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Molecular Hybridization

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ABSTRACT

Molecular hybridization is a new technique in drug design and development based on the combination of pharmaco moieties of different bioactive substances to produce a new hybrid compound with improved affinity and efficacy. Hybridization is the process of combination of two complementary single-stranded DNA / RNA molecules and allowing them to form a single double-stranded molecule through base pairing.

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INTRODUCTION

Molecular hybridization is the link between strands of DNA and RNA whose kinetics are govern by the degree of intra- and inter strand affinity, solvent type, solvent ionic strength, temperature, and time These bases stack in the vertical plane of the helix by hydrophobic interactions and associate in the horizontal plane according to "base pairing" rules: purine bases, guanine (G) and adenine (A), must base pair with pyrimidine bases, cytosine (C) and thymidine (T), respectively. The rate-limiting step in molecular hybridization is the initial association between two antiparallel strands of nucleic acid that are capable of base pairing.

Single-stranded DNA can hydridize to either single-stranded DNA or single-stranded RNA. Two complementary single-stranded DNA molecules can reform the double helix after annealing. In DNA-RNA hybridization, the RNA base uracil pairs with adenine in DNA. Single-stranded RNA that is complementary to a messenger RNA (mRNA) sequence is called "antisense" RNA. Antisense RNA and mRNA form a double helix that is slightly different from a DNA double helix.

LABORATORY TEST AND THERAPY OF INFECTIOUS DISEASE

NUCLEIC ACID DETECTION:

Molecular hybridization techniques using probes directed at a unique, well-maintained portion of a viral genome are highly specific and bind only to complementary DNA or RNA sequences. DNA replication and transcription of DNA into RNA both rely upon nucleotide hybridization, as do molecular biology techniques including Southern blots and Northern blots,[2] the polymerase chain reaction (PCR), and most approaches to DNA sequencing. Unincorporated nucleotides, enzyme, crosslinking reagents, buffer components, and the like, may cause high backgrounds or interfere with downstream experiments. Hybridization experiments where the volume of the probe labeling reaction is negligible in comparison to the hybridization buffer volume do not always require probe cleanup.

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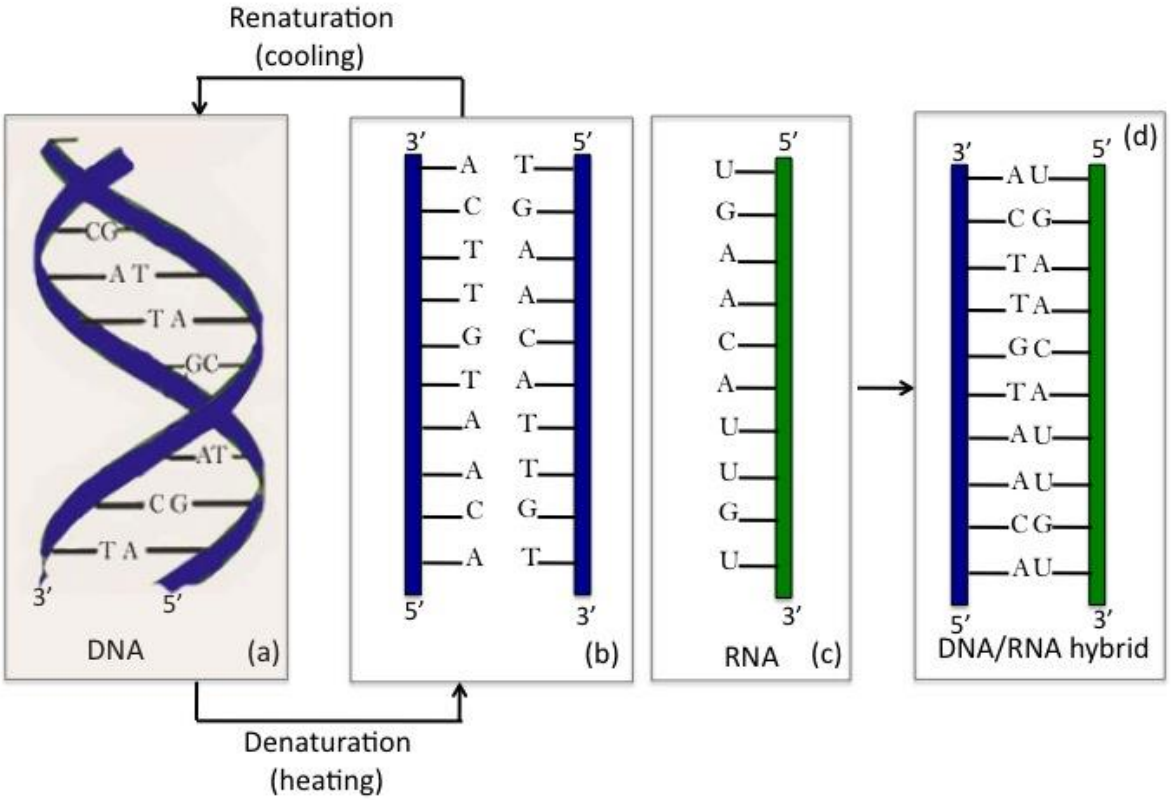


NUCLEIC ACID TRANSFER

Nucleic acid hybridization is a basic technique in molecular biology which takes advantage of the ability of individual single-stranded nucleic acid molecules to form double-stranded molecules.

ACTIVE PASSIVE TECHNIQUES

Vacuum, electrophoretic, and downward gravity transfer methods are fast and efficient Transfer efficiency depends on thickness and percentage of the gel and nucleic acid concentration Capillary blotting of RNA larger than 2.5kb takes more than 12 hours, and downward transfer only 1 to 3 hours.

BUFFER TRANSFER

Transfer buffer pH can also affect the surface charge of the membrane. Nylon membranes are polyamides. The net charge of unmodified nylon is zero, but the polyamide backbone will become more positive when lowering the pH.

DEPURINATION

Breakdown of nucleic acids via depurination increases transfer efficiency.

➢ HYBRIDISATION PRINCIPLES:

- SOUTHERN HYBRIDISATION
- NORTHERN HYBRIDISATION

SOUTHERN HYBRIDISATION

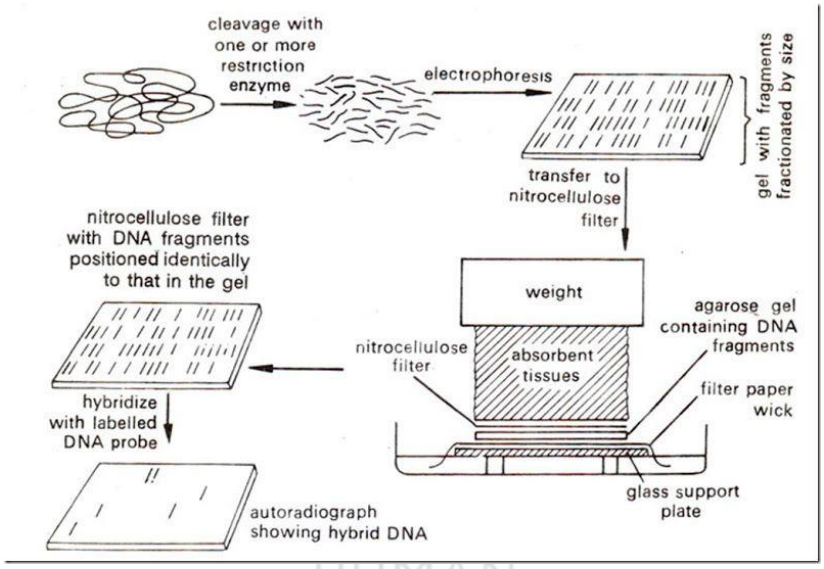
The basic principle behind the southern hybridization is the nucleic acid hybridization. Southern hybridization commonly known as southern blot is a technique employed for detection of a specific DNA sequence in DNA samples that are complementary to a given RNA or DNA sequence.

PROCEDURE

- The high-molecular-weight DNA strands are fractioned using restriction enzymes.
- The DNA fragments are then electrophoresed on an agarose gel to separate them by size.
- A sheet of nitrocellulose membrane is placed on top of the gel. Pressure is applied evenly to the gel to ensure good and even contact between gel and membrane. If transferring by suction buffer is used to ensure a seal and prevent drying of the gel.
- The membrane is then heated in a vacuum or regular oven at 80 °C for 2 hours or opens to ultraviolet radiation to permanently attach the transferred DNA to the membrane.
- The membrane is then exposed to a hybridization probe a single DNA fragment with a specific sequence whose presence in the target DNA is to be determined. The probe DNA is labelled so that it can be detected, usually by joining radioactivity or tagging the molecule

with a fluorescent dye. In some cases, the hybridization probe may be made from RNA, rather than DNA. To ensure the specificity of the binding of the probe to the sample DNA, most common hybridization methods use salmon or herring sperm DNA for blocking of the membrane surface and target DNA, deionized formamide, and detergents such as SDS to reduce non-specific binding of the probe.

- After hybridization excess material on probe is washed from membrane and hybridization is visualized on X-ray film by various means such as audiography.



MOLECULAR GENETIC TEST

Genetic testing is a type of diagnostic test that identifies changes in chromosomes, genes, or proteins. The results of a genetic test can confirm out a suspected genetic condition or help to determine a person's chance of developing on a genetic disorder.

➢ **METHODS FOR GENETIC TESTING.**

▪ GENE TEST: study single genes or short lengths of DNA to identify variations or mutations that lead to a genetic disorder.

- CHROMOSOMAL GENETIC TEST: analyze whole chromosomes or long lengths of DNA to see if there are large genetic changes, such as an extra copy of a chromosome, that cause a genetic condition.

- BIOCHEMICAL GENETIC TEST: study the amount or activity level of proteins; abnormalities in either can indicate changes to the DNA that result in a genetic disorder.

APPPLICATIONS:

- Diagnostic test of alpha thalassemia
- Host taxonomy of RNA viruses
- Measuring nucleic acid sequencing
- Clinical medicines
- Prenatal diagnosis of pregnancy
- Detection of plant RNA virus

REFERENCES

1. YuetWai Kan, M.B., et al, prenatal diagnosis of alpha thalassemia- clinical application of molecular hybridization, the new England Journal of Medicine, Nov. 18 1976,295:1165-67.
2. Daniel L. Stoler, Nelson. L. Michel, Molecular Detection of Viruses,1995, Science Direct.
3. Escaig-Haye, F,Etal, Detection ultrastructuraled'ARN ribosomal par hybridation in situ à l'aided'unesondebiotinyléesur coupes ultrafines de cellules animales en culture. *C. R. Acad. Sci., Paris, D* **429**, 429–434.
4. Jesus A., et al, simultaneous detection of five carnation virues by nonisotopic molecular hybridization, Journal of Virological Methods, Elsevier,167-175.
5. V. Pallas, F. Figgoli, et al, molecular hybridization technique for detecting and studying fruit tree virus and viroids, Research gate.
6. Vicidi RP, Yolken RG, molecular diagnosis of infectious diseases by nucleic acid hybridization, NCBI, Pubmed.gov,1987, Mar (1) 3-4.
7. Sklar J, DNA hybridization in diagnostic pathology, NCBI, Pubmed.gov,1985,jul16(7)654-8.