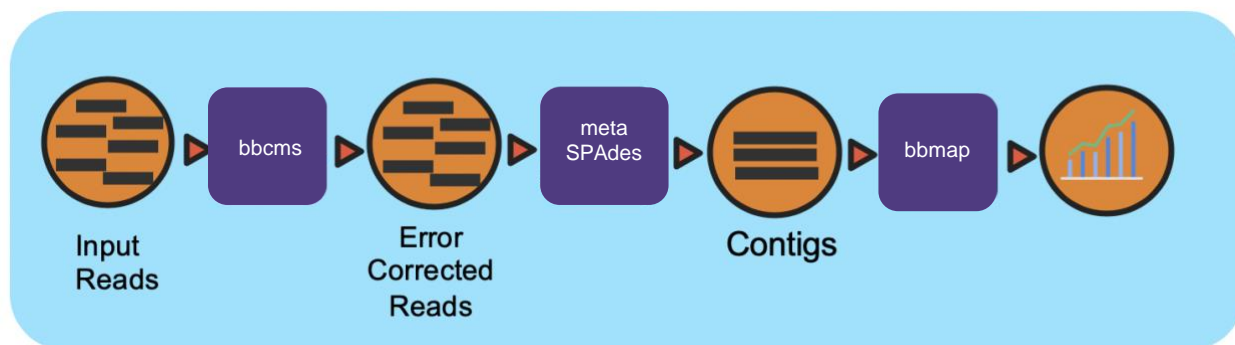


Metagenome Assembly Workflow (v1.0.1)



Overview

This workflow takes in paired-end Illumina data, runs error correction, assembly, and assembly validation.

Running the Workflow

Currently, this workflow can be run in [NMDC EDGE](#) or from the command line (CLI instructions and requirements are found [here](#)).

Tutorial videos on how to run each workflow in NMDC EDGE are found [here](#).

Input

Metagenome Assembly requires paired-end Illumina data as an interleaved file or as separate pairs in FASTQ files. The recommended input is the output from the ReadsQC NMDC workflow.

- **Acceptable file formats:** .fastq, .fq, .fastq.gz, .fq.gz

Details

This workflow takes in paired-end Illumina reads and performs error correction using bbcms (BBTools). Then the corrected reads are assembled using metaSPAdes. After assembly, the reads are mapped back to the contigs by bbmap (BBTools) for coverage information.

Software Versions

- bbcms (BBTools:38.94)
- metaSpades (v3.15.0)
- bbmap (BBTools:38.94)

Output

The main output is the assembled contigs file (assembly_contigs.fna).

Primary Output Files	Description
Assembly Contigs	Final assembly contigs (assembly.contigs.fna)
Assembly Scaffolds	Final assembly scaffolds (assembly_scaffolds.fna)

Assembly AGP	An AGP format file which describes the assembly
Assembly Coverage BAM	Sorted bam file of reads mapping back to the final assembly
Assembly Coverage Stats	Assembled contigs coverage information

Running the Metagenome Assembly Workflow in NMDC EDGE

Select a workflow

1. From the Metagenomics category in the left menu bar, select 'Run a Single Workflow'.
2. Enter a **unique** project name with no spaces (underscores are fine).
3. A description is optional, but helpful.
4. Select 'Metagenome Assembly' from the dropdown menu under Workflow.

The screenshot shows the NMDC EDGE web interface for running a single workflow. The left sidebar is dark blue with a white 'M' logo. It has a 'Metagenomics' dropdown menu that is expanded, showing options like 'Run a Single Workflow' (highlighted in blue), 'Run Multiple Workflows', 'Metatranscriptomics', 'Organic Matter', 'Viruses and Plasmids', and 'Metaproteomics'. The main content area is white and titled 'Run a Single Workflow'. It contains four input fields: 'Project/Run Name' (required, 3-30 characters), 'Description' (optional), 'Workflow' (a dropdown menu with 'Metagenome Assembly' selected), and 'ReadsQC' (optional). Four orange arrows with numbers 1 through 4 point to these elements: 1 points to the 'Run a Single Workflow' menu item, 2 points to the 'Project/Run Name' field, 3 points to the 'Description' field, and 4 points to the 'Metagenome Assembly' option in the 'Workflow' dropdown menu.

Input

This workflow accepts Illumina data in FASTQ format as the input; the file can be interleaved and can be compressed. This input can be the output from the ReadsQC workflow, and this is recommended. **Acceptable file formats:** .fastq, .fq, .fastq.gz, .fq.gz

5. The default setting is for the raw data to be in an interleaved format (paired reads interleaved into one file). If the raw data is paired reads in separate files (forward and reverse), click 'No'.
6. Additional data files (of the same type—interleaved or separate) can be added with the button below.
7. Click the button to the right of the input blank for data to select the data file for the analysis. (If there are separate files, there will be two input blanks.) A box called 'Select a File' will open to allow the user to find the desired file(s) from previously run projects, the public data folder, or files uploaded by the user.
8. Then click 'Submit'.

The screenshot shows the 'Input' section of a web interface. It contains the following elements with numbered arrows pointing to them:

- Arrow 5:** Points to the 'No' button in the 'Is interleaved?' toggle.
- Arrow 6:** Points to the 'Add interleaved fastq' button.
- Arrow 7:** Points to the file selection icon (three horizontal lines) next to the 'interleaved FASTQ #1' input field.
- Arrow 8:** Points to the 'Submit' button at the bottom of the section.

Output

The General section of the output shows which workflow and which tools were run and the run time information.

General					
Workflow	Run	Status	Running Time	Start	End
Metagenome Assembly	On	Done	02:02:22	2021-10-13 22:51:35	2021-10-14 00:53:57
"Project Configuration" : { ... }					

The Metagenome Assembly Result section has all the statistics from the assembly.

Metagenome Assembly Result	
Name	Status
scaffolds	25,324
contigs	25,726
scaf_bp	52,206,897
contig_bp	52,201,077
gap_pct	0.011
scaf_N50	691
scaf_L50	4,103
ctg_N50	724
ctg_L50	3,971
scaf_N90	14,186
scaf_L90	726
ctg_N90	14,473
ctg_L90	716
scaf_logsum	645,093
scaf_powsum	120,098
ctg_logsum	638,015
ctg_powsum	116,432
asm_score	33.765
scaf_max	1,491,105
ctg_max	859,644
scaf_n_gt50K	96
scaf_l_gt50K	20,678,937
scaf_pct_gt50K	39.61
gc_avg	0.473
gc_std	0.062
filename	assembly_scaffolds.fna

The Browser/Download Output section provides output files available to download. The primary result is the assembly_contigs.fna file which can also be the input for the Metagenome Annotation workflow. The pairedMapped_sorted.bam file along with the assembled contigs file can be the input for the MAGs Generation workflow.

Browser/Download Outputs		
File	Size	Last Modified
 MetagenomeAssembly		
assembly.agp	1.72 MB	21 days ago
assembly_contigs.fna	51.30 MB	21 days ago
assembly_scaffolds.fna	51.22 MB	21 days ago
covstats.txt	1.92 MB	21 days ago
pairedMapped.sam.gz	2338.54 MB	21 days ago
pairedMapped_sorted.bam	2130.75 MB	21 days ago
stats.json	619 B	21 days ago