<table>
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<th>PI Name</th>
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<td>Grant # (CHOP)/PO # (External)</td>
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<tr>
<td>Contact Name</td>
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<td>Contact Phone</td>
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<td>Date Submitted</td>
<td>No. of Samples</td>
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**Service Requested**
- □ 16S Tag Sequencing: V1-V2
- □ ITS Tag Sequencing:
- □ Shotgun Metagenomics
- □ Other: __________________________

**Sequencing Chemistry**
- □ MiSeq 500 Cycle
- □ MiSeq 600 Cycle
- □ HiSeq 250 Cycle
- □ Other: __________________________

**Sample Type**
- □ Primary Sample
- □ Premade Library
- □ Genomic DNA
- □ Other: __________________________

**Sample Format**
- □ Tube
- □ 96-well Plate (preferred)

**For customers submitting DNA or premade library**

**DNA Extraction Method** __________________________

**DNA Quantification**
- □ In-house: additional charges will apply
- □ Customer: please add relevant information to the sample metadata form

**DNA Quantification Method**
- □ Qubit/PicoGreen (preferred)
- □ NanoDrop

**For customers submitting premade library**

**DNA Quality Check**
- □ In-house: additional charges will apply
- □ Customer: please provide traces

**DNA Visualization Method**
- □ BioAnalyzer
- □ TapeStation

Premade Library Concentration: __________________________

Premade Library Average Fragment Size: __________________________

If run on MiSeq/HiSeq, loading conc. and cluster density: __________________________
Sample Submission Guidelines

The following guidelines are to ascertain sample concentration, purity, integrity, and fragment size. If you are unable to provide sample this information, we can perform the assays on a fee-for-service basis.

We can still perform sequencing if your sample does not meet the quality requirements of the core. However, run performance is longer guaranteed. For some applications, sub-optimal sample quality may be sufficient to perform sequencing and obtain useful data.

If you are providing samples in a 96-well plate, please follow a column-wise pattern from left to right as indicated below. If possible, leave the indicated wells empty for the addition of controls.

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For nucleic acid sample submissions, please include the following:

1. Nucleic acid concentration determined by Qubit or PicoGreen. Please add to the sample metadata form.

2. Gel, BioAnalyzer, TapeStation, or Fragment Analyzer image in PDF.

   Please indicate the marker/ladder sizes and the amount of nucleic acid loaded in each well. For RNA samples, please submit the RNA Integrity Number (RIN) with BioAnalyzer traces. The RIN should be greater than 7.

3. Nucleic acid purity measurements by UV spectrophotometry. Please add to the sample metadata form.

   For DNA: A260/280 ratio of 1.8 - 2.0 and A260/230 ratio of 1.8 - 2.4.

For pre-made library submissions, please include the following:

Please note, we cannot guarantee the sequencing performance of user-generated libraries.

1. BioAnalyzer, TapeStation, or Fragment Analyzer traces of the library.

2. DNA concentration determined by Qubit.
General considerations during submission

Number of samples – We prepare samples for sequencing in 96-well microtiter plates and we offer sequencing in half- and full-plate formats. A half-plate will hold a maximum of 40 samples and a full-plate will hold a maximum of 88 samples. This leaves us room for eight controls per experiment.

Sample Buffers - Please do not provide samples in buffers containing EDTA as this will interfere with most of the downstream processing steps. Provide samples in sterile H₂O or EB (10 mM Tris-HCl, pH 8.5).

For Nextera XT Submissions - Nextera methods are very sensitive to the quality and concentration of DNA. Even providing DNA at half or twice the desired concentration can have drastic effects on performance. Use a fluorometric method such as Invitrogen´s Qubit or PicoGreen to determine concentration, not spectral methods such as NanoDrop.

Library Preparation – Other than 16S and ITS tag-sequencing, we do not offer library preparation methods other than those from Illumina. However, we will consider requests to perform such preps on a collaborative basis, so please inquire.

Sample Multiplexing (a.k.a. indexing/barcoding) - It is important to specify the read depth requirement that will satisfy your study objective so that we can determine a strategy for barcode and pooling. The read depth requirement may vary depending on the host DNA percentage, biomass abundance, etc. For example in our recent shotgun metagenomic study of gut samples, we found 6 million read pairs per sample to be optimal.

PhiX Spiking: It is a general practice to add PhiX DNA to sequencing mixtures, which helps identify clusters and assess the run parameters. If you are submitting low diversity samples it is your responsibility to inform us so that we can spike higher levels of PhiX to improve cluster identification. Please note that the number of sample reads will decrease proportionally. We are happy to consult on optimal spiking strategies.