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EcoRI

EcoRI (pronounced "eco R one") is a restriction endonuclease enzyme isolated from species <u>E. coli</u>. The <u>Eco</u> part of the enzyme's name originates from the species from which it was isolated, while the R represents the particular strain, in this case RY13. The last part of its name, the I, denotes that it was the first enzyme isolated from this strain. <u>EcoRI</u> is a restriction enzyme that cleaves DNA double helices into fragments at specific sites. It is also a part of the <u>restriction modification</u> system.

In molecular biology it is used as a restriction enzyme. *EcoRI* creates 4 nucleotide sticky ends with 5' end overhangs of AATT. The nucleic acid recognition sequence where the enzyme cuts is G/AATTC, which has a palindromic, complementary sequence of CTTAA/G. The / in the sequence indicates which phosphodiester bond the enzyme will break in the DNA molecule. Other restriction enzymes, depending on their cut sites, can also leave 3' overhangs or blunt ends with no overhangs.

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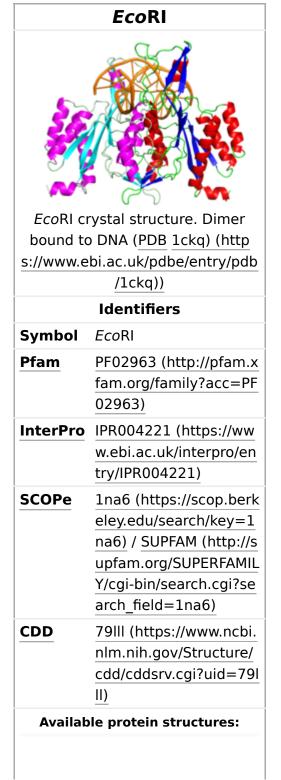
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Structure

Primary structure

EcoRI contains the PD..D/EXK motif within its active site like



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many restriction endonucleases.

Tertiary and quaternary structure

The enzyme is a homodimer of a 31 kilodalton subunit consisting of one globular domain of the α/β architecture. Each subunit contains a loop which sticks out from the globular domain and wraps around the DNA when bound. [1][2]

EcoRI has been cocrystallized with the sequence it normally cuts. This crystal was used to solve the structure of the complex 1QPS (https://www.rcsb.org/structure/1QPS). The solved crystal structure shows that the subunits of the enzyme homodimer interact with the DNA symmetrically. In the complex, two α-helices from each subunit come together to form a four-helix bundle. On the interacting helices are residues Glu144 and Arg145, which interact together, forming a crosstalk ring that is believed to allow the enzyme's two active sites to communicate.

<u>Pfam</u>	structures (http://pfam.xf am.org/family/PF02963?t ab=pdbBlock) / ECOD (ht tp://prodata.swmed.edu/e cod/complete/search?kw =PF02963)
PDB	RCSB PDB (http://www.rcsb.org/pdb/search/smartSubquery.do?smartSearchSubtype=PfamIdQuery&pfamID=PF02963); PDBe (https://www.ebi.ac.uk/pdbe/entry/search/index?pfam_accession:PF02963); PDBj (https://pdbj.org/searchFor?query=PF02963)
PDBsum	structure summary (http s://www.ebi.ac.uk/thornto n-srv/databases/cgi-bin/p dbsum/GetPfamStr.pl?pfa m_id=PF02963)

Uses

Restriction enzymes, such as EcoRI, are used in a wide variety of molecular genetics techniques including cloning, DNA screening and deleting sections of DNA in vitro. Restriction enzymes, like EcoRI, that generate sticky ends of DNA are often used to cut DNA prior to ligation, as the sticky ends make the ligation reaction more efficient. EcoRI can exhibit non-site-specific cutting, known as star activity, depending on the conditions present in the reaction. Conditions that can induce star activity when using EcoRI include low salt concentration,



EcoRI recognition site with cutting pattern indicated by a green line

high glycerol concentration, excessive amounts of enzyme present in the reaction, high pH and contamination with certain organic solvents. The cut made by the Eco RI enzyme produces sticky ends on the vector mainly plasmids or viral DNA's. [5]

See also

- EcoRII, another nuclease enzyme from *E. coli*.
- EcoRV, another nuclease enzyme from *E. coli*.

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External links

- Eco-RI (https://meshb.nlm.nih.gov/record/ui?name=Eco-RI) at the US National Library of Medicine Medical Subject Headings (MeSH)
- Overview of all the structural information available in the <u>PDB</u> for <u>UniProt</u>: <u>P00642 (https://www.ebi.ac.uk/pdbe/pdbe-kb/proteins/P00642)</u> (Type-2 restriction enzyme EcoRI) at the PDBe-KB.

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