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Sub. Biology

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Chapter 11 Biotechnology Principles and Processes

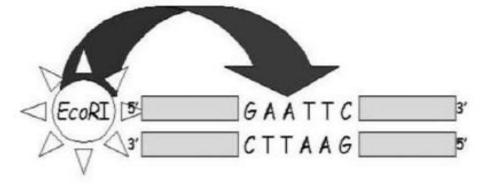
Basic tools of gene cloning.

Names of restriction endonucleases

Titles of restriction enzymes are derived from the first letter of the genus + the first two letters of the species of organism from which they were isolated.

Source microorganism	Enzyme	Reco	Recognition Site AGICT GIGATCC GIAATTC GACGC(N)51		produced
Arthrobacter luteus	Alu I				
Bacillus amyloiquefaciens H	Bam HI				
Escherichia coli	Eco RI				
Haemophilus gallinarum	Hga I				
Haemophilus infulenzae		Hind III A		CTT	Sticky
Providencia stuartii 164		t I	CTGCAIG		Sticky
Nocardia otitiscaviaruns No		t I	GCIGGCCGC		Sticky
Staphylococcus aureus 3A Sa		Sau 3A GAT		TC	Sticky
Serratia marcesans		Sma I CC		GGG	Blunt
Thermus aquaticus		Tag I TiC		SA	Sticky

Restriction enzymes recognise a specific short nucleotide sequence



This is known as a Restriction Site The phosphodiester bond is cleaved between specific bases, one on each DNA strand

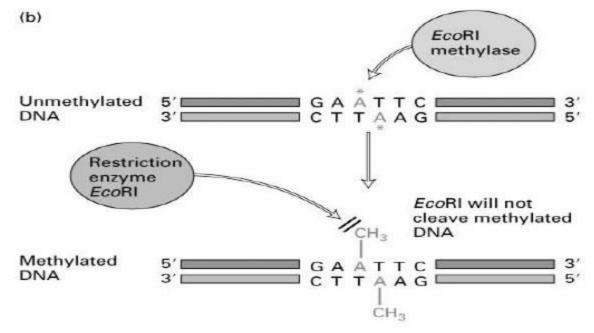
The product of each reaction is two double stranded DNA fragments Restriction enzymes do not discriminate between DNA from different organisms Restriction endonucleases are a natural part of the bacterial defence system

• Part of the restriction/modification system found in many bacteria

• These enzymes **RESTRICT** the ability of foreign DNA (such as bacteriophage DNA) to infect/invade the host bacterial cell by cutting it up (degrading it)

• The host DNA is **MODIFIED** by **METHYLATION** of the sequences these enzymes recognise

o Methyl groups are added to C or A nucleotides in order to protect the bacterial host DNA from degradation by its own enzymes



Types of restriction enzymes

• Type I Recognise specific sequences but then track along DNA (~1000-5000 bases) before cutting one of the strands and releasing a number of nucleotides (~75) where the cut is made. A second molecule of the

endonuclease is required to cut the 2nd strand of the DNA