

# Sumário

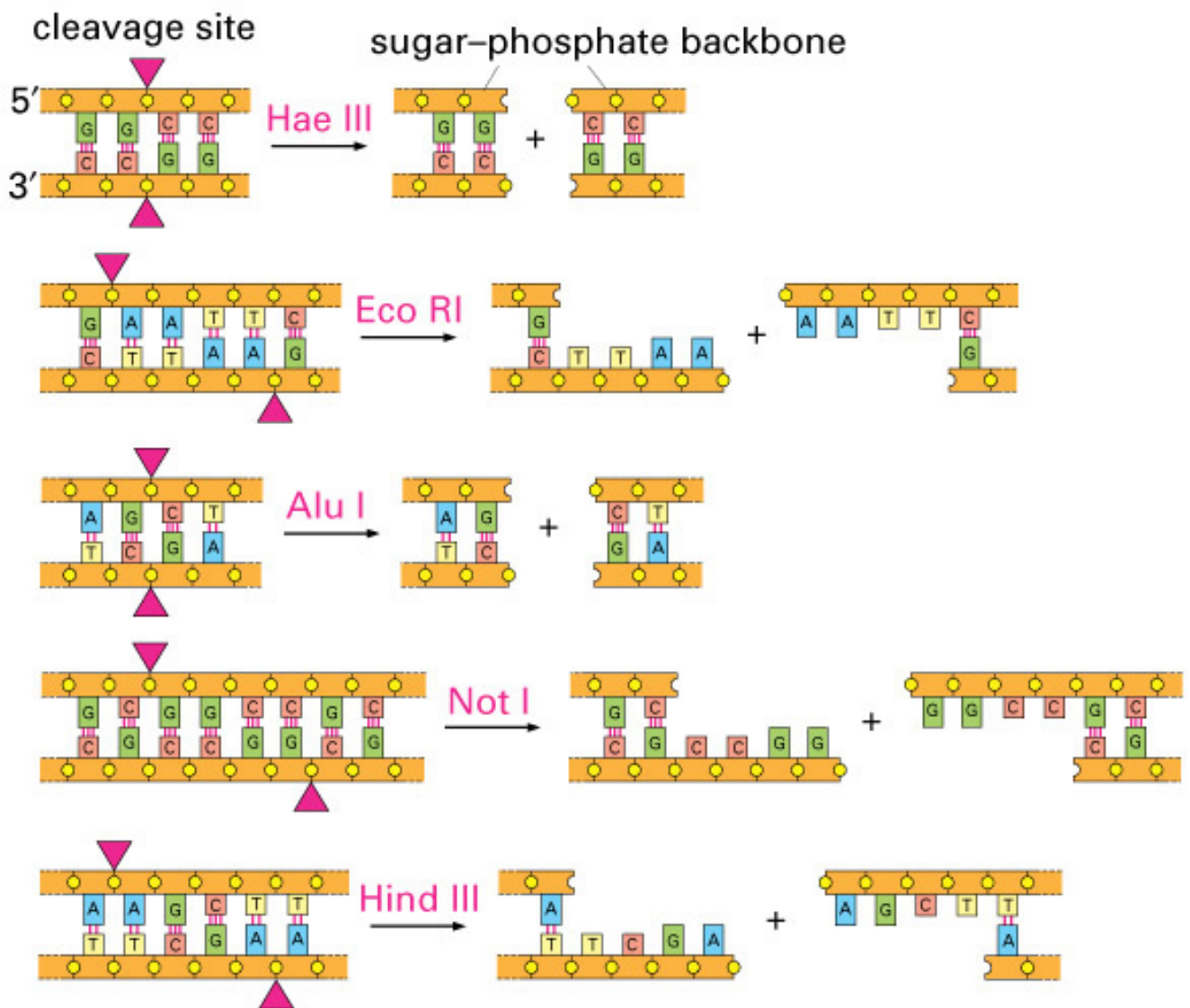
## Breves Noções de Engenharia Genética

- Engenharia genética: as metodologias do DNA recombinante.
- Enzimas de restrição.
- Clonagem molecular
- Hibridação molecular.
- Sequênciação de ácidos nucleicos.
- A reacção em cadeia da polimerase – PCR.
- Análises forenses e 'DNA fingerprinting'

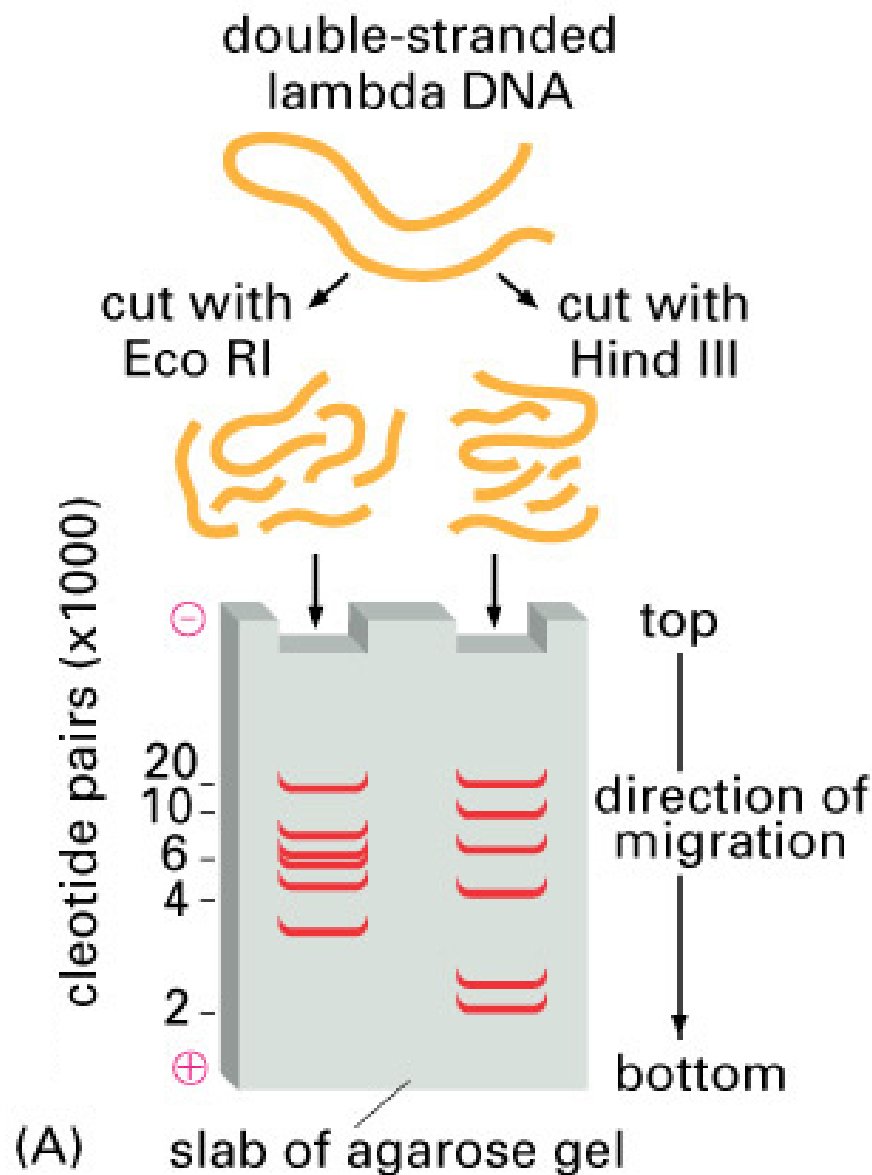
## Bibliografia

- Bruce Alberts, Dennis Bray, Karen Hopkin, Alexander Johnson, Julian Lewis, Martin Raff, Keith Roberts and Peter Walter (2004) *Essential Cell Biology* 2ª ed. Garland Science Publishing. Cap.10

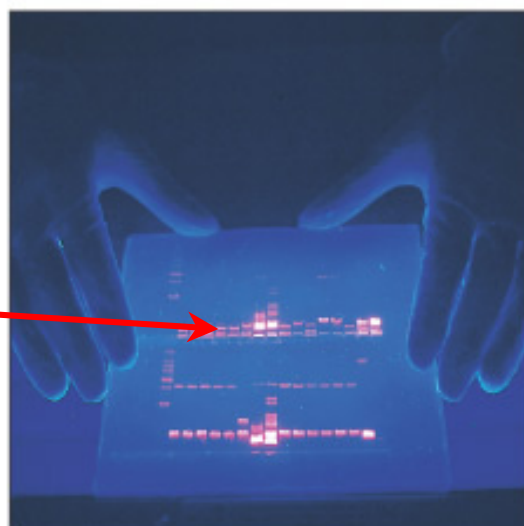
# Enzimas de Restrição: 'Tesouras' Moleculares



# Visualização em Gel de Agarose

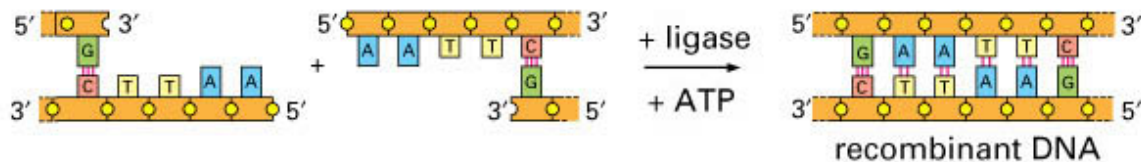


**Gel corado com  
Brometo de Etídio  
visualizado sob luz  
UV**

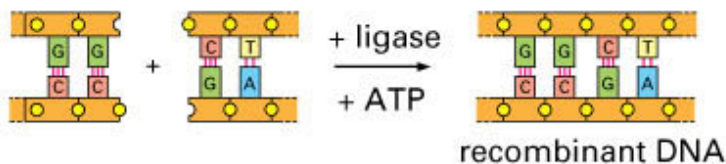


# Clonagem Molecular

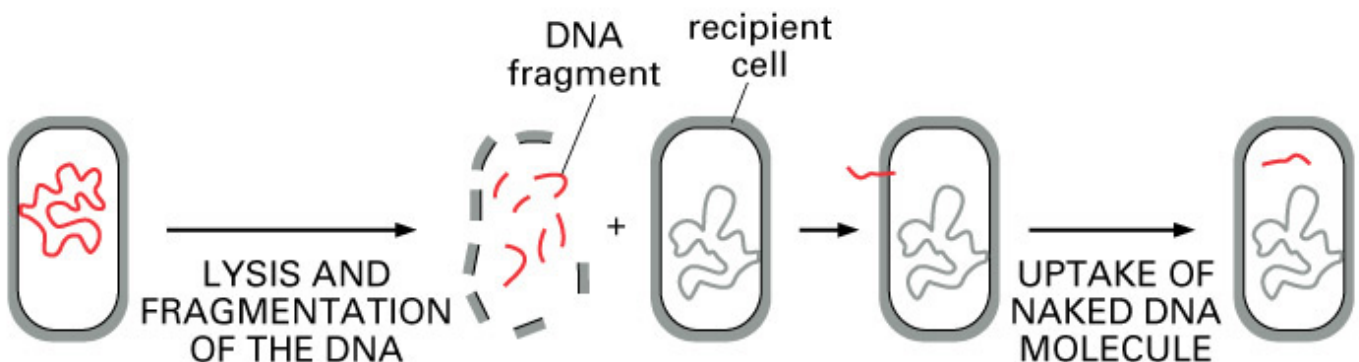
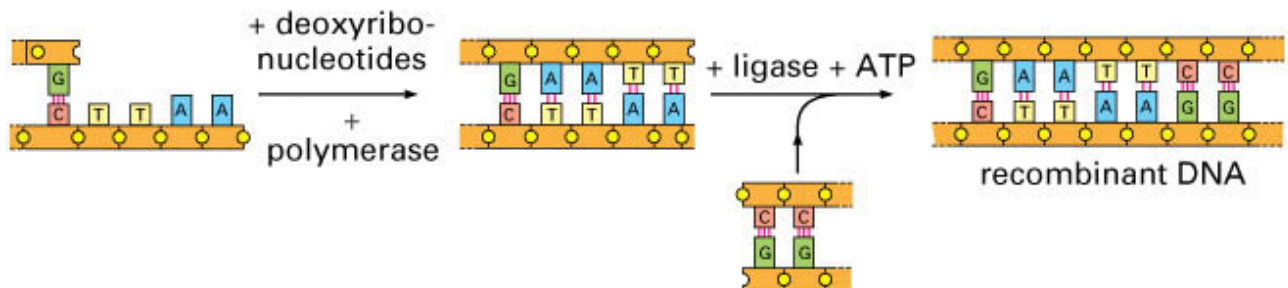
## (A) JOINING TWO COMPLEMENTARY STAGGERED ENDS



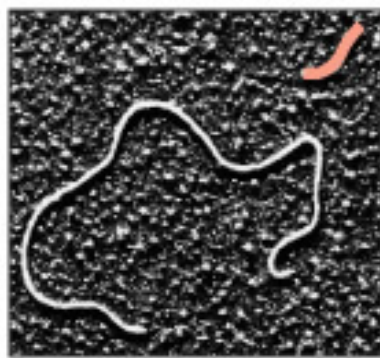
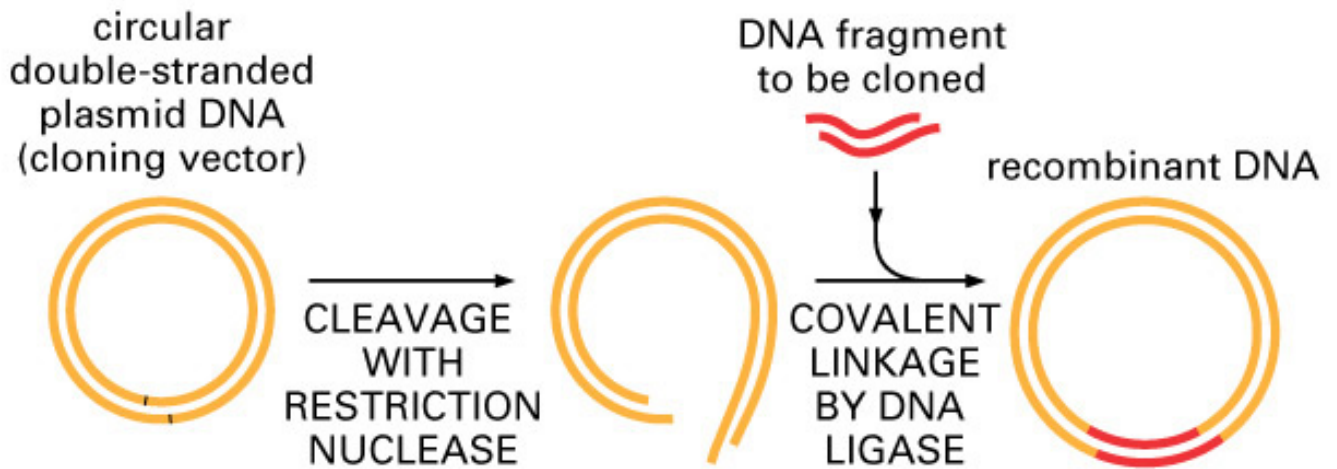
## (B) JOINING TWO BLUNT ENDS



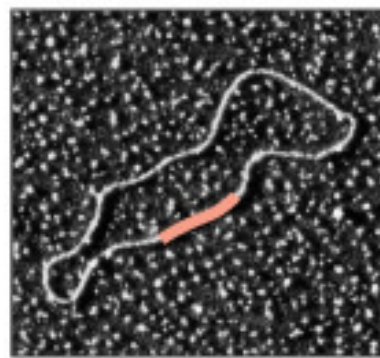
## (C) JOINING A BLUNT END WITH A STAGGERED END



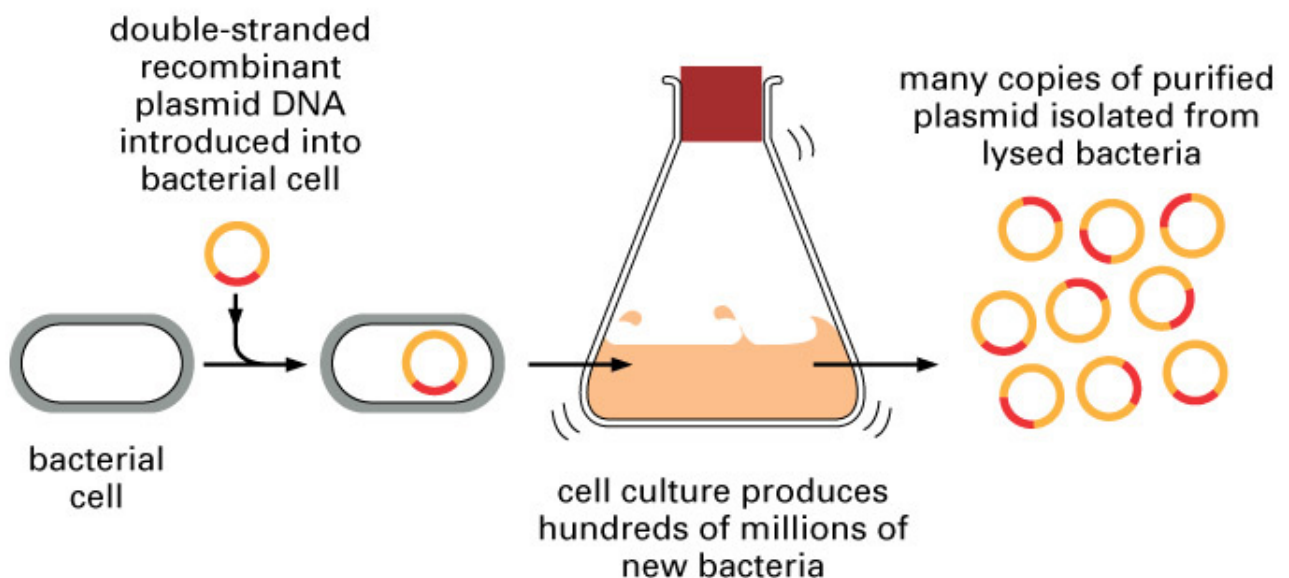
# Clonagem num Plasmídeo



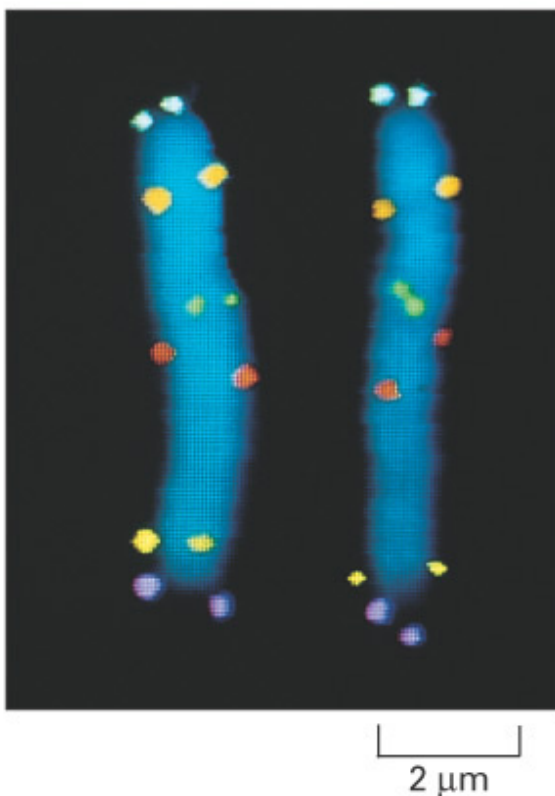
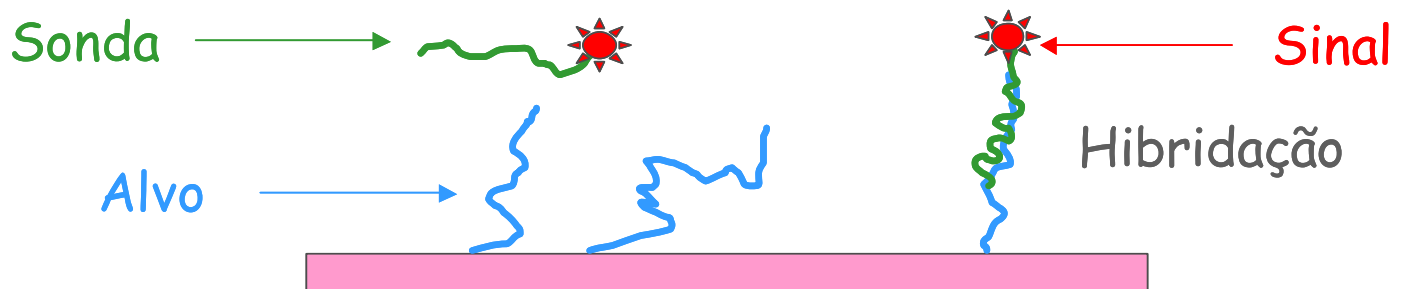
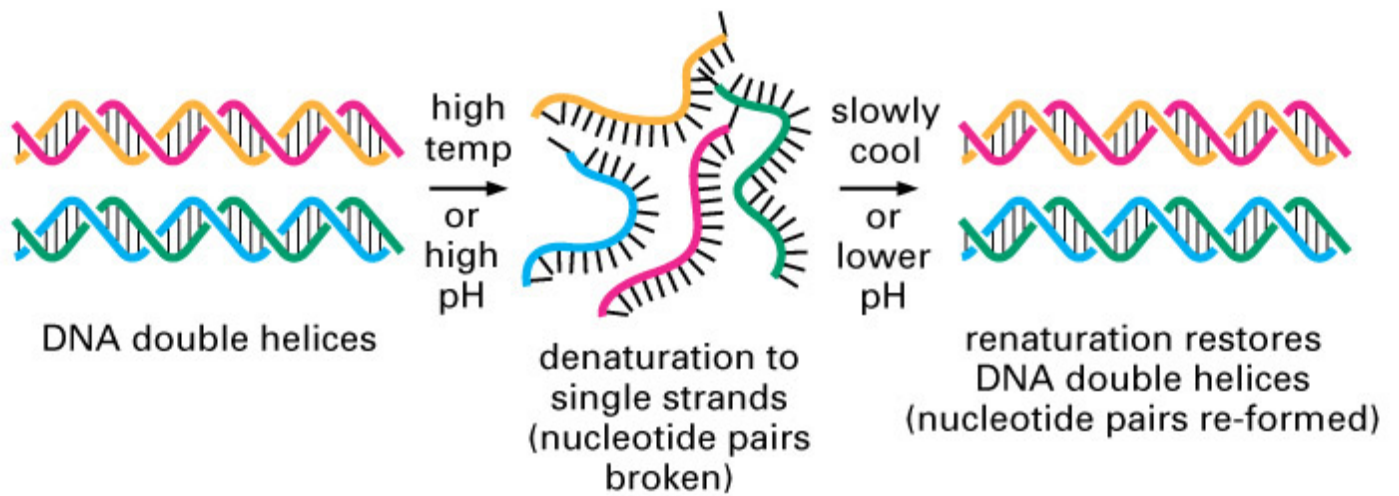
200 nm



200 nm



# Hibridação Molecular

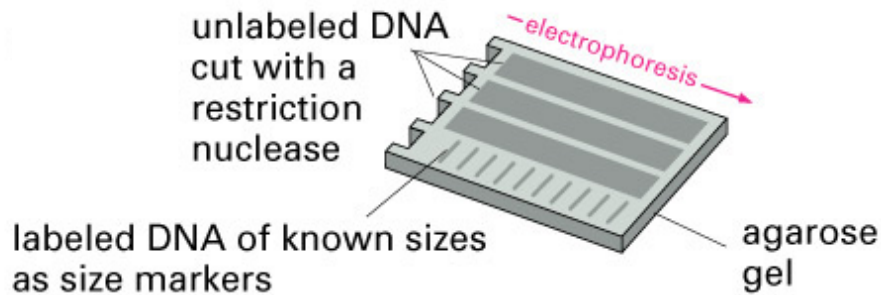


Fluorescent In Situ Hybridization (FISH)



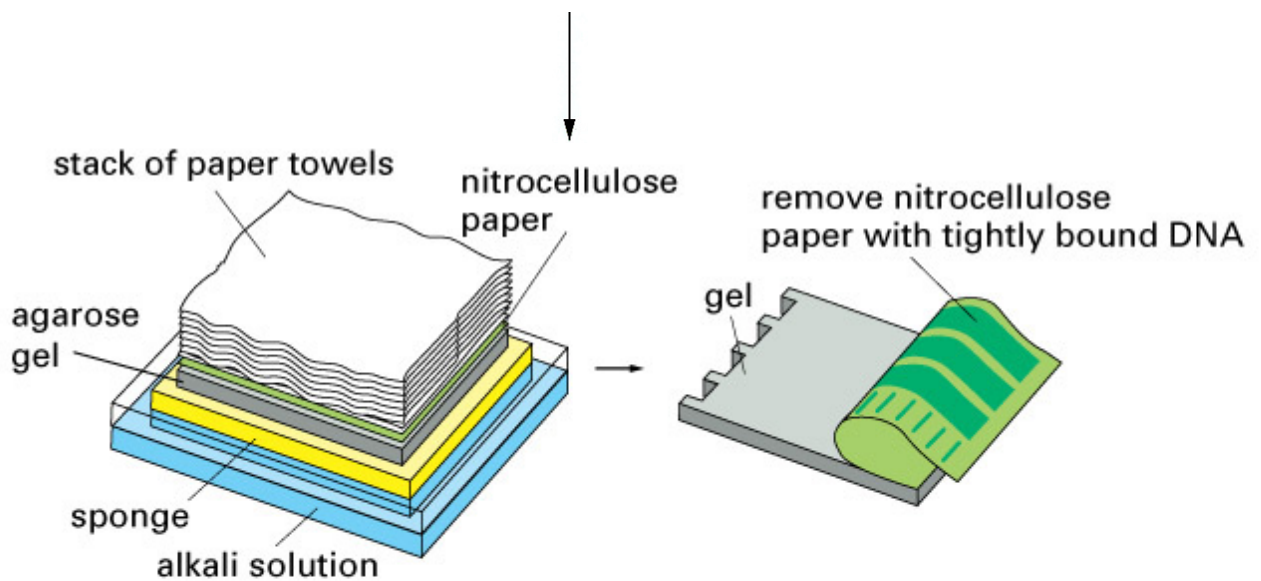
# Southern Blotting

**A**



DNA FRAGMENTS SEPARATED BY AGAROSE GEL ELECTROPHORESIS

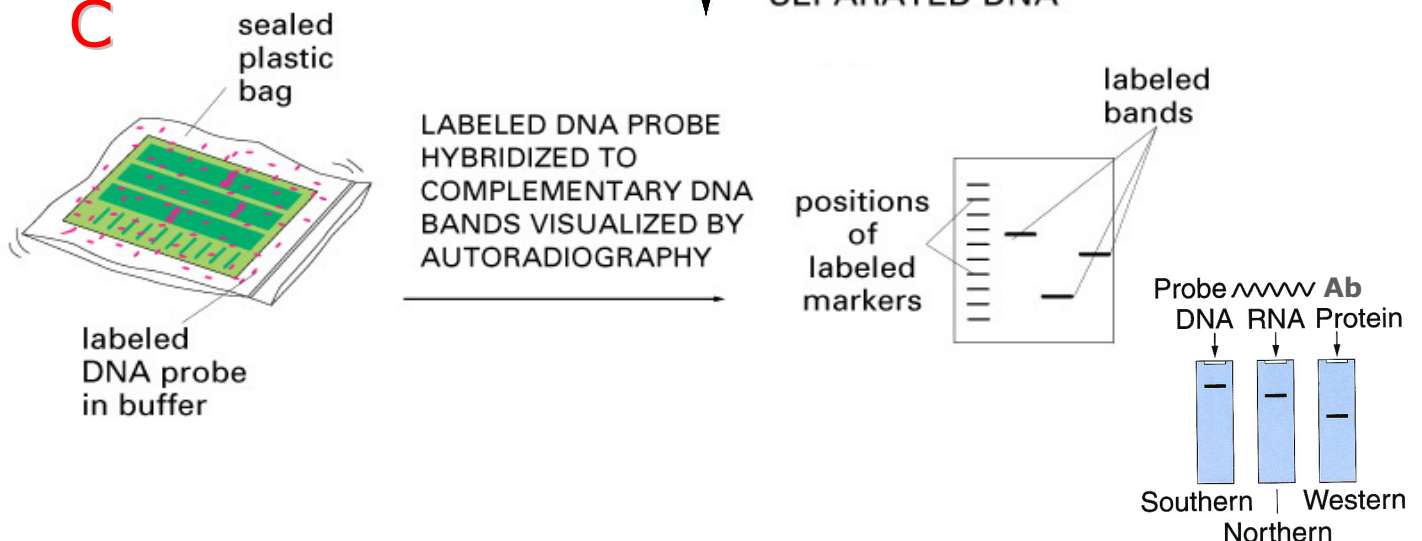
**B**



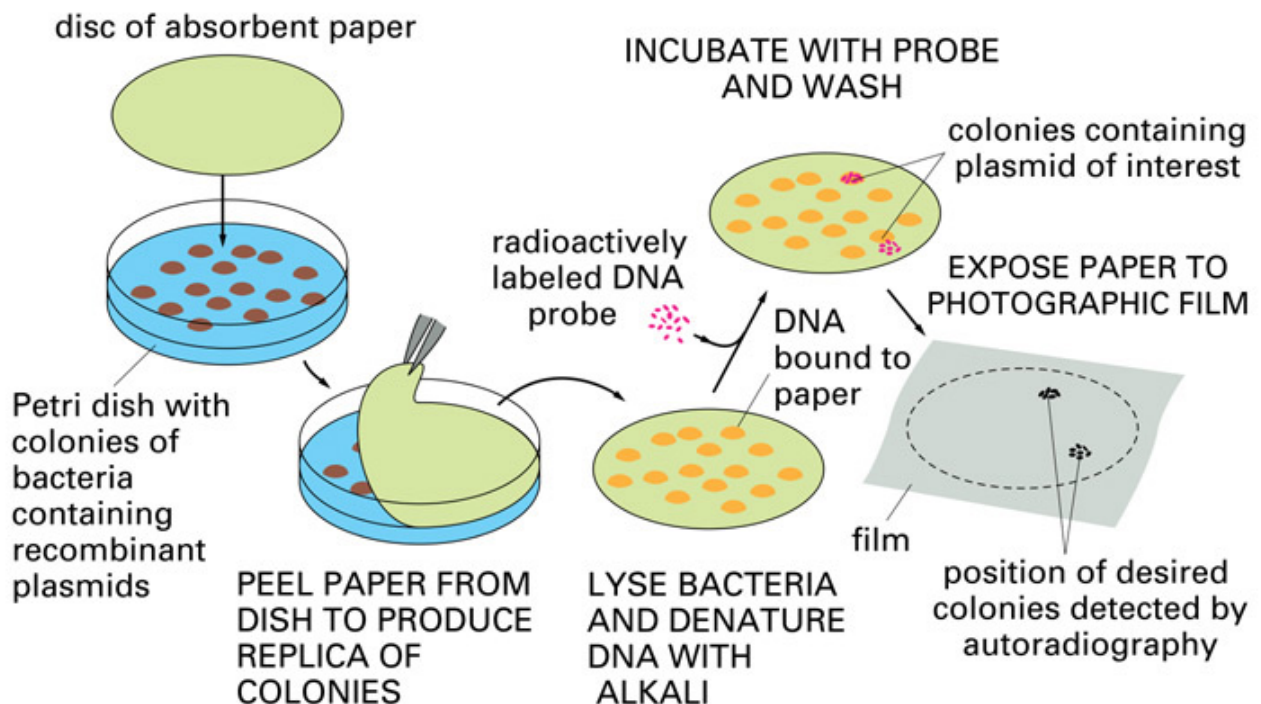
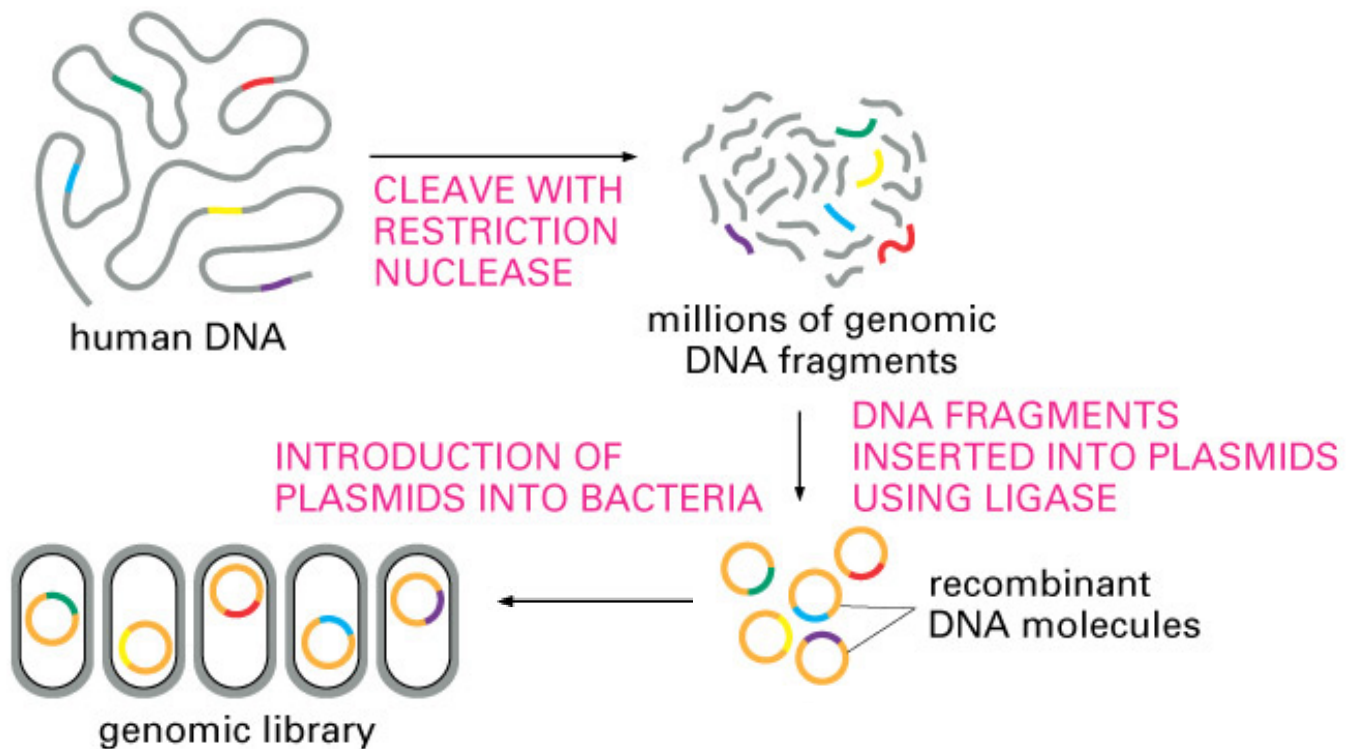
SEPARATED DNA FRAGMENTS  
BLOTTED ONTO NITROCELLULOSE PAPER

LABELLED DNA PROBE  
HYBRIDIZED TO  
SEPARATED DNA

**C**



# Banco Genómico

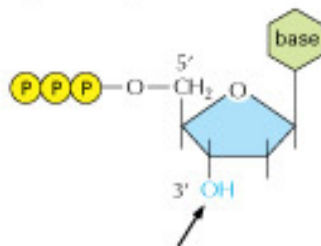




# Sequênciação com ddNTPs

dNTP (normal)

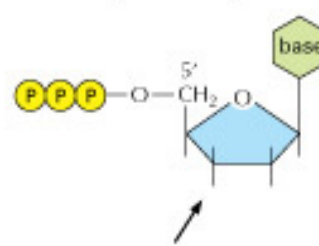
deoxyribonucleoside triphosphate



allows strand extension at 3' end

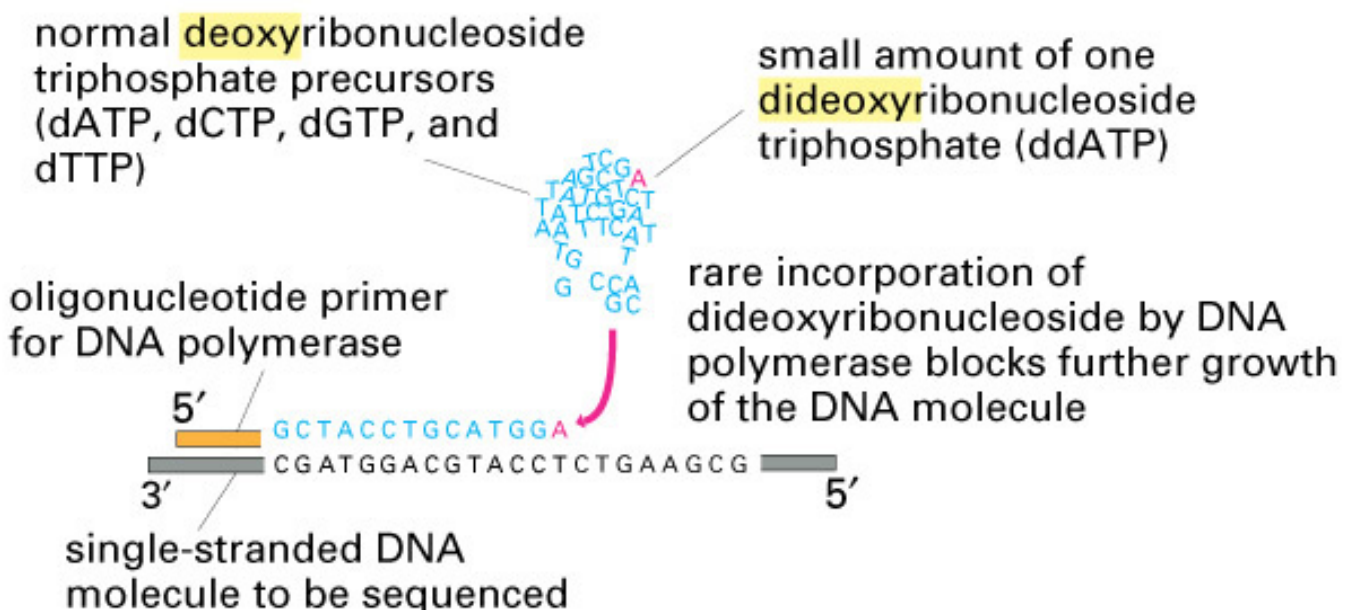
ddNTP (sequênciação)

dideoxyribonucleoside triphosphate

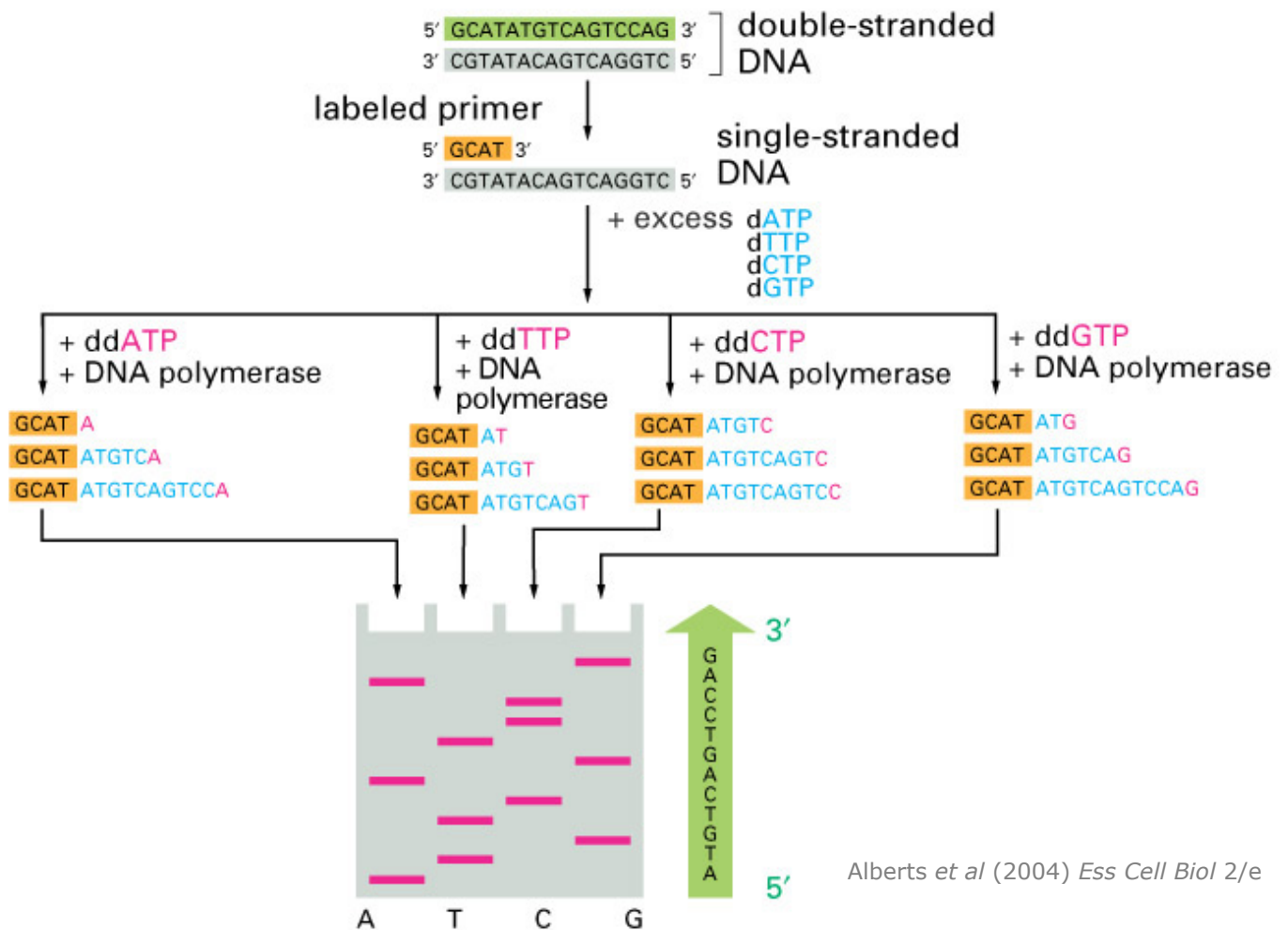


prevents strand extension at 3' end

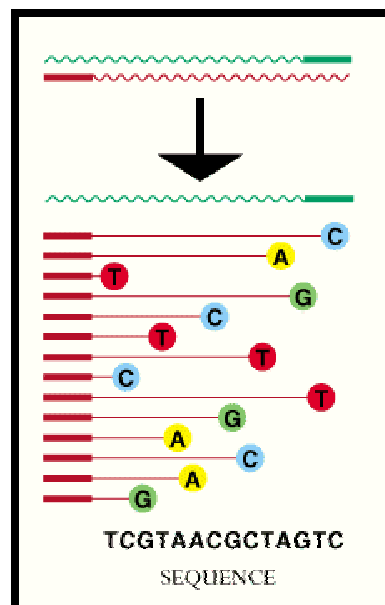
## Reacções de Sequênciação



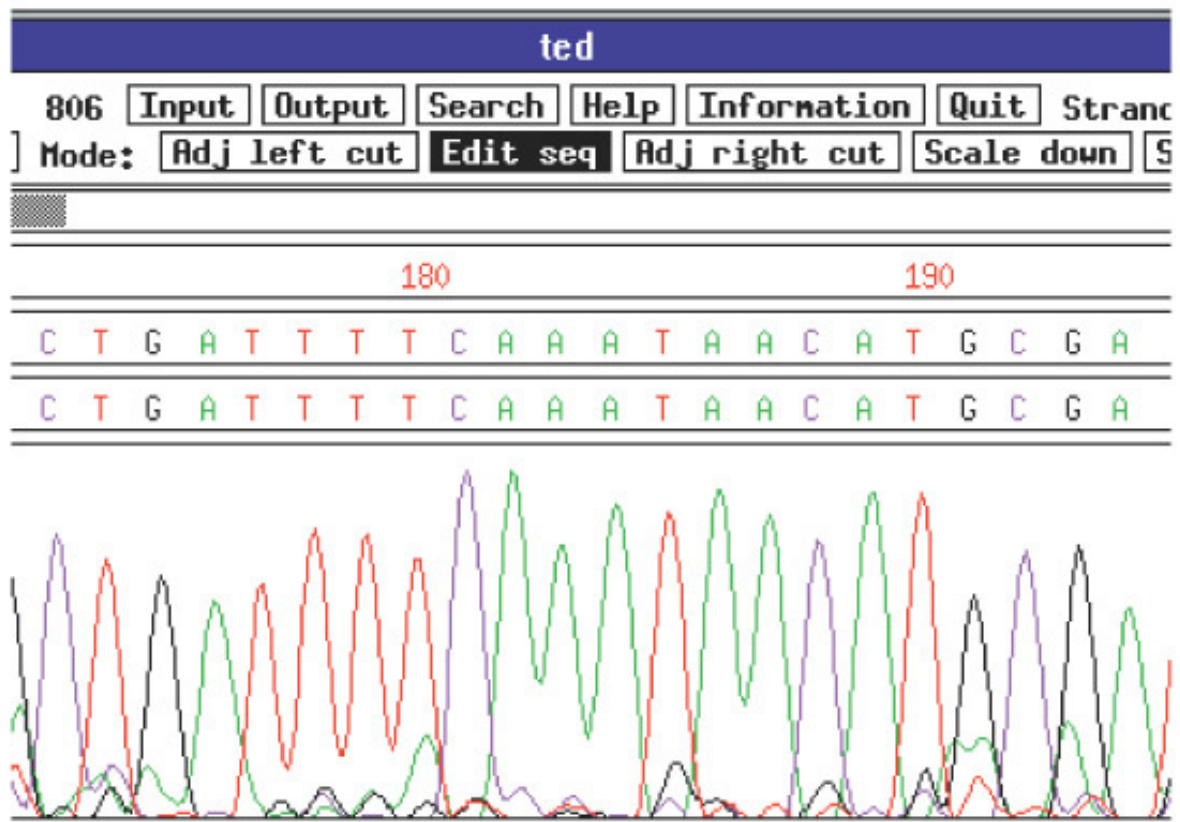
# Método de Sanger: Quatro Reacções de Sequênciação



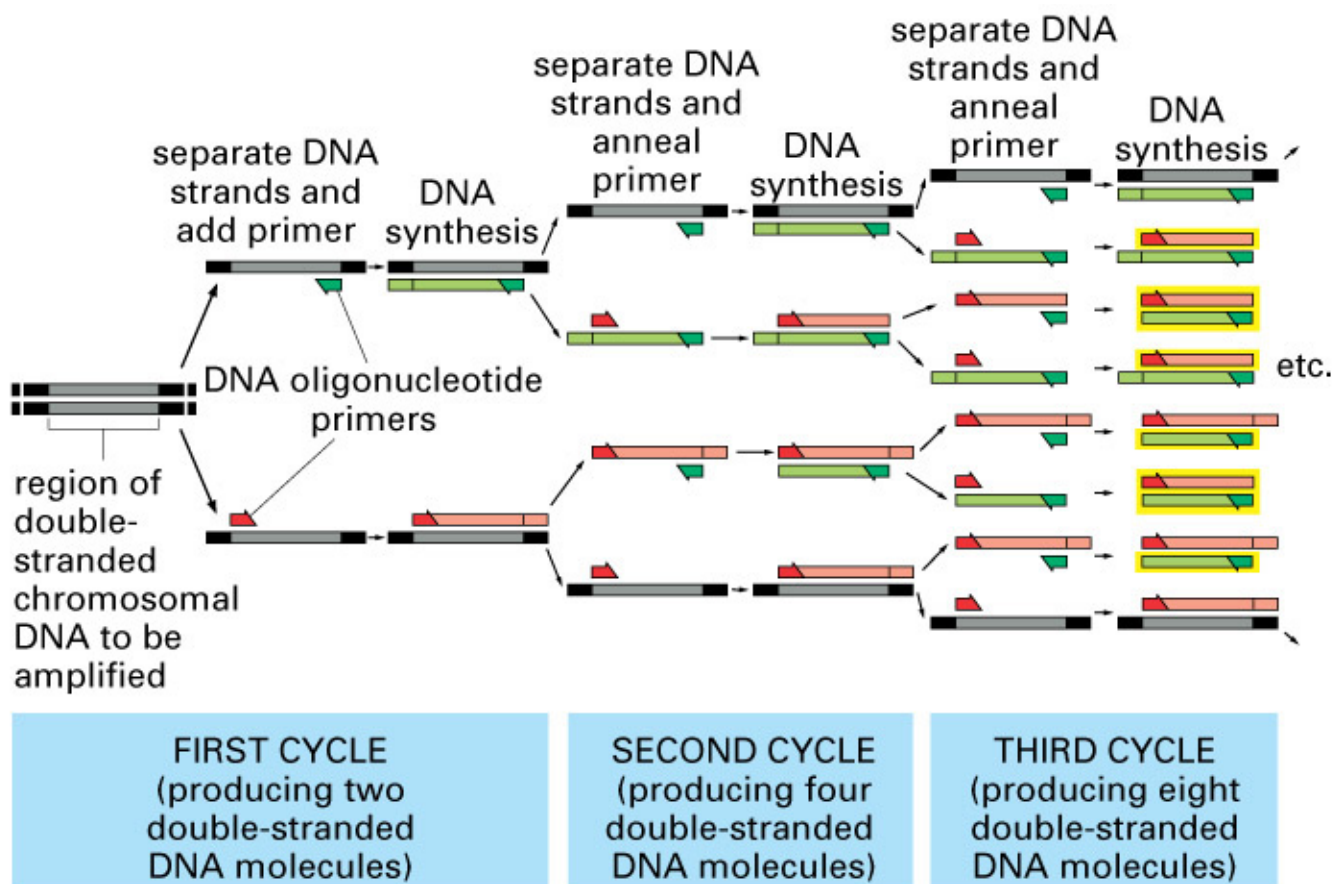
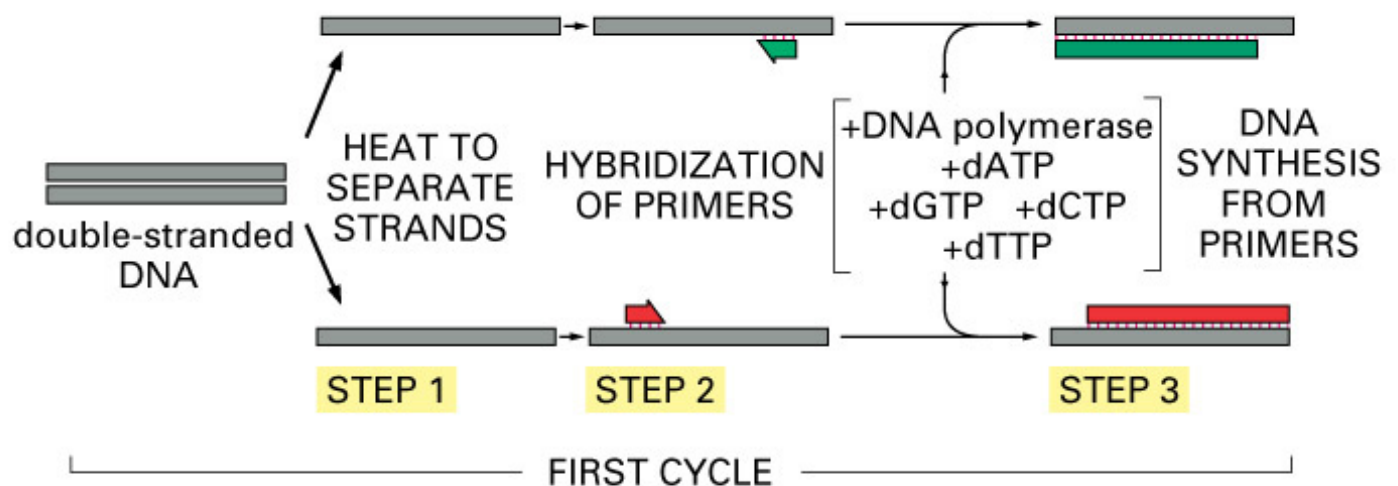
Alberts *et al* (2004) *Ess Cell Biol* 2/e



# Sequencição Automática



# Reacção em Cadeia da Polimerase (PCR)



# Análises Forenses: 'DNA Fingerprinting'

