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Commentary

Importance of Gene Cloning through Bacterial Transformation

Emily R. Smith*

Department of Biotechnology, Isfahan University of Medical Sciences, Isfahan, Iran

DESCRIPTION

Transformation involves introducing DNA (usually in the form of a plasmid) into a competent bacterial strain so that the bacterium can replicate the sequence of interest at levels suitable for further analysis and manipulation. Bacterial transformation is the process of horizontal gene transfer in which some bacteria take up foreign genetic material (naked DNA) from the environment. The process of gene transfer by transformation does not require living donor cells, only the presence of persistent DNA in the environment is enough for the process. Bacteria are highly versatile organisms with the unique ability to take up and replicate (or copy) foreign DNA. This gives them an evolutionary advantage and allows them to survive environmental changes. For example, bacteria can acquire DNA that makes them resistant to antibiotics. Transformation is the process of introducing foreign DNA into cells. Transformation of bacteria with plasmids is important not only for the study of bacteria, Bacteria are used as a means of both storage and replication of plasmids. During transformation, bacterial cells take up plasmids. This bacterial cell can then contain the genes on this plasmid and express these genes. Examples of transgenic microorganisms are bacterial strains that produce human insulin. A human insulin gene was inserted into the plasmid. Not all bacteria can naturally take up DNA, but chemical manipulations in the laboratory can enhance their ability. This is usually done with calcium chloride, which permeabilizes the cell membrane, so the bacteria can easily take up the plasmid of interest. Bacterial transformation is a key step in molecular cloning, the purpose of which is to generate multiple copies of a recombinant DNA molecule. The steps prior to generating recombinant plasmids are described in Conventional Cloning Basics and require the insertion of the DNA sequence of interest into the vector backbone. A prerequisite for bacterial transformation is the ability of bacteria to absorb free extracellular genetic material. Such bacteria are called as competent cells. Factors that regulate natural abilities vary from genus to genus. Once the DNA enters the cytoplasm, it can be degraded by nucleases if it differs from bacterial DNA. If the exogenous genetic material resembles bacterial DNA, it can be integrated into the chromosome. Exogenous genetic material may coexist with chromosomal DNA as a plasmid. Under such conditions, several genera of bacteria spontaneously release DNA from the cell into the environment for uptake by competent cells. Competent cells also respond to environmental changes and control the level of gene uptake through the natural transformation process. Some basic steps of the transformation process includes

- Specially prepared bacteria are mixed with DNA.
- Heat shocking the bacteria causes the plasmid to be taken up by some of the bacteria.
- Plasmids used for cloning contain antibiotic resistance genes. Therefore, place all bacteria on antibiotic plates and select those that have taken up the plasmid.
- Each plasmid-bearing bacterium gives rise to a collection of bacteria with the same plasmid known as colony. Bacteria which do not contain plasmids usually die.
- Several colonies are screened to identify those with the correct plasmid
- Colonies containing the correct plasmid are propagated to bulk and used for plasmid or protein production.

It can also increase the permeability of cells and increase the efficiency of transformation. Bacterial transformation is a key step in molecular cloning, the purpose of which is to generate multiple copies of a recombinant DNA molecule. The phenomenon of transformation is widespread in molecular biology. A practical approach to obtaining competent cells is to artificially render bacterial cells competent using chemicals or electrical impulses. The phenomenon of natural transformation has enabled bacterial populations to overcome large fluctuations in population dynamics and the overcome the challenges of maintaining population numbers during extreme environmental changes.

Correspondence to: Emily R. Smith, Department of Biotechnology, Isfahan University of Medical Sciences, Isfahan, Iran, E-mail: siavash.i@gva.ir

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