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Oligo Pool Resuspension and PCR

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ABSTRACT

2023 NUS-Singapore iGEM team followed this protocol to resuspend the dried oligo pool and perform PCR to amplify the DNA fragments in the oligo pool.

MATERIALS

KOD OneTM PCR Master Mix (Blue)

PROTOCOL MATERIALS



In 2 steps

SAFETY WARNINGS



Proper lab PPE must be worn at all times.

guidelines

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Protocol status: Working We use this protocol and it's

working

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Oligo Pool Resuspension

- 1 Centrifuge the oligo pool for a few seconds before opening the tube.
- 2 Buffer Calculate the volume of required for the oligo pool (the EB Qiagen Catalog #19086 final concentration of the sample must be at least 20ng/µL).
- 3 into the tube with the Add the respective volume of EB Qiagen Catalog #19086 oligos and resuspend the dried oligos by pipetting the solution.

Oligo Pool PCR

Prepare a PCR tube, and add the following reagents:

Item	Volume
KOD OneTM PCR Master Mix (Blue)	25μL
Each Primer (Two Primers In Total)	1.5µL
Oligo Pool Sample	1μL
DI Water	21μL

5 Run a PCR with the following setting:

Temperature	Duration	
98°C	10s	
60°C	5s	
68°C	10s	
Go to step 1, repeat the cycle 10 times		
68°C	5 minutes	
12°C	Infinite Loop	

Follow the PCR clean-up protocol to isolate the oligos from the PCR product and keep it at Room temperature.

