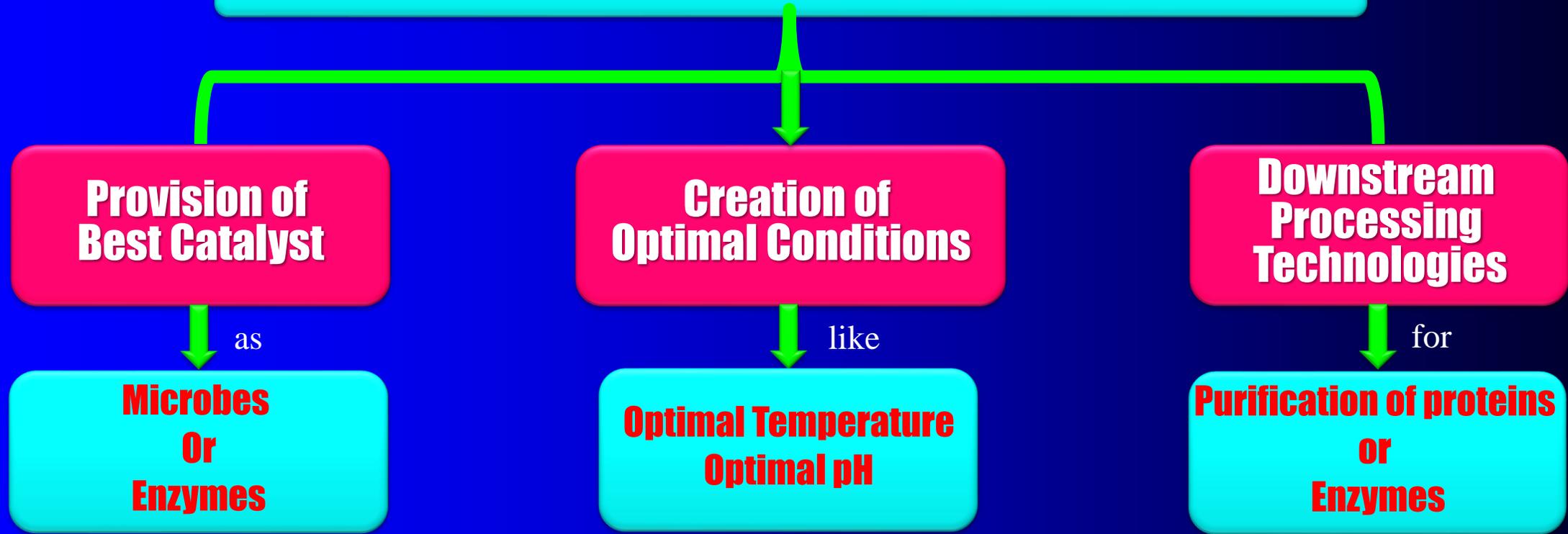


**Biotechnology and its
Applications**

Three Critical Research Areas of Biotechnology



Three Options to Increase Food Production

```
graph TD; A[Three Options to Increase Food Production] --> B[Agrochemical based Agriculture]; A --> C[Organic Agriculture]; A --> D[Genetically Engineered Crop based Agriculture];
```

**Agrochemical based
Agriculture**

Organic Agriculture

**Genetically Engineered
Crop based Agriculture**

Green Revolution

Advantages of Green Revolution

The **Green Revolution** succeeded in

- **Tripling the food supply** but yet it was not enough to feed the growing human population.
- Increased yield is

Partly due to the use of improved crop varieties. but

Mainly due to the use of better management practices and use of agrochemicals (fertilizers and pesticides).



Green Revolution

Limitations of Green Revolution

- Agrochemicals are **too expensive**.
- **Further increase in yield** with existing varieties are not possible using conventional breeding.



Advantages of GM Crops

Tailor made plants

Supply

**Starch, Fuels,
Pharmaceuticals**

Pest Resistant Plants

To reduce the amount of

Pesticides

Minimize the use of

Fertilizers



Bt Cotton

Bt Cotton

Why does this toxin not kill the Bacillus?

- The Bt toxin protein exists as inactive *protoxins*.
- When an insect ingests the inactive toxin, it is converted into an active form of toxin due to the alkaline pH of the gut which solubilise the crystals.
- The activated toxin binds to the surface of midgut epithelial cells and create pores that cause cell swelling and lysis and eventually cause death of the insect.



Bt Cotton

Bt toxin protein exists as **Inactive Protoxins**

↓
Converted into

Active form of toxin due to the **alkaline pH** of the gut
which solubilise the crystals.



Binds to the surface of **midgut epithelial cells**



Cause **cell swelling and lysis** and leads to **death** of the insect.



Bt Cotton

B. thuringiensis forms protein crystals during a particular phase of their growth. These crystals contain a toxic **insecticidal protein**.

Some strains of *Bacillus thuringiensis* produce proteins that kill certain insects such as lepidopterans (tobacco budworm, armyworm), coleopterans (beetles) and dipterans (flies, mosquitoes).



Some strains of *Bacillus thuringiensis*

Produce

Toxic insecticidal protein crystals during a particular phase of their growth

Kill insects such as

Lepidopterans (tobacco budworm, armyworm)

Coleopterans (beetles)

Dipterans (flies, mosquitoes).



Bt toxins are coded by cry genes

**Proteins encoded by the genes
cry I Ac and cry II Ab**

**Proteins encoded by the gene
cry I Ab**

Control

Controls

Cotton bollworms

Corn borer



Pest Resistant Plants

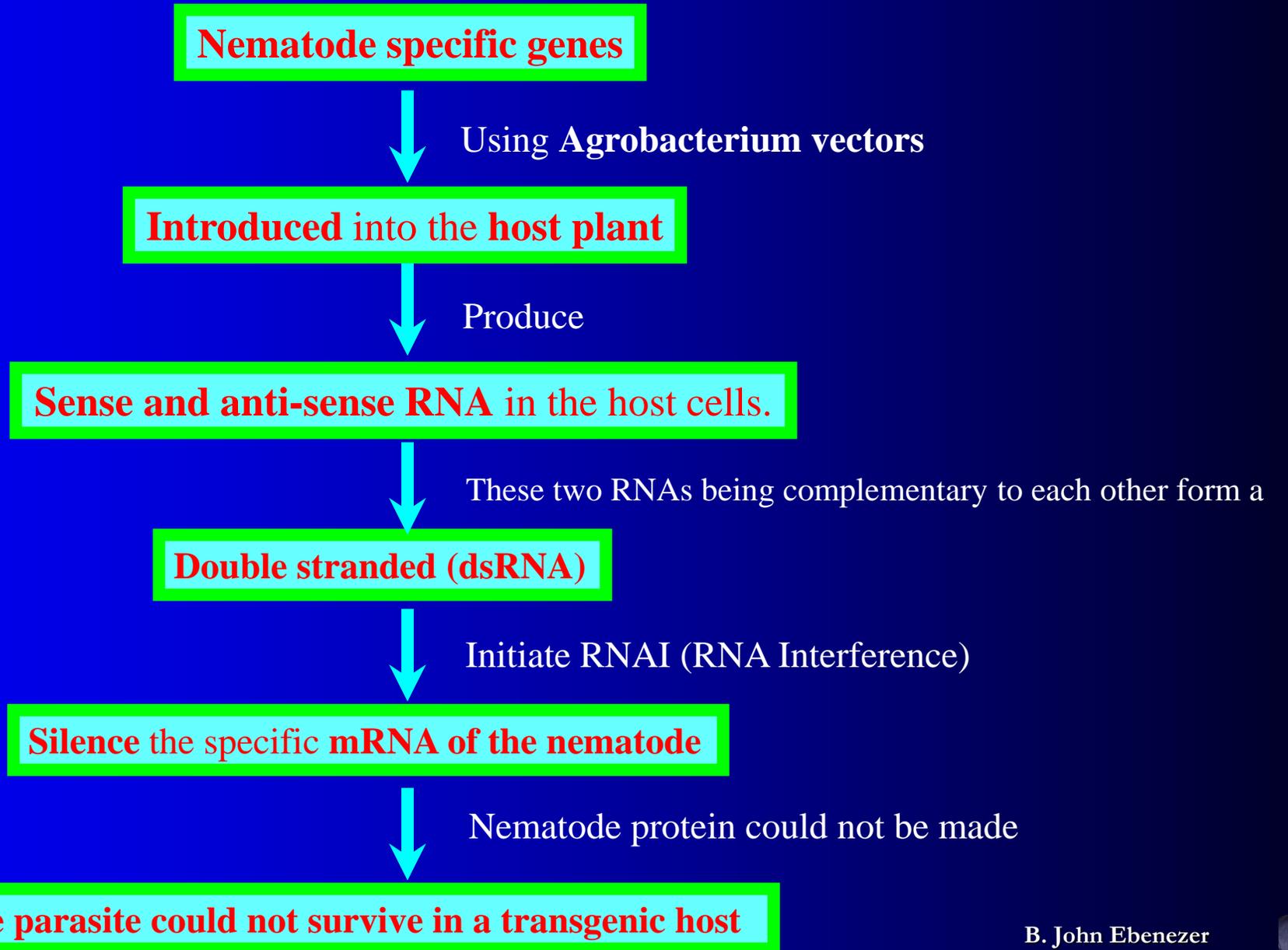
Source of RNA for RNAi

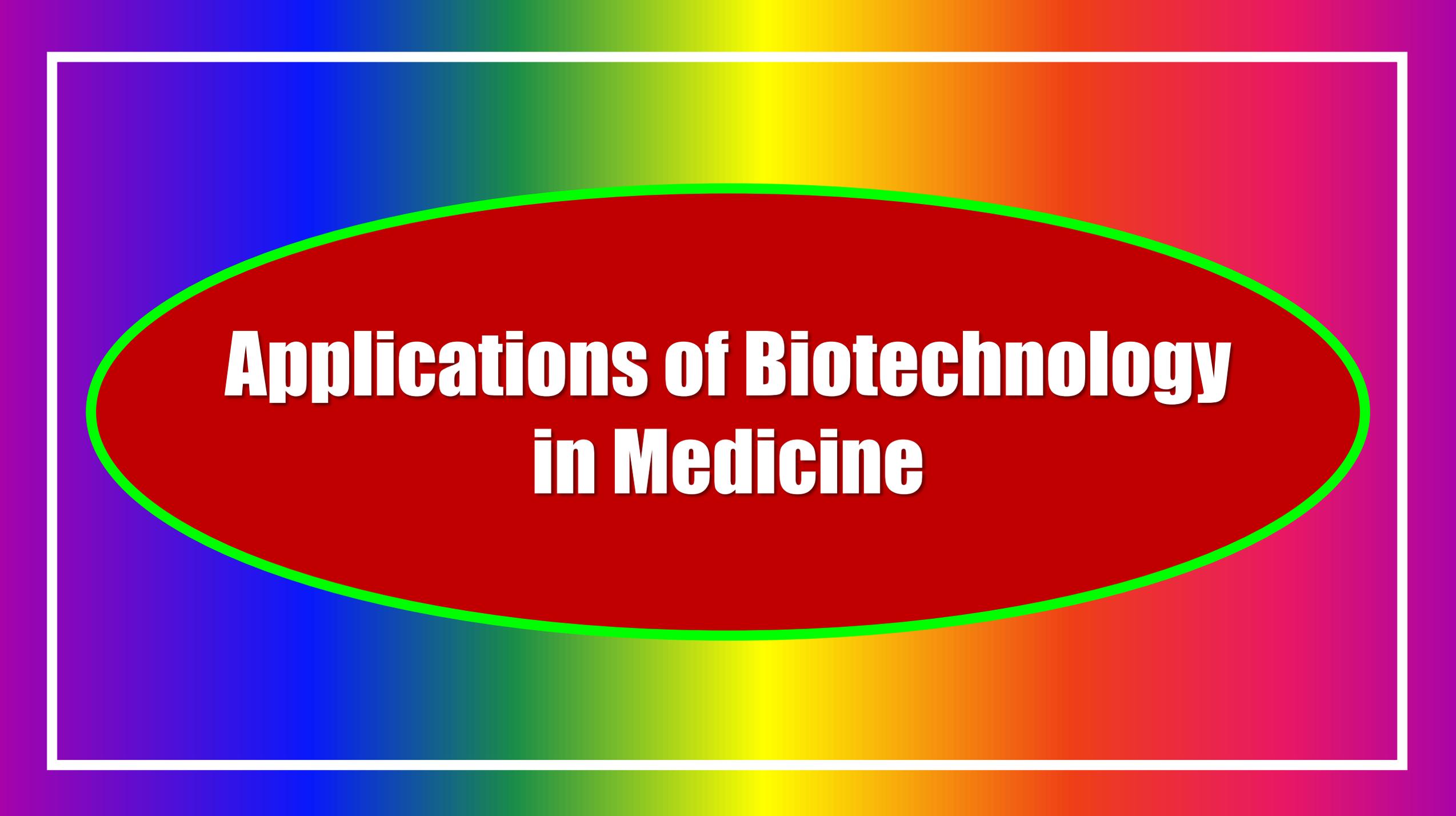
```
graph TD; A[Source of RNA for RNAi] --> B[Infection by RNA viruses]; A --> C[Transposons Mobile Genetic Materials Jumping Genes];
```

Infection by RNA viruses

**Transposons
Mobile Genetic Materials
Jumping Genes**

Strategy adopted to prevent the infestation of Nematode *Meloidogyne incognita*





**Applications of Biotechnology
in Medicine**

Biotechnological Applications in Medicine

The recombinant DNA technological processes have made **immense impact** in the area of healthcare by enabling **mass production of safe and more effective** therapeutic drugs.

The recombinant therapeutics **do not induce unwanted immunological responses** as is common in case of similar products isolated from non-human sources.

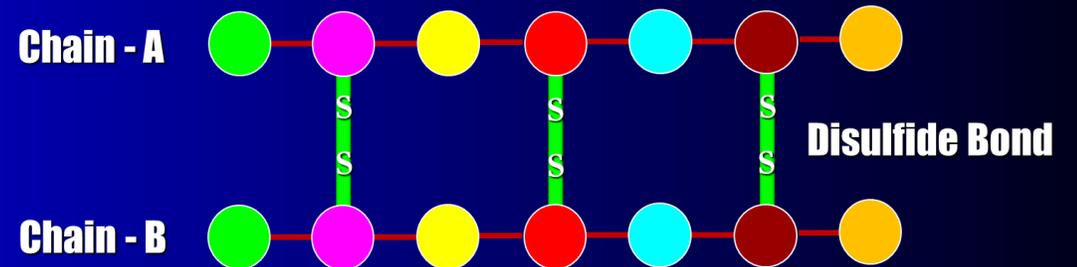
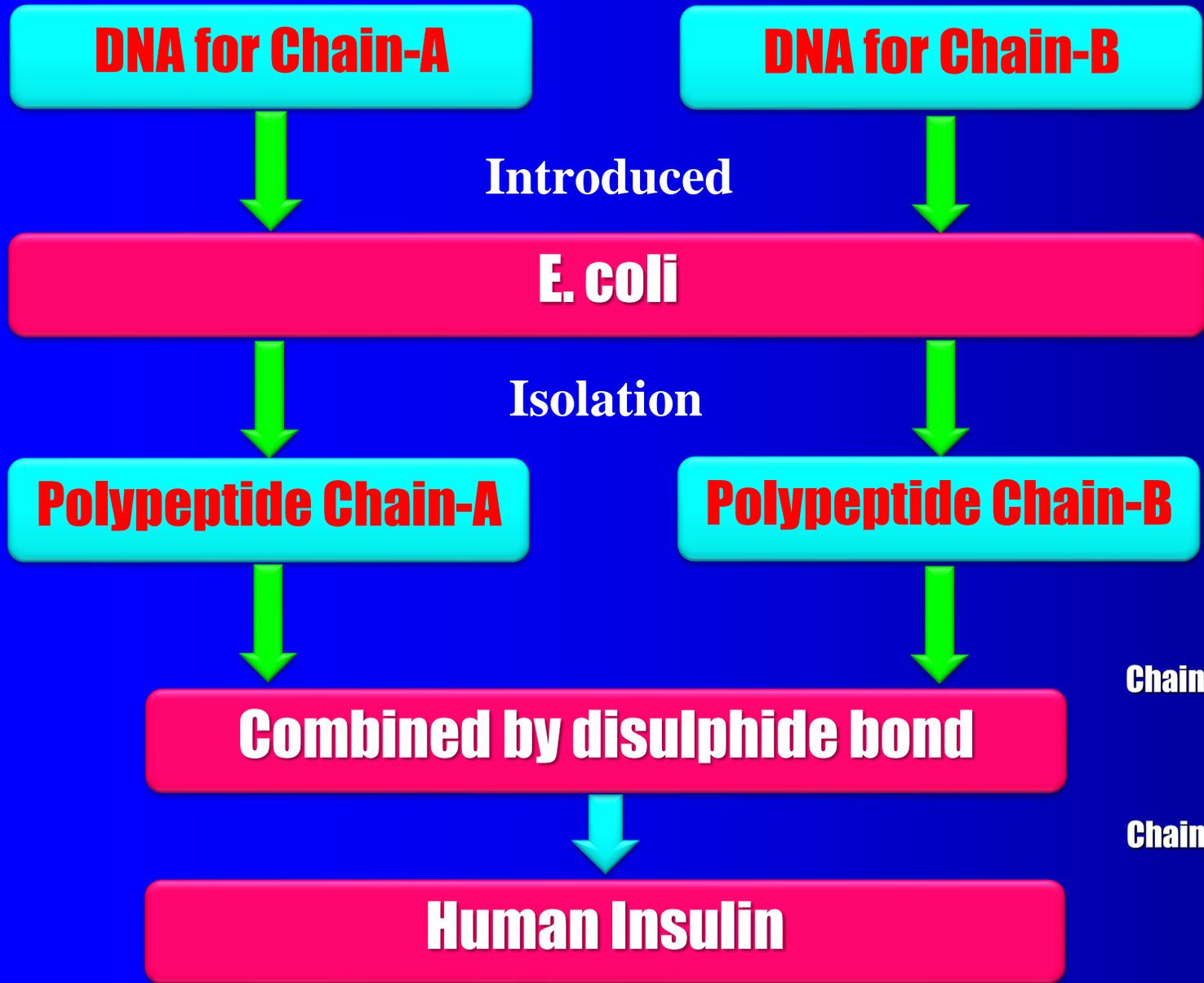
At present, about **30 recombinant therapeutics** have been approved for human use the world over.

In India, **12** of these **recombinant therapeutics** are presently being marketed.

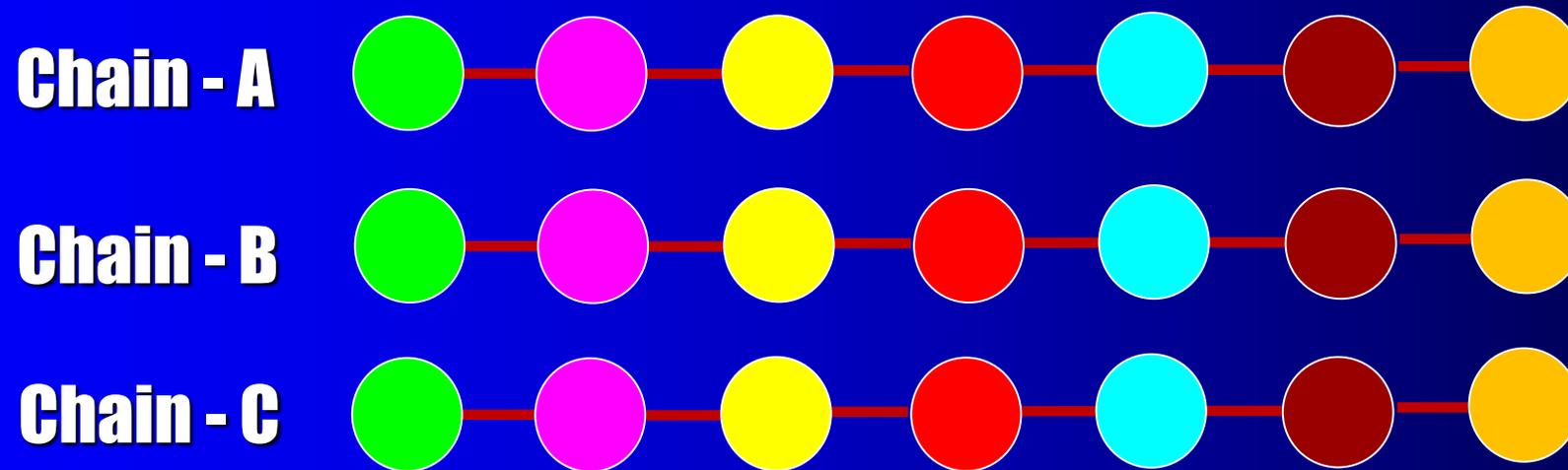


Genetically Engineered Insulin

Genetically Engineered Insulin



Structure of Insulin as a Pro-Hormone



Genetically Engineered Insulin

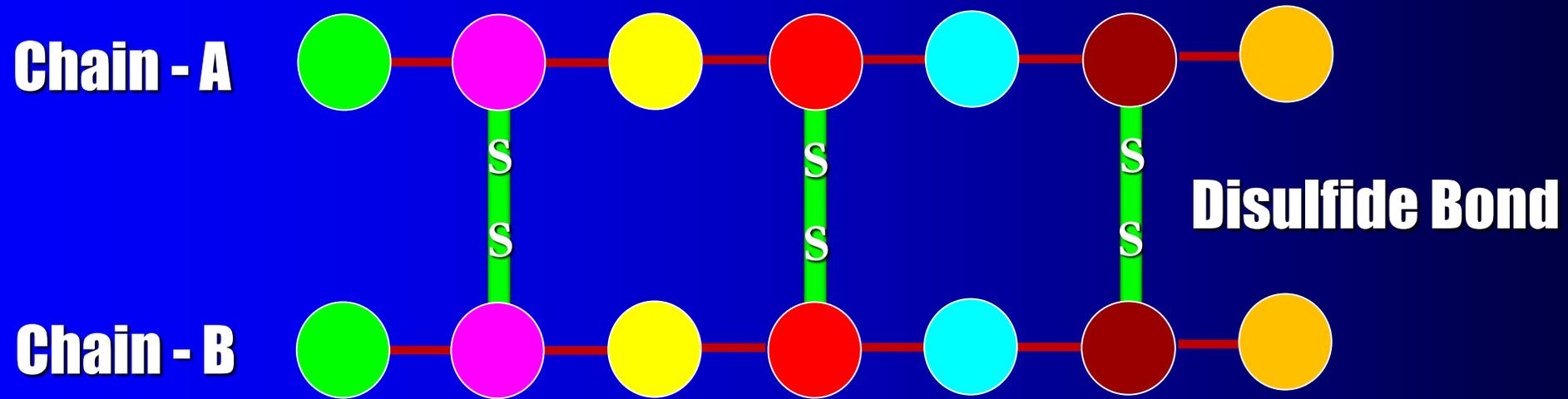
The main **challenge for production of insulin using rDNA techniques** was getting **insulin assembled into a mature form**.

In 1983, Eli Lilly an American company **prepared two DNA sequences** corresponding to the **chains A and B of human insulin** and introduced them in plasmids of *E. coli* to produce insulin chains.

Chains A and B were produced separately, which were isolated and combined by creating **disulfide bonds** to form human insulin.



Genetically Engineered Insulin



Human Insulin





Gene Therapy

Gene Therapy

Gene therapy is a collection of methods that allows correction of a gene defect that has been diagnosed in a child/embryo.



Gene Therapy

The first clinical gene therapy was given in 1990 to a 4-year old girl with **Adenosine deaminase (ADA) deficiency**.

This enzyme is **crucial** for the **immune system to function**.

The disorder is caused due to the **deletion of the gene** for adenosine deaminase.



Adenosine Deaminase Enzyme Disorder - ADA Gene Disorder



Treatment for ADA Deficiency

```
graph TD; A[Treatment for ADA Deficiency] --> B[Bone Marrow Transplantation]; A --> C[Enzyme Replacement Therapy]; A --> D[Gene Therapy];
```

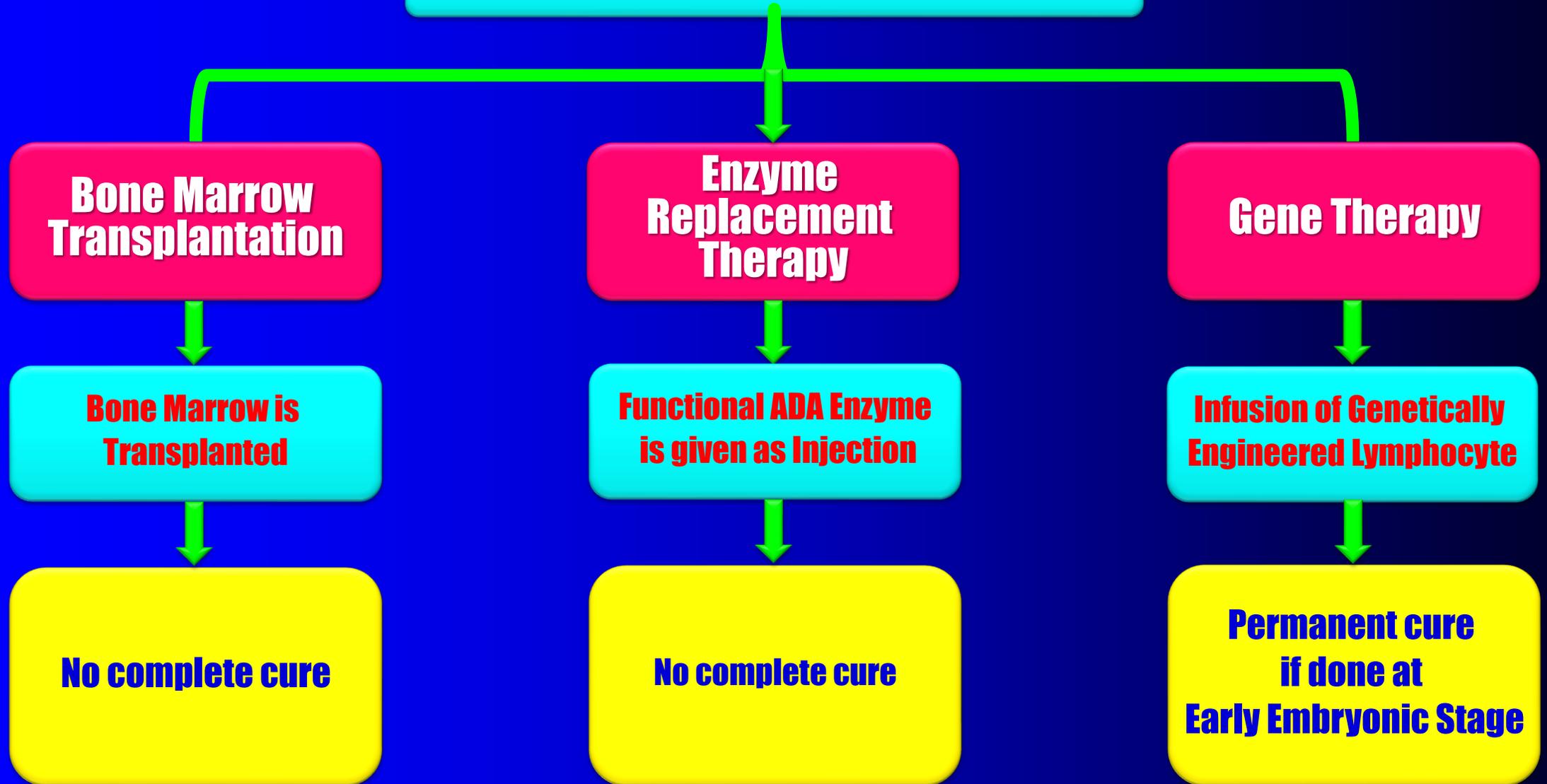
**Bone Marrow
Transplantation**

**Enzyme
Replacement
Therapy**

Gene Therapy

1. Bone marrow transplantation
2. Enzyme replacement therapy
3. Gene therapy

Treatment for ADA Deficiency



Isolation of Lymphocyte

From the
Blood of the
Patient

Functional ADA cDNA

Introduction
using
Retrovirus

Gene Therapy to cure ADA deficiency

Culture of Lymphocyte

Returned

Patient

Cure of the disease

A **functional ADA cDNA** is introduced into the lymphocytes using a retroviral vector, which are subsequently returned to the patient.

As these cells are not **immortal**, the patient requires **periodic infusion** of such genetically engineered lymphocytes.

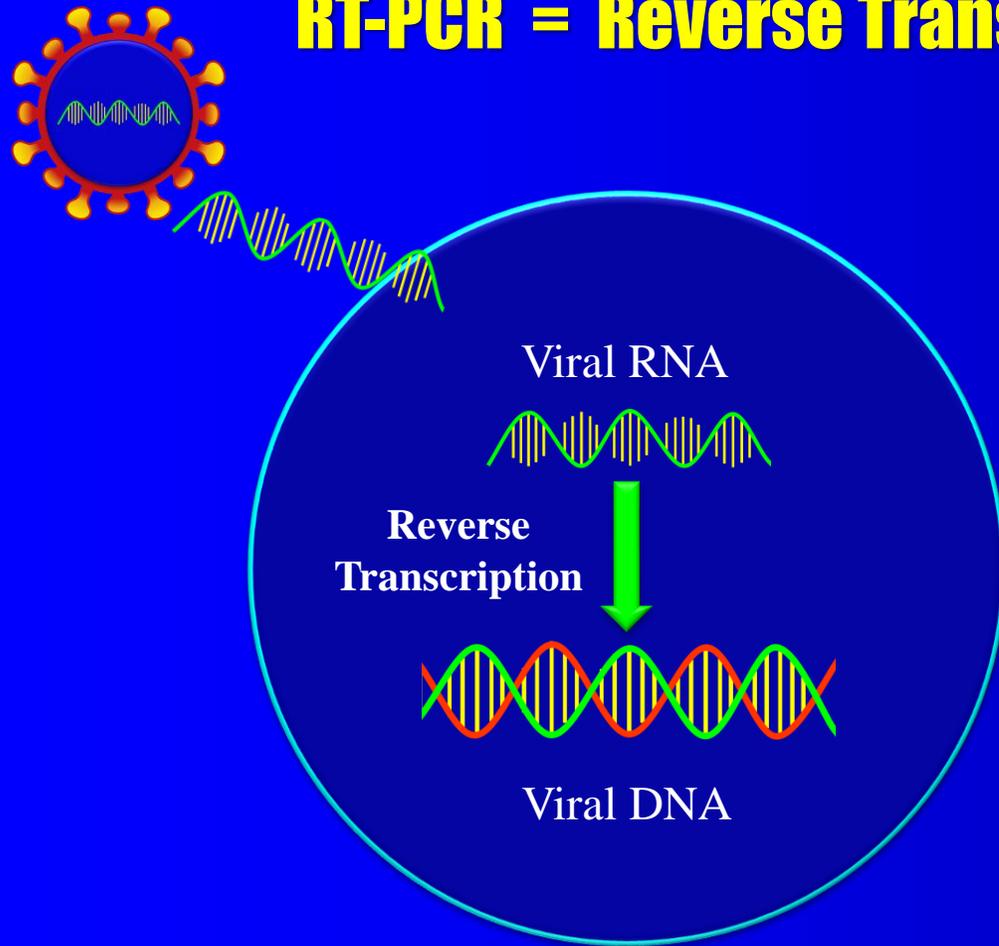
If the ADA gene isolate from marrow cells is introduced into cells of the patient at **early embryonic stages**, it could be a permanent cure.



Molecular Diagnosis

PCR - Polymerase Chain Reaction

RT-PCR = Reverse Transcription Polymerase Chain Reaction

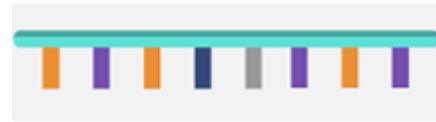


- PCR is now routinely used to detect HIV in suspected AIDS patients.
- It is being used to detect mutations in genes in suspected cancer patients too.
- It is a powerful technique to identify many other genetic disorders.



PCR

A single stranded DNA or RNA, tagged with a radioactive molecule (probe)



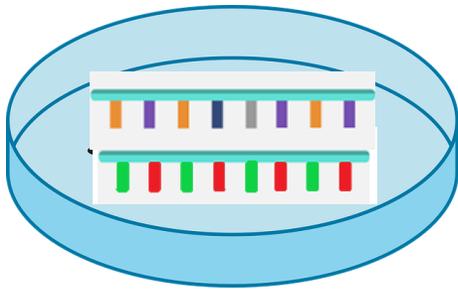
Probe

Hybridized with

Complementary DNA in a clone of cells



Mutated Gene



Detection using autoradiography

The probe does not have complementarity with the mutated gene in the clone.

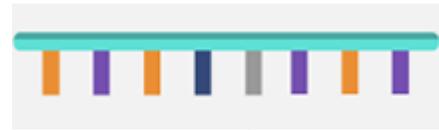
Positive PCR

DNA does not appear on photographic film



PCR

A single stranded DNA or RNA, tagged with a radioactive molecule (probe)



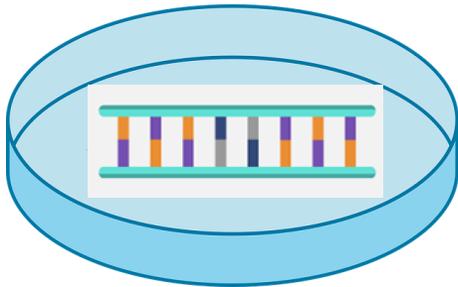
Probe

Hybridized with

Complementary DNA in a clone of cells

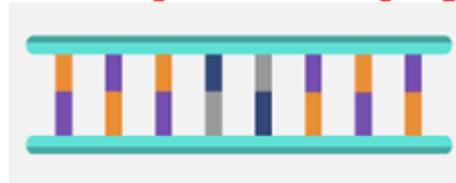


Non Mutated Gene



Detection using autoradiography

DNA

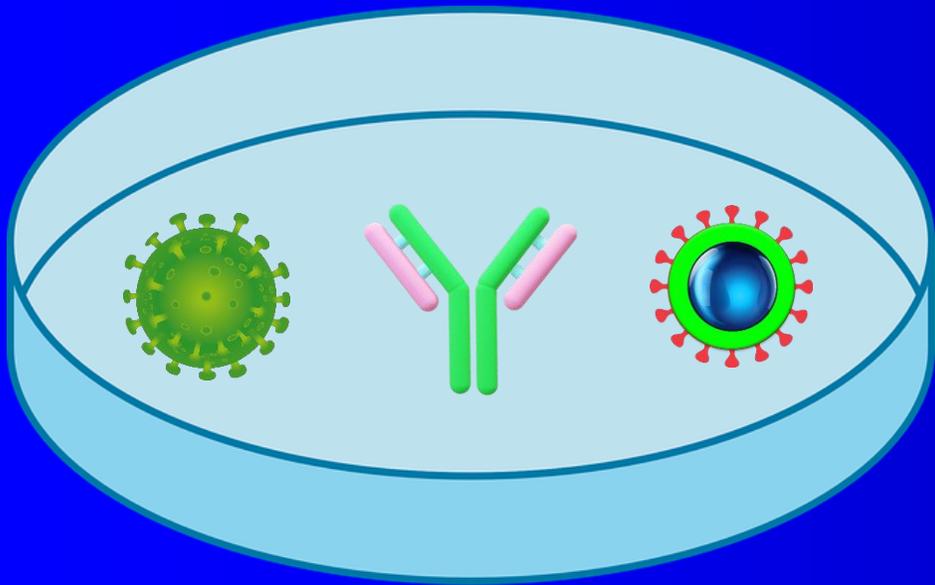


DNA appear on photographic film

Negative PCR



Enzyme Linked Immunosorbent Assay



ELISA is based on the principle of antigen-antibody interaction.

Infection by pathogen can be detected by the **presence of antigens** (proteins, glycoproteins, etc.) or by detecting the **antibodies** synthesised against the pathogen.



Transgenic Animals

Transgenic Animals

Animals that have manipulated DNA to possess and express an extra (foreign) gene are known as **transgenic animals**.

Transgenic rats, rabbits, pigs, sheep, cows and fish have been produced.

Over 95 per cent of all existing transgenic animals are mice.



Transgenic Animals

Normal physiology and development:

Transgenic animals can be specifically designed to allow the **study of gene regulation and how they affect the normal functions of the body and its development**, e.g., study of complex factors involved in growth such as insulin-like growth factor.

The **biological role of the genes** can be studied in the body by **introducing genes** from other species that alter the formation of this gene.



Transgenic Animals

Study of disease:

They increase our understanding about the **contribution of genes to the development of disease.**

They serve as models for human diseases so that investigation of new treatments for diseases is made possible.

Transgenic models exist for many human diseases such as cancer, cystic fibrosis, rheumatoid arthritis and Alzheimer



Transgenic Animals

Biological products:

Medicines required to treat certain human diseases can contain biological products, but such products are often expensive to make.

Biological products such as **human protein (α -1 -antitrypsin)** used to treat **emphysema** can be created by the introducing the portion of a DNA into the transgenic animals.

Similar attempts are being made for treatment of **phenylketonuria (PKU)** and cystic fibrosis.



Transgenic Animals

Vaccine safety:

Transgenic mice are being developed for testing the safety of vaccines before they are used on humans.

Transgenic mice are being used to test the safety of the **polio vaccine**.

If successful and found to be reliable, they could replace the use of monkeys to test the safety of batches of the vaccine.



Transgenic Animals

Chemical safety testing:

This is known as toxicity/safety testing. The procedure is the same as that used for testing toxicity of drugs.

As the transgenic animals carry genes, **they are more sensitive to toxic substances** than non-transgenic animals.

They are exposed to the toxic substances and the effects are studied.

Toxicity testing in such animals will allow us to obtain results in less time.





Ethical issues

Ethical Issues

Genetic modification may have **unpredictable results** in the ecosystem.

It has **created problems with patents** granted for the same.



Ethical Issues

There is growing public anger that certain companies are being granted patents for **products and technologies** that have long been **identified, developed and used** by farmers and indigenous people of a specific region/country.

GEAC (Genetic Engineering Approval Committee), will **make decisions** regarding the **validity of GM research** and the **safety of introducing GM organisms** for public services.



Ethical Issues

Rice is an important food grain, the presence of which goes back thousands of years in Asia's agricultural history.

There are an estimated **200,000 varieties of rice in India** alone.

The diversity of rice in India is one of the richest in the world.

Basmati rice is distinct for its **unique aroma and flavour** and **27 documented varieties of Basmati** are grown in India.

There is reference to Basmati in ancient texts, folklore and poetry, as it has been grown for centuries.



Ethical Issues

In 1997, an American company got **patent rights** on **Basmati rice** through the **US Patent and Trademark Office**.

This allowed the company to sell a **'new' variety of Basmati in the US and abroad**.

This 'new' variety of Basmati had actually been derived from Indian farmer's varieties.

Indian Basmati was crossed with **semi-dwarf varieties** and claimed as an invention or a novelty.



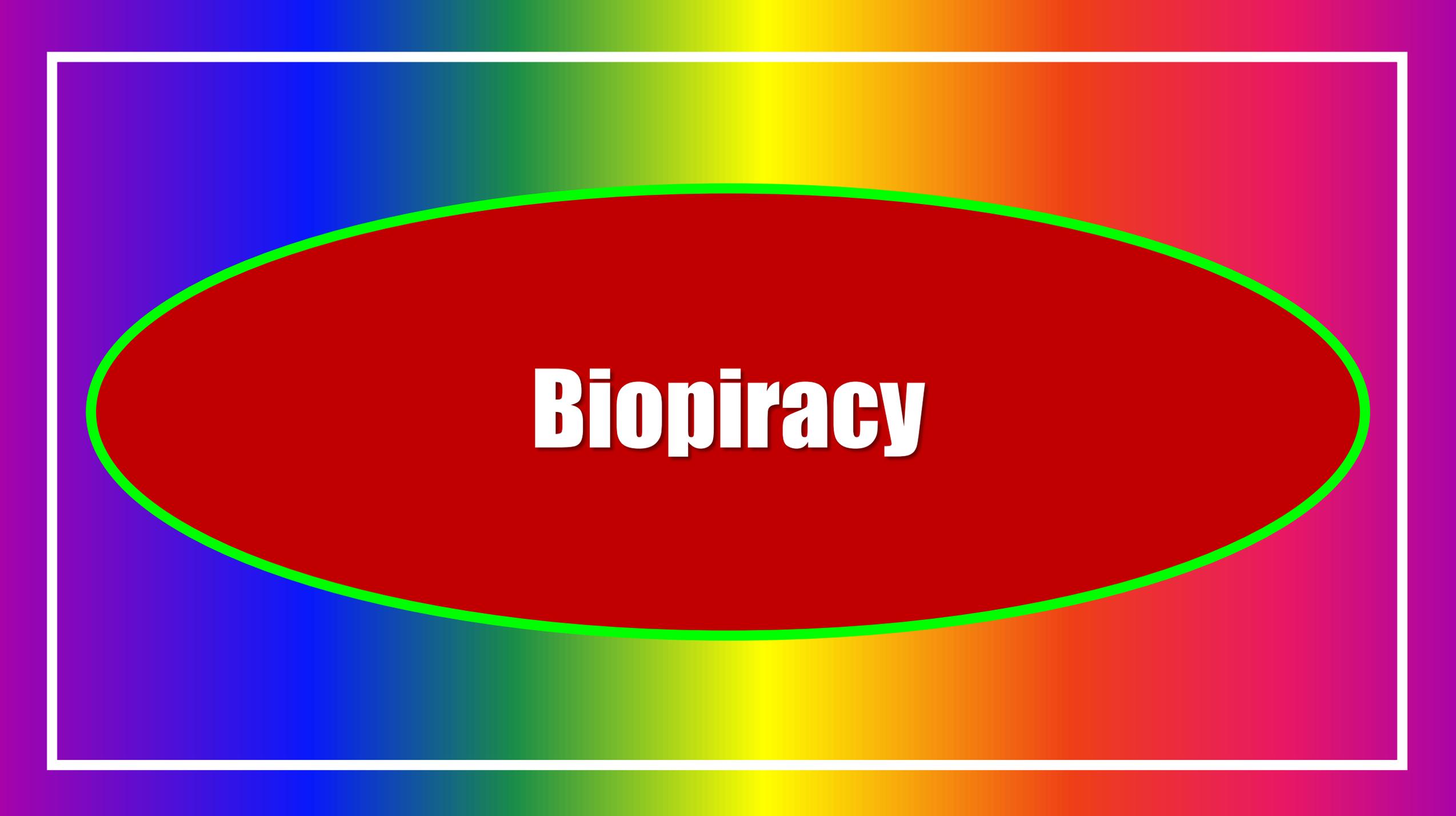
Ethical Issues

The patent could restrict other people selling Basmati rice.

Several attempts have also been made to **patent uses, products and processes** based on Indian traditional herbal medicines, e.g., **turmeric neem.**

If we are not vigilant and we do not immediately counter these patent applications, **other countries/individuals may encash on our rich legacy** and we may not be able to do anything about it.





Biopiracy

Biopiracy

Biopiracy is the **use of bio-resources** by **multinational companies** and other organisations **without proper authorisation** from the countries and people concerned without compensatory payment.

Most of the industrialised nations are rich financially but poor in biodiversity and traditional knowledge.

In contrast the developing and the underdeveloped world is rich in biodiversity and traditional knowledge related to bio-resources.



Biopiracy

Traditional knowledge related to bio-resources can be exploited to develop modern applications and can also be used to save time, effort and expenditure during their commercialisation.

There has been growing realisation of the **injustice, inadequate compensation and benefit sharing** between developed and developing countries.

Therefore, some nations are developing **laws to prevent such unauthorised exploitation** of their **bio-resources and traditional knowledge**.





God Bless You!