

Sample requirements for submission of prepared Illumina amplicons

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

Sample submission requirements

We ask for the following:

- Amplification conditions should be optimised to ensure synthesis of the expected product(s) of the desired size. In addition, we strongly recommend size selection of the samples *before* purification
- Amplicons are free from contaminants, as enzymatic steps could be inhibited. Purify (size selected) samples, using columns or Agencourt AMPure XP beads, 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.
- Determine sample concentration(s) by a dye-based method such as the PicoGreen or Qubit assays (ThermoFisher).
- 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).
- Please supply **≥100 ng amplified DNA in a maximal volume of 30 µl nuclease-free water** per sample. This will allow us to further size select the sample(s) by use of a Pippin prep instrument, if needed to remove traces of primer-dimers and/or other non-target PCR products.

Experimental design

Please open this link [Standard Illumina sequencing primers](#) for an overview of the standard sequencing primers used for the Illumina platforms. Additional information on primers and adapter sequence can be found on the [Illumina website](#). If you wish to use custom sequencing primers, please contact us at CGR.Enquiries@liverpool.ac.uk to confirm their suitability for the Illumina platforms.

When the samples are submitted [online](#) we request that you upload a document that outlines the entire sequence of the forward and reverse primers (adaptor, index/barcode and template specific primer). Please highlight the barcodes/indexes on the document or provide them as a separate column for each sample.

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.