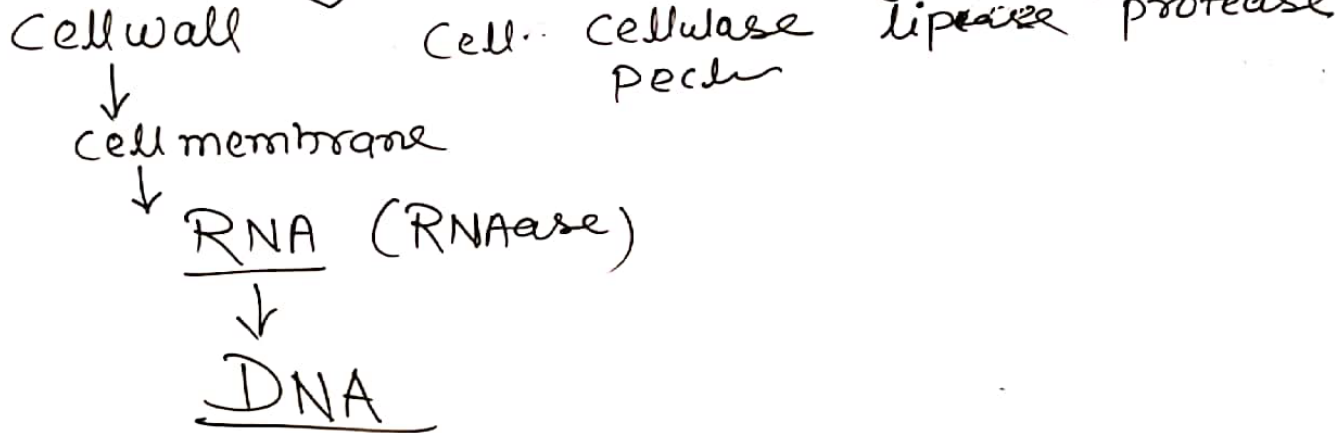
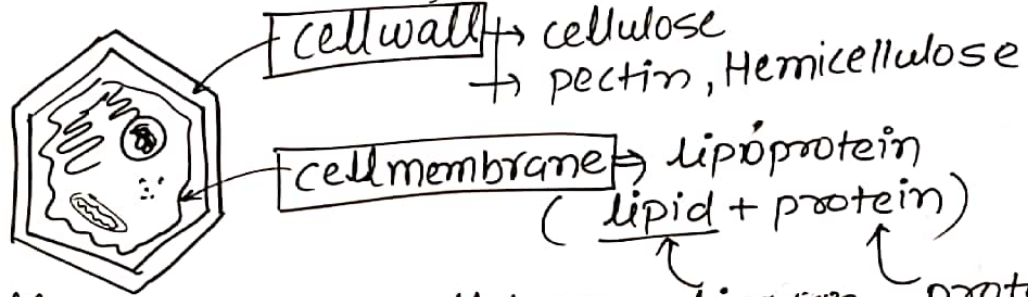


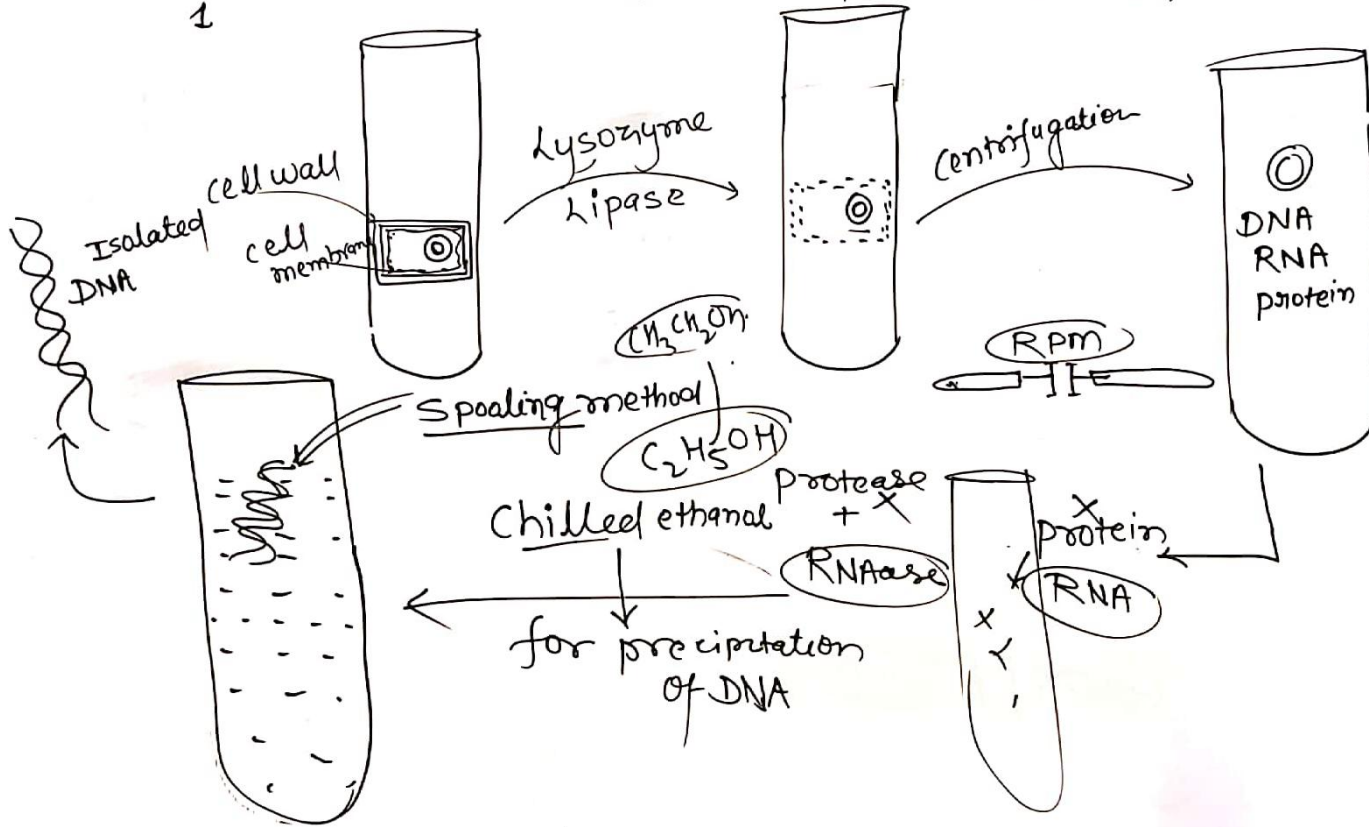
Isolation of DNA → from bacterial cells.

1

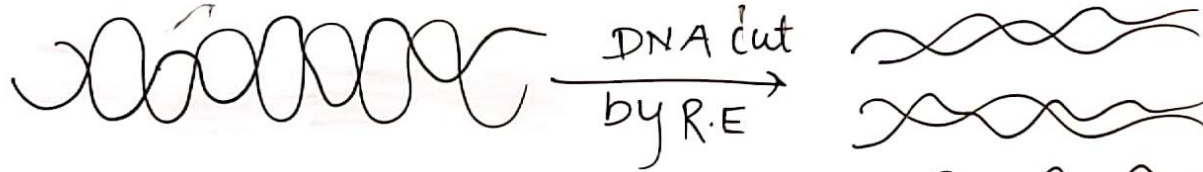


Isolation of DNA from bacterial cells.

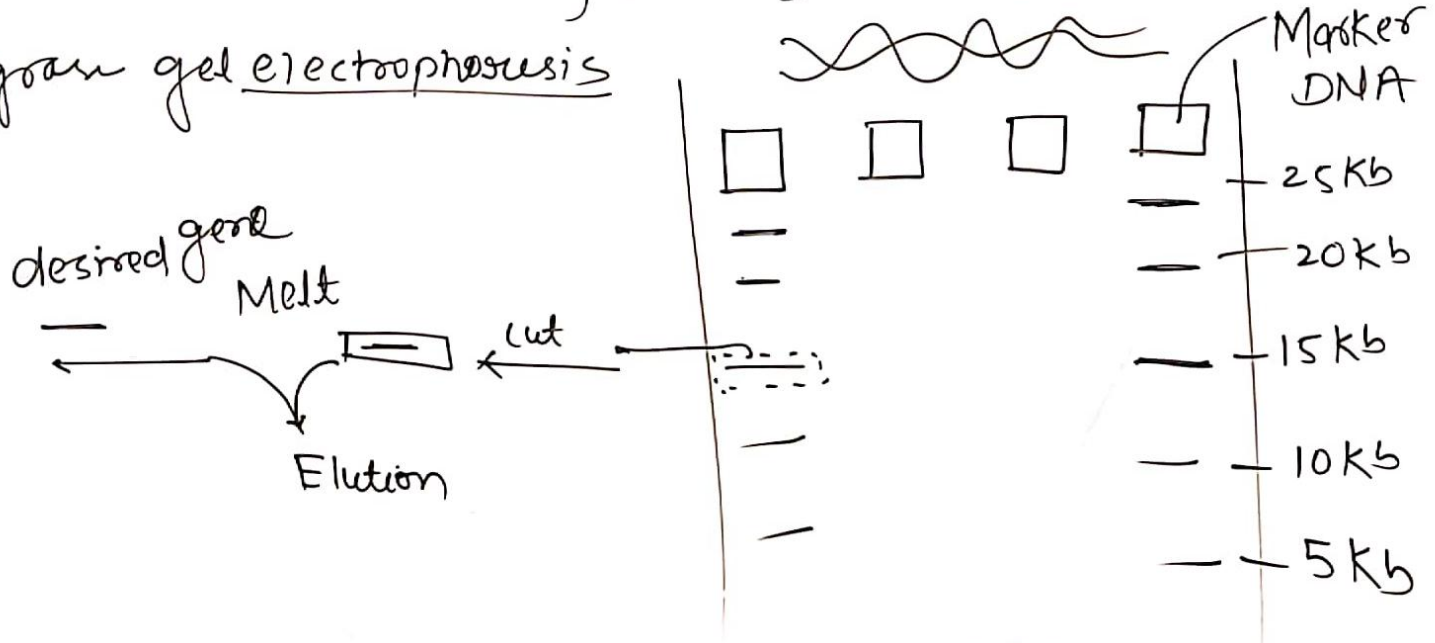
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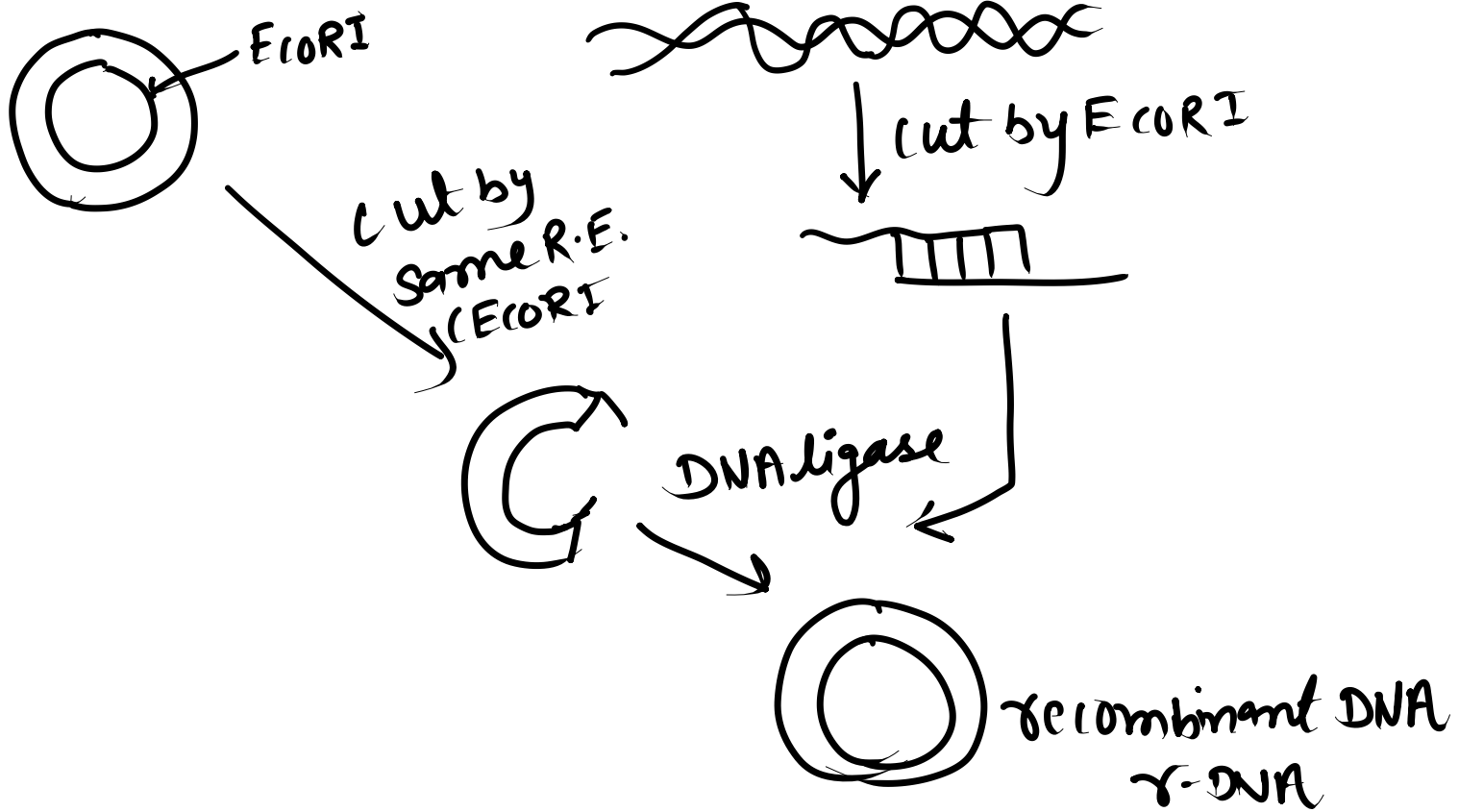
Fragmentation of DNA by Restriction Endonuclease.



Agarose gel electrophoresis

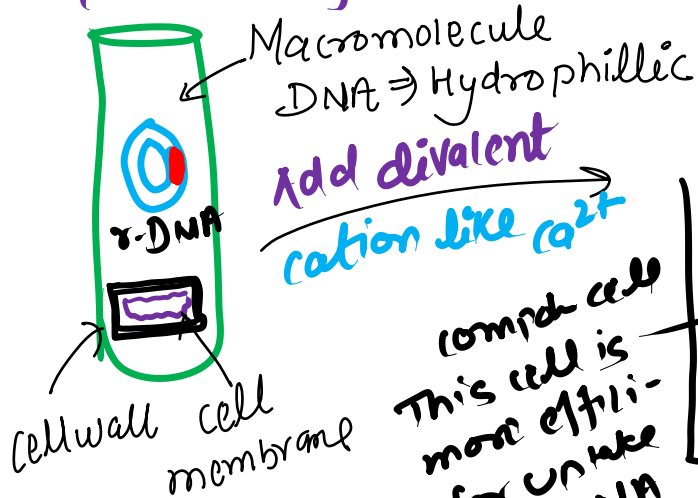


Ligation of the DNA fragment into a Vector



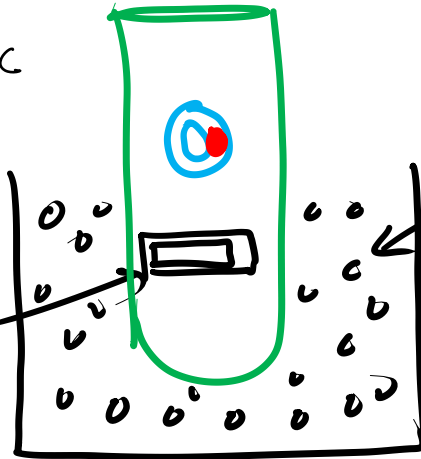
GENE TRANSFER

Indirect (vector) Gene transfer in bacteria

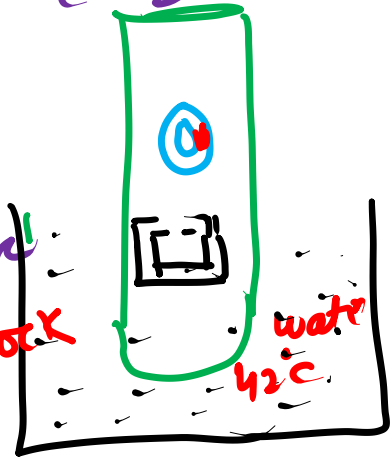


Add divalent cation like Ca^{2+}

competent cell
This cell is more efficient for uptake of DNA



Sudden transfer at 42°C for 2 minutes
heat shock



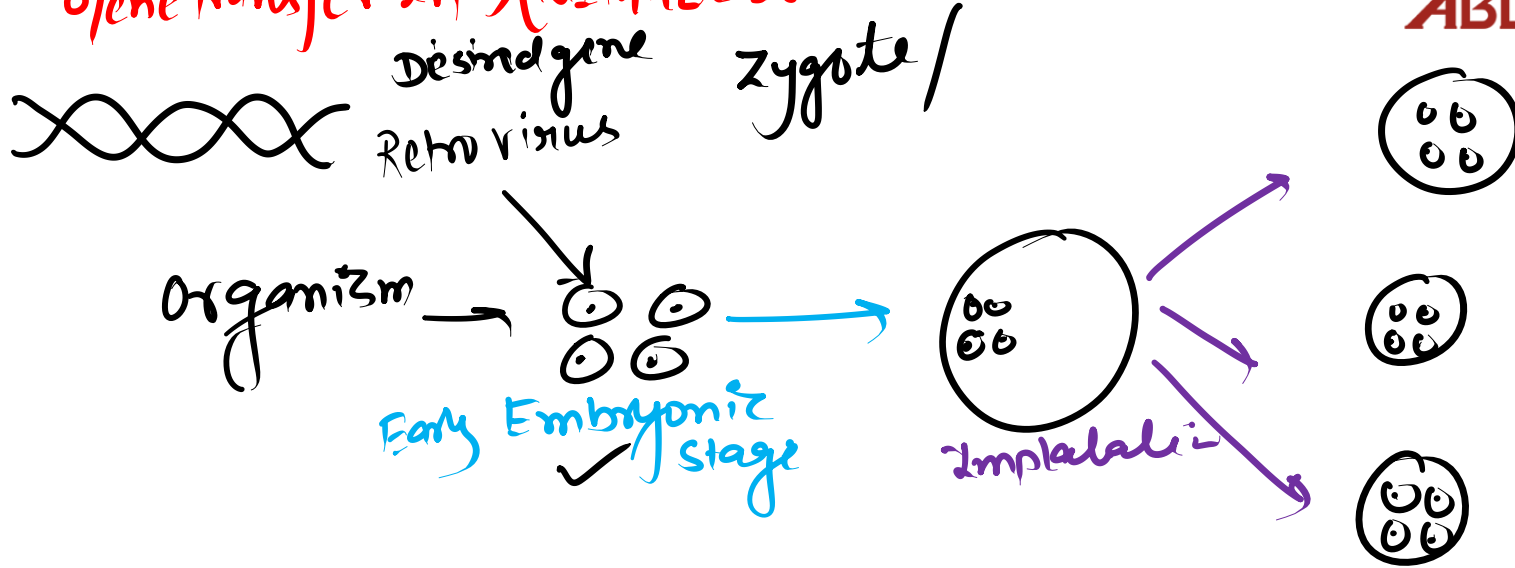
Recombinant DNA

ice 0°C

Sudden transfer at 0°C

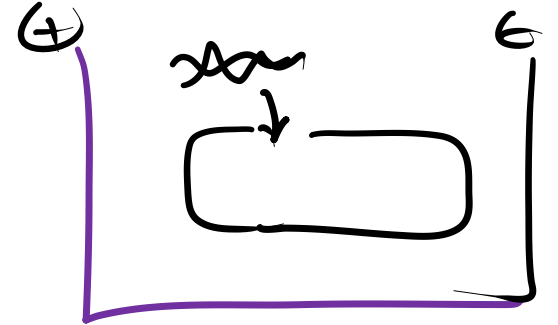
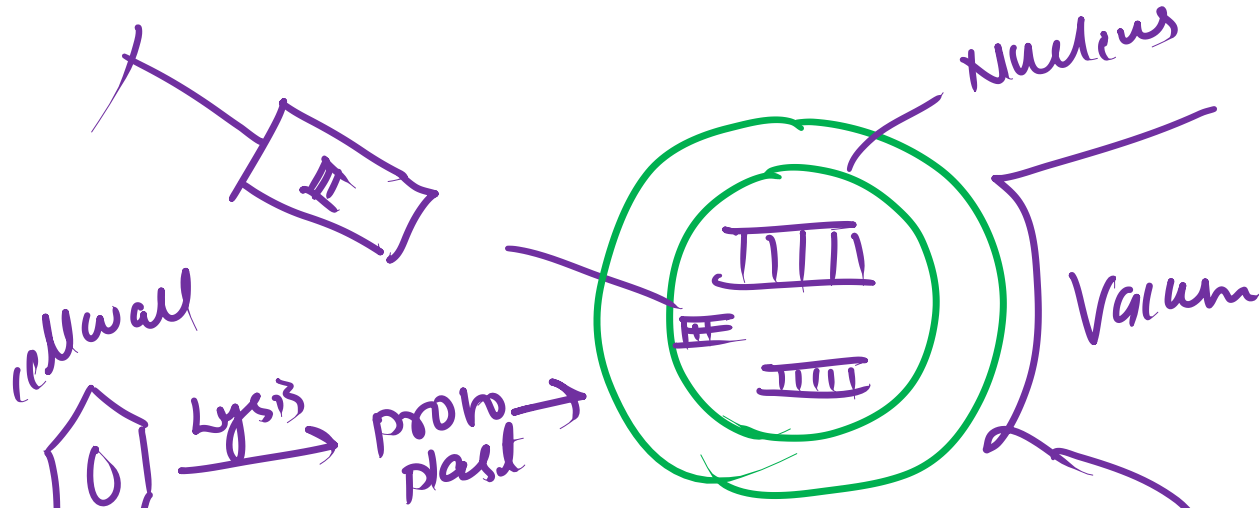
Direct method (vectorless) Host cell are treated with $CaCl_2$

Gene transfer in ANIMAL CELL

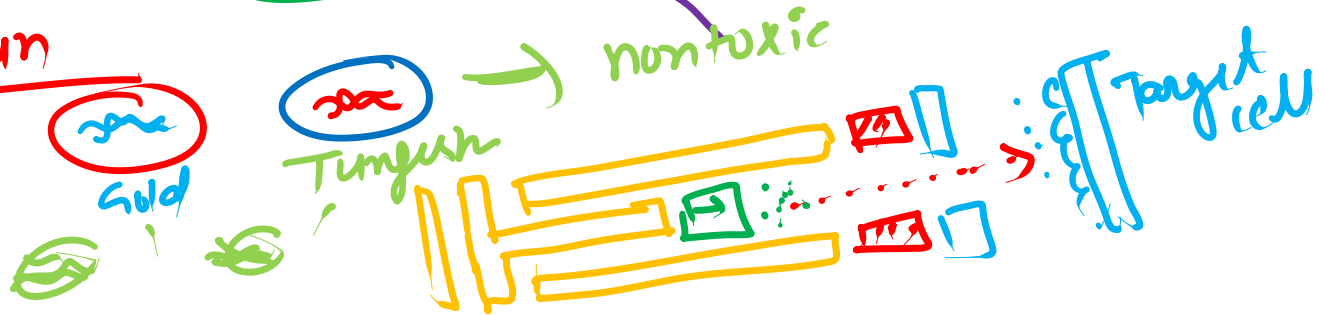


Direct method

Microinjection
↳ Animal cell



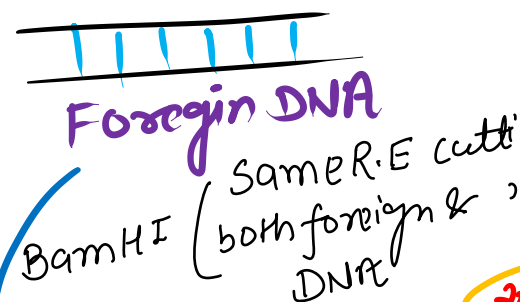
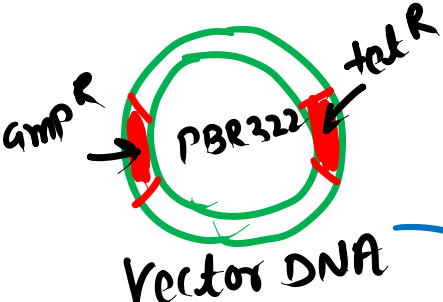
Gene gun



Recombinant DNA (selection of transformant with recombinant DNA)

① Selection by two antibiotic resist gene

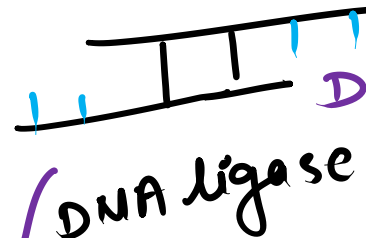
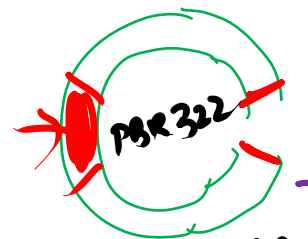
+ transform of plasmid DNA
 Death:



transformants growing on tetracycline

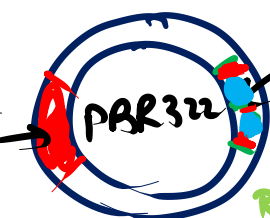


Death



Transformant with non recombinant DNA
 Transform with recombinant DNA
 Non trans (without plasmid)

non Recombinant DNA
 ampR



desired gene
 Transformation