Biotechnology-Based Vaccines

Dr. Aws Alshamsan
Department of Pharmaceutics
Office: AA87
Tel: 4677363
aalshamsan@ksu.edu.sa

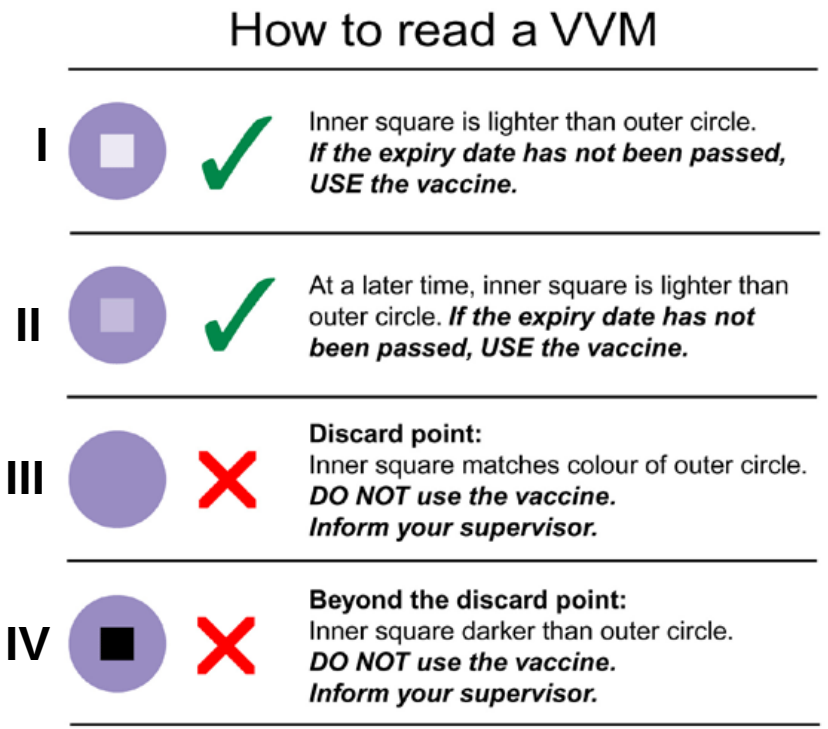
Objectives of this lecture

By the end of this lecture you will be able to:

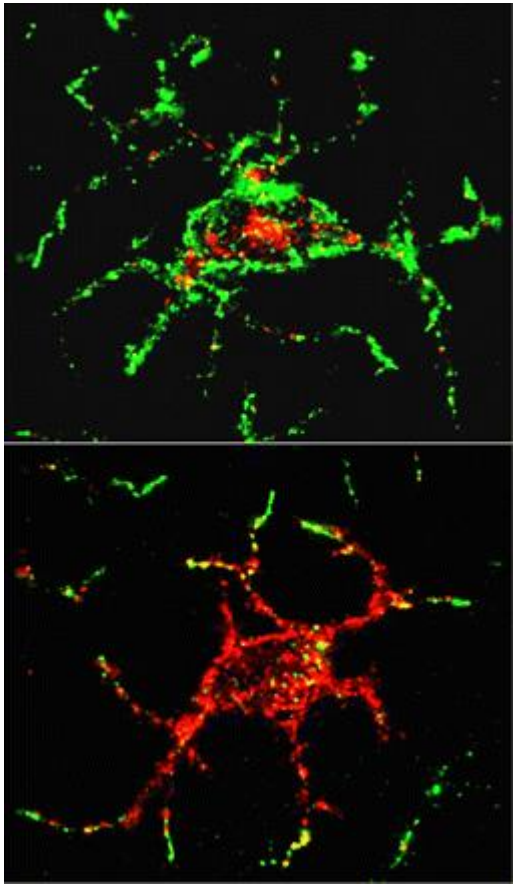
1. Describe how vaccines work
2. Realize the significance of vaccination
3. Understand the technology vaccine manufacturing
4. Compare between traditional and biotechnology-based vaccines

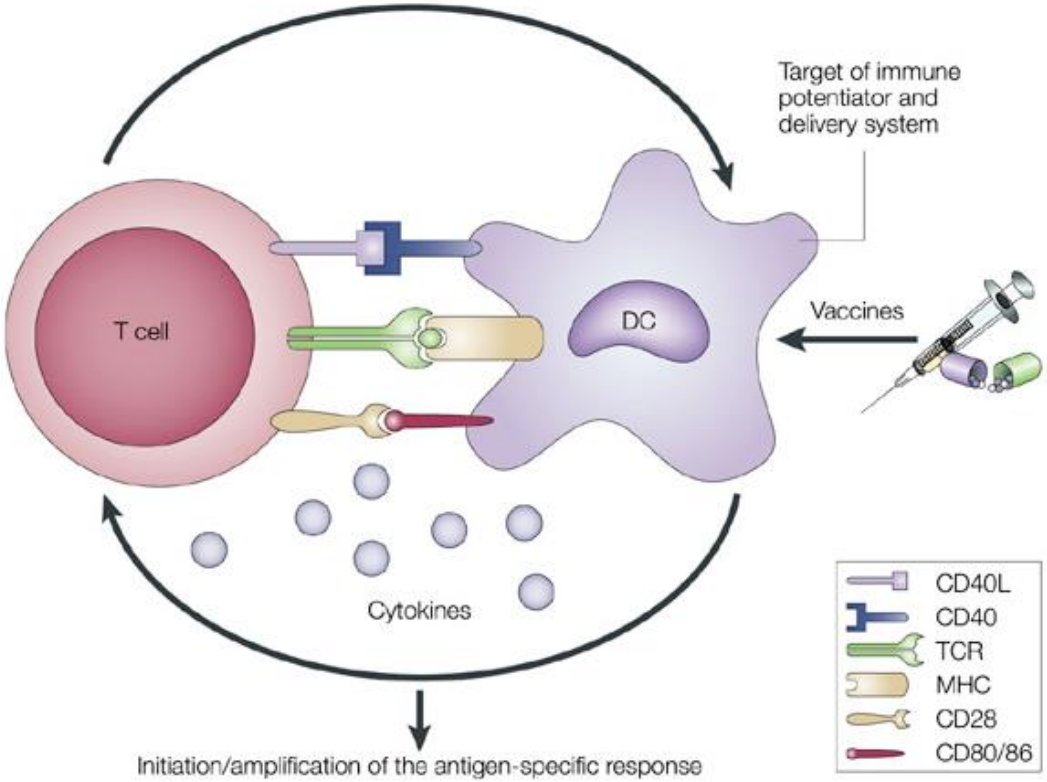
What are vaccines?

- Biological preparations that direct the immune system toward a particular disease in a specific manner



T cell activation by DC





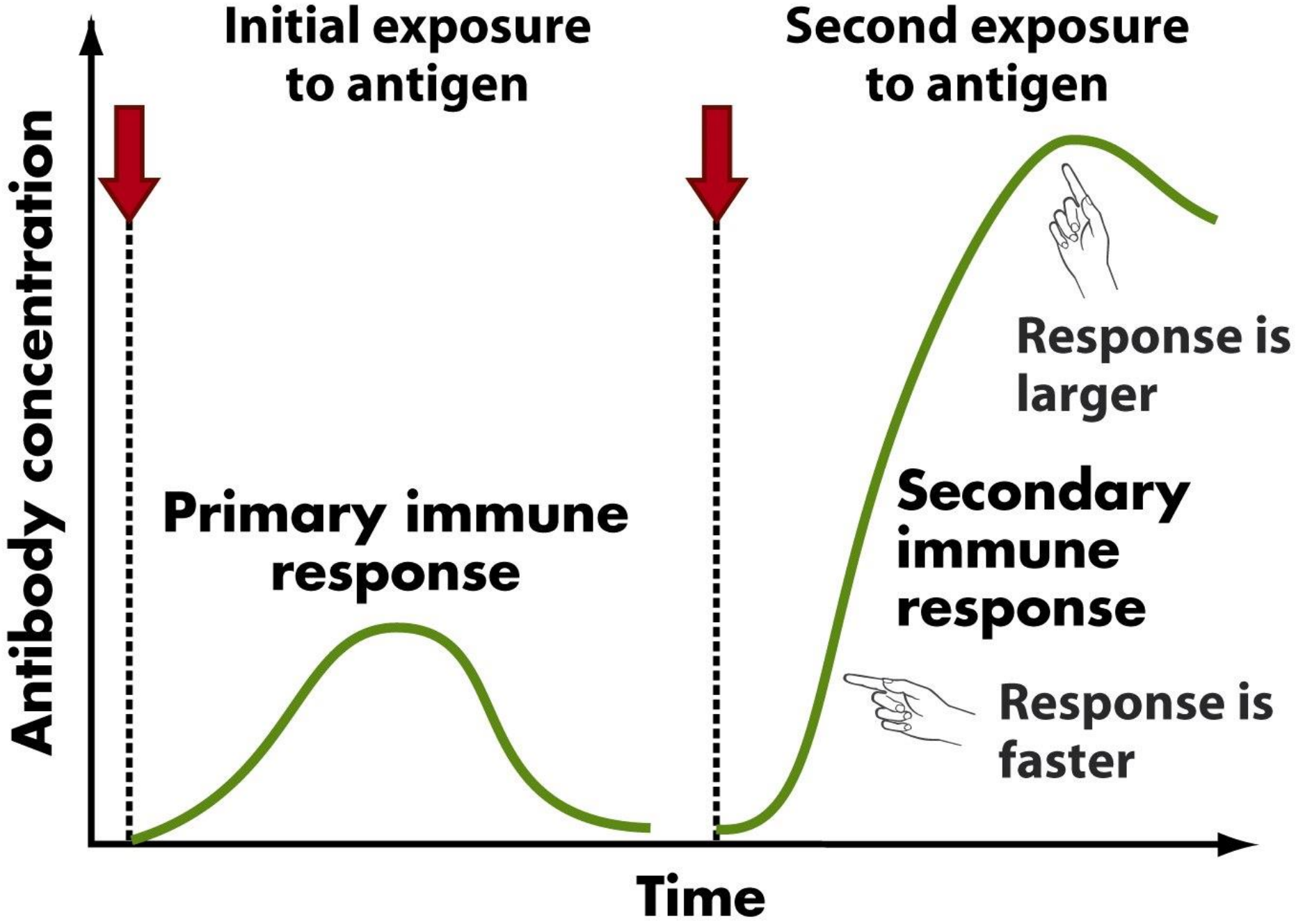


Figure 49-16 Biological Science, 2/e

Vaccination vs. Immunization

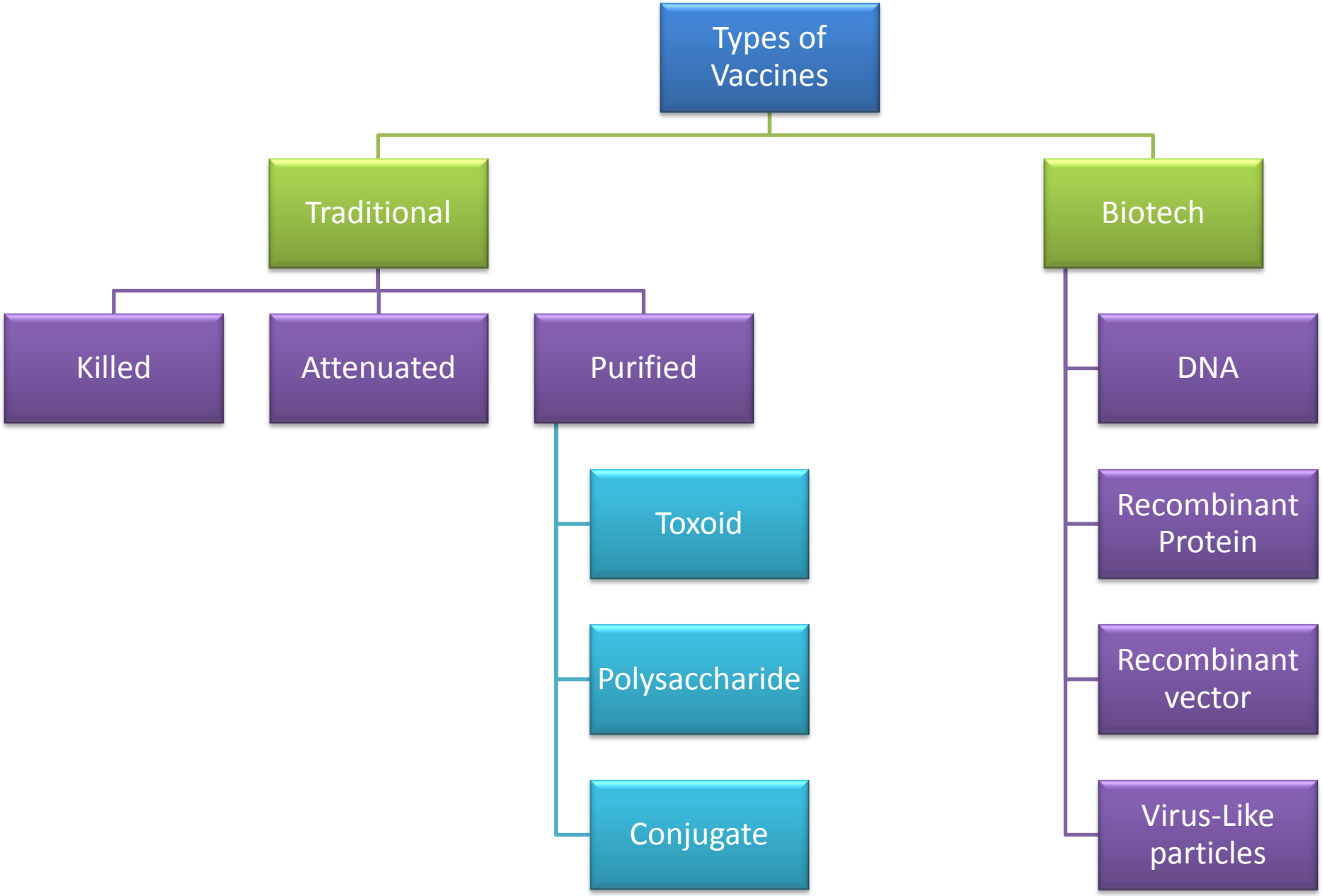
- **Immunization is the process of eliciting a robust, specific, and long-lasting response against any given antigen.**

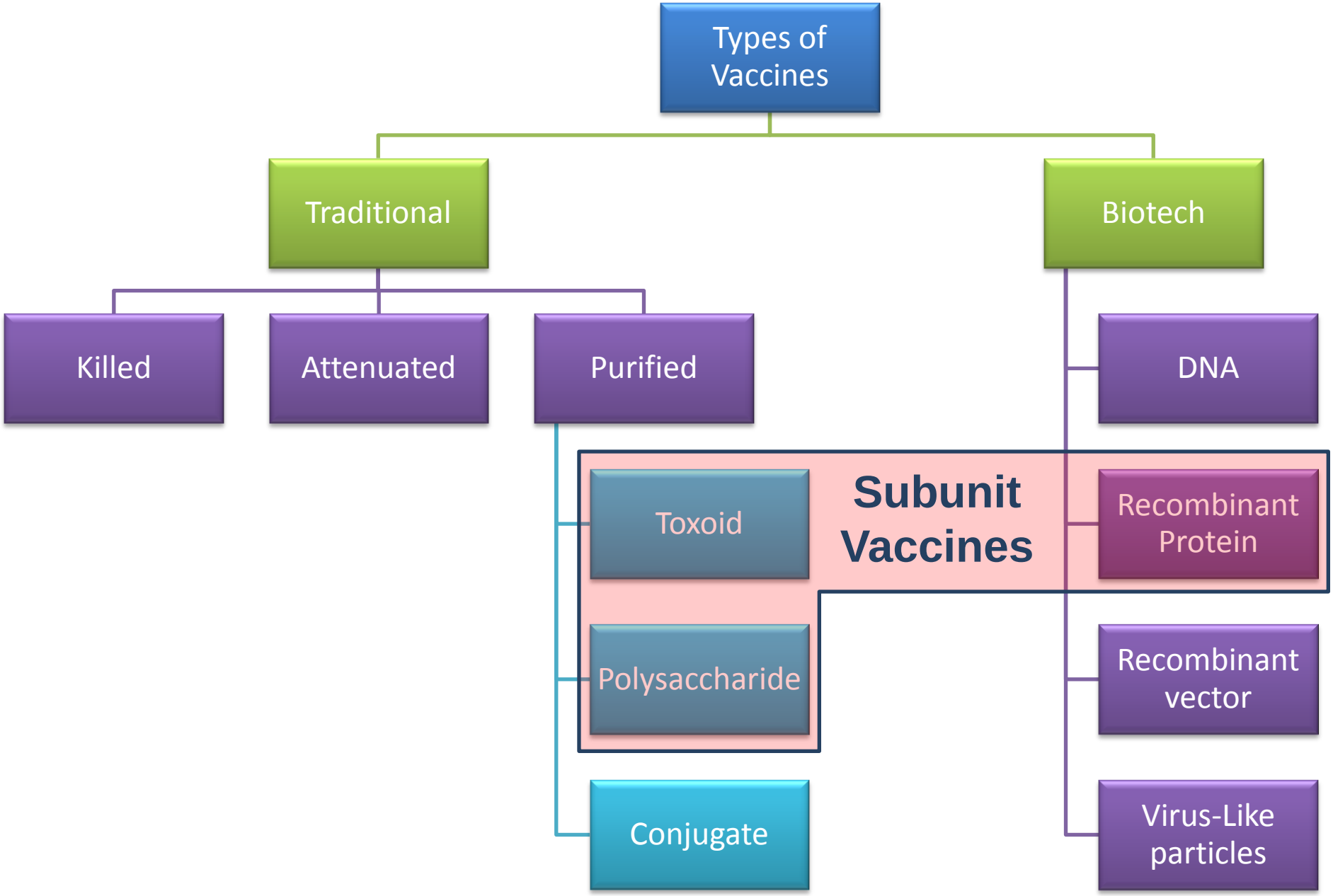
- **Passive immunization:** transfer of some immune system elements (Abs or cells) to a living body

- **Active immunization:** induction of immune response against a specific antigen ***(Vaccination)***

The Ideal Vaccine

- **100% efficient in all individuals of any age**
- **Provides life-long protection after single administration**
- **Does not evoke adverse reactions or cause diseases**
- **Physically stable under various conditions (temp., light, transportation)**
- **Genetically stable**
- **Easy to administer (orally vs. parenterally)**
- **Available in unlimited quantities**
- **Affordable (<$1, WHO)**


Types of Vaccines
Traditional
Biotech
Killed
Attenuated
Purified
Toxoid
Polysaccharide
Conjugate
DNA
Recombinant Protein
Recombinant vector
Virus-Like particles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Conjugate
Virus-Like particles

Traditional Vaccines

1. **Non-living whole organism**

   Heat inactivated or chemically inactivated

2. **Live attenuated organism**

   Bacteria or viruses attenuated in culture

3. **Purified Macromolecules**

   Extraction and purification of pathogen components

Non-Living Whole Organism

- **Heat or formaldehyde inactivation "killing" of a pathogen**
- **The pathogen raises an immune response but not capable of replication in the host**
- **Heat inactivation causes protein denaturation i.e. loss of the antigenic 3D structure**
- **Formaldehyde inactivation is preferable**
- **Salk polio vaccine (IPV)**
- **Flu shot**

Live Attenuated Organism

- **Attenuation means reducing "virulence" while maintaining the capability of transient growth and immunogenicity.**
- **Attenuated vaccines elicit a vigorous long-lasting immune response from a single dose**
- **They can induce cell-mediated immune response**
- **Attenuated pathogens can mutate and revert virulent forms**
- **Sabin polio vaccine (OPV)**
- **Bacille Calmette-Guérin**

Traditional Attenuation

- **Pathogens grown in unnatural hosts, unusual media, or exposure to harsh chemicals for extended periods**

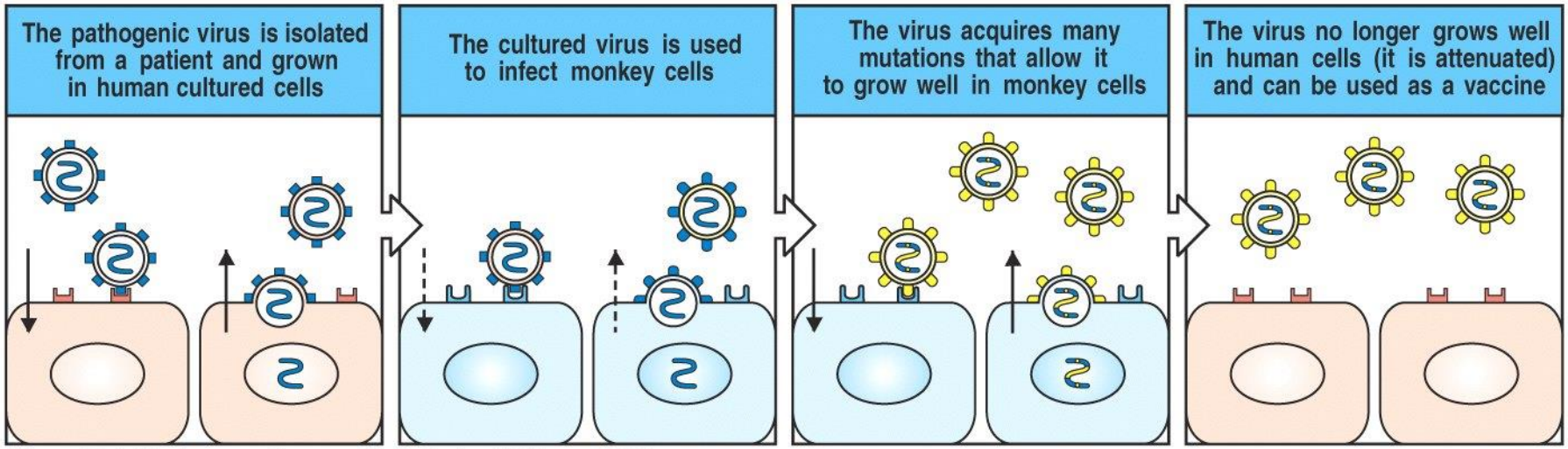
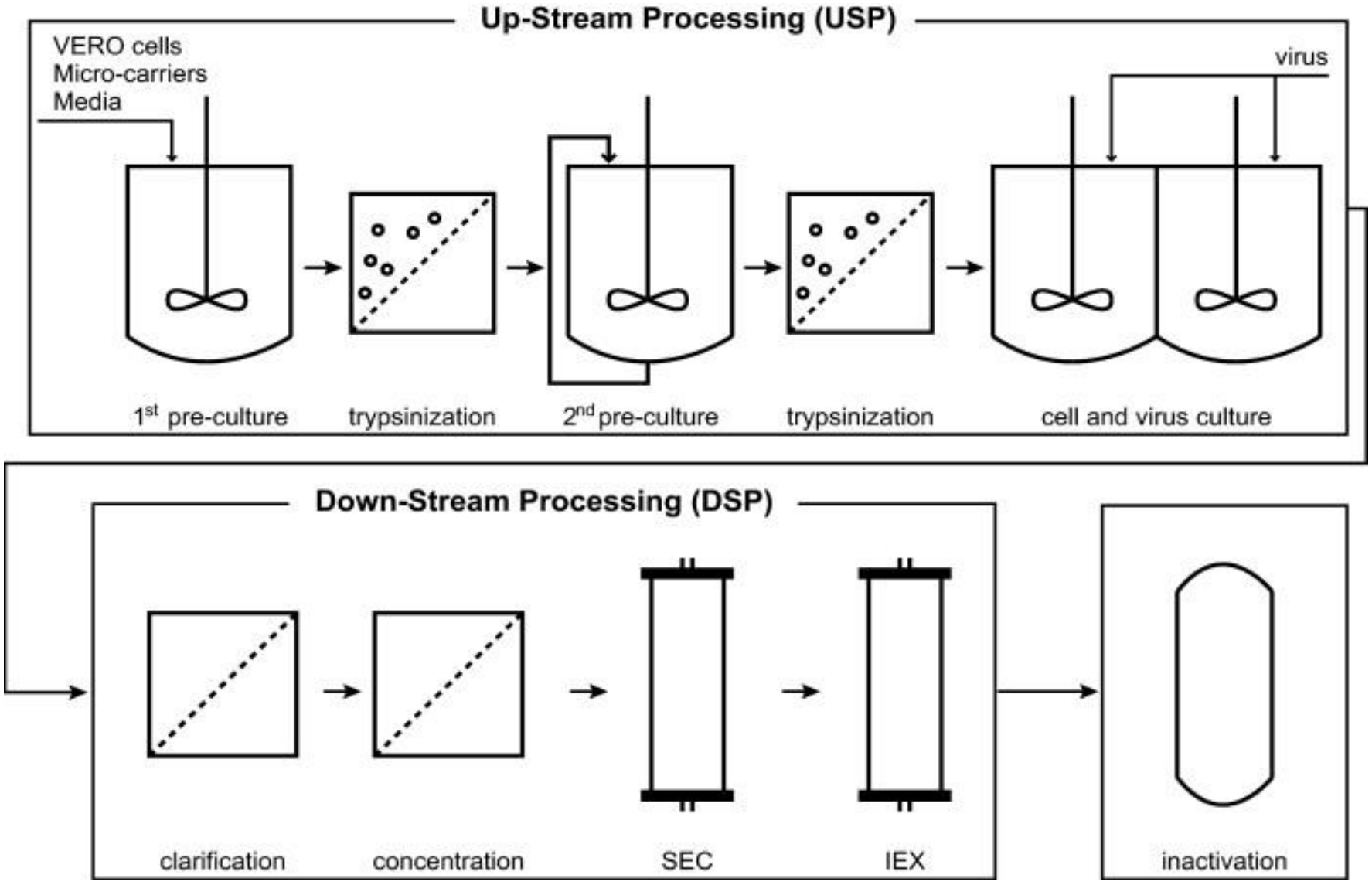


Figure 12-2 The Immune System, 2/e (© Garland Science 2005)

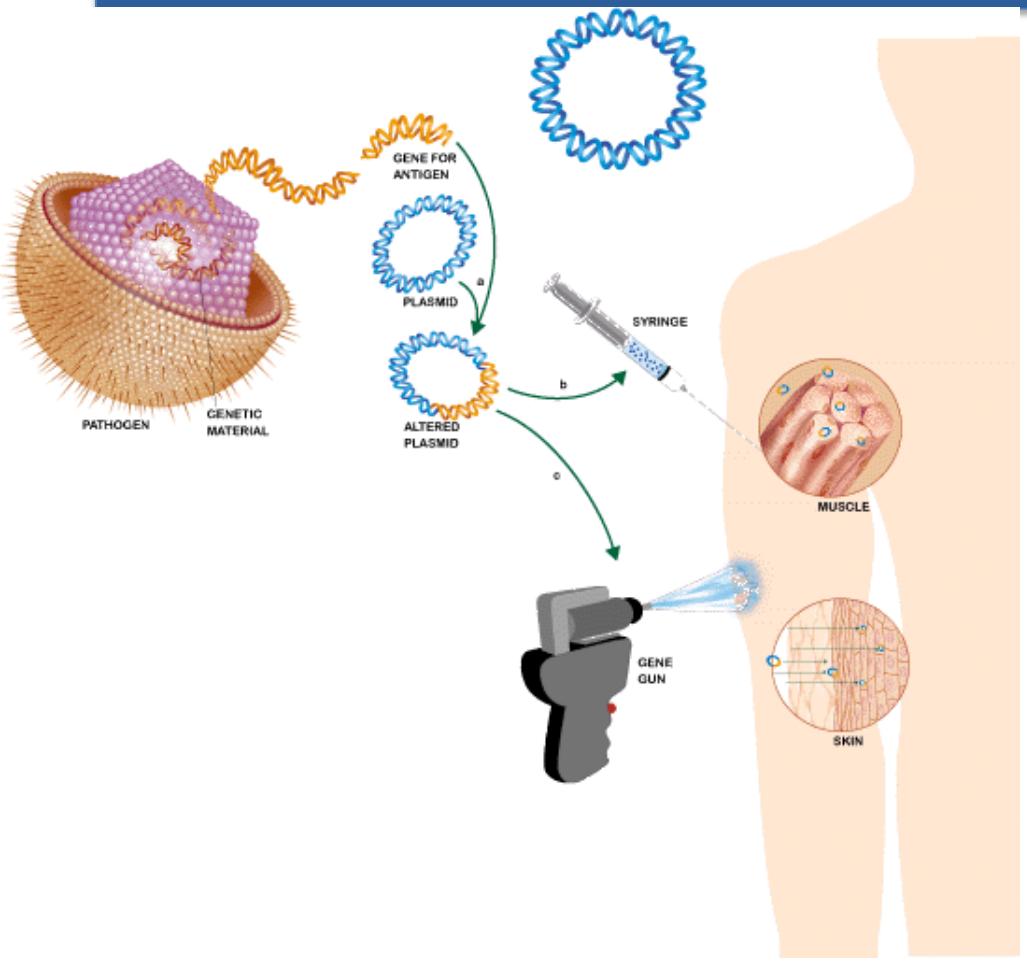

Up-Stream Processing (USP)
VERO cells
Micro-carriers
Media
virus
1st pre-culture
trypsinization
2nd pre-culture
trypsinization
cell and virus culture
Down-Stream Processing (DSP)
clarification
concentration
SEC
IEX
inactivation

Purified Macromolecules

- ***Toxoid:*** **modified bacterial exotoxins. Toxins treated with iodine, pepsin, ascorbic acid, or formalin to reduce toxicity while retaining immunogenicity. Toxoids generate neutralizing antibodies.**
- **Diphtheria toxoid, tetanus toxoid**

- ***Conjugate:*** **polysaccharide capsule conjugated with toxoid to elicit type II thymus-independent antigen and generate memory cells.**
- **HiB (+tetanus toxoid), PCV (+diphtheria toxoid)**

Recombinant DNA Vaccine

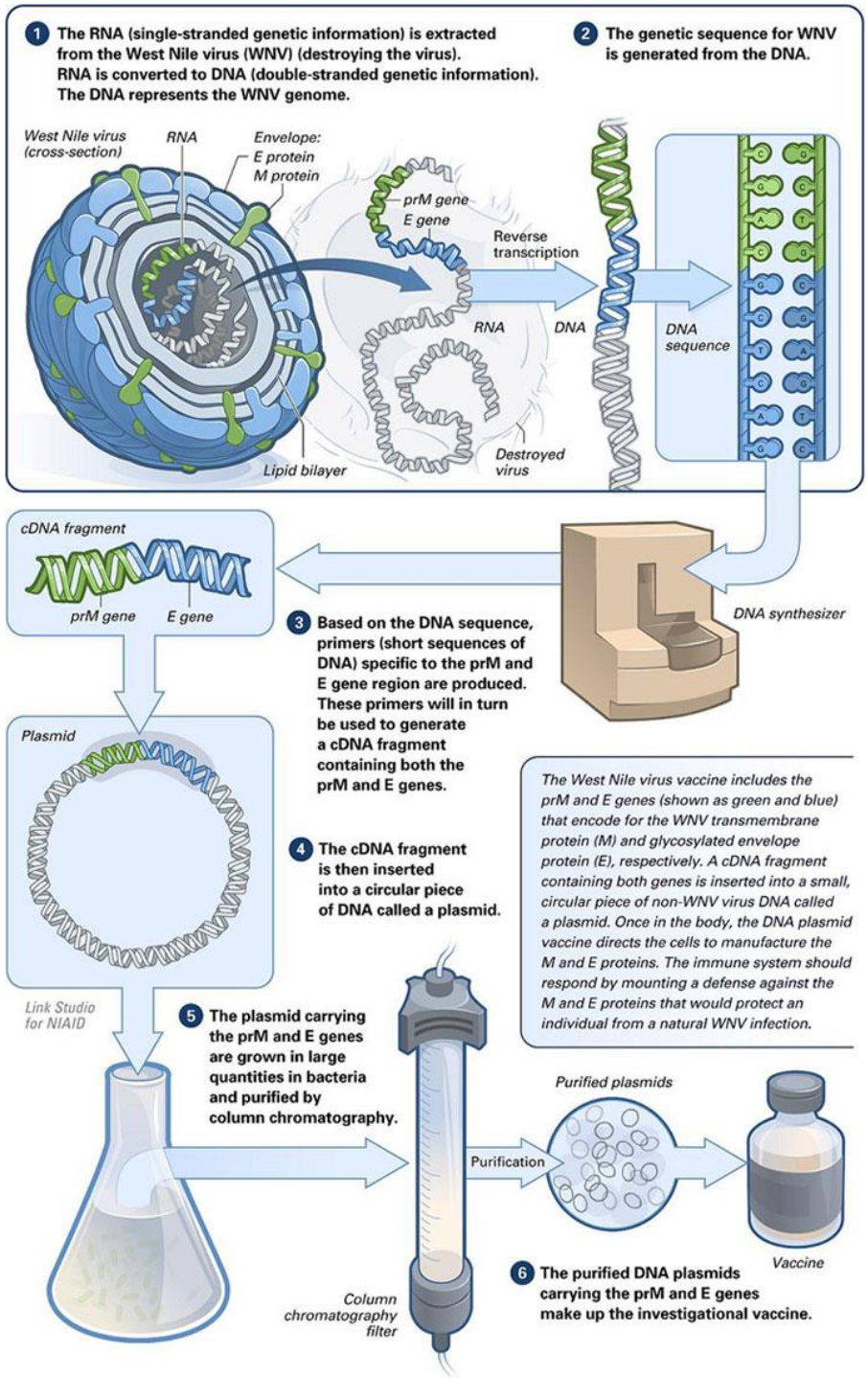


- **Plasmid DNA encoding the desired antigen is administered parenterally. The foreign protein is expressed by the host cell and generate an immune response.**

Scientific American: Feature Article: Genetic Vaccines: July 1999

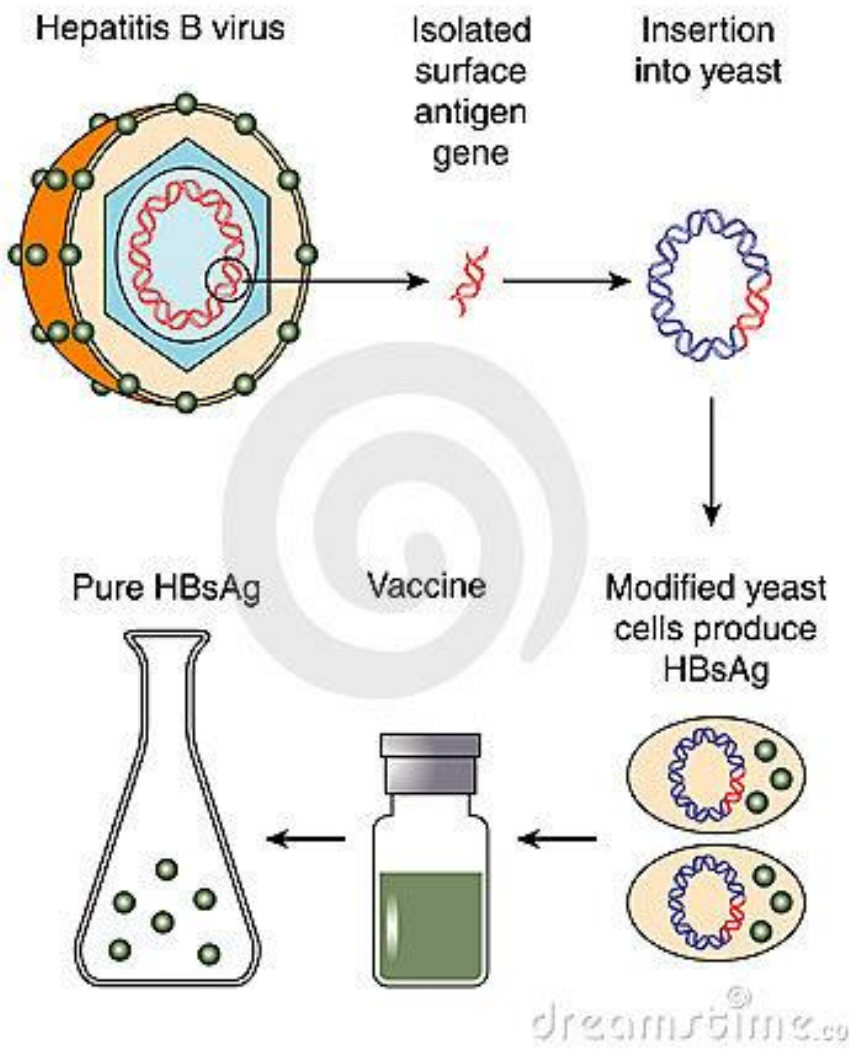
Recombinant DNA Vaccine

Advantages	Disadvantages
Low intrinsic immunogenicity	Unknown effect of long-term expression
Induction of long-term immunity	Formation anti-DNA antibodies
Induction of both humoral and cellular response	Possible genome integration
Possibility of constructing multiple-epitope plasmid	
Heat stability	
Ease of large-scale production	


1 The RNA (single-stranded genetic information) is extracted from the West Nile virus (WNV) (destroying the virus). RNA is converted to DNA (double-stranded genetic information). The DNA represents the WNV genome.
2 The genetic sequence for WNV is generated from the DNA.
West Nile virus (cross-section)
RNA
Envelope: E protein M protein
Lipid bilayer
prM gene
E gene
Reverse transcription
RNA
DNA
Destroyed virus
DNA sequence
cDNA fragment
prM gene
E gene
DNA synthesizer
3 Based on the DNA sequence, primers (short sequences of DNA) specific to the prM and E gene region are produced. These primers will in turn be used to generate a cDNA fragment containing both the prM and E genes.
Plasmid
4 The cDNA fragment is then inserted into a circular piece of DNA called a plasmid.
The West Nile virus vaccine includes the prM and E genes (shown as green and blue) that encode for the WNV transmembrane protein (M) and glycosylated envelope protein (E), respectively. A cDNA fragment containing both genes is inserted into a small, circular piece of non-WNV virus DNA called a plasmid. Once in the body, the DNA plasmid vaccine directs the cells to manufacture the M and E proteins. The immune system should respond by mounting a defense against the M and E proteins that would protect an individual from a natural WNV infection.
Link Studio for NIAID
5 The plasmid carrying the prM and E genes are grown in large quantities in bacteria and purified by column chromatography.
Purification
Purified plasmids
Vaccine
Column chromatography filter
6 The purified DNA plasmids carrying the prM and E genes make up the investigational vaccine.

Recombinant Protein Vaccine

- **Incorporation of the corresponding peptide sequence into a plasmid and expressed in host cells.**

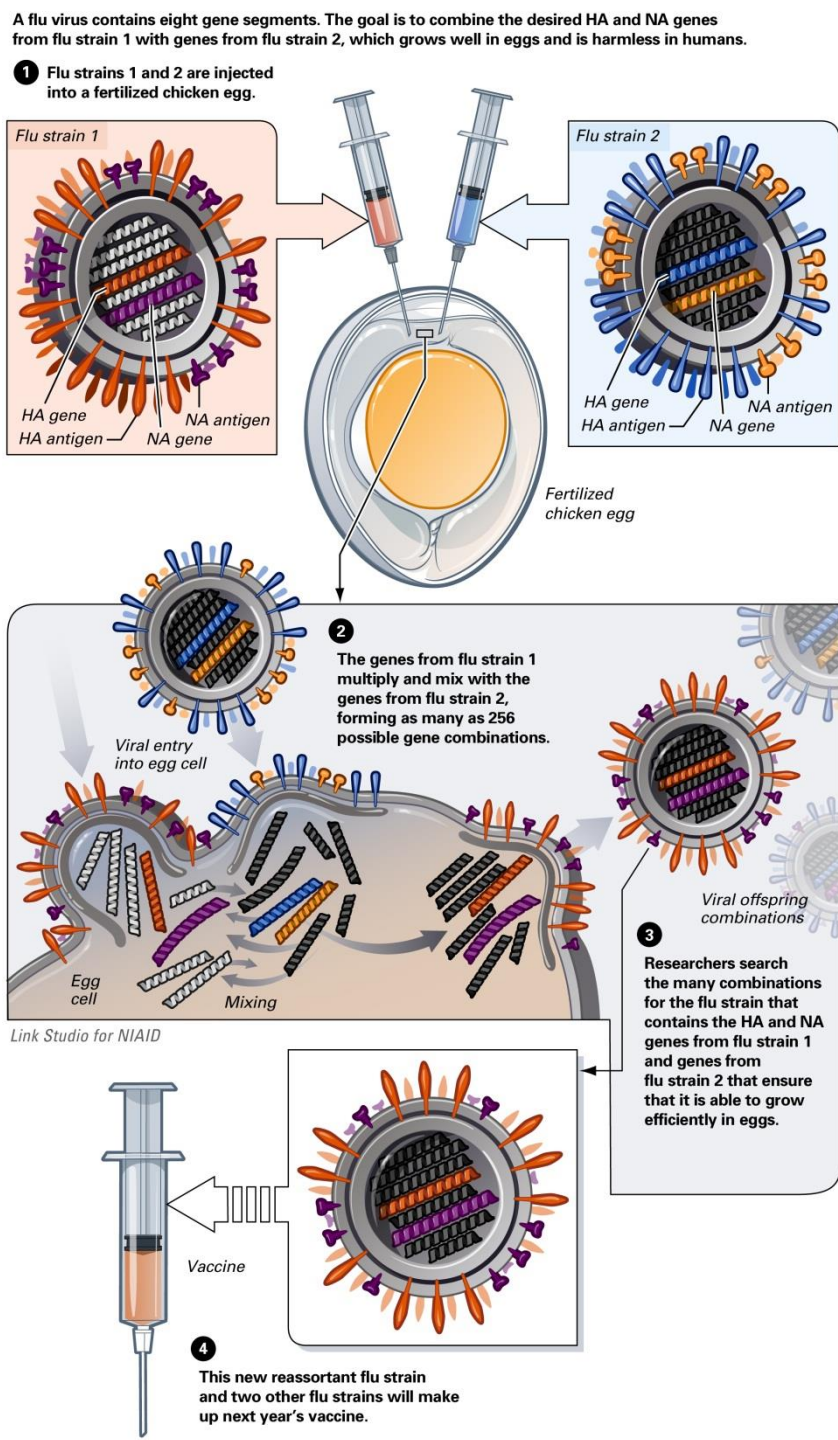


Recombinant Vector Vaccine

- There are several drawbacks in traditional strategy of attenuation e.g:
  1. **The virulence and life cycle of the pathogen must be known in detail**
  2. **Traditional attenuation may result in reduced immunogenicity**
  3. **Reversion of attenuated microorganism is possible either during its production or presence in the host**

Reassortment

The swapping and mixing of gene segments between two different viral strains inside a fertilized chicken egg, to create a new vaccine.



Credit: *These images are curtsey of the National Institute of Allergy and Infectious Diseases (NIAID).*

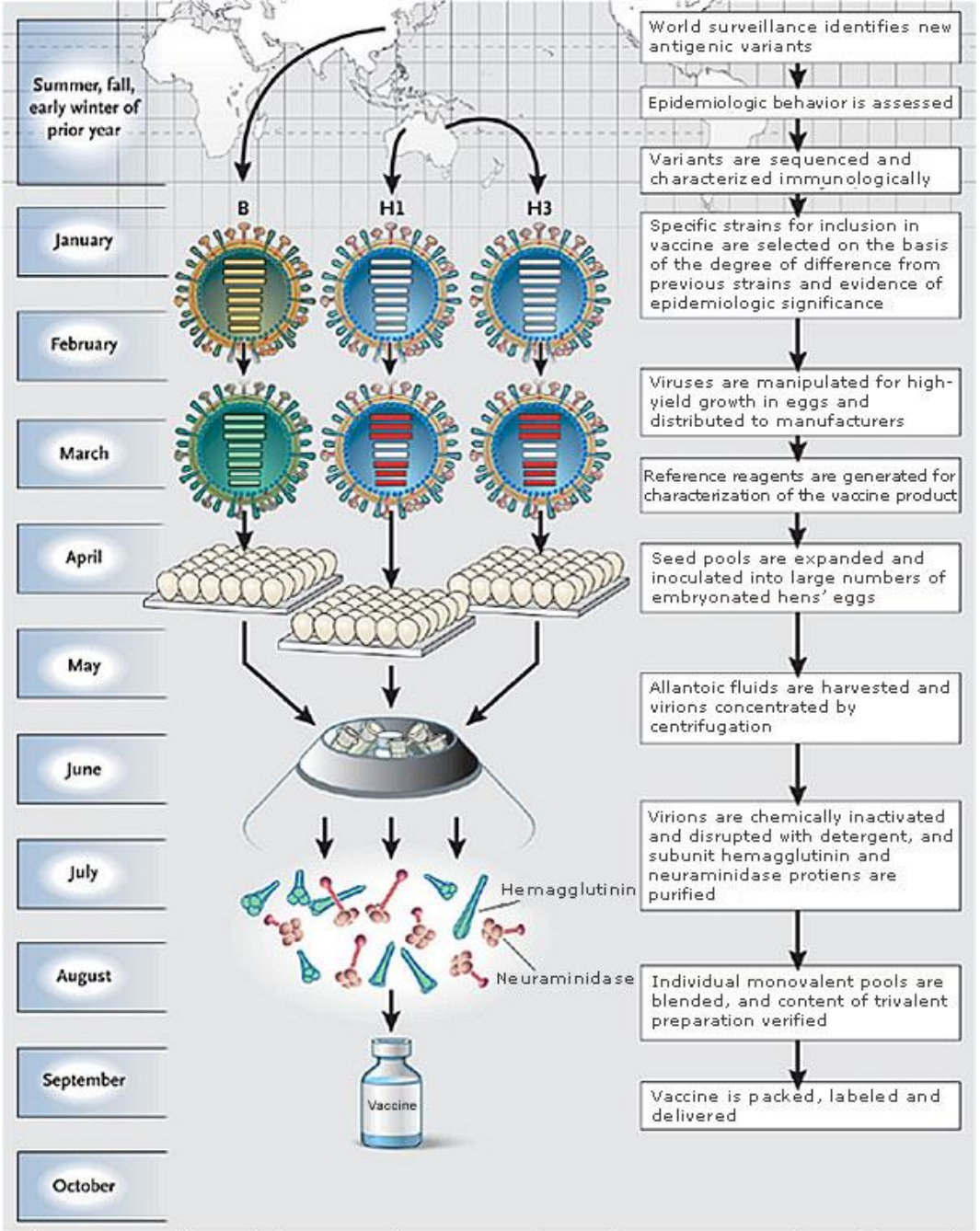


Diagram: Outline of the Annual Process of Development, Manufacturing and Distribution of Influenza Vaccines

N Engl J Med 2004;351:2037-40

World Health Organization

International Surveillance Network

Detects important antigenic variations of influenza virus in humans and animals (especially birds & pigs)

Data Summarised

Allows WHO to propose new recommendations for vaccine formulations for each Northern and Southern hemisphere influenza season annual vaccination and pandemics

WHO distributes seed strain to vaccine manufacturers including GSK

Start of 5-6 month vaccine production process

Month 1

Vaccine composition must match global epidemiological surveillance data, so an updated vaccine formulation is developed each time

Optimise/ establish working seed bank

Month 2

Innoculation of eggs, incubation

Month 3

Harvesting, purification

Month 3

Reagent standardisation

Month 3

Characterisation, sterility testing

Month 3

Filling, safety testing, release

Month 4

Clinical Trial (in Europe)

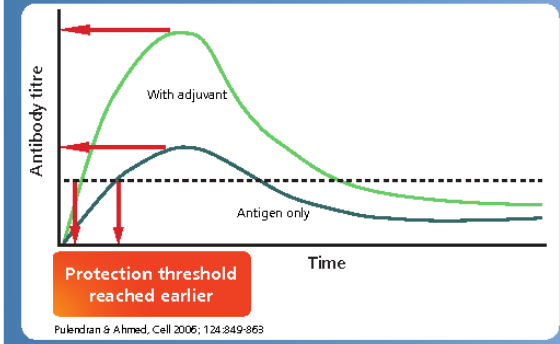
Month 4

Regulatory agency review and release

Month 5

Final vaccine = antigen + adjuvant

Month 5



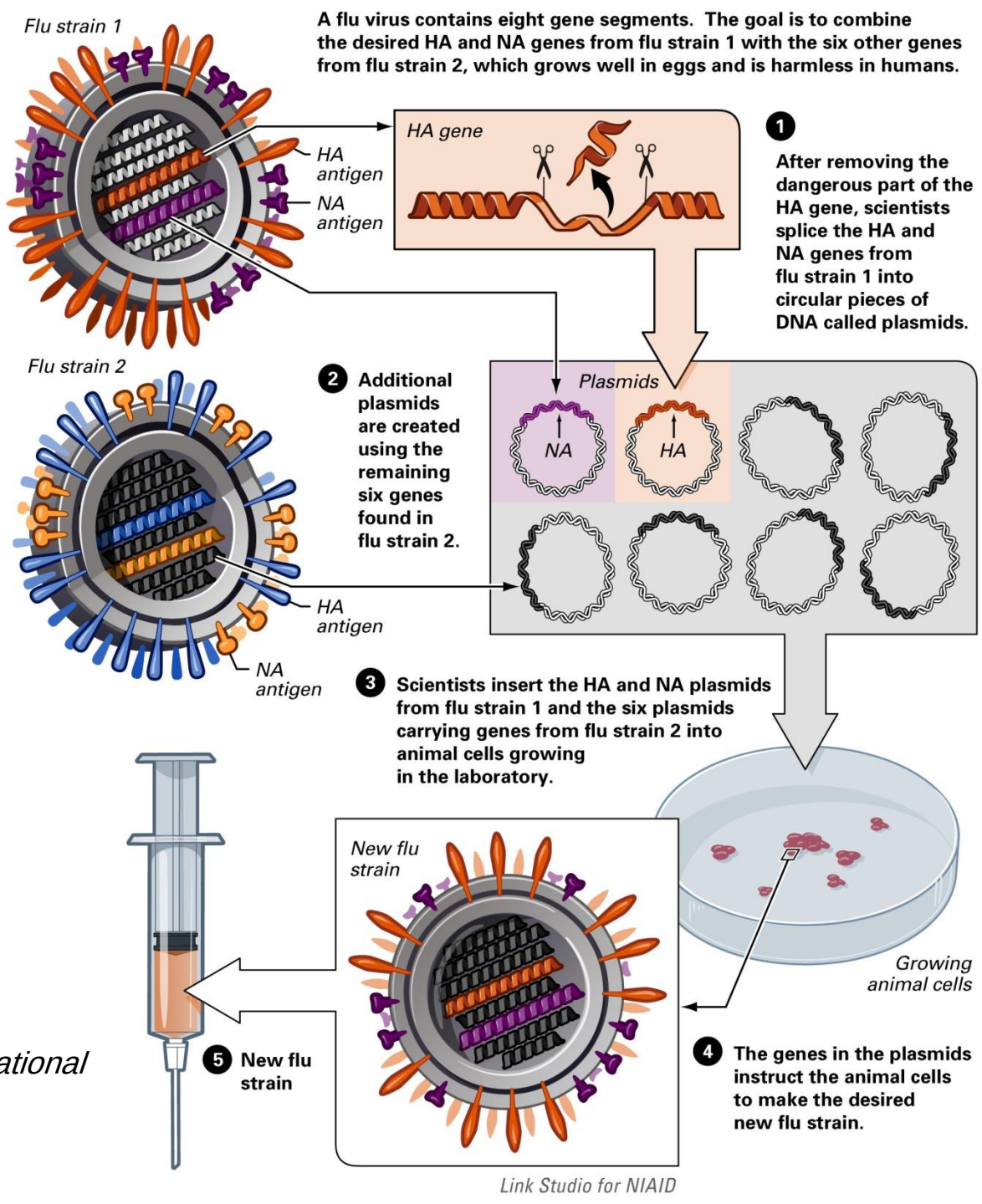
Increased efficiency of vaccines using adjuvant systems

GSK's flu vaccine development process

gsk GlaxoSmithKline

Reverse Genetics

Custom-make of a strain to be incorporated into a vaccine



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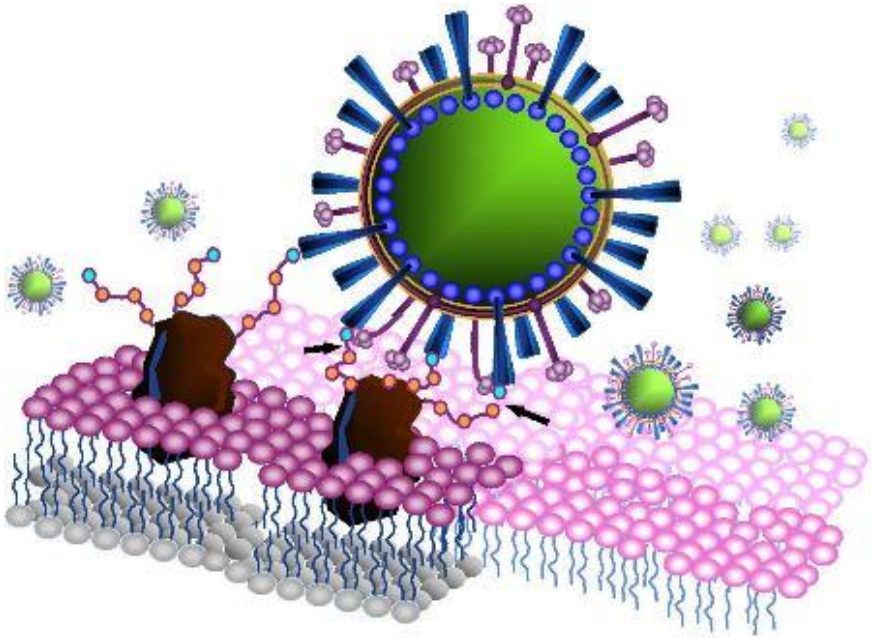
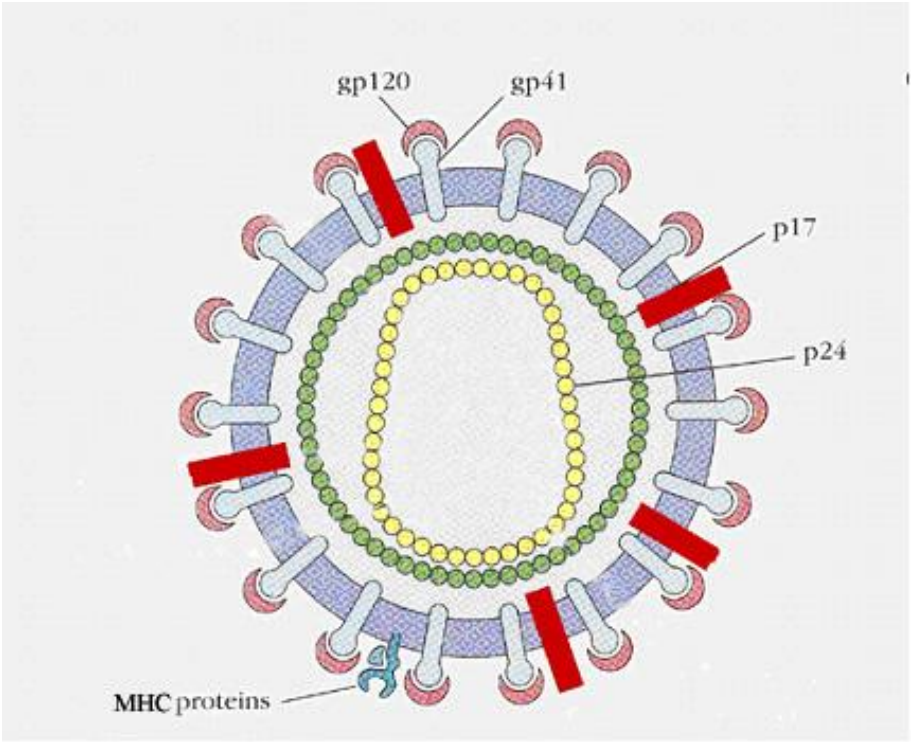
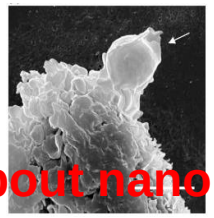
Virus-Like Particles (VLPs)

- **It is comprised of multiple copies of protein antigens that when assembled together mimic the appearance of a virus**

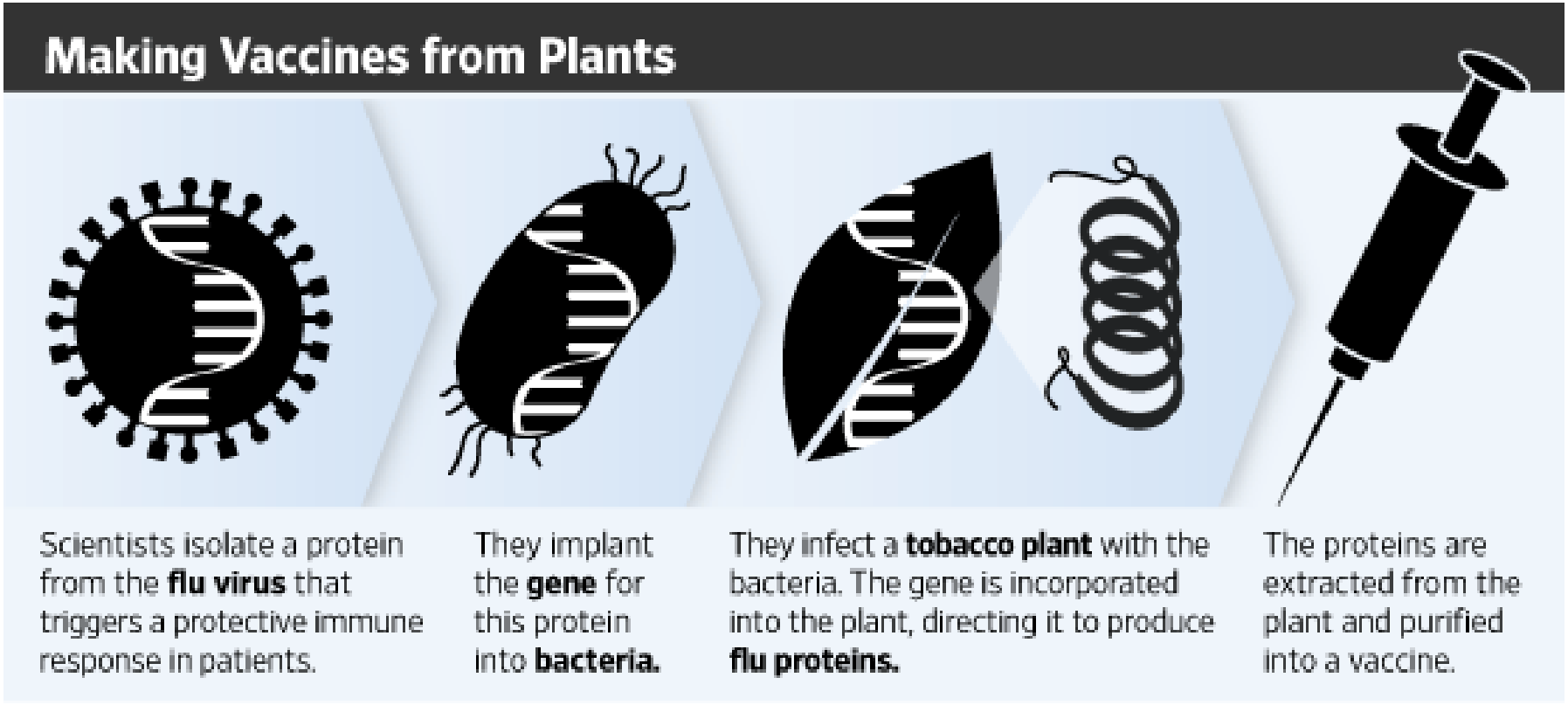






What about nanoparticles?

Making vaccines from Plants



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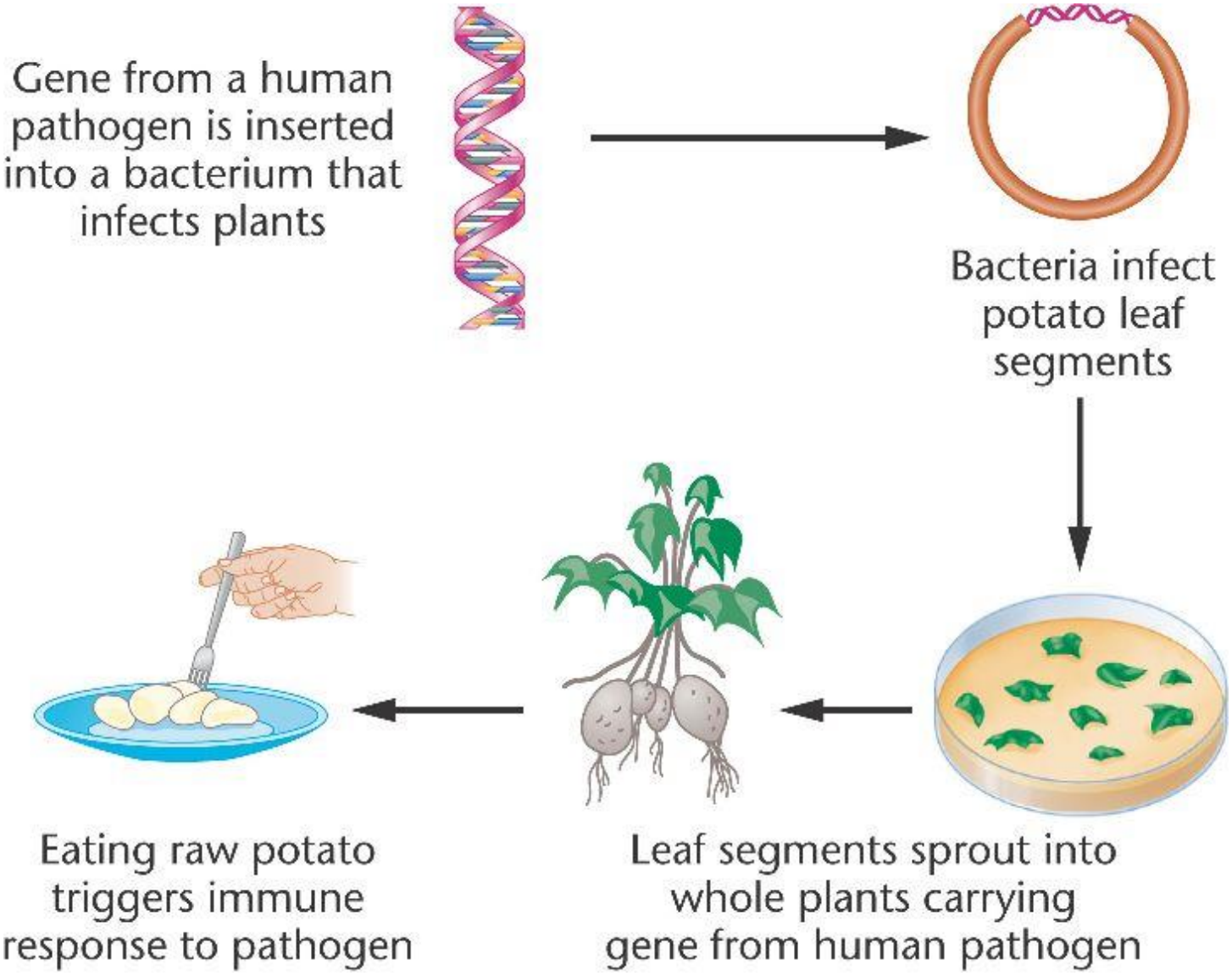

You are now able to:

- ✓ Describe how vaccines work
- ✓ Realize the significance of vaccination
- ✓ Understand the technology vaccine manufacturing
- ✓ Compare between traditional and biotechnology-based vaccines