CENTER FOR MICROBIAL SYSTEMS
UNIVERSITY OF MICHIGAN

Internal
Protocol for submitting DNA for Illumina sequencing genomes in the background of low-diversity runs

Version: 1

Summary:

This protocol is for the submission of DNA for the generation of paired-end libraries for sequencing on the Illumina MiSeq platform. DNA is quantified using a flourometric-based method and diluted to 0.2 ng/µL. Libraries are prepared using the NexteraXT Library prep kit, which uses an enzymatic reaction called tagmentation to fragment the DNA and add adapter sequences. After libraries are prepared they are quantified with another low-diversity library and run together on the MiSeq. The "Preparing Libraries for Sequencing on the MiSeq" (part 15039740, Rev. D) protocol was used to prepare libraries with a final load concentration of 5.5 pM, spiked with 85% low diversity library, 13% genomic library and 2% PhiX. FASTQ files are distributed to the client when the 2 x 250 bp sequencing completes.

Reagents and Materials:

Reagent/Material	Vendor	Stock Number
Eppendorf LoBind tubes	Fisher Scientific	13-698-791

Protocol:

1. In a clean hood, add at least 10 µL of DNA to each of the LoBind tubes. Clearly label plates with PI, Reference ID, and date.

2. Fill out submission form.

3. Send a list of the genome names and shipment tracking information to msmblcore@umich.edu.

4. Deliver on ice to:

Host Microbiome Initiative
University of Michigan Medical School
Internal Medicine/Infectious Diseases
1500 MSRB1
1150 W. Medical Center Drive
Ann Arbor, MI 48109-5666

Please include reference ID on package documentation.