Instructions for submitting samples for Illumina Library preparation

1. Download the *Illumina Library Sample Submission Form*, and fill it in completely. Sample names must be simple, short and should not contain period (.), coma (,), star signs (*) or spaces ( ).
2. Samples gel image or Agilent Bioanalyzer, TapeStation or GX traces of the DNA/RNA sample(s) were requested (see Table below).
3. Submit the Sample Submission Form and Gel image or traces on line at: *https://mcic.osu.edu/genomics/illumina-sequencing/submit-samples.*
4. Sample tubes need to be carefully labeled. More than 16 samples have to be submitted in a microtiter, conical bottom plate properly sealed and in the order indicated in the submission form. Use strip caps, not adhesive seals, if you have raised top plates.
5. Bring or ship samples in carefully sealed tubes or plates, on dry ice (RNA) or with ice packs (DNA) to:
   MCIC genomic core
   Selby Hall, room 009
   1680 Madison Avenue
   Wooster, OH
6. MCIC staff can coordinate sample transport from Columbus to Wooster.

Amounts of DNA or RNA sample and quality required:

Library type	Quality and amounts needed
Genomic DNA-Seq*	50uL of RNAase treated gDNA at 30ng-40ng/uL (OD260/280=1.8; OD260/230=2) and gel image or traces
RNA-Seq*	50uL of total RNA at 40ng-50ng/uL and gel image or traces
GBS, ddRAD, RAD libraries	50uL of RNAase treated gDNA at 20ng/uL [GBS], 40ng/uL [RAD, ddRAD] (OD260/280=1.8; OD260/230=2) and gel image or traces
HT plexWell HT LP plexWell	20 ul of RNAase treated gDNA at 1.7 ng/ul, (OD260/280=1.8; OD260/230=2)
Ribosomal 16S/ITS or custom amplicons	25ul of 5 ng/ul RNAase treated genomic DNA (OD260/280=1.8; OD260/230=2), as starting concentration for each amplicon target

⇒ Samples need to be submitted at the required concentrations. If not, we charge $100.00 per plate or samples set for normalization.

⇒ Avoid resuspending nucleic acids in EDTA-containing buffer.

DNA and RNA library preparation. Nucleic acid need to be quantified using a fluorescence based method (qBit, picogreen, …), and purity needs to be assessed using the nanodrop.

*Low input DNA or RNA library preparation. The amounts of sample needed for the library preparation listed in the table above are for standard library preparation protocols. If you do not have the required amounts of nucleic acid, please let the MCIC staff know, so that we can apply protocols for library preparation with very low amounts of input DNA or RNA.

GBS, ddRAD, RAD,… library preparation. gDNA needs to be quantified using a fluorescence based method (qBit, picogreen), and purity needs to be assessed using the nanodrop. It is critical for a successful library and pool preparation that samples are all at the same concentrations,

otherwise samples will be sequenced to different depth. Customer is responsible to verify concentrations.

HT plexWell and HT LP plexWell library preparation: This is a low-cost high-throughput (HT) library preparation method based on tagmentation (sequential transposition reaction). Samples are submitted in 96-well plates or multiples. HT plexWell version is optimized for sequencing of whole genome smaller than 20Mb, while LP plexWell has higher coverage and is optimized for genomes up to 50Mb (*plexWell comparison chart*). Sample gDNA concentration and purity are critical for successful library preparation and sequencing. Concentrations must be validated using a fluorescence method (qBit, picogreen), and purity needs to be assessed using nanodrop. Customer is responsible for verifying concentration and purity. Due to the nature of the library preparation we are not able to offer troubleshooting or re-sequencing for individual samples.

Ribosomal (or other) custom amplicons: samples need to be submitted in microtiter plates (no culture plates and no empty wells, as we use a liquid handlers to prepare libraries). For 16S and ITS amplicons the sequence primers available at *http://mcic.osu.edu/genomics/documents/AmpliconPrimers.pdf*. Let us know if your samples have been already tested with the amplicon primers. If needed we can optimize the conditions for a fee.