CHEMICAL VERSUS ENZYMATIC DNA SYNTHESIS

From synthetic biology and gene therapy to data storage and biofuel production, the demand for synthetic DNA is rising.

CHEMICAL SYNTHESIS

Phosphoramidite oligonucleotide synthesis is the gold-standard method for generating synthetic DNA. Scientists use the method to assemble modified nucleotides—phosphoramidite nucleosides—with the help of chemicals.1

- The nucleosides are rich in exposed hydroxyl and amino groups, so scientists use protecting groups to prevent unintended reactions with compounds used in DNA synthesis. Chemicals remove the protecting groups to facilitate nucleoside coupling.
- Continued exposure to harsh chemicals damages the DNA and reduces yields. Scientists use short oligonucleotides as primers for polymerase chain reaction and next-generation sequencing, in DNA microarrays and fluorescence in situ hybridization, and in antisense therapies.

ENZYMATIC SYNTHESIS

Enzymatic DNA synthesis is an emerging technology that offers several advantages over chemical methods. Scientists use two main approaches to achieve template-independent oligonucleotide assembly.2

- Protected deoxynucleotide triphosphate (dNTP) and tethered dNTP approaches both use terminal deoxynucleotidyl transferase (TdT), a unique and specialized DNA polymerase that does not require a primer template to construct DNA.
- Reactions occur in mild, aqueous conditions, which limits the use of harsh chemicals that cause DNA damage. Therefore, enzymatic approaches can generate longer strands, such as gene fragments, with a lower error rate.

A flowchart comparing the processes of chemical and enzymatic DNA synthesis is provided, illustrating the steps for each method.