

NGS sample submission guide **PCR amplicons**



- Follow the instructions below for your sample type and application.
- BaseClear can supply sample collection kits, tubes, barcode stickers, barcoded plates or racks and DNA/RNA Shield solution upon request. Contact your account manager.
- Before sending samples please visit the Order Portal and submit the order form. Provide a PRINTED version of your order form with the shipped samples. Samples received without an order form may cause project delay and extra fees may apply.

Biosafety regulations:

BaseClear can only process micro-organisms of biosafety Class I and II according to European regulations.

PCR AMPLICONS

LIGATION / PCR-BASED

PREPARATION

Amplicon length 100-600 bp

At least 200 ng DNA with a concentration of > 10 ng/ μl

Final volume: Min. 40 µl

ILLUMINA NEXTERA

PREPARATION



Amplicon length > 600 bp

At least 200 ng DNA with a concentration of > 10 ng/ µl



The customer has the responsibility not to ship samples that might contain class III or IV organisms according to EU directive 2000/54/ EU. For Class III and IV organisms, only deactivated cells (in Shield solution) or DNA and RNA can be provided. RNA from RNA viruses cannot be send intact but can be send in Shield solution. Provide samples in clean tubes. Extra fees may apply for additional decontamination work. Possibly contaminated samples will be destroyed and a additional fee will apply.

Postal address:

BaseClear B.V. Attn. Expedition Galileiweg 4 2333 BE Leiden, The Netherlands.

Notes

- Provide material in excess whenever possible.
- DNA concentrations must be measured with an assay specific for doublestranded DNA, i.e. Qubit with the dsDNA Broad Range assay. Spectrophotometric measurements (i.e. Nanodrop) are not specific and can result in overestimated concentrations.
- We recommend that customers check size and concentration by agarose gel electrophoresis or BioAnalyzer (or equivalent) and provide the results.
- Amplicon DNA must be purified and provided in molecular- grade water.
- DNA must not contain RNA inhibitors or other inhibitors.
- Amplicons can be provided in 96 well plates (see requirements below).
- Primers used for amplicon PCR require addition of Illumina adapters.
 Forward 5' TCGTCGGCAGCGTCAGATGTGTATAAGAGACAG-[target specific sequence forward] 3'.

Reverse 5' GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAG-[target specific sequence - reverse] 3'.

Delivery of samples in 96-well format

- For delivery of samples for high throughput analysis.
- Use a high quality seal for the plate and make sure the seal is properly pressed between the wells to avoid cross contamination during shipment.
- Baseclear advices the following 96 well plates and seals: Hard-Shell® 96-Well PCR Plates, low profile, thin wall, skirted, white/clear (HSP9601 BioRad) and MicroAmp[™] Clear Adhesive Film (4306311 Thermofisher).
- Samples must be contiguous in a plate in column format. Do not leave empty wells between wells with samples.
- The maximum number of samples per rack is 94. Leave 2 spaces (G12 and H12) empty for our internal controls (PTC (Positive Template Control) and NTC (No Template Control).
- For multiple plates, completely fill the first plate (up to 94 samples) before filling the next plate.
- Fill out the F089-NG-06-2 form and upload it together with the weborder in the order. Ask at info@basclear.com in case you do not have this document. In our online order portal we create 1 sample line per rack. The name should match the name on the rack and of the F089-NG-06-2 form.