WHITEPAPER

Probing Questions: What are the 5' and 3' ends of DNA?

EMPIRE GENOMICS
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Probing Questions: What are the 5’ and 3’ ends of DNA?

The 5’ and 3’ ends of DNA are fundamental to its structure and function. These structural ends are also significant in a variety of molecular biology techniques, including Fluorescence in Situ Hybridization (FISH), a clinical testing method used to detect and study genetic abnormalities.

The 5’ end refers to the five-prime end of a single DNA strand, which has a phosphate group attached to the fifth carbon.[3] The 3’ end of DNA refers to the three-prime end, which has a hydroxyl group attached to the third carbon.[3] This polarity is critical because DNA synthesis and other cellular processes occur in a directional manner from 5’ to 3’.[2]

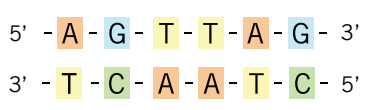
Most of the time, DNA exists in a double-stranded form as two single strands bound together through complementary base pairing with their 5’ and 3’ ends running in opposite (antiparallel) directions.[2,4] When DNA undergoes replication, the double-stranded DNA being replicated opens into two single strands in an “unzipping” configuration, referred to as the replication fork.[1]

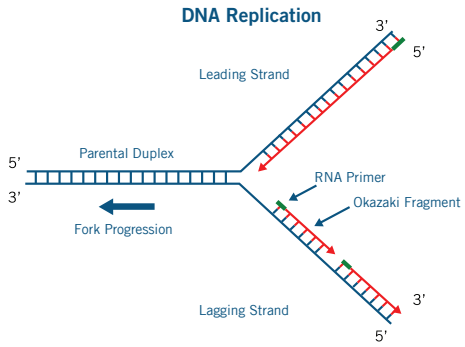
As the replication fork advances, the leading strand is synthesized continuously in the 5’ to 3’ direction, while the lagging strand is synthesized discontinuously in small fragments called Okazaki fragments.[2] This directionality ensures that the DNA molecule grows consistently and correctly.[1,2] The two duplicate strands produced from this process each contain one “old” strand and one newly synthesized strand.[2] This makes DNA replication a semi-conservative process since each new DNA molecule conserves one of the previous DNA strands.[2]

The 5’ and 3’ ends are also relevant in FISH testing. In clinical FISH testing, DNA probes that are complementary to the target DNA sequences are labeled with fluorescent tags.[5] These probes are then hybridized to the target sequences in the patient’s chromosomes.[5] Once the DNA probes have hybridized with the patient’s chromosomes, the fluorescent tags allow for the visualization of the target DNA sequences under a fluorescence microscope.[5] The 5’ to 3’ directionality of DNA ensures that the probes bind in the correct orientation.[5] If the probes were to bind in the wrong direction, the results may be inaccurate, and the clinical utility of FISH testing may be compromised.[5]

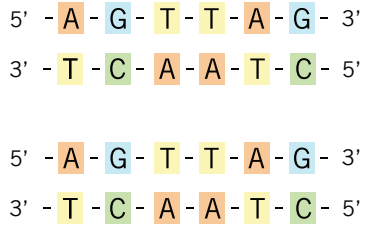
The presence, absence, or abnormal arrangement of these sequences may provide critical information about genetic disorders, such as chromosomal abnormalities, gene amplifications, or translocations.[5]

DNA Illustrations





Each Strand of DNA acts as a template for synthesis of a new, complementary strand



Replication produces two identical DNA double helices, each with one new and one old strand

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