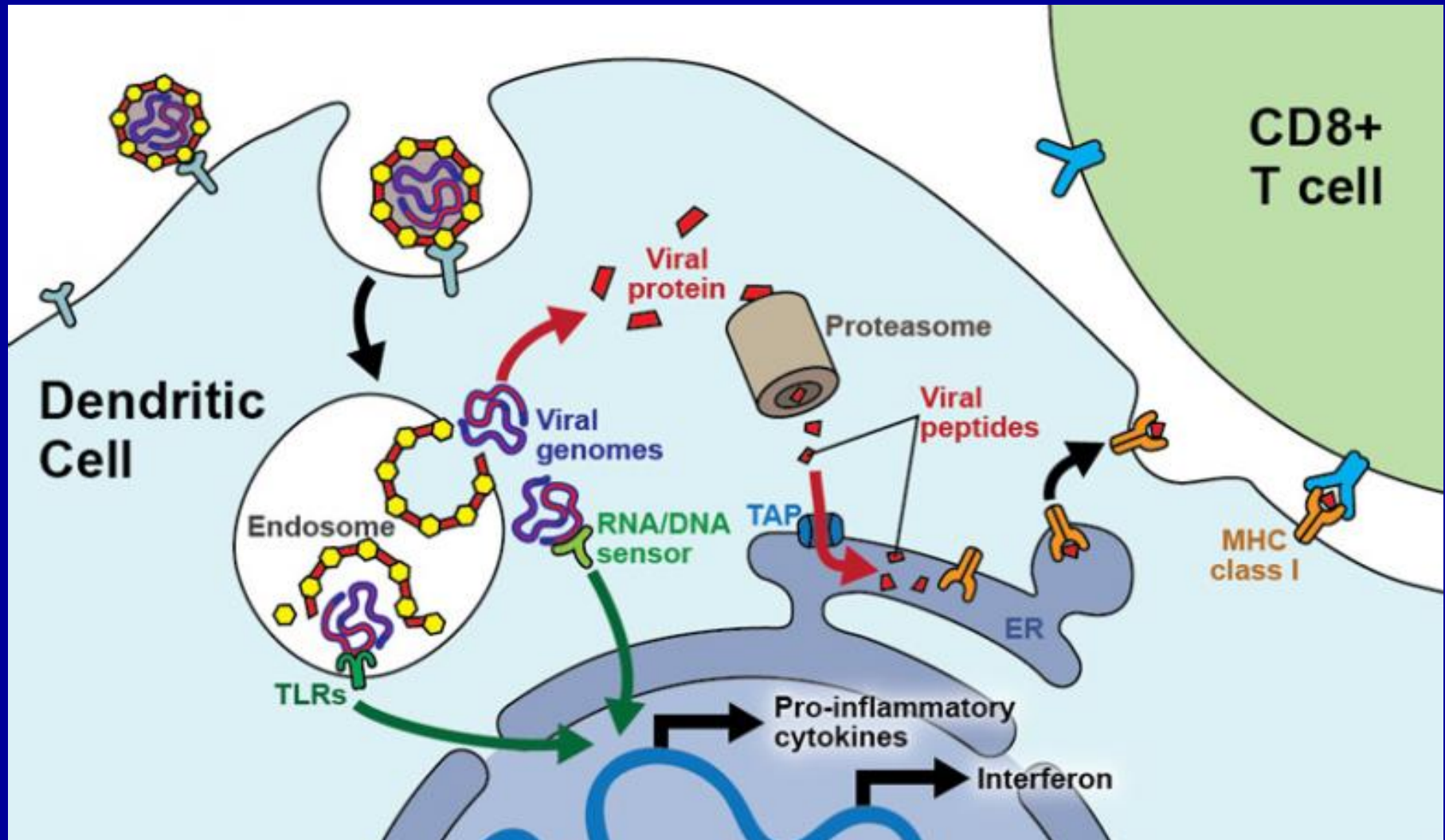
Virus-like particles



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# Mechanisms of immune activation by vaccine vector particles (VVP) through two pathways (Advantage)



- VVP infect DCs of a vaccinee, taken up by receptor-mediated endocytosis and release their genome into the cytoplasm of the DC. TLRs sense it in endosome or/and by cytoplasmic sensors of viral nucleic acids (“RNA/DNA sensor”). Both pathways signal through common pathways such as the NF $\kappa$ B and MAPK pathways, resulting in the transcriptional activation of pro-inflammatory cytokines but also in type I interferon production. These events lead to functional activation of the DCs dendritic cell as APCs. Simultaneously, the viral genomic information will be expressed, leading to synthesis of viral proteins and endosomal processing. Viral peptides are loaded onto MHC class I molecules, which are then exported to the cell surface for presentation to virus-specific CD8+ T

## DNA vaccine

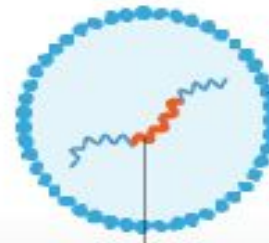
## RNA vaccine

Electroporation

Coronavirus spike gene



DNA



RNA

RNA is often encased in a lipid coat so it can enter cells

A process called electroporation creates pores in membranes to increase uptake of DNA into a cell

## NUCLEIC-ACID VACCINES

Coronavirus spike peptide

Immune response

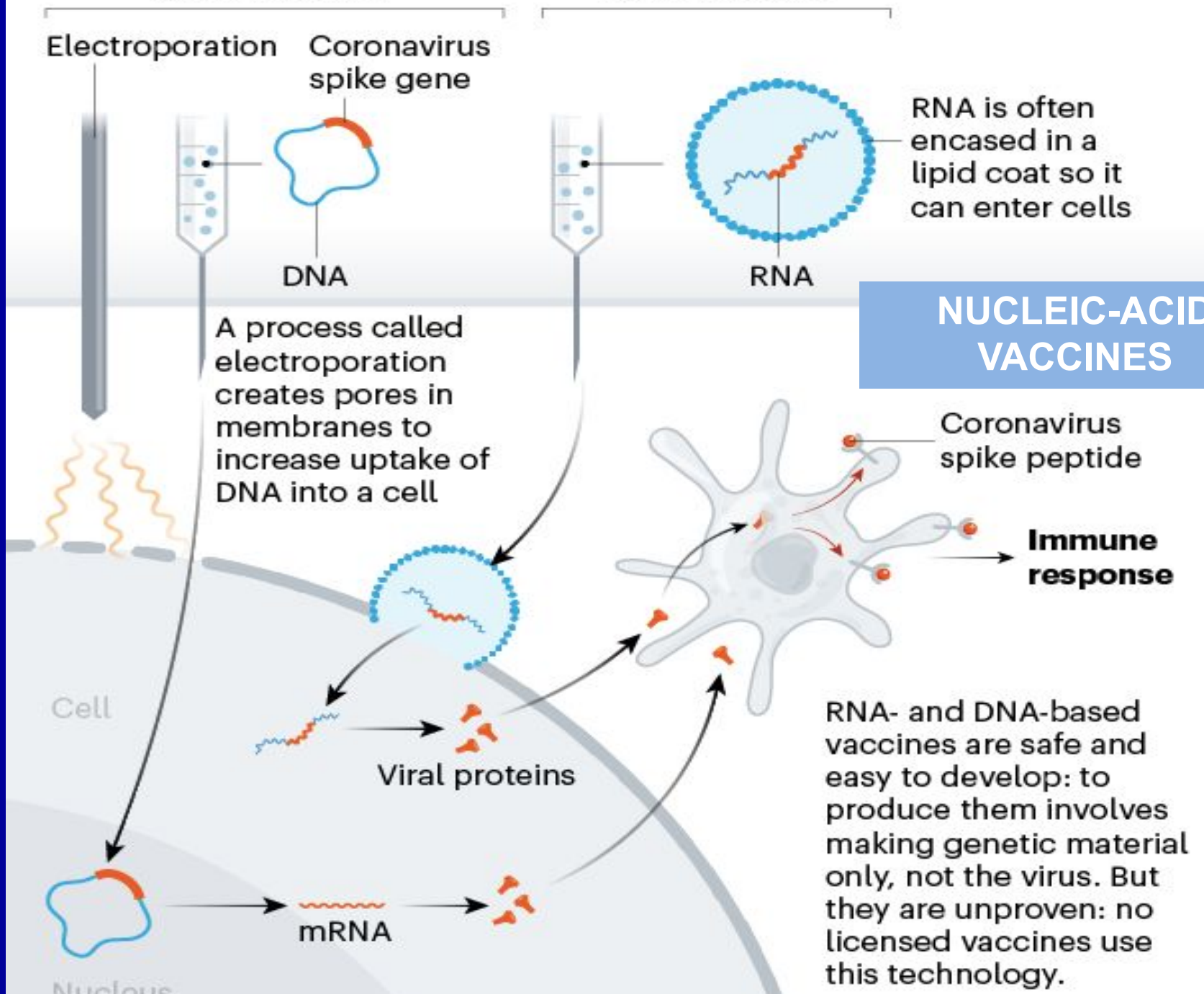
Viral proteins

RNA- and DNA-based vaccines are safe and easy to develop: to produce them involves making genetic material only, not the virus. But they are unproven: no licensed vaccines use this technology.


Cell

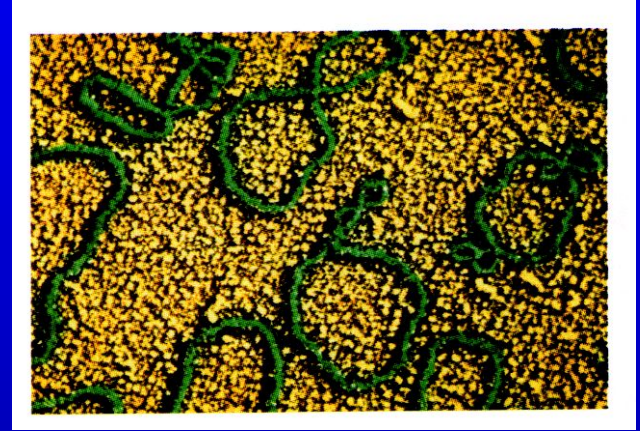
mRNA

Nucleus



# DNA vaccines

- DNA vaccines consist of **naked DNA** code for a **gene for vaccinal protective antigen**. This construct is produced by cloning gene, code for protective antigen, into a **bacterial plasmid**. 
- The use of **DNA vaccines** makes possible developing vaccines against infectious agents such as **HIV**, **herpes** virus, **malaria**, and others, which **require** not only **humoral** but also **cellular** immune responses for **protection**.



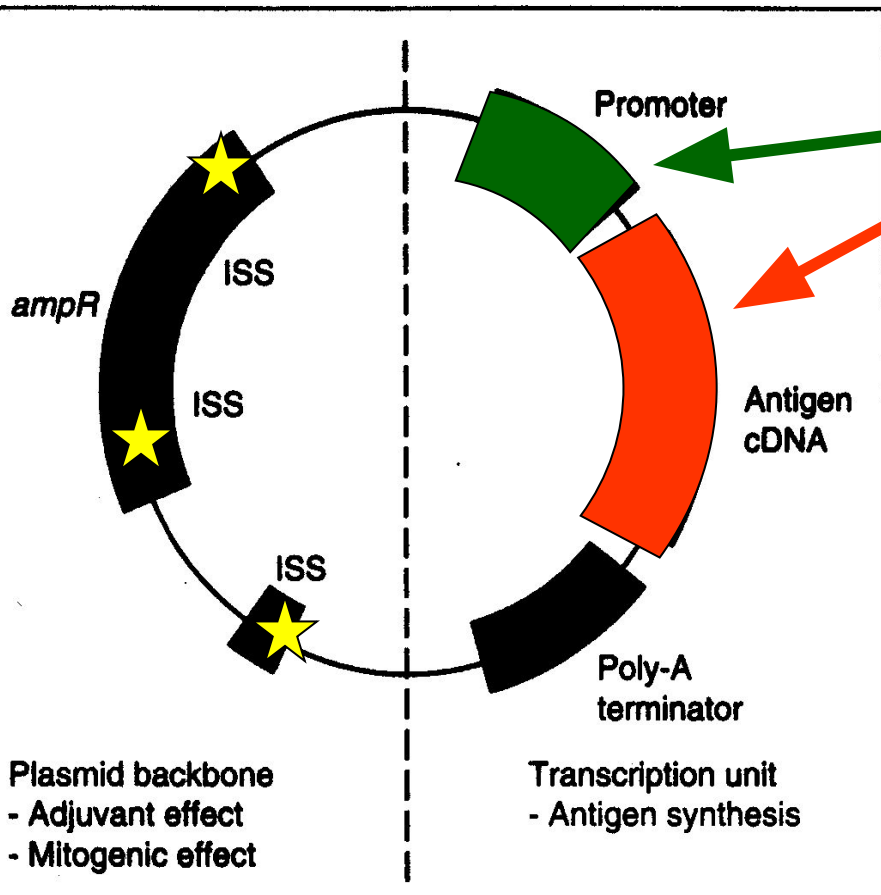
# Plasmid DNA for gene vaccination

has two major units:

(1) A **transcription unit** comprising **promoter**, an **antigen cDNA**, and **poly-adenylation (A) addition sequence**, which together direct protein synthesis.

(2) A plasmid backbone **delivers adjuvant and mitogenic activity via immunostimulatory**

**sequences (ISS)**. ISS are located within the ampicillin antibiotic resistance gene (ampR). ISS are the noncoding region of the plasmid.



## Immune responses elicited by DNA vaccines

The **DNA** plasmid is **injected** into the **muscle cell** or **skin** of the vaccine recipient.

The plasmid can be **uptaken** by both **muscle cell** and **antigen-presenting cell (APC)**.

The **gene for the antigen (Ag)** will be **expressed in muscle cell** and this antigen will be **produced by** the recipient **muscle cells** in large amounts.

(1) When **uptaken by APC**, the **Ag** can be **presented on the APC together with class MHC-II** to activate T helper cells to mediate **humoral immunity**.

(2) When the **Ag** is produced and presented **as endogenous Ag together with class MHC-I** on the surface of the **muscle cell**, it can elicit TH1 **cell-mediated immune response**.

# DNA vaccines: Disadvantages

<b>Vaccines</b>	<b>Advantages</b>	<b>Disadvantages</b>
Viral vectored vaccines	Stimulation of innate immune response; induction of T and B cell immune response.	induction of anti-vector immunity: cell based manufacturing
DNA vaccines	Non-infectious; stimulation of innate immune response; egg and cell free; stable, rapid and scalable production; induction of T and B cell immune response.	Potential integration into human genome; poor immunogenicity in humans.