Sample preparation for Nanopore sequencing

1. DNA isolation
2. library preparation

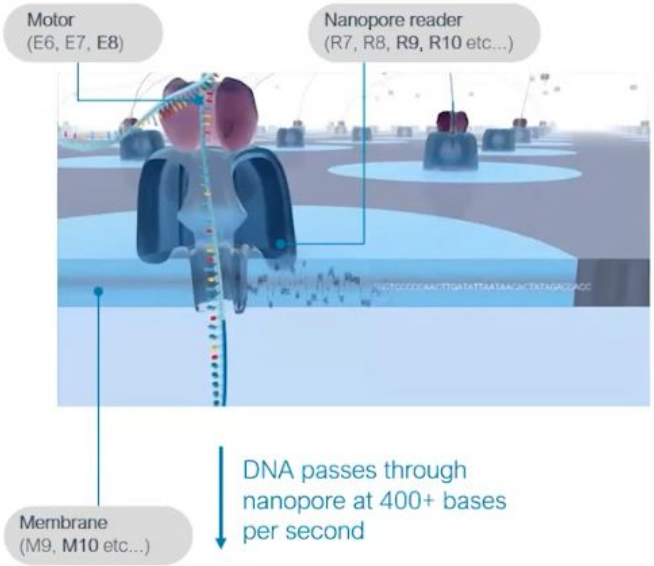
DNA isolation

- homogenise sample, lyse cells
- spin down (soil and cell) debris
- precipitate or degrade proteins, RNA
- bind DNA to a membrane
- wash chemical residues
- elute DNA from membrane

Library preparation (source)

Nanopore sequencing chemistry

Library preparation: converting sample into a format compatible with nanopore sequencing



Experimental set-up

- Ions flow through nanopore embedded within electrically resistant membrane
- DNA/RNA bases perturb current flow
- Motor controls movement

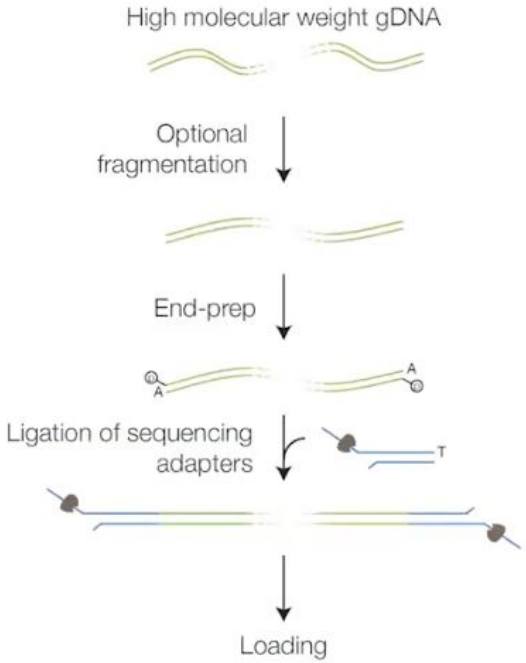
Library prep for nanopore sequencing

- Attachment of "sequencing adapter"
- Motor protein is pre-bound to adapter
- Adapter facilitates tethering

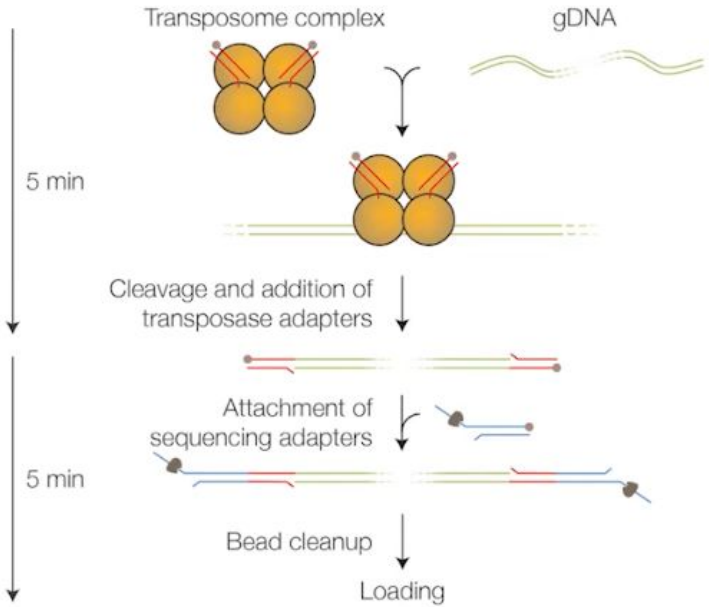
Oxford NANOPORE Technologies

WGS DNA sequencing + optional multiplexing

Ligation Kit



Rapid Barcoding Kit



Other possibilities for library preparation

- direct RNA sequencing, cDNA sequencing
- PCR amplification (for higher throughput or targeted sequencing - 16S)
- Cas9 targeted sequencing