

Sample requirements for submission of prepared Illumina libraries

Libraries prepared by non-core Centre for Genomic Research staff, performance and throughput cannot be guaranteed.

Sample submission requirements

We ask for the following:

- Libraries are free from contaminants, as enzymatic steps could be inhibited. Purify samples, using columns or Agencourt AMPure XP beads (Beckman Coulter), and elute in nuclease-free water.
- After purification, run each sample on an agarose gel or automated electrophoresis instrument such as Bioanalyzer, TapeStation, Fragment Analyzer or equivalent using High Sensitivity chip to evaluate quality. Please attach a copy of the gel image(s) or trace(s) on the order form, including the relevant ladder. If available, please provide Bioanalyzer traces of the (pooled) libraries and calculate the average fragment size for the entire range of fragments using the Bioanalyzer software
- Quantify the samples by use of a dye-based method such as Qubit (ThermoFisher Scientific) rather than a spectrophotometric method such as NanoDrop.
- Library must be provided free from traces of primer and/or adaptor dimers
- Please supply each sample in a tube labelled with the sample number and/or name exactly as given on the online order form. If more than a tube is provided, labelled them in numerical order for ease of sample identification. Please underline any numbers that could be misread upside-down (e.g. 6/9, 16/91).

Library submission requirements

The concentrations required will depend on the type of platform and run configuration for your project (please check the details in your quote, if you have already obtained one) and will determine the method used to load your samples.

Sequencing Platform and chemistry	Total number of lanes/run required per pool of libraries	Required amount of library*
NovaSeq 6000 SP or S1	1	≥ 2.5 nM in a volume of ≥ 20 µl
NovaSeq 6000 SP or S1	2 (full flowcell)	≥ 5 nM in a volume of ≥ 60 µl
NovaSeq 6000 S2	1	≥ 2.5 nM in a volume of ≥ 20 µl
NovaSeq 6000 S2	2 (full flowcell)	≥ 5 nM in a volume of ≥ 80 µl
NovaSeq 6000 S4	1	≥ 3 nM in a volume of ≥ 20 µl
NovaSeq 6000 S4	2	≥ 3 nM in a volume of ≥ 40 µl
NovaSeq 6000 S4	3	≥ 3 nM in a volume of ≥ 60 µl
NovaSeq 6000 S4	4 (full flowcell)	≥ 10 nM in a volume of ≥ 100 µl
MiSeq	1	≥ 2.5 nM in a volume of ≥ 20 µl
MiSeq (sample requires size selection to remove traces of primer and/or adaptor dimers)	1	≥ 100 ng in a volume of ≤ 30 µl

* These quantities are expected to include sufficient material for library QC and qPCR. In order to convert concentrations from ng/µl to nM, please use the following equation: $nM = (\text{Concentration in ng/}\mu\text{l} \times 1,000,000) / (\text{Average size of library in base pairs} \times 660)$.

If you are unable to meet the stated requirements for your library type, please contact us at CGR.Enquiries@liverpool.ac.uk and we will be happy to offer further advice.