TdT Panel for DNA and RNA Synthesis

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INTRODUCTION

In the last decade, there has been a growing interest to shift from outsourced chemical phosphoramidite synthesis to a less toxic and longer DNA and RNA synthesis method. A promising solution for DNA synthesis is using an enzymatic approach with Terminal deoxynucleotidyl Transferases (TdTs) an enzyme that can polymerase nucleotides in an untemplated fashion.

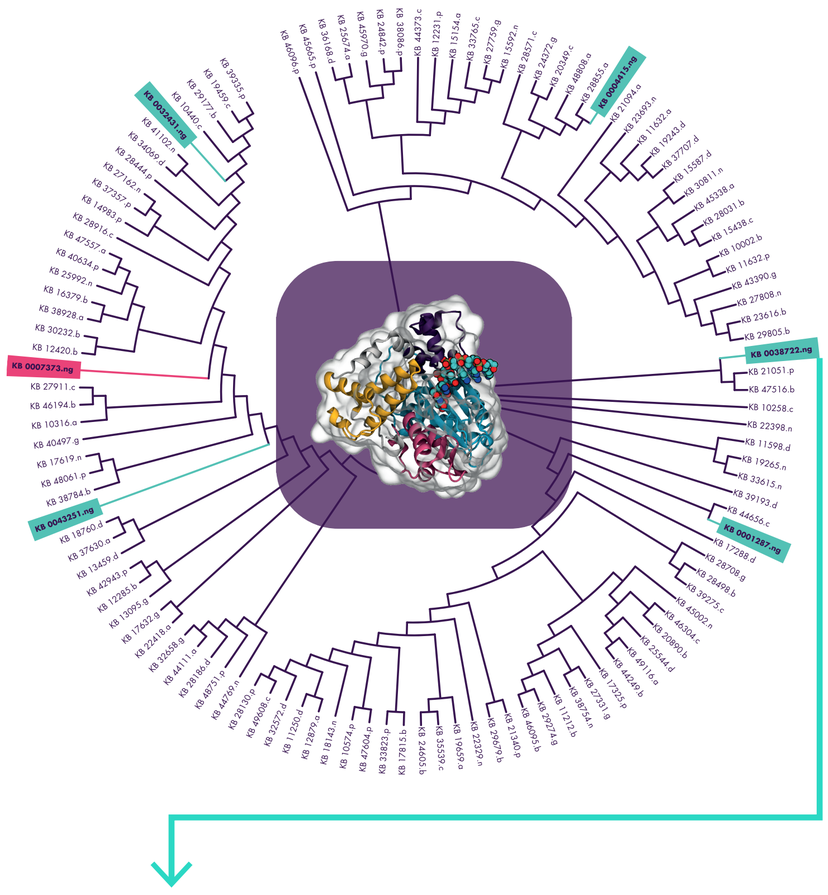
Nonetheless, there are few commercially available TdT enzymes intended for DNA and RNA synthesis. This prompted us to create a panel of six proprietary TdT candidates for DNA/RNA synthesis sourced from diverse origins, both natural and artificial, aiming for optimized features. These six proprietary candidates, strong by themselves, also comprise initial points of exploration for our Protein Engineering team, allowing DNA and RNA Synthesis companies to set the course for us to navigate the sequence landscape and identify the best fit for their needs.

OBJECTIVE

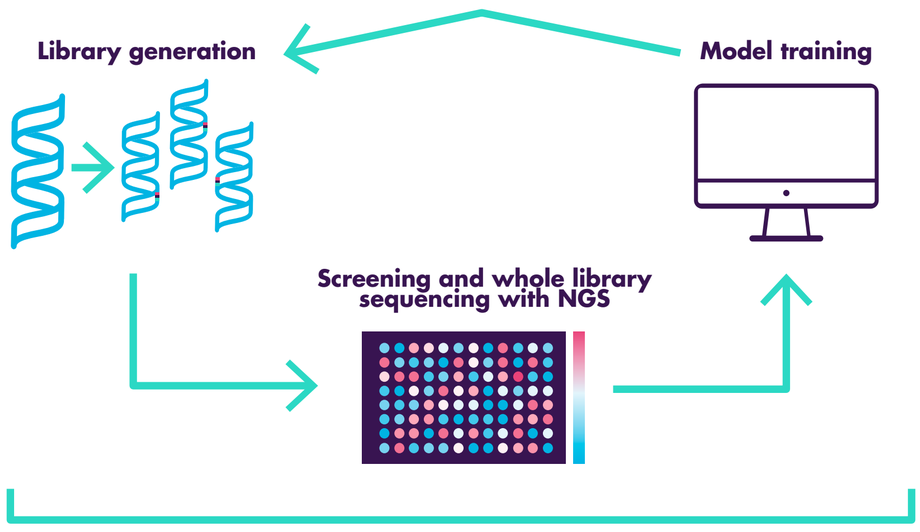
Leverage the Kura's enzyme engineering platform to develop a TdT panel that includes novel proprietary TdT for DNA and RNA synthesis with the following characteristics:

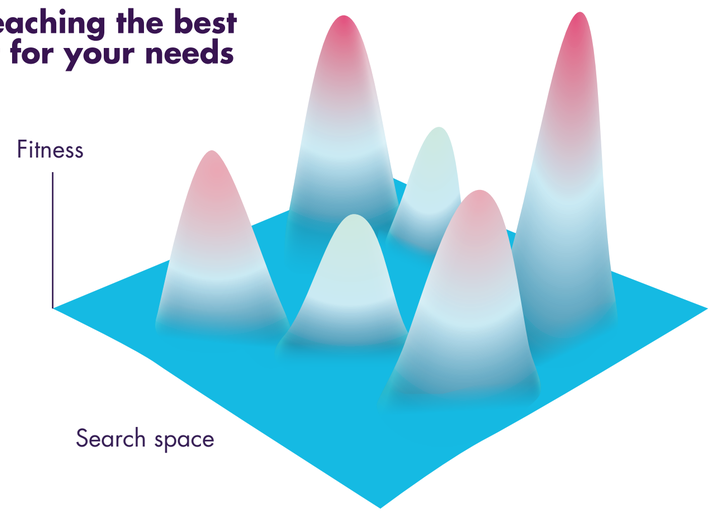
- High coupling efficiency for natural DNA nucleotides
- High coupling efficiency for 3'-reversibly-blocked DNA nucleotides
- High coupling efficiency for natural RNA nucleotides
- Thermostability
- pH Resistance

6 Proprietary TdT enzymes with different features to explore: Sourced from different origins



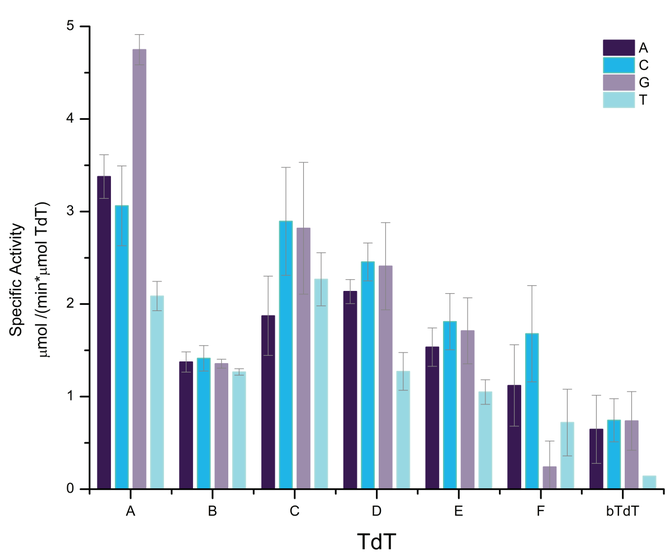
Protein engineering: Tailor a TdT enzyme to your exact needs.





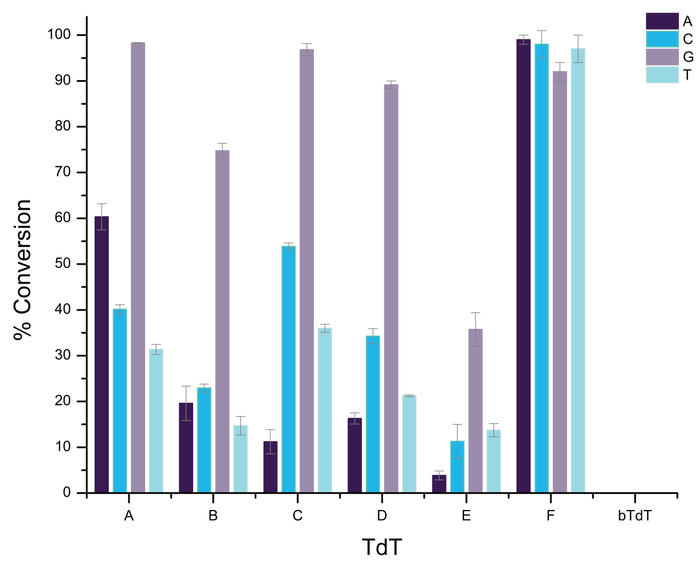
OUR RESULTS

Incorporation of DNA natural nucleotides



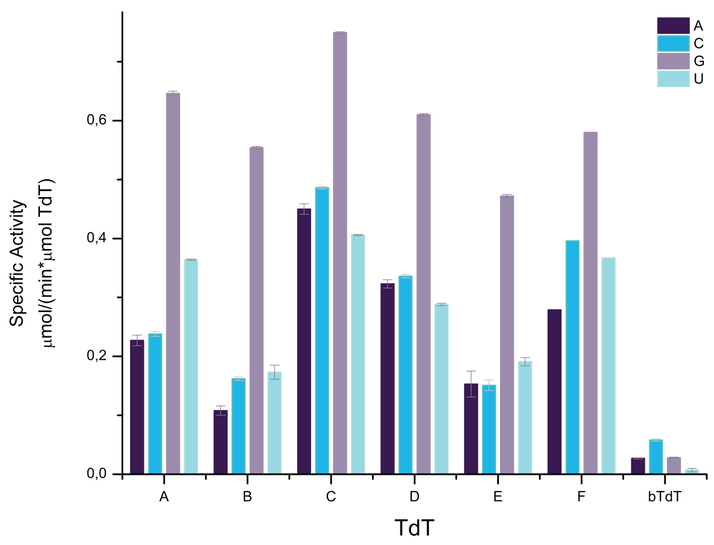
Addition of natural deoxyribonucleotides. Enzymes were incubated with a 1:10 oligo:dNTP ratio for 1 minute at 37°C.

Incorporation of DNA modified nucleotides



Addition of modified nucleotides. Enzymes were incubated with a 1:10 oligo:dNTP ratio for 30 minutes at 37°C. Bovine TdT didn't show activity.

Incorporation of RNA natural nucleotides



Addition of natural ribonucleotides. Enzymes were incubated with a 1:10 oligo:dNTP ratio for 30 minute at 37°C.

TdT Synthesis Panel Enzymes	Thermostability (Tm)
TdT A	54°C
TdT B	46°C
TdT C	51°C
TdT D	52°C
TdT E	38°C
TdT F	48°C

NEXT STEPS

Many companies involved in oligo and gene synthesis are developing proprietary methodologies and instrumentation for EDS. One of the main barriers is the commercial availability of TdT enzymes. To address this challenge, we developed comprehensive TdT panel with a range of distinct features. This panel provides companies with the opportunity to test and select TdT enzymes that best suit their specific synthesis needs. By facilitating access to these critical enzymes, we aim to empower companies to overcome barriers, accelerate their research, and drive innovation in EDS.

Kura intends to:

- Offer a commercial panel of TdT enzymes with different features:
  - Incorporation of natural DNA nucleotides
  - Incorporation of modified DNA nucleotides
  - Incorporation of natural RNA nucleotides
  - Higher thermostability
  - Resistance to pH
- In co-development with strategic partners developing enzymatic DNA or RNA synthesis, engineer TdT to do high fidelity synthesis in customized workflow

Explore our TdT panel today and revolutionize your approach to enzymatic DNA synthesis.