

# Illumina PCR-Free library preparation

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## Abstract

This protocol describes the library preparation for Illumina sequencing. It accompanies the *GigaScience* publication:

Benjamin Istace, et al. (2017) De novo assembly and population genomic survey of natural yeast isolates with the Oxford Nanopore MinION sequencer. *GigaScience*...

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## Protocol

### DNA fragmentation

#### Step 1.

Fragment the DNA (6µg): 100 to 1500bp size using a Covaris E210 sonicator

### End Repair

#### Step 2.

DNA fragments were End-repaired by using NEBNext® End Repair Module

### Clean Up

#### Step 3.

The end-repair reaction was cleaned up with Agencourt AMPure XP beads (1x)

### dA-tailing

#### Step 4.

DNA fragments were dA-tailed by using the NEBNext® dA-Tailing Module

### Clean Up

#### Step 5.

The dA-tail reaction was cleaned up with AMPure beads (1x)

### Adaptors ligation

#### Step 6.

Adaptors were ligated to the dA-tailed DNA fragment using NEBNext® Ligation Module

### Clean Up

#### Step 7.

The ligation reaction was cleaned up with AMPure beads (1x)

## Clean Up

### **Step 8.**

The ligation reaction was cleaned up with AMPure beads (0,6x)

## Quantification

### **Step 9.**

The library was quantified by qPCR by using the KAPA Library Quantification Kit for Illumina Libraries

## Library profile

### **Step 10.**

The library profile was assessed by using a DNA High Sensitivity LabChip kit and an Agilent Bioanalyzer