

1. Project Information

Program	Microbial/CSP 2012
PMO Project	0
Seq Proj ID	1027109
Sequencing Project Name	Leptolyngbya sp. HL7711_P1F1 JGI 000150CP-N13
JGI Project ID	0

2. Read Statistics

Illumina Std PE Statistics

File name	7666.6.80850.ATGAGC.fastq
Library	TGTH
Number of reads	25,892,744
Sequencing depth [†]	777X
Read type	2x150 bp

[†] A genome size of 5.0 Mbp was assumed in this calculation.

3. Read QC Results

The following are the results of reads screened against contaminants. Pairs of matching reads were removed from the dataset.

Illumina Std PE Read Filter Statistics

Description	Num Reads	Pct Reads
Input	25,892,744	100
Contam removed	406	0.0
Artifact removed	911,516	3.5
Total removed	5,892,744	22.8
Total remaining	20,000,000	77.2

List of Contaminants Removed

Description	Num Reads	Pct Reads
gi 357579550 Canis.lupus_familiaris_chr12	214	0.00
gi 357579577 Canis.lupus_familiaris_chr3	74	0.00
human_chr2	66	0.00
gi 357579566 Canis.lupus_familiaris_chr6	34	0.00
gi 362110642 Felis_catus_breed_Abyssinian_chrC2	34	0.00
human_chr4	18	0.00
human_chr5	16	0.00

gi 357579535 Canis_lupus_familiaris_chr20	16	0.00
human_chr7	16	0.00
gi 357579571 Canis_lupus_familiaris_chr5	16	0.00
human_chr8	6	0.00
human_chr20	6	0.00
human_chr17	2	0.00
human_chr14	2	0.00
human_chr10	2	0.00
gi 357579523 Canis_lupus_familiaris_chr27	2	0.00

The following are the results of reads screened against potential reagent and process contaminants but were not removed from the dataset.

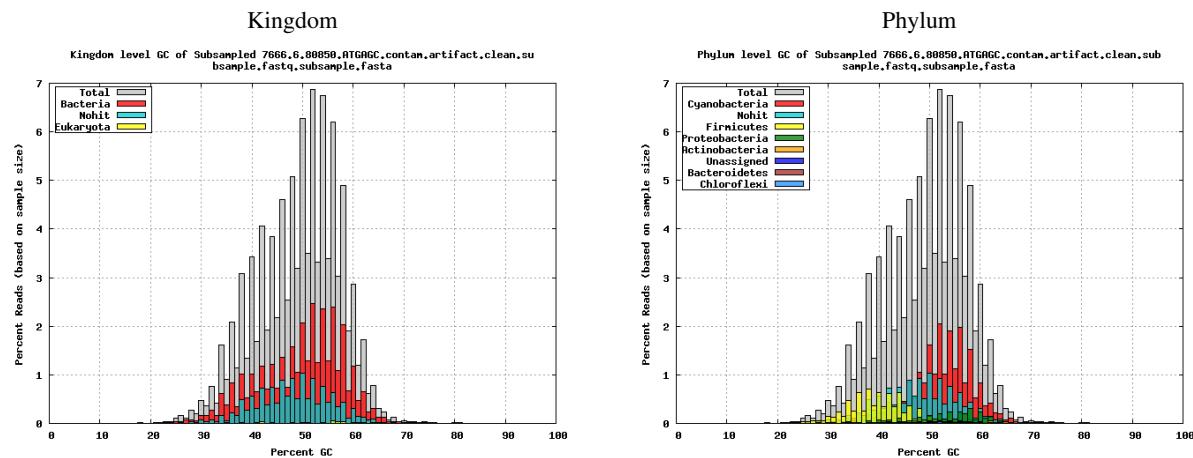
Illumina Std PE Contamination Identification Statistics

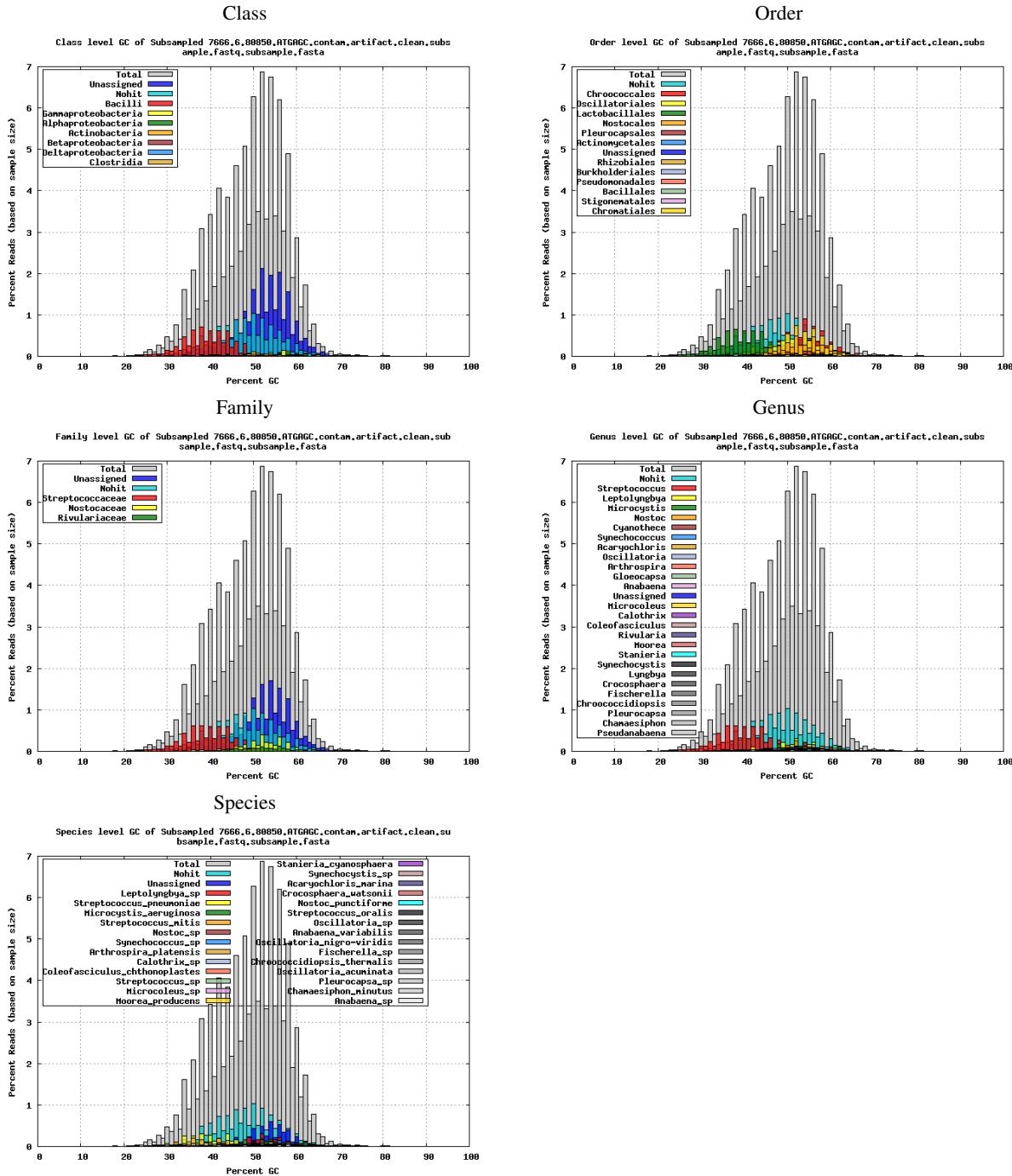
Description	Num Reads	Pct Reads
Input	25,892,744	100
Contam identified	4	0.0

List of Contaminants Identified

Description	Num Reads	Pct Reads
<i>Escherichia</i>	2	0.00
<i>Ralstonia</i>	2	0.00

GC histogram of the reads subsampled to 10k, overlaid with GC of hits based on BLASTX, shown for different taxonomic levels.



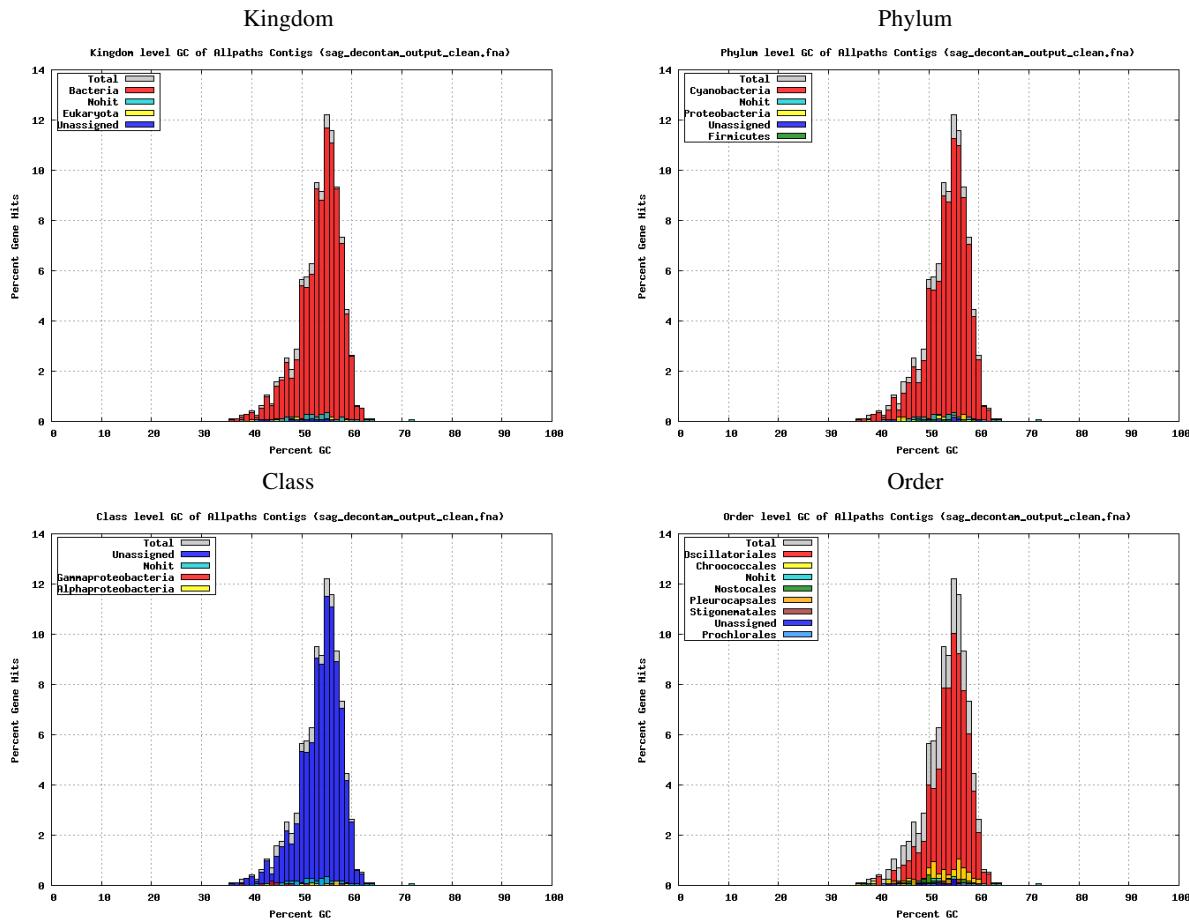


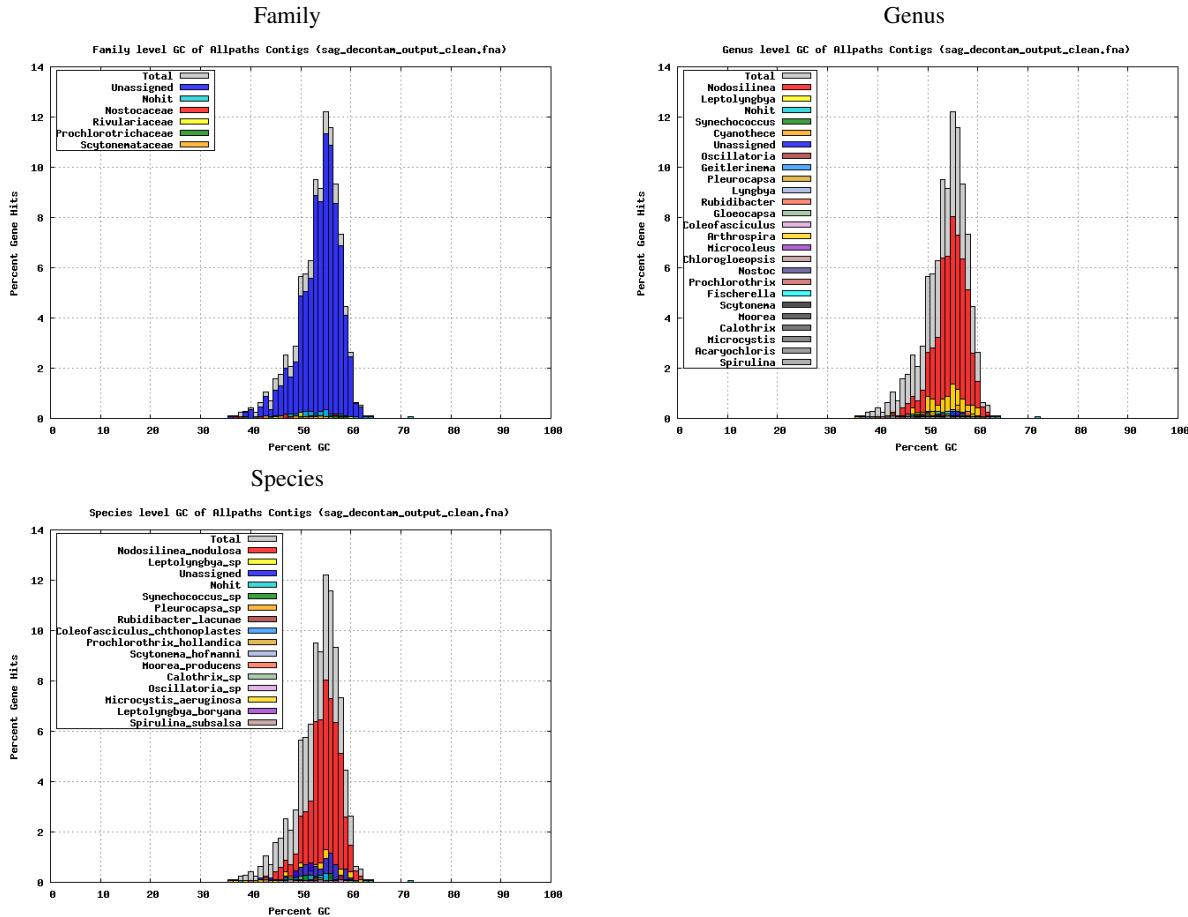
4. Assembly Statistics

Assembly method	SPAdes with auto decontamination
Scaffold total	89
Contig total	89
Scaffold sequence length	1.8 Mb
Contig sequence length	1.8 Mb (0.0% gap)
Scaffold N/L50	22/27.8 kb
Contig N/L50	22/27.8 kb
Largest Contig	112.7 kb
Number of scaffolds >50 kb	3
Pct of genome in scaffolds >50 kb	13.3
Pct of reads assembled (raw)	86.1
Pct of reads assembled (decontam)	18.4

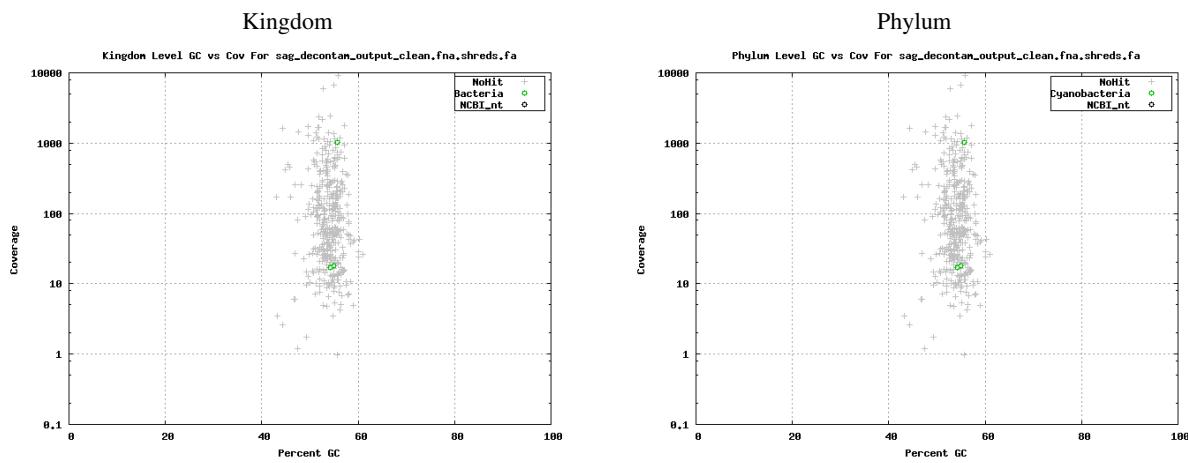
5. Assembly QC Results

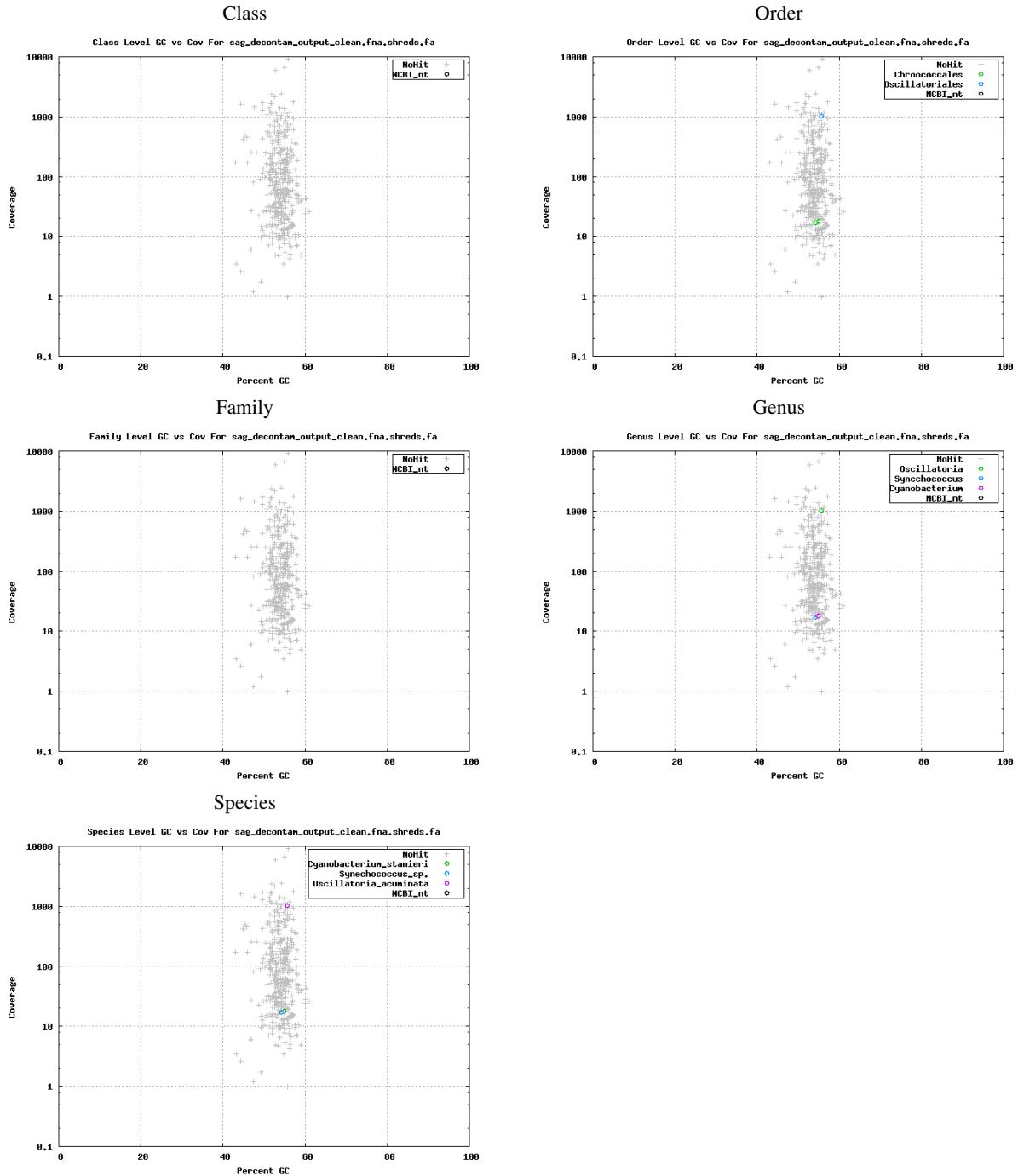
GC histogram of the predicted genes on each contig, overlaid with GC of hits based on BLASTP, shown for different taxonomic levels.



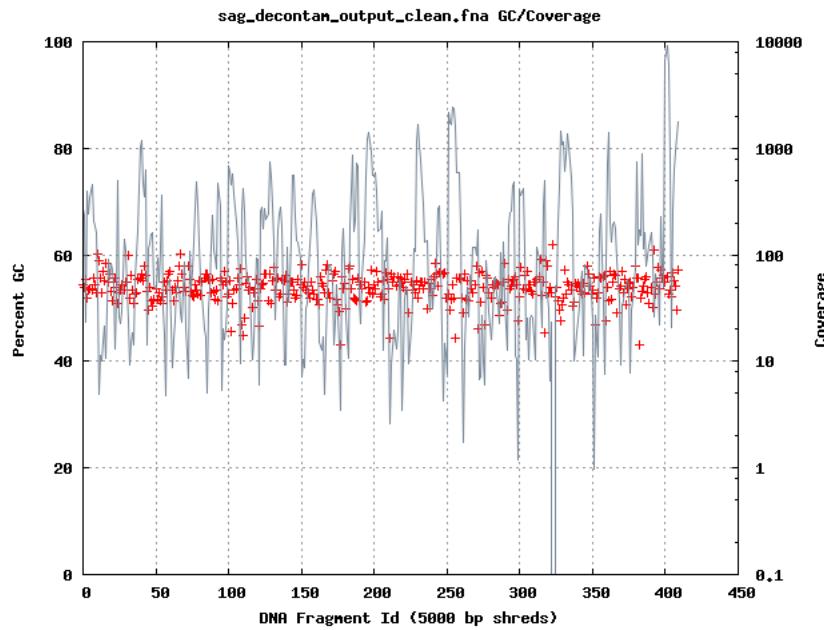


GC vs coverage based on GC of NCBI nt and Greengenes 16S rRNA gene hits to the assembly using megablast, shown for different taxonomic levels.

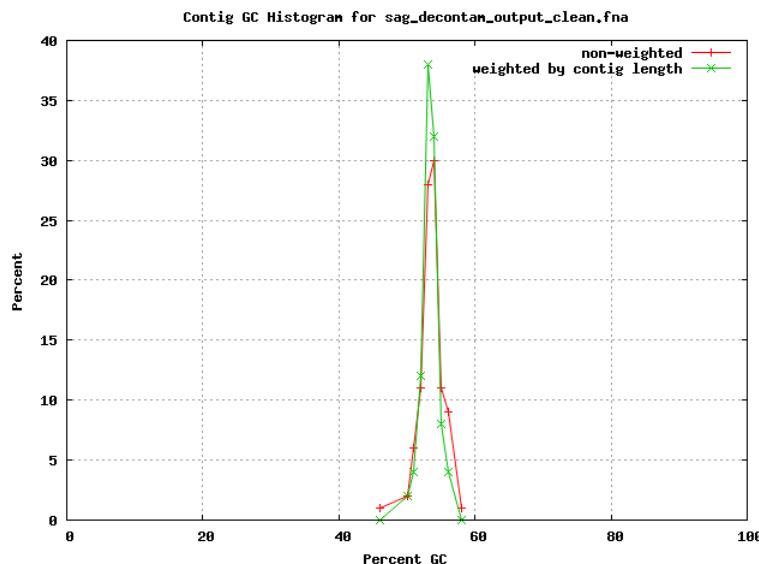




Coverage vs GC. Contigs were shredded into non-overlapping 5kbp and the GC of each shred was plotted as a point, colored by scaffold id. Coverage was calculated by mapping the fragment library to the final assembly and plotted as connected points.



GC histogram of the contigs, including contig length weighted distribution.



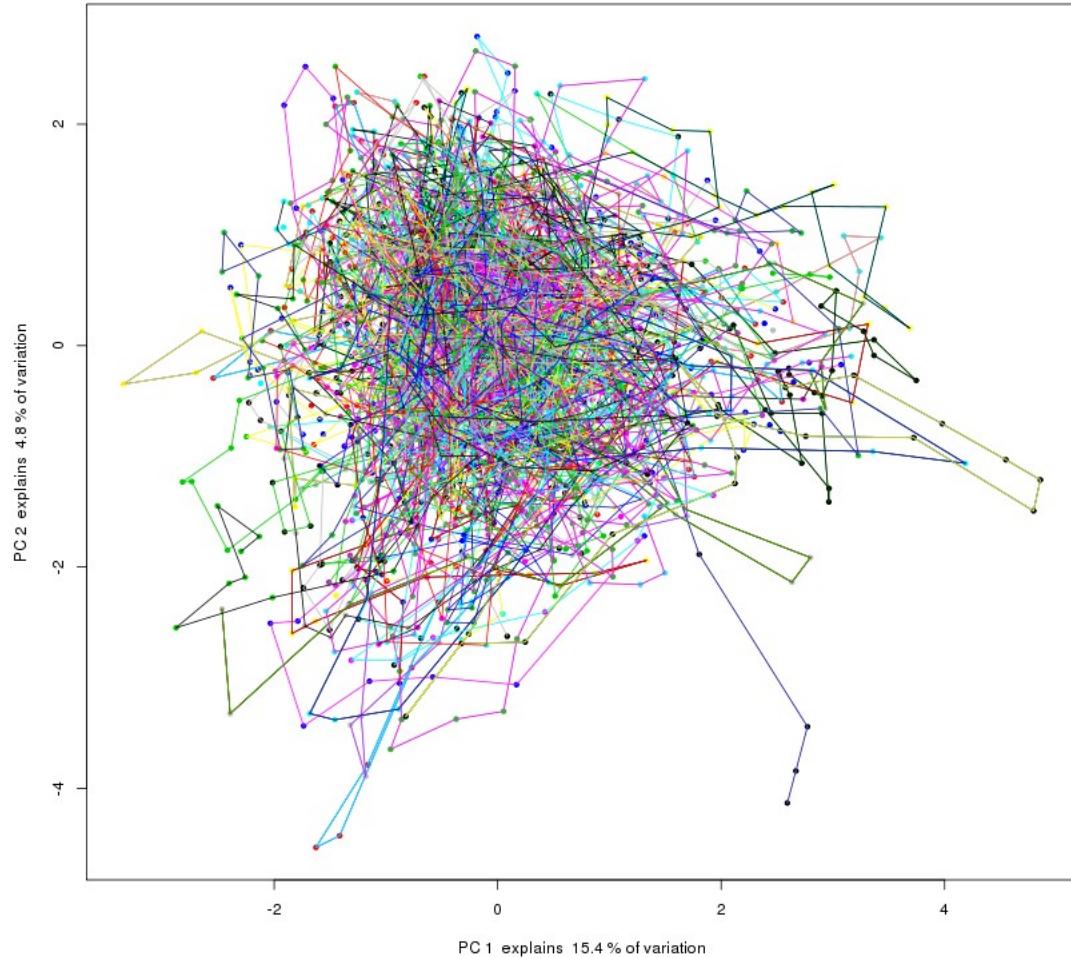
List of contigs and average percent GC, grouped in bins of 5:

Pct GC Bin	Contig Name
45	NODE_370.length_4109 cov_23.4595.ID_761
50	NODE_1.length_112716 cov_337.799.ID_1, NODE_2.length_78255 cov_173.62.ID_3, NODE_5.length_50128 cov_892.299.ID_9, NODE_6.length_47984 cov_585.106.ID_11, NODE_7.length_47831 cov_289.411.ID_13, NODE_8.length_43507 cov_239.441.ID_15, NODE_9.length_41592 cov_461.835.ID_17, NODE_11.length_39809 cov_150.281.ID_21, NODE_12.length_38661 cov_113.403.ID_23, NODE_17.length_36535 cov_172.756.ID_33, NODE_18.length_35864 cov_111.637.ID_35, NODE_20.length_35490 cov_188.719.ID_39, NODE_22.length_35144 cov_31.9722.ID_43, NODE_23.length_33127 cov_69.1028.ID_45,

	NODE_25.length_31681_cov_33.3651.ID_49, NODE_27.length_31165_cov_22.4463.ID_53, NODE_28.length_31096_cov_81.6598.ID_55, NODE_29.length_30495_cov_91.0686.ID_57, NODE_31.length_29442_cov_23.1106.ID_61, NODE_40.length_27752_cov_28.6984.ID_79, NODE_43.length_26896_cov_117.437.ID_85, NODE_44.length_26819_cov_52.0024.ID_87, NODE_45.length_26749_cov_93.7587.ID_89, NODE_47.length_24824_cov_13.8339.ID_93, NODE_51.length_23594_cov_45.0261.ID_101, NODE_52.length_23442_cov_26.4064.ID_103, NODE_54.length_22939_cov_209.589.ID_107, NODE_62.length_21925_cov_22.5763.ID_127, NODE_63.length_21831_cov_579.735.ID_129, NODE_64.length_21769_cov_33.0418.ID_131, NODE_66.length_21207_cov_3567.19.ID_139, NODE_70.length_20600_cov_39.0933.ID_147, NODE_73.length_20347_cov_54.0533.ID_137, NODE_76.length_19988_cov_408.194.ID_157, NODE_78.length_19934_cov_242.145.ID_161, NODE_89.length_18820_cov_160.303.ID_183, NODE_90.length_18636_cov_20.9363.ID_185, NODE_93.length_18489_cov_56.1142.ID_195, NODE_96.length_18158_cov_42.2007.ID_201, NODE_105.length_17574_cov_35.1555.ID_219, NODE_108.length_17442_cov_48.8376.ID_225, NODE_114.length_16806_cov_57.2095.ID_235, NODE_125.length_15639_cov_256.505.ID_261, NODE_129.length_15360_cov_13.1574.ID_269, NODE_130.length_15250_cov_96.6511.ID_271, NODE_137.length_14175_cov_9.3313.ID_189, NODE_141.length_13911_cov_82.9161.ID_291, NODE_150.length_13524_cov_129.163.ID_307, NODE_151.length_13449_cov_82.541.ID_309, NODE_154.length_13175_cov_8.2702.ID_315, NODE_163.length_12650_cov_49.1755.ID_333, NODE_183.length_11041_cov_15.0459.ID_373, NODE_184.length_10842_cov_249.717.ID_375, NODE_189.length_10515_cov_11.3779.ID_385, NODE_194.length_10217_cov_7.15174.ID_395, NODE_204.length_9734_cov_12.7251.ID_415, NODE_207.length_9436_cov_152.862.ID_425, NODE_222.length_8796_cov_54.7775.ID_455, NODE_224.length_8780_cov_9.22029.ID_459, NODE_252.length_7749_cov_13.4426.ID_517, NODE_263.length_7091_cov_26.1867.ID_539, NODE_276.length_6911_cov_15.4377.ID_569, NODE_286.length_6247_cov_9.87403.ID_589, NODE_292.length_6039_cov_30.7418.ID_607, NODE_327.length_5134_cov_15.4054.ID_119, NODE_421.length_3101_cov_4.97242.ID_859, NODE_424.length_3079_cov_15.2622.ID_865, NODE_451.length_2782_cov_6.96883.ID_917 NODE_489.length_2354_cov_26.1096.ID_991
55	NODE_21.length_35229_cov_87.8306.ID_41, NODE_32.length_29068_cov_34.5693.ID_63, NODE_67.length_20985_cov_25.4636.ID_141, NODE_80.length_19804_cov_35.3215.ID_165, NODE_102.length_17817_cov_54.0754.ID_213, NODE_107.length_17503_cov_9.84726.ID_223, NODE_119.length_16254_cov_92.5644.ID_249, NODE_136.length_14237_cov_37.7756.ID_283, NODE_168.length_12102_cov_8.66265.ID_343, NODE_182.length_11054_cov_96.6539.ID_371, NODE_257.length_7622_cov_5.58081.ID_527, NODE_304.length_5818_cov_30.0344.ID_631, NODE_318.length_5324_cov_6.21959.ID_659, NODE_391.length_3642_cov_8.90605.ID_801, NODE_412.length_3220_cov_15.7134.ID_841, NODE_428.length_3051_cov_8.74132.ID_873, NODE_463.length_2670_cov_7.97935.ID_941, NODE_499.length_2265_cov_3.41674.ID_1011 NODE_512.length_2121_cov_4.37561.ID_1037

Principal component analysis of tetramer frequencies of contigs. Detectable variations are highlighted in color.

sag_decontam_output_clean.fna - PC1 vs PC2



Estimated genome recovery derived from analysis of universal single-copy genes detected in final assembly.

HMM	Pct Recovered
bacteria	55.16 %
archaea	30.18 %

6. Sequence Data Availability

The following sequence fasta files can be downloaded from our JGI portal website.
<http://www.jgi.doe.gov/genome-projects>

Filename	Description
sag_decontam_output_clean.fna	SPAdes with auto decontamination

7. Annotation Data Availability

The annotation of the assembled contigs can be found within IMG.
<http://img.jgi.doe.gov>

8. Methods

Single Cell Minimal Draft

Genome sequencing and assembly

The draft genome of was generated at the DOE Joint genome Institute (JGI) using the Illumina technology [1]. An Illumina std shotgun library was constructed and sequenced using the Illumina HiSeq 2000 platform which generated 25,892,744 reads totaling 3,883.9 Mb. All general aspects of library construction and sequencing performed at the JGI can be found at <http://www.jgi.doe.gov>. All raw Illumina sequence data was passed through DUK, a filtering program developed at JGI, which removes known Illumina sequencing and library preparation artifacts [2]. Following steps were then performed for assembly: (1) artifact filtered Illumina reads were assembled using SPAdes [3] (version 3.0.0), (3) Parameters for assembly steps were `-t 16 -m 120 --sc --careful --12`. The final draft assembly contained 89 contigs in 89 scaffolds, totalling 1.8 Mb in size. The final assembly was based on 3,000.0 Mb of Illumina data. Based on a presumed genome size of 5.0 Mb, the average input read coverage used for the assembly was 600.0X.

Genome annotation

Genes were identified using Prodigal [4], followed by a round of manual curation using GenePRIMP [5] for finished genomes and Draft genomes in fewer than 20 scaffolds. The predicted CDSs were translated and used to search the National Center for Biotechnology Information (NCBI) nonredundant database, UniProt, TIGRFam, Pfam, KEGG, COG, and InterPro databases. The tRNAscanSE tool [6] was used to find tRNA genes, whereas ribosomal RNA genes were found by searches against models of the ribosomal RNA genes built from SILVA [7]. Other non-coding RNAs such as the RNA components of the protein secretion complex and the RNase P were identified by searching the genome for the corresponding Rfam profiles using INFERNAL [8]. Additional gene prediction analysis and manual functional annotation was performed within the Integrated Microbial Genomes (IMG) platform [9] developed by the Joint Genome Institute, Walnut Creek, CA, USA [10].

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8. INFERNAL. Inference of RNA alignments. <http://infernal.janelia.org>.
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10. Markowitz VM, Mavromatis K, Ivanova NN, Chen IMA, Chu K, Kyrpides NC. IMG ER: a system for microbial genome annotation expert review and curation. *Bioinformatics* 2009; 25:2271–2278.