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DNA library

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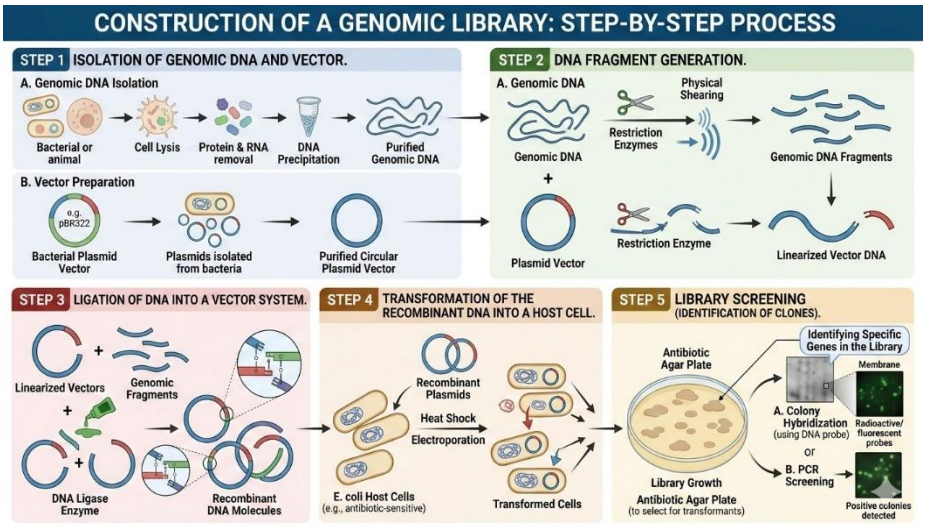
INTRODUCTION

A DNA library, also referred to as a gene library, is a well-organized collection of DNA fragments that are cloned into suitable vectors and maintained within host organisms. Since the genome of an organism is vast and highly complex, studying it as a whole is difficult. To overcome this limitation, the genome is broken down into smaller, manageable fragments. DNA libraries make the entire genome accessible in this fragmented form, enabling researchers to identify, isolate and study specific genes of interest with ease. A genomic DNA library is a type of DNA library that contains DNA fragments representing the complete genetic information of an organism. It includes both coding and non-coding regions of DNA, such as introns and regulatory sequences. This comprehensive nature allows detailed study of regulatory elements and non-coding regions that play an essential role in gene expression and regulation. Owing to these features, genomic libraries are widely used in genome mapping, comparative genomics and studies related to gene regulation. However, genomic libraries also have certain limitations. Handling large DNA fragments often ranging from thousands to millions of base pairs is technically challenging, and the construction and maintenance of such libraries require significant time, labour and resources. In a genomic library, the source of DNA is total genomic DNA extracted from the cells or tissues of the organism. Genomic libraries thus provide a complete representation of the organism's genome and are essential tools for genomic mapping, comparative genomics and the study of gene regulation.

A cDNA (complementary DNA) library is a specialized type of DNA library that consists of cDNA molecules synthesized from messenger RNA (mRNA). Unlike genomic libraries, cDNA libraries represent only the expressed genes of an organism and exclude non-expressed regions such as introns and other non-coding sequences. Because of this, cDNA libraries provide a focused and practical representation of functional genes that are actively transcribed in a particular tissue or at a specific developmental stage. They are especially useful for studying gene expression, understanding protein function, and producing recombinant proteins. However, cDNA libraries also have certain limitations. Since they lack regulatory and non-coding sequences, they are less suitable for studying gene regulation. In addition, they may show limited gene diversity and are often biased toward genes that are highly expressed. cDNA libraries are mainly constructed from eukaryotic organisms to study their expressed genes. Characteristically, cDNA libraries contain only coding regions, consist of relatively smaller DNA fragments, and are derived from mRNA that has been reverse-transcribed into cDNA.

Construction and screening of genomic library

The construction of a genomic library became possible with advances in molecular biology, particularly recombinant DNA (rDNA) technology, the development of cloning vectors and techniques for transforming host organisms. The process begins with the isolation of genomic DNA from the organism of interest. Both coding and non-coding regions of DNA can be isolated using methods such as cell lysis, protein digestion and phenol-chloroform extraction. The isolated genomic DNA is usually too large to be cloned directly, so it is fragmented into smaller pieces suitable for cloning. Fragmentation may be achieved by physical methods such as mechanical shearing or by enzymatic methods using restriction enzymes. The fragmented DNA is then cloned into suitable vectors. Vectors capable of carrying large DNA inserts such as bacteriophages, plasmids, bacterial artificial chromosomes (BACs) and yeast artificial chromosomes (YACs) are employed in construction of genomic library. Before cloning, vectors are treated with restriction enzymes to generate compatible sticky ends. DNA ligase is used to join the DNA fragments with the vectors, forming recombinant DNA molecules. These recombinant vectors are subsequently introduced into suitable host organisms, commonly *Escherichia coli* or yeast, through a process known as transformation. The transformed host cells are grown on selective media, allowing only those cells carrying recombinant DNA to form colonies. Collectively, these colonies constitute a genomic DNA library. Once the library is constructed, screening is performed to identify and isolate recombinant clones containing the DNA insert of interest. Common screening methods include hybridization-based screening, PCR screening, immunological assays and sequencing-based approaches. In hybridization-based screening, labelled DNA or RNA probes that are complementary to the target sequence are used. The colonies or plaques from the library are transferred onto a solid support, such as a nitrocellulose membrane, where the probe hybridizes with the target DNA sequence.



Construction and screening of cDNA library

The construction of a cDNA library begins with the isolation of mRNA from eukaryotic cells. This is typically achieved using column purification methods that employ oligo-dT-coated resins, which selectively bind mRNA molecules containing a poly-A tail at their 3′ end. When a cell lysate is passed through the poly-T column, mRNA molecules bind to the oligo-dT sequences, while other cellular components are washed away. The bound mRNA is then eluted using an appropriate buffer. The isolated mRNA serves as a template for cDNA synthesis. Initially, an oligo-dT primer anneals to the poly-A tail of the mRNA, initiating first-strand cDNA synthesis by the enzyme reverse transcriptase, resulting in an RNA–DNA hybrid. The mRNA strand is partially degraded by RNase H, leaving small RNA fragments that act as primers for second-strand synthesis. DNA polymerase I then synthesizes the second DNA strand, replacing the RNA fragments, and DNA ligase seals the gaps to form a complete double-stranded cDNA molecule. The newly synthesized cDNA is then cloned into suitable vectors. Since cDNA molecules generally have blunt ends, restriction site linkers or adapters are added to make them compatible with vector DNA. These linkers contain specific restriction enzyme recognition sites and are digested accordingly. Plasmid and phage vectors are most commonly used for cDNA cloning, making these libraries particularly suitable for analysing gene expression patterns and the functions of expressed genes. Both the cDNA and vector are cut with the same restriction enzyme and joined together to form recombinant DNA molecules. In the final step, these recombinant DNA molecules are introduced into suitable host cells through transformation. The transformed cells are cultured on selective media containing antibiotics, ensuring that only cells carrying the recombinant vectors survive and form colonies. Each colony represents a different cDNA insert, and collectively these colonies constitute the cDNA library. Screening of the cDNA library is carried out using labelled probes that hybridize with the target DNA sequence. Positive clones are identified by detecting hybridization signals through autoradiography or fluorescence-based methods.

Applications of DNA library

Genomic libraries have wide-ranging applications in biological research. They are used for the construction of physical maps of genomes, the study of non-coding regions and regulatory elements and the analysis of gene sequences that may not be actively expressed. They also play a vital role in understanding evolutionary relationships among different species through comparative genomic studies. Additionally, genomic libraries aid in the identification of genetic mutations and disease-associated genes, contributing significantly to our understanding of the genetic basis of various diseases.

cDNA libraries have wide applications in molecular biology and biotechnology. They are used to study actively expressed genes in different tissues under specific physiological or environmental conditions, to identify and clone functional genes, and to compare gene expression profiles across species in evolutionary studies. Additionally, cDNA libraries are essential tools for the production of recombinant proteins and play a significant role in the development of diagnostic markers for various diseases.

Suggested reading

Wu CC, Xu Z, and Zhang H-B (2004). DNA libraries. In: Meyers RA (ed.) Encyclopedia of Molecular Cell Biology and Molecular Medicine ,3(2): 385-425. Weinheim, Germany: Wiley-VCH Verlag GmbH.

Schmitz JE, Daniel A, Collin M (2008) Rapid DNA Library Construction for Functional Genomic and Metagenomic Screening. *Applied Environmental Microbiology* **74**:1649-1652.