



FEB 27, 2024

Rapid Sequencing gDNA

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ABSTRACT

ONT Rapid sequencing kit use in a classroom setting.

OPEN  ACCESS



DOI:

dx.doi.org/10.17504/protocols.io.14egn27eyg5d/v1

Protocol Citation: Carlos Goller 2024. Rapid Sequencing gDNA. **protocols.io**
<https://dx.doi.org/10.17504/protocols.io.14egn27eyg5d/v1>

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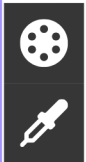
Protocol status: In development
We are still developing and optimizing this protocol


Created: Oct 24, 2022

Library Preparation

2m

1 DNA tagmentation




Thaw kit components at  Room temperature , spin down briefly using a microfuge and mix by pipetting as indicated below:

- Lambda DNA (50 µg/ml): thaw at RT, briefly spin down, mix well by pipetting
- Fragmentation Mix (FRA): not frozen, briefly spin down, mix well by pipetting
- Rapid Adapter (RAP): not frozen, briefly spin down, mix well by pipetting
- Sequencing Buffer (SQB): thaw at RT, briefly spin down, mix well by pipetting*
- Loading Beads (LB): thaw at RT, briefly spin down, mix by pipetting or vortexing immediately before use
- Sequencing Tether (SQT): thaw at RT, briefly spin down, mix well by pipetting

2 Prepare the DNA in Nuclease-free water

2.1 Transfer ~  200 ng genomic DNA into a 1.5 ml Eppendorf DNA LoBind tube

2.2 Adjust the volume to  3.75 µL with Nuclease-free water

2.3 Mix by flicking the tube to avoid unwanted shearing

2.4 Spin down briefly in a microfuge



2.5 In a 0.2 ml thin-walled PCR tube, mix the following:

3.75 μ L 200 ng template DNA

1.25 μ L FRA

2.6 Mix gently by flicking the tube, and spin down.



Incubate the tube at 30 °C for 00:01:00 and then at 80 °C for 00:01:00 . 2m



Briefly put the tube on ice to cool it down.