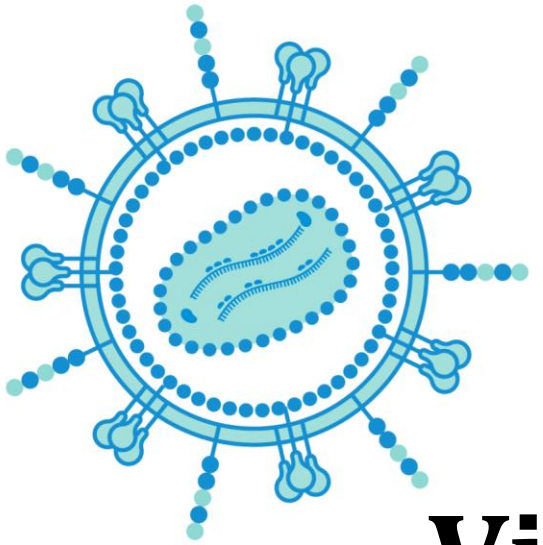
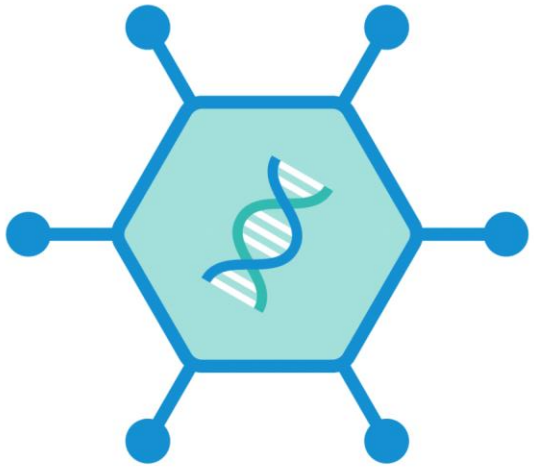




Viral Vectors and Gene Therapy

Summer Term
Lecture no. 7 & 8

Asst. Prof. Dr. Tola A. Faraj

Viral Vectors and Gene Therapy

→ Learning Objectives

- Define **viral vectors** and **gene therapy**.
- Explain why viruses are useful for delivering therapeutic genes.
- Distinguish between **viral and non-viral gene-delivery systems**.
- Describe major viral vectors and their characteristics.
- Explain the basic mechanism of viral-vector-mediated gene therapy.
- Compare **in vivo** and **ex vivo** gene therapy.
- Discuss advantages, limitations, and safety concerns.
- Give examples of approved or clinically important gene therapies.

→ What is Gene Therapy?

Gene therapy is the treatment or prevention of disease by modifying genetic material in a patient's cells.

The therapeutic strategy may involve:

- **Gene addition** _ introducing a functional copy of a gene.
- **Gene replacement** _ replacing a defective gene.
- **Gene editing** _ precisely modifying DNA, for example using CRISPR-based systems.
- **Gene silencing** _ reducing expression of a harmful gene.
- **Cellular reprogramming** _ modifying cells to acquire a therapeutic function.

Basic concept

Disease-causing genetic defect → therapeutic genetic modification → improved cellular function → clinical benefit

→ Why Use Viruses?

Viruses have evolved to perform one task extremely efficiently:

- **Deliver genetic material into host cells.**

A viral vector uses this natural ability while removing or disabling much of the viral machinery responsible for causing disease and replication.

Simplified concept

✔ **Natural virus**

- Virus → enters cell → releases genome → replicates → produces new viruses

✔ **Viral vector**

- Modified virus → enters cell → delivers therapeutic genetic material → therapeutic effect
- The vector is therefore essentially used as a **biological delivery vehicle**.

→ What Is a Viral Vector?

- A **viral vector** is a genetically modified virus used to deliver genetic material to target cells without causing the normal viral disease.
- Typically, genes required for viral replication or pathogenicity are removed or modified and replaced, partially or functionally, with a therapeutic genetic load.

- **Important distinction**

A viral vector is **not necessarily a complete infectious virus**.

1. Many vectors are designed so that:
2. they cannot replicate efficiently in the patient's cells;
3. pathogenic genes are removed;
4. only selected viral components are kept;
5. the therapeutic gene is delivered to the target cell.

→ Major Viral Vectors

The most important viral-vector platforms include:

Vector	Major feature	Common applications
Adenovirus	High gene-expression capacity; generally non-integrating	Cancer therapy, vaccines, gene delivery
AAV	Small, relatively low immunogenicity; mostly episomal	Inherited genetic diseases
Lentivirus	Integrates into host genome; can transduce non-dividing cells	Ex vivo cell therapies
Retrovirus	Integrates into host genome; mainly dividing cells	Earlier gene-therapy approaches
Herpes simplex virus (HSV)	Large genome/payload; neurotropic	Nervous-system applications
Vaccinia/poxvirus	Large genetic capacity	Vaccines and cancer immunotherapy

Viral vectors in approved gene therapies

Adenovirus

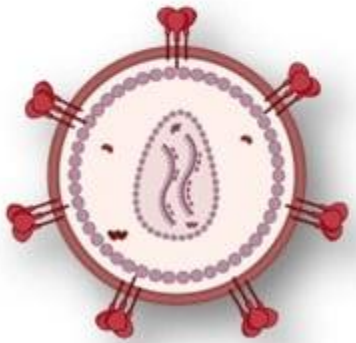
- 36 kb genome size
- Non-integrating vector
- High immunogenicity
- Used in vaccines, oncolytic viral therapy, localized gene delivery

AAV

- 4.8 kb genome size
- Non-integrating vector
- Low immunogenicity
- Used in *in vivo* gene therapy
- Serotype varieties for tissue specificity

Retrovirus

- 8 kb genome size
- Integrating vector
- Risk for insertional mutagenesis
- Used in *ex vivo* cell-based gene therapies



Lentivirus

- 8 kb genome size
- Integrating vector
- Low immunogenicity
- Lower risk of mutagenesis
- Widely used in cell-based gene therapies

HSV

- 150 kb genome size
- Non-integrating vector
- Low immunogenicity
- Natural neuronal tropism

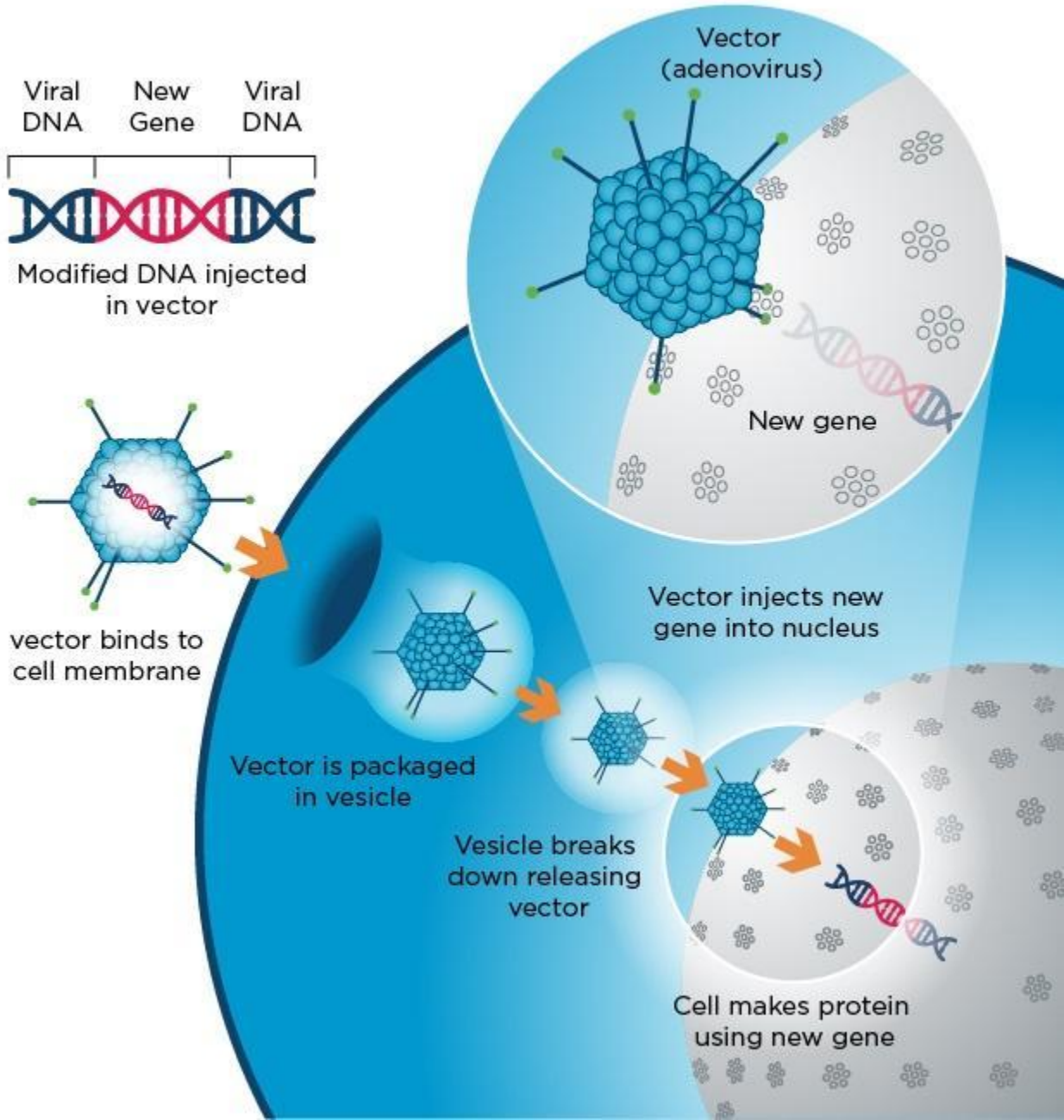
→ Adenoviral Vectors

Characteristics

- Double-stranded DNA genome
- Relatively large genetic capacity
- Efficient entry into many cell types
- Usually remain **episomal** rather than integrating into host DNA
- Can produce strong immune responses

Advantages

- Efficient gene delivery
- Broad tissue tropism
- High-level temporary gene expression
- Large payload compared with some other vectors



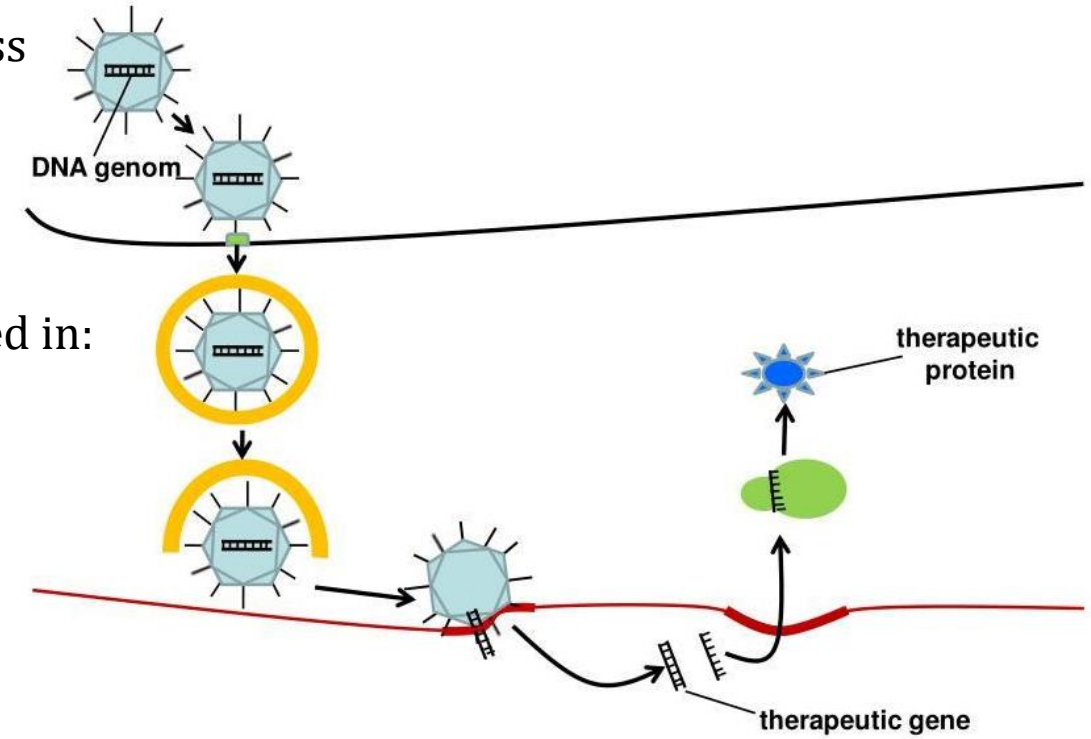
Limitations

- Pre-existing immunity can reduce effectiveness
- Strong inflammatory/immune responses
- Expression is often transient (short-lived)

Applications

Adenoviral vectors have been investigated or used in:

- Cancer therapy
- Vaccine development
- Experimental gene replacement
- Immunotherapy



→ Adeno-Associated Virus (AAV)

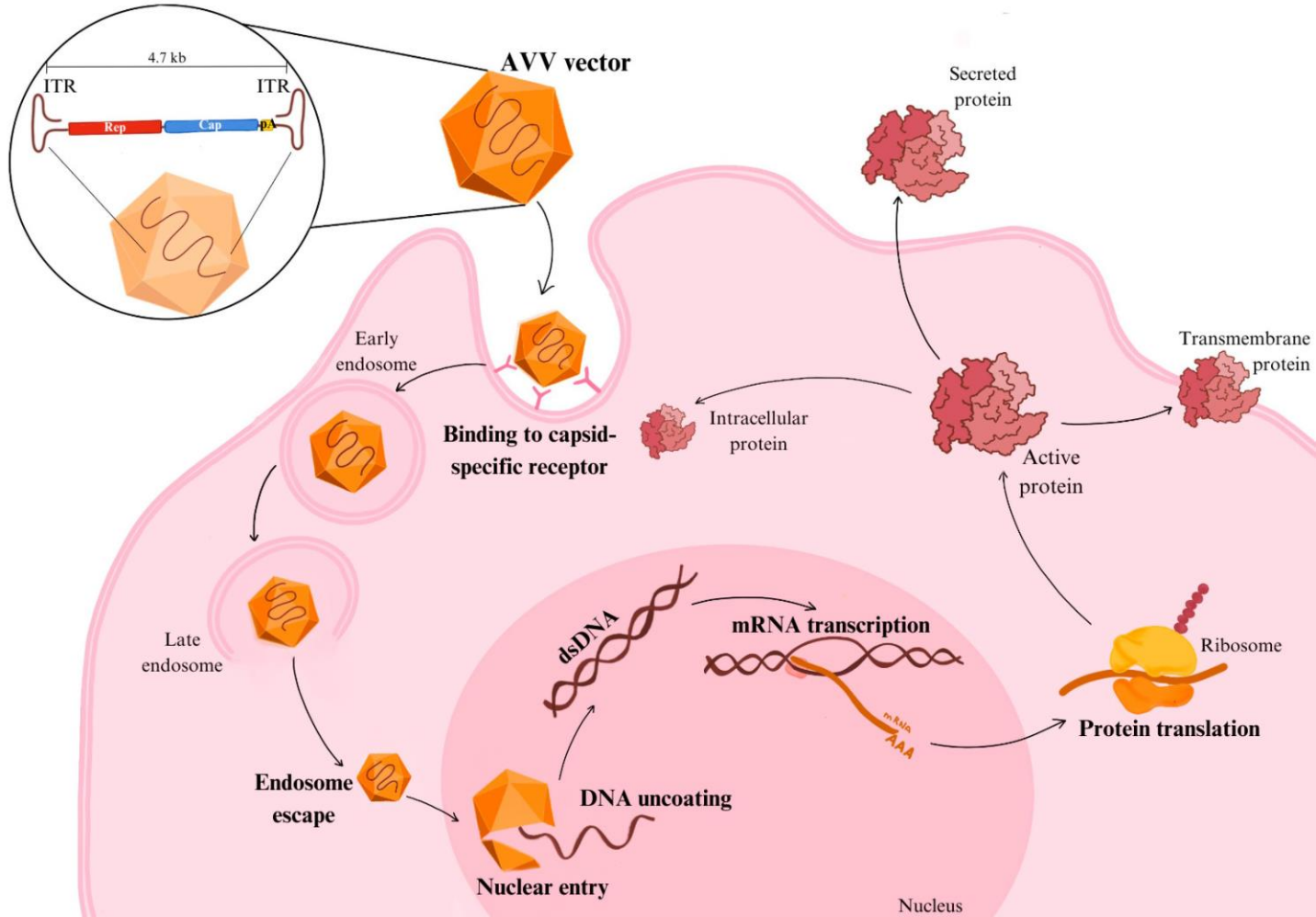
✔ **AAV** has become one of the most important platforms for modern gene therapy.

Characteristics

- Small, non-enveloped DNA virus
- Relatively small genetic payload
- Can infect many tissues depending on the serotype/capsid
- Therapeutic DNA generally continues mainly as **episomal DNA** in target cells

Major advantage

- AAV can provide **long-lasting expression** in some non-dividing tissues.



Major limitation

- Its packaging capacity is small.
- This means that very large therapeutic genes can be difficult to deliver using conventional AAV.

Examples of applications

AAV-based therapies have been developed for diseases involving:

- Retina
- Liver
- Muscle
- Nervous system
- Blood-clotting pathways

→ In Vivo *vs* Ex Vivo Gene Therapy

In vivo
The vector is administered **directly into the patient**.
Vector → patient → target tissue → therapeutic gene expression

Examples can involve administration through:

- Intravenous delivery
- Local injection
- Intramuscular injection
- Intrathecal administration
- Subretinal administration

Ex vivo
Cells are **removed from the patient and modified outside** the body.
Patient → cell collection → genetic modification → cell expansion/testing → reinfusion

Why use ex vivo therapy?
It allows researchers to:

- Select or improve modified cells
- Test the cells before administration
- Expand the therapeutic cell population
- Manipulate immune cells outside the patient

→ Gene Therapy for a Genetic Disease

- Imagine a patient has a mutation resulting in production of a defective protein.

Normal situation

- **Normal gene → normal protein → normal cellular function**

Disease

- **Mutated gene → defective/absent protein → abnormal cellular function → disease**

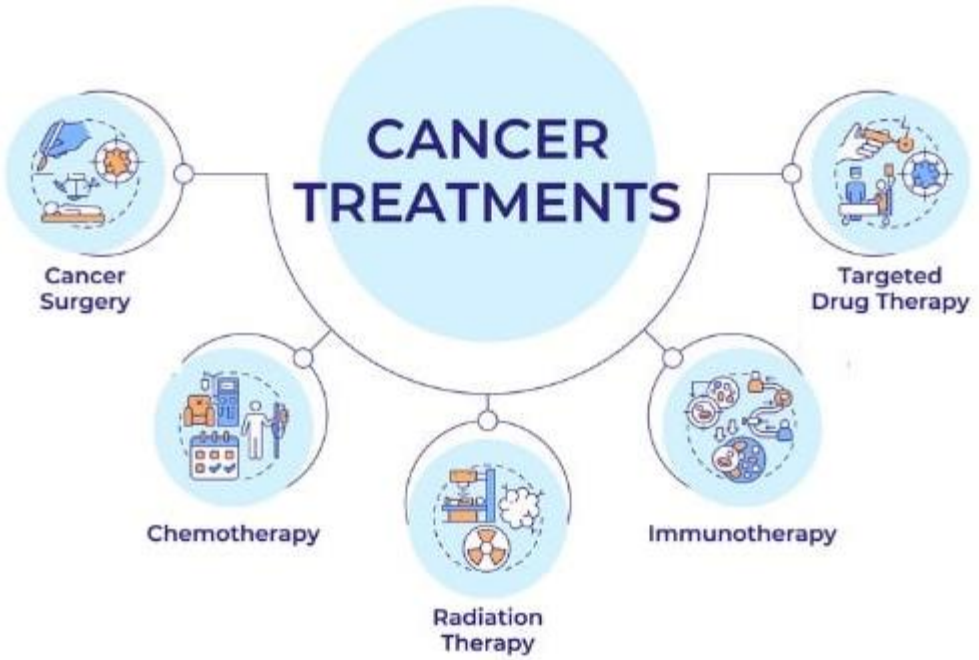
Gene therapy

- A viral vector delivers a functional gene:
- **Viral vector → target cell → therapeutic gene → functional protein → improved cellular function**
- This is the fundamental concept behind many gene-replacement therapies.

→ Viral Vectors and Cancer Therapy

Viral vectors are also important in **cancer treatment**.
They can be designed to:

- Deliver tumor-suppressor genes
- Stimulate anti-tumor immunity
- Deliver genes that cause selective tumor-cell killing
- Modify immune cells
- Express therapeutic proteins
- Function as oncolytic viruses



<u>Oncolytic viruses</u>

- Some therapeutic viruses are designed to **specially infect and destroy tumor cells**.

- This represents an important connection between:

Virology + Immunology + Oncology + Gene therapy

Viral Vectors and Immunotherapy

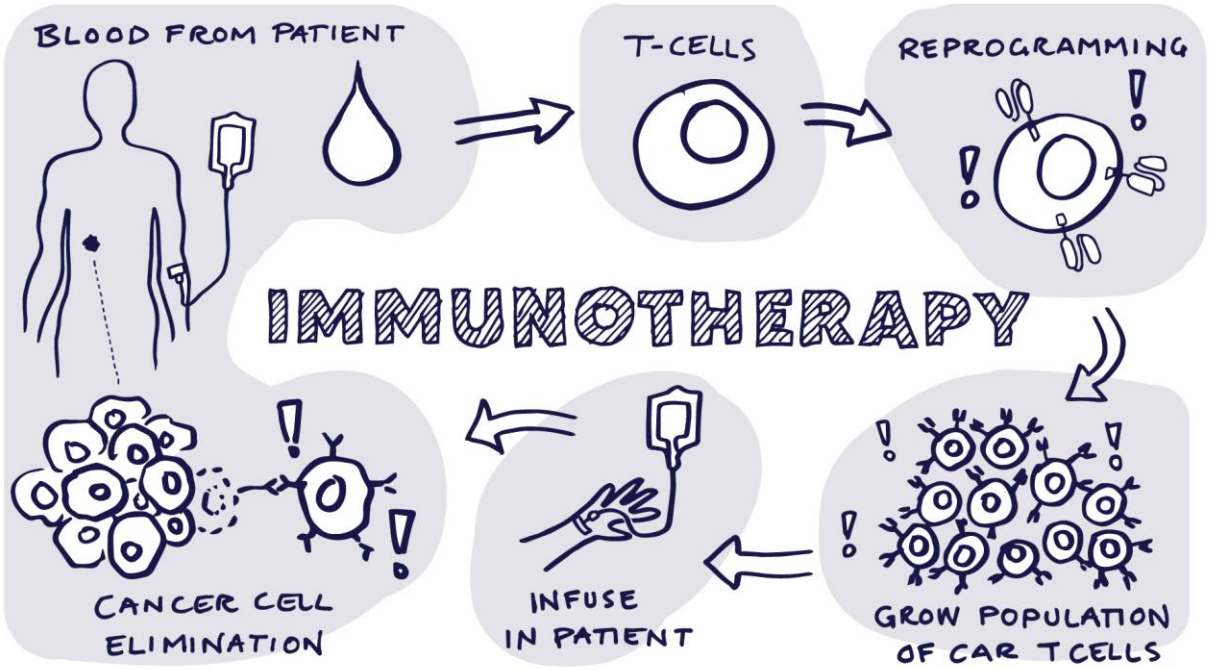
- Viral vectors can deliver genes encoding molecules that modify immune responses.

For example:

- **T cell → genetic modification → enhanced tumor recognition → expansion → administration to patient**
- This concept is central to several forms of modern cellular immunotherapy.

The broader field therefore connects:

- **Viral vector technology → genetic engineering → immune-cell manipulation → cancer treatment**



Thank You!