# Metagenomics Assemblers Evaluation [Or Whatever Titus suggests:)]

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#### Abstract

# **Author Summary**

# Introduction

# Materials and Methods

Podar (write correct name) datasets where downloaded from XX. The dataset represent XX brief description for the data.

## **Preparing Reads Treatments**

- 1. Quality Filtering: In this treatment, low quality bases were trimmed and low quality reads were removed using trimmomatic [?]. After quality trimming reads were either directly assembled, or first preprocessed with digital normalization and then assembled.
- 2. **Digital Normalization:** Digital normalization works after sequencing data has been generated, progressively removing high-coverage reads from shotgun data sets. This normalizes average coverage to a specified value, reducing sampling variation while removing reads, and also removing the many errors contained within those reads. This data and error reduction results in dramatically decreased computational requirements for de novo assembly. Moreover, unlike experimental normalization where abundance information is removed prior to sequencing, in digital normalization this information can be recovered from the unnormalized reads [?]
- 3. Partitioning: In this treatment, we partitioned the filtered data set based on de Bruijn graph connectivity and assembled each partition independently. Subsequently, partitioning separates reads based on transitive connectivity, resulting in easily assembled subsets of reads.

#### 4. Reinflation:

After Quality trimming, we either preprocess the reads using digital normalization, partitioning, or reinflation. We assembled the reads using four different assemblers; Velvet [?], Idba [?], Spades [?], and Megahit [?] and using the various reads treatments.

#### Results

### **Evaluating Assembly Quality Using Various Reads Treatments**

Table 1 shows . Table 2 shows .

Table 1. Evaluating Assembly Using Different Treatments

Treatment/Quality Metric	Quality Filtering	Digital Normalization	Partition	Reinflation			
(1) Velvet							
Genome Fraction	72.949	89.043	88.879	-			
Unaligned Length	8,977,149	10,909,693	11,317,834	-			
Misassembled contigs length							
(2) Idba							
Genome Fraction	90.969	91.003	90.082	88.346			
Unaligned Length	10,709,716	10,637,811	10,644,357	10,288,486			
Misassembled contigs length							
(3) Spades							
Genome Fraction	90.424	90.173	89.272	89.798			
Unaligned Length	10,597,529	10,621,398	10,500,235	10,461,672			
Misassembled contigs length							
(4) Megahit							
Genome Fraction	89.961		88.769				
Unaligned Length	10,525,444		10,565,036				
Misassembled contigs length							

# Time and Memory Comparisons

Table 3 shows the running time and memory utilizations for four assemblers and different reads treatments. There is no significant enhancement difference in Genome fraction and unaligned contains length. However, the results show a significant decrease in running time and memory utilization based on some treatments.

# **Assembly Validation**

# Raw Reads Mapping

We estimated the percentage of unaligned sequences by each assembly treatment and using the four assemblers. We mapped the raw reads to each assembly. Then we extracted the unaligned sequences to each assembly. Table XX shows the percentages of unaligned sequences from raw reads to each assembly treatment using the four assemblers under study.

For all treatments assemblies, the full set of trimmed reads were used for mapping. Default parameters were used, and both paired ends and singletons were mapped. Samtools [?] was used for format conversion from SAM to BAM format, and also to calculate the percentage of mapped reads.

# Discussion

Main points Explain the significance of each treatment

Table 2. Raw Reads Mapping to Assembly With Different Treatment Analysis

Treatment/Quality Metric	Quality Filtering	Digital Normalization	Partition	Reinflation			
(1) Velvet							
No. of Unaligned Sequences				-			
(2) Idba							
No. of Unaligned Sequences	2092523	2761871	3602590				
(3) Spades							
No. of Unaligned Sequences	3013782		3579651				
(4) Megahit							
No. of Unaligned Sequences							

Table 3. Running Time and Memory Utilization

Treatment/Quality Metric	Quality Filtering	Digital Normalization	Partition	Reinflation			
(1) Velvet							
Running Time	60:42:52	6:48:46	4:30:36	-			
Memory Utilization in KB	1594851536	827412304	1156729920	-			
(2) Idba							
Running Time	33:53:46	6:34:24	6:34:24	2:33:17			
Memory Utilization in KB	129853424	104736448	93584624	393938608			
(3) Spades							
Running Time	67:02:16	15:53:10	7:54:26	6:50:46			
Memory Utilization in KB	400340512	127423856	129715072	434531888			
(4) Megahit							
Running Time			0:00:27				
Memory Utilization in KB			7548668				

# Acknowledgments

# References

@articleBrown2012, Author = C Titus Brown and Adina Howe and Qingpeng Zhang and Alexis B Pyrkosz and Timothy H Brom, Journal = , Month = May, Number = , Title = A reference-free algorithm for computational normalization of shotgun sequencing data, Volume = , Year = 2012

# Figure Legends

# **Tables**

# **Supporting Information Legends**