Applications of Recombinant DNA technologies :-

① Insulin :- Used in treatment of diabetes mellitus

Insulin is produced via the following steps :-

i) Gene Isolation :- human insulin gene is identified & isolated

ii) Insertion of this gene into ~~Plasmid~~ Plasmid :- The human insulin gene is inserted into plasmid of E.coli bacteria

iii) Introduction to Bacteria :- The plasmid (which get cut by restriction enzyme) combines with insulin gene

The insulin gene is inserted into the open site on the plasmid

iv) Ligation :- An enzyme called DNA ligase is used to seal the insulin gene into the plasmid forming → recombinant plasmid. DNA ligase acts like molecular glue, binding DNA fragments together.

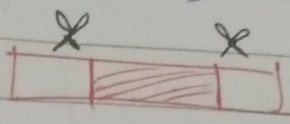
v) Alkaline lysis :- The recombinant plasmid is isolated from E.coli bacteria through a process called plasmid extraction/purification

vi) Reintroduction into E.coli bacteria :- This recombinant plasmid is re-introduced into E.coli bacteria during process called transformation where some bacteria/E.coli(s) may/may not pick up or take up the recombinant plasmid

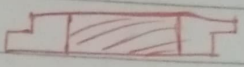
vii) Production of Insulin :- The bacteria(s) which do take up this recombinant plasmid will start using insulin gene to produce insulin proteins as they grow & multiply.

Process of Insulin Production :-

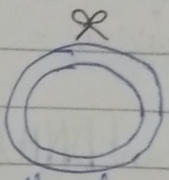
1) From human pancreatic cell, human insulin is extracted.



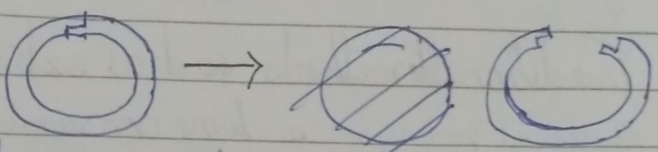
2) Restriction enzyme is used to cut the DNA



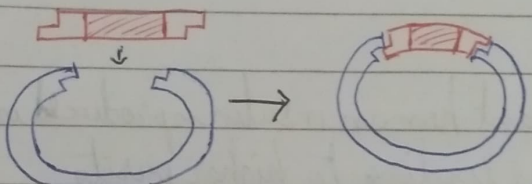
3) From *E. coli*, plasmid is extracted.



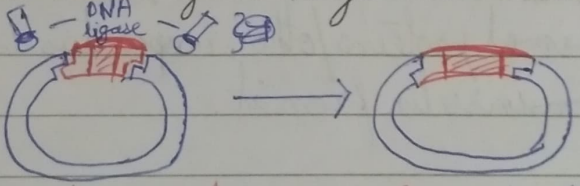
4) Restriction enzyme is used to cut this plasmid



5) Now, the insulin gene is introduced into this plasmid

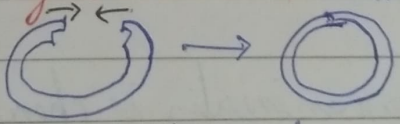


6) DNA ligase (molecular glue) is used to stick the plasmid and the insulin gene together.

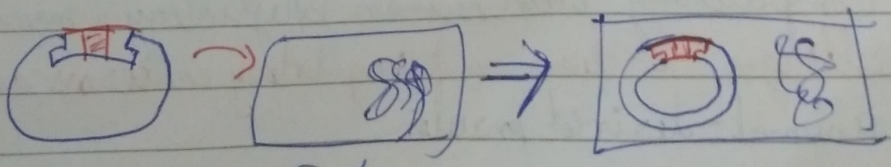


(Recombinant plasmid)

Some plasmids may not combine with insulin DNA and combine with itself :-



7) The ones which combine with insulin → tranformants.
These *E. coli* that take up the recombinant plasmid are the ones capable of expressing insulin gene.



(2) Hepatitis B vaccine :- Used in treatment of hepatitis B

(3) DNA vaccines :- Used in treatment of malaria, influenza etc

(*) Insulin is cheaper and safer compared to animal insulin :-

1) Cost-Effectiveness :-

Recombinant DNA technology allows large scale production of human insulin using E.coli or yeast.

Traditional methods such as extracting insulin from animal pancreas is time consuming & higher cost.

2) Purity :-

Recombinant human insulin is produced in controlled laboratory conditions, leading to higher purity.

In case of animal pancreas, there is risk of contamination with animal proteins/other impurities when extracting ~~DNA from~~ insulin from it.

Summary :-

- Recombinant human insulin is cheaper due to its efficient production processes and reduced resource use.
- It is also safer because of its purity, consistency and compatibility with human physiology, minimizing allergic reactions and regulatory concerns associated with animal-derived insulin.

(4) Gene Therapy - Replacement of defective or missing gene with a corrected gene.

Gene variants :- Are changes in genetics due to the cell's age, exposure to certain chemicals/environmental factors. Cells often recognise these changes and repair them. Other times, if left unchecked can cause diseases & treatment is needed. This is where gene therapy comes into play.

If genes are blueprint to our body, then gene therapy can fill in missing parts/errors in the drawing/blueprint.

Hence, Gene therapy is the use of genetic material to treat or prevent disease from happening.

Fixing a defective gene :-

Step 1:- Viral Vector :- Virses are very good at entering and harming our body. Therefore, Modified virses are used for delivery of genetic material. These modified viral vectors don't harm body.

Step 2:- Gene Modification :- The defective gene is identified, and a functional copy of this gene is cultured in the lab. This involves using synthetic DNA or cloning normal gene from a healthy individual/human.

Step 3:- Packaging :- The cultured gene is then included into the viral vector and this vector is responsible for transporting the gene to the target cells.

Step 4:- Cell Restoration :- Once inside cell, viral vector releases its genetic material into host cell's nucleus. The host cell transcribes and translates the new gene producing functional protein