**Guidelines for Ready-to-Sequence Illumina libraries**

1. Ready-to-run libraries must not contain primer-dimers or adapter-dimer peak (around 120 bp) on the Bioanalyzer trace.
2. We will not perform any additional clean up or library size selection for your libraries.
3. The standard requirements for library submission are at least 15 ul volume at a concentration of 5 nM. Specific instrument requirements are listed below.
4. Please recommend PhiX spike-ins for libraries containing sequences with low base diversity.
5. The best buffer to ​store and submit libraries is 10 mM Tris​/0.01% Tween-20 ph=8.0 or 8.4​, but EB buffer is also acceptable​​.
6. Use 1.5 ml – 2.0 ml low-bind tubes to ship your libraries.
7. Please note, the DNA insert size(s) should not exceed 700 bp and most Illumina adapters add about 120 bases to the fragment length as observed on the Bioanalyzer. When submitting your libraries for sequencing, provide the bioanalyzer profile, library prep methods, and index sequences used. We will measure the quantity of your libraries using qPCR.
8. It is recommended to measure concentration using fluorescence-based assays such as Qubit. We strongly advise not to use Nanodrop since UV-based measurements are in general inaccurate and may result in significant overestimation, even by orders of magnitude!
9. We cannot guarantee the run yield if your libraries do not meet any of the above criteria.
10. Lower sequencing yield is the likely outcome for library concentrations 1 nM or less, and we cannot guarantee the data quantity or quality for such libraries.
11. We will only rerun sequencing for your libraries if we notice an issue with the sequencer or the sequencing reagents.

**Required information**

Library type:

Library kit used:

% Phix :

Custom primer needed: Yes  No 

Any special run set up parameters:

|  |  |  |
| --- | --- | --- |
| **Sequencing platform** | **Volume** | **Library concentration** |
| NovaSeq Xplus | 150 ul | 4-5 nM |
| MiSeq | 10 ul | 2-3 nM |
| NextSeq 2000 | 10 ul | 2-3 nM |