

16S NGS METAGENOME ANALYSIS REPORT



2022

1. Analysis Content

1.1 DNA isolation from samples

Genomic DNA isolation from environmental soil samples was done using SuSpin Bacterial Fecal/Soil DNA Isolation Kit, Cat. No.: NA01B100 “SuGenomik Biotechnology”. DNA quality and quantity were measured spectrophotometrically and fluorometrically.

1.2 16s rRNA V3-V4 region amplification

V4 regions -which will be used in species identification- from 16s rRNA gene were amplified with Bacteria-16s-V4-Seq kit Cat. No.: NGS01A025 by SimpliAmp Thermal Cycler as the cycling program below;

- 95°C 10 minutes – initial denaturation (HotStart activity)
- 35 cycles:
 - 95°C for 45 sec - denaturation
 - 53-50°C for 45 sec – annealing
 - 72°C for 60 sec – extension
- 72°C for 3 min – final extension

1.3 NGS library preparation and sequencing

16s rRNA V4 amplicons were purified using ABMGood “Column-Pure PCR Clean-Up Kit, Cat. No.: D509”. NGS libraries and indexing respectively were prepared using Illumina “Nextera XT DNA Library Prep Kit, Cat. No.: FC-131-1096” and “TG Nextera XT Index Kit v2 Set A (96 Indices, 384 Samples), Cat. No.: TG-131-2001”. Sequencing was carried out with Illumina MiSeq platform as paired-end (PE) 2x150 bases.

1.4 Bioinformatics analysis

Raw NGS reads were quality checked, trimmed (if necessary) and classified based on OTU (operational taxonomic unit) criteria using Kraken metagenomics system. This system assigns taxonomic labels to the short DNA sequences with high precision and speed. For more information, refer to the “Wood, D. E., & Salzberg, S. L. (2014). Kraken: ultrafast metagenomics sequence classification using exact alignments. Genome biology, 15(3), R46.”

1.5 Preparing a report

Bacterial distributions in samples were prepared as a report as well as additional files such as complete OTU distribution per sample, various diversity analyses were provided.

2. Taxonomic Classification Report

2.1 Sequencing Statistics

Sample	Reads	Avg Read Length	Reads Classified
HT-1	128052	114.0	128052 / 100.00%
HT-2	218420	116.4	218420 / 100.00%
HT-3	218420	116.4	218420 / 100.00%
HT-4	378008	115.6	378008 / 100.00%
HT-5	51351	116.5	51351 / 100.00%
HT-6	205532	116.7	205532 / 100.00%
HT-7	35709	136.4	35709 / 100.00%
HT-8	93208	115.3	93208 / 100.00%
HT-10	45067	108.5	45067 / 100.00%
HT-11	339980	107.3	339980 / 100.00%
R2_GT9	179050	106.7	179050 / 100.00%
R2_GT12	35307	108.8	35307 / 100.00%

2.2 Taxonomy Statistics

α-Diversity at Species Level		
Sample	Shannon Index (H) / (H / LN(N))	Simpson Index (1-D)
HT-1	4.822 / 0.7173	0.9789
HT-2	4.874 / 0.7038	0.9763
HT-3	4.874 / 0.7038	0.9763
HT-4	4.75 / 0.6678	0.9777
HT-5	4.98 / 0.7737	0.9834
HT-6	4.869 / 0.6985	0.9748
HT-7	0.651 / 0.1427	0.2535
HT-8	5.215 / 0.7688	0.9849
HT-10	5.23 / 0.7838	0.9824
HT-11	5.25 / 0.7059	0.9798
R2_GT9	5.32 / 0.7392	0.9844
R2_GT12	4.972 / 0.7551	0.9776

* Indicates species diversity in samples. Simpsons index takes a value between 0 and 1. 1 indicates diversity while 0 indicates no diversity. Shannon index usually takes a value between 1.5 and 3.5, and as this index increases, the diversity increases.

2.3 Diversity Analysis

At species level,

- Diversity curve,
- Principal coordinate analysis (PCoA) and,
- Rarefaction curves analyses were done and produced plots were given in the attached “plot” file.

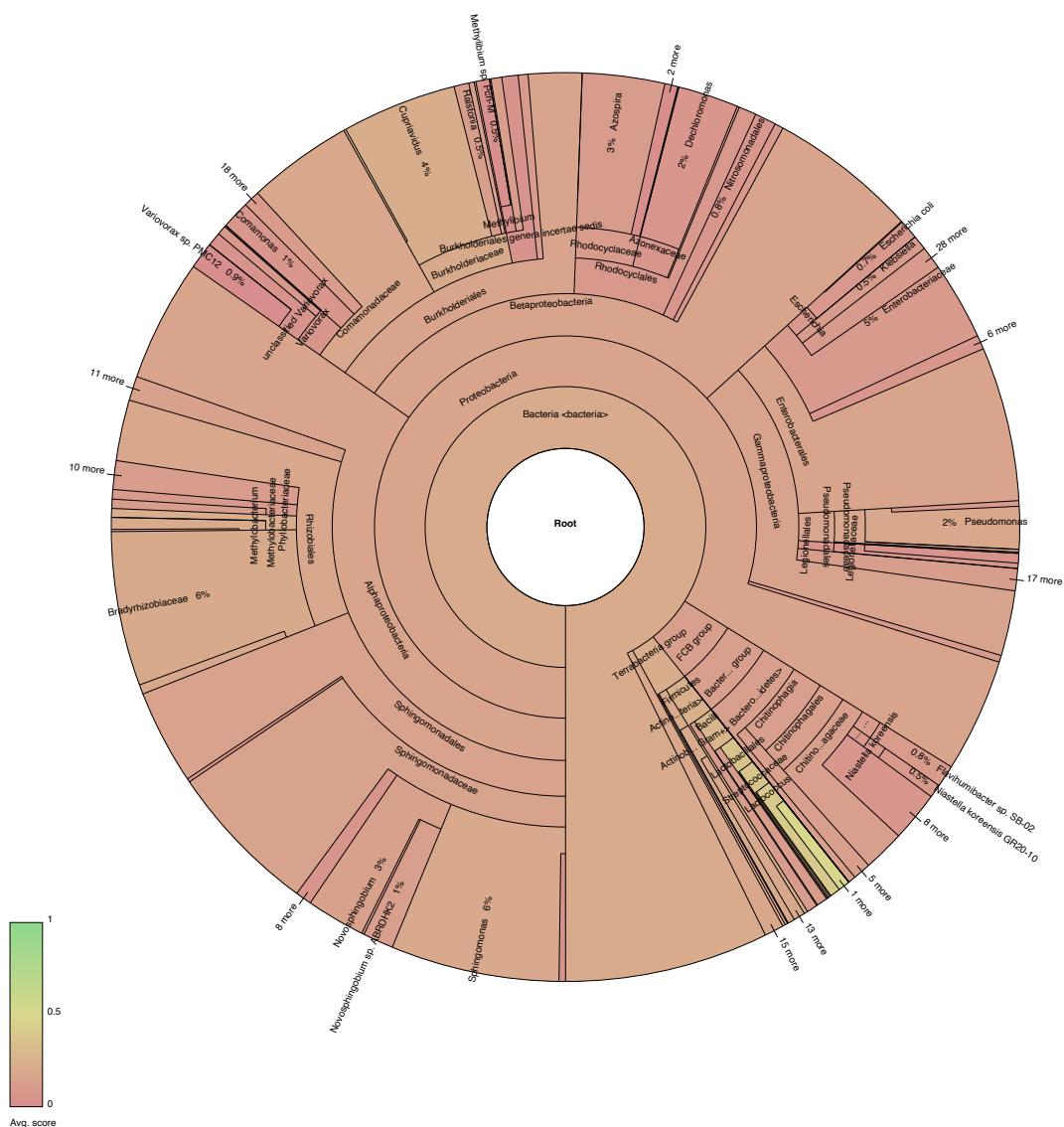
Diversity curve shows the minimum, average and maximum number of total OTUs in the samples. This chart can be used to assess the diversity between similar or different data sets. This analysis can be applied on 2 samples and more.

PCoA plot is a 2-dimensional graph showing similarities and differences between samples. This analysis can be applied on 2 samples and more.

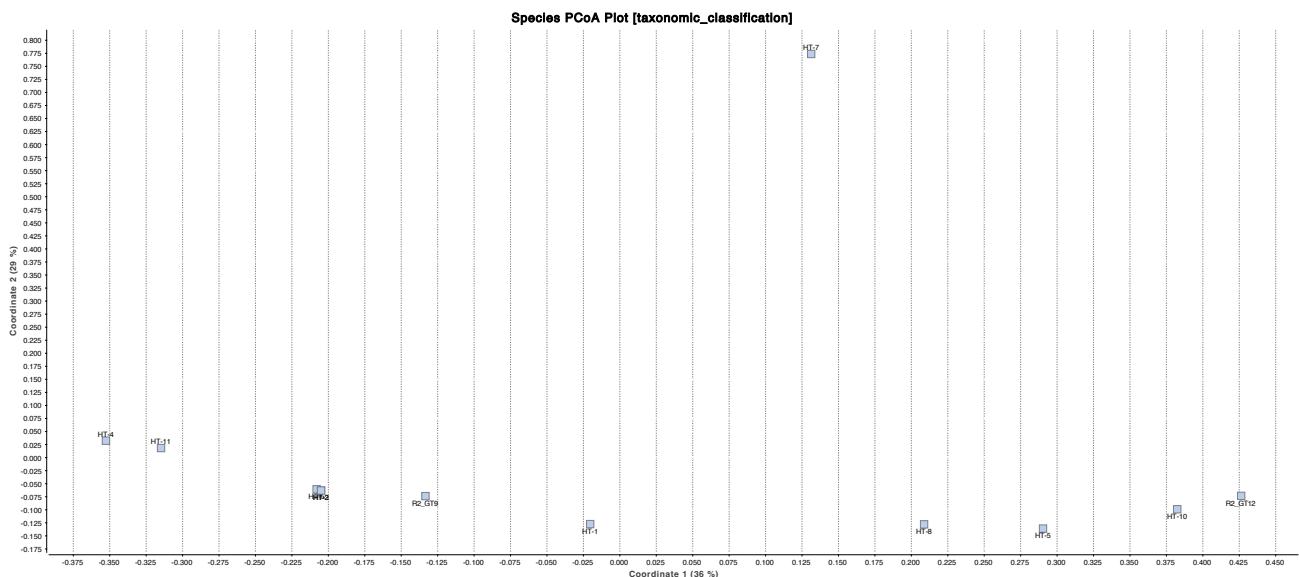
Rarefaction curve is a widely used technique in OTU analysis in ecological studies. This plot shows the expected number of OTUs (Y-axis) in n NGS readings (X-axis). Purpose of this curve is to show whether the sequencing coverage is depth enough to determine the total number of OTUs in a given sample. If this curve reaches the plateau, it indicates the species diversity in given samples. This analysis can be applied on 3 samples and more.

3. Species Level Taxonomic Distributions

Note: Below are given the tables and pie chart distributions of individual samples, each represented with top 10 species at highest reading counts. In addition, entire bacterial distributions in samples are given in the attached “table” file.



Sample Korona picture for HT-1



Species diversity PcoA curve

Species lists (10 mostly represented), please see attached full species lists.

HT-1		
Taxa	Count	%
Novosphingobium sp. ABRDHK2	1399	6.87
Variovorax sp. PMC12	1209	5.93
Flavihumibacter sp. SB-02	1003	4.92
Escherichia coli	923	4.53
Methylibium sp. Pch-M	621	3.05
Niastella koreensis	609	2.99
Oryzomicrobium terrae	552	2.71
Lactococcus lactis	533	2.62
Ralstonia pickettii	494	2.42
Filimonas lacunae	466	2.29

HT-2		
Taxa	Count	%
Escherichia coli	3243	9.96
Novosphingobium sp. ABRDHK2	1840	5.65
Salmonella enterica	1406	4.32
Ralstonia pickettii	1128	3.47
Clostridium estertheticum	1126	3.46
Pseudoclostridium thermosuccinogenes	943	2.9
Klebsiella pneumoniae	795	2.44
Clostridium sp. SYSU GA15002T	715	2.2
Lactococcus lactis	693	2.13
Variovorax sp. PMC12	656	2.02

HT-3		
Taxa	Count	%
Escherichia coli	3243	9.96
Novosphingobium sp. ABRDHK2	1840	5.65
Salmonella enterica	1406	4.32

HT-3		
Taxa	Count	%
Ralstonia pickettii	1128	3.47
Clostridium estertheticum	1126	3.46
Pseudoclostridium thermosuccinogenes	943	2.9
Klebsiella pneumoniae	795	2.44
Clostridium sp. SYSU GA15002T	715	2.2
Lactococcus lactis	693	2.13
Variovorax sp. PMC12	656	2.02

HT-4

Taxa	Count	%
Sorangium cellulosum	4776	6.21
Sandaracinus amyloyticus	4688	6.09
Escherichia coli	3951	5.14
Variovorax sp. PMC12	3496	4.54
Niastella koreensis	3070	3.99
Variovorax paradoxus	2794	3.63
Flavihumibacter sp. SB-02	2781	3.61
Novosphingobium sp. ABRDHK2	2245	2.92
Filimonas lacunae	1908	2.48
Salmonella enterica	1798	2.34

HT-5

Taxa	Count	%
Novosphingobium sp. ABRDHK2	497	5.43
Niastella koreensis	435	4.75
Variovorax sp. PMC12	426	4.66
Escherichia coli	391	4.27
Oryzomicrobium terrae	290	3.17
Variovorax paradoxus	254	2.78
Ralstonia pickettii	201	2.2
Salmonella enterica	187	2.04
Methylibium sp. Pch-M	186	2.03
Flavihumibacter sp. SB-02	179	1.96

HT-6

Taxa	Count	%
Mesorhizobium terrae	4218	10.6
Escherichia coli	2531	6.36
Novosphingobium sp. ABRDHK2	1565	3.93
Salmonella enterica	1267	3.18
Klebsiella pneumoniae	1267	3.18
Ralstonia pickettii	1206	3.03
Niastella koreensis	1129	2.84
Variovorax sp. PMC12	1044	2.62
Variovorax paradoxus	1041	2.61
Filimonas lacunae	774	1.94

HT-7

Taxa	Count	%
Bifidobacterium longum	24124	86.03

HT-7		
Taxa	Count	%
Escherichia coli	1836	6.55
Pantoea ananatis	1282	4.57
Klebsiella pneumoniae	194	0.69
Salmonella enterica	55	0.2
Bacillus tropicus	32	0.11
Staphylococcus aureus	28	0.1
Streptococcus pyogenes	28	0.1
Bacillus cereus	28	0.1
Gibbsiella quercinecans	25	0.09
HT-8		
Taxa	Count	%
Escherichia coli	928	6.39
Ralstonia pickettii	656	4.52
Lactococcus lactis	656	4.52
Legionella pneumophila	452	3.11
Novosphingobium sp. ABRDHK2	389	2.68
Salmonella enterica	351	2.42
Variovorax sp. PMC12	332	2.29
Gilliamella apicola	284	1.96
Tessaracoccus sp. T2.5-30	265	1.82
Nocardioides sp. JS614	255	1.76
HT-10		
Taxa	Count	%
Escherichia coli	766	8.44
Gilliamella apicola	382	4.21
Salmonella enterica	377	4.15
Desulfosporosinus meridiei	367	4.04
Novosphingobium sp. ABRDHK2	292	3.22
Niastella koreensis	254	2.8
Klebsiella pneumoniae	192	2.12
Ferrovibrio terrae	161	1.77
Tessaracoccus sp. T2.5-30	152	1.67
Ralstonia pickettii	133	1.47
HT-11		
Taxa	Count	%
Desulfosporosinus meridiei	7620	10.1
Novosphingobium sp. ABRDHK2	3137	4.16
Escherichia coli	3047	4.04
Niastella koreensis	2502	3.32
Ferrovibrio terrae	2023	2.68
Tessaracoccus sp. T2.5-30	1795	2.38
Desulfotomaculum reducens	1769	2.35
Salmonella enterica	1765	2.34
Desulfosporosinus youngiae	1431	1.9
Klebsiella pneumoniae	1192	1.58
R2_GT9		

Taxa	Count	%
Novosphingobium sp. ABRDHK2	2283	6.76
Escherichia coli	1791	5.3
Desulfosporosinus meridiei	1275	3.78
Salmonella enterica	1090	3.23
Lactobacillus ruminis	959	2.84
Niastella koreensis	904	2.68
Klebsiella pneumoniae	829	2.46
Variovorax paradoxus	715	2.12
Ralstonia pickettii	595	1.76
Ferrovibrio terrae	571	1.69

R2_GT12		
Taxa	Count	%
Desulfosporosinus meridiei	662	7.49
Mycoavidus cysteinexigens	659	7.46
Escherichia coli	482	5.45
Gilliamella apicola	399	4.52
Paraburkholderia rhizoxinica	326	3.69
Ferrovibrio terrae	230	2.6
Niastella koreensis	223	2.52
Salmonella enterica	221	2.5
Desulfosporosinus youngiae	138	1.56
Arachidicoccus ginsenosidivorans	126	1.43

4. Other Taxonomic Distributions

Superkingdom

HT-1		
Taxa	Count	%
Bacteria	128052	100.0
HT-2		
Taxa	Count	%
Bacteria	218420	100.0
HT-3		
Taxa	Count	%
Bacteria	218420	100.0
HT-4		
Taxa	Count	%
Bacteria	378008	100.0
HT-5		
Taxa	Count	%
Bacteria	51351	100.0

HT-6		
Taxa	Count	%
Bacteria	205532	100.0
HT-7		
Taxa	Count	%
Bacteria	35709	100.0
HT-8		
Taxa	Count	%
Bacteria	93208	100.0
HT-10		
Taxa	Count	%
Bacteria	45067	100.0
HT-11		
Taxa	Count	%
Bacteria	339980	100.0
R2_GT9		
Taxa	Count	%
Bacteria	179050	100.0
R2_GT12		
Taxa	Count	%
Bacteria	35307	100.0

Phylum

HT-1		
Taxa	Count	%
Proteobacteria	107369	90.52
Bacteroidetes	6941	5.85
Firmicutes	2433	2.05
Actinobacteria	878	0.74
Acidobacteria	380	0.32
Verrucomicrobia	364	0.31
Cyanobacteria	51	0.04
Tenericutes	38	0.03
Planctomycetes	32	0.03
Deferribacteres	22	0.02
HT-2		
Taxa	Count	%
Proteobacteria	187025	92.63
Firmicutes	7655	3.79
Bacteroidetes	4441	2.2
Actinobacteria	1133	0.56
Verrucomicrobia	630	0.31
Acidobacteria	554	0.27
Tenericutes	184	0.09
Cyanobacteria	39	0.02
Planctomycetes	35	0.02
Thermodesulfobacteriia	29	0.01

HT-3		
Taxa	Count	%
Proteobacteria	187025	92.63
Firmicutes	7655	3.79
Bacteroidetes	4441	2.2
Actinobacteria	1133	0.56
Verrucomicrobia	630	0.31
Acidobacteria	554	0.27
Tenericutes	184	0.09
Cyanobacteria	39	0.02
Planctomycetes	35	0.02
Thermodesulfobacteria	29	0.01
HT-4		
Taxa	Count	%
Proteobacteria	312500	88.87
Bacteroidetes	23399	6.65
Verrucomicrobia	5408	1.54
Firmicutes	4805	1.37
Actinobacteria	3775	1.07
Acidobacteria	809	0.23
Cyanobacteria	183	0.05
Tenericutes	143	0.04
Planctomycetes	108	0.03
Chloroflexi	99	0.03
HT-5		
Taxa	Count	%
Proteobacteria	42143	88.04
Bacteroidetes	3537	7.39
Firmicutes	1158	2.42
Actinobacteria	565	1.18
Verrucomicrobia	174	0.36
Acidobacteria	158	0.33
Cyanobacteria	26	0.05
Planctomycetes	18	0.04
Tenericutes	17	0.04
Spirochaetes	14	0.03
HT-6		
Taxa	Count	%
Proteobacteria	160335	84.41
Firmicutes	16299	8.58
Bacteroidetes	9603	5.06
Actinobacteria	2167	1.14
Verrucomicrobia	710	0.37
Acidobacteria	348	0.18
Cyanobacteria	106	0.06
Tenericutes	95	0.05
Planctomycetes	59	0.03
Chloroflexi	31	0.02

HT-7		
Taxa	Count	%
Actinobacteria	24133	76.59
Proteobacteria	4498	14.27
Firmicutes	2861	9.08
Bacteroidetes	14	0.04
Tenericutes	2	0.01
Chlamydiae	1	0.0
Nitrospirae	1	0.0
Chlorobi	0	0.0
Fusobacteria	0	0.0
Balneolaeota	0	0.0

HT-8		
Taxa	Count	%
Proteobacteria	77145	90.09
Firmicutes	3180	3.71
Actinobacteria	2481	2.9
Bacteroidetes	2406	2.81
Acidobacteria	130	0.15
Cyanobacteria	72	0.08
Verrucomicrobia	38	0.04
Tenericutes	37	0.04
Chloroflexi	16	0.02
Gemmatimonadetes	16	0.02

HT-10		
Taxa	Count	%
Proteobacteria	32169	84.79
Bacteroidetes	2398	6.32
Firmicutes	2057	5.42
Actinobacteria	999	2.63
Verrucomicrobia	72	0.19
Acidobacteria	52	0.14
Cyanobacteria	40	0.11
Tenericutes	25	0.07
Planctomycetes	18	0.05
Synergistetes	16	0.04

HT-11		
Taxa	Count	%
Proteobacteria	236822	78.11
Firmicutes	28607	9.44
Bacteroidetes	24427	8.06
Actinobacteria	10365	3.42
Verrucomicrobia	801	0.26
Acidobacteria	467	0.15
Cyanobacteria	457	0.15
Planctomycetes	200	0.07
Tenericutes	160	0.05
Spirochaetes	99	0.03

R2_GT9		
Taxa	Count	%
Proteobacteria	135430	84.83
Bacteroidetes	10609	6.65
Firmicutes	9735	6.1
Actinobacteria	2494	1.56
Verrucomicrobia	339	0.21
Acidobacteria	327	0.2
Cyanobacteria	123	0.08
Tenericutes	88	0.06
Planctomycetes	71	0.04
Spirochaetes	50	0.03

R2_GT12		
Taxa	Count	%
Proteobacteria	23964	80.12
Firmicutes	2734	9.14
Bacteroidetes	2146	7.18
Actinobacteria	760	2.54
Verrucomicrobia	91	0.3
Acidobacteria	62	0.21
Cyanobacteria	36	0.12
Planctomycetes	21	0.07
Synergistetes	10	0.03
Fusobacteria	9	0.03

Class

HT-1		
Taxa	Count	%
Alphaproteobacteria	44457	39.41
Betaproteobacteria	36545	32.4
Gammaproteobacteria	20877	18.51
Chitinophagia	5789	5.13
Bacilli	1775	1.57
Actinobacteria	845	0.75
Flavobacteriia	490	0.43
Clostridia	466	0.41
Acidobacteriia	365	0.32
Verrucomicrobiae	282	0.25

HT-2		
Taxa	Count	%
Gammaproteobacteria	66901	35.12
Alphaproteobacteria	62540	32.83
Betaproteobacteria	46137	24.22
Clostridia	4981	2.62
Chitinophagia	3191	1.68
Bacilli	2166	1.14
Actinobacteria	1107	0.58
Deltaproteobacteria	573	0.3

HT-2		
Taxa	Count	%
Flavobacteriia	511	0.27
Acidobacteriia	506	0.27
HT-3		
Taxa	Count	%
Gammaproteobacteria	66901	35.12
Alphaproteobacteria	62540	32.83
Betaproteobacteria	46137	24.22
Clostridia	4981	2.62
Chitinophagia	3191	1.68
Bacilli	2166	1.14
Actinobacteria	1107	0.58
Deltaproteobacteria	573	0.3
Flavobacteriia	511	0.27
Acidobacteriia	506	0.27
HT-4		
Taxa	Count	%
Alphaproteobacteria	101551	30.33
Betaproteobacteria	96246	28.74
Gammaproteobacteria	83794	25.02
Chitinophagia	20776	6.2
Deltaproteobacteria	16111	4.81
Actinobacteria	3647	1.09
Bacilli	2364	0.71
Verrucomicrobiae	2255	0.67
Opitutae	2189	0.65
Clostridia	1907	0.57
HT-5		
Taxa	Count	%
Betaproteobacteria	16951	37.08
Alphaproteobacteria	14249	31.17
Gammaproteobacteria	8610	18.84
Chitinophagia	3057	6.69
Clostridia	563	1.23
Actinobacteria	551	1.21
Bacilli	481	1.05
Deltaproteobacteria	386	0.84
Flavobacteriia	170	0.37
Acidobacteriia	132	0.29
HT-6		
Taxa	Count	%
Gammaproteobacteria	57874	32.27
Betaproteobacteria	49129	27.39
Alphaproteobacteria	44202	24.64
Bacilli	11267	6.28
Chitinophagia	8112	4.52
Clostridia	2539	1.42

HT-6		
Taxa	Count	%
Actinobacteria	2103	1.17
Deltaproteobacteria	1497	0.83
Flavobacteriia	487	0.27
Opitutae	347	0.19
HT-7		
Taxa	Count	%
Actinobacteria	24133	76.67
Gammaproteobacteria	4423	14.05
Bacilli	2858	9.08
Betaproteobacteria	24	0.08
Alphaproteobacteria	15	0.05
Sphingobacteriia	9	0.03
Flavobacteriia	4	0.01
Clostridia	3	0.01
Mollicutes	2	0.01
Epsilonproteobacteria	2	0.01
HT-8		
Taxa	Count	%
Betaproteobacteria	28416	35.7
Gammaproteobacteria	23030	28.93
Alphaproteobacteria	19859	24.95
Bacilli	2484	3.12
Actinobacteria	2446	3.07
Chitinophagia	2023	2.54
Clostridia	464	0.58
Cytophagia	127	0.16
Deltaproteobacteria	125	0.16
Flavobacteriia	108	0.14
HT-10		
Taxa	Count	%
Alphaproteobacteria	10815	30.6
Gammaproteobacteria	9752	27.59
Betaproteobacteria	9359	26.48
Chitinophagia	1744	4.93
Actinobacteria	984	2.78
Clostridia	929	2.63
Bacilli	851	2.41
Flavobacteriia	160	0.45
Cytophagia	141	0.4
Bacteroidia	132	0.37
HT-11		
Taxa	Count	%
Alphaproteobacteria	94472	33.44
Betaproteobacteria	71283	25.24
Gammaproteobacteria	54995	19.47
Chitinophagia	19203	6.8

HT-11		
Taxa	Count	%
Clostridia	18851	6.67
Actinobacteria	10065	3.56
Bacilli	6396	2.26
Bacteroidia	1145	0.41
Cytophagia	1031	0.36
Flavobacteriia	898	0.32
R2_GT9		
Taxa	Count	%
Alphaproteobacteria	53429	35.89
Betaproteobacteria	38249	25.69
Gammaproteobacteria	34878	23.43
Chitinophagia	7843	5.27
Bacilli	4919	3.3
Clostridia	3330	2.24
Actinobacteria	2429	1.63
Flavobacteriia	641	0.43
Bacteroidia	570	0.38
Cytophagia	526	0.35
R2_GT12		
Taxa	Count	%
Betaproteobacteria	10238	36.72
Alphaproteobacteria	6179	22.16
Gammaproteobacteria	5924	21.25
Chitinophagia	1629	5.84
Clostridia	1373	4.92
Bacilli	1013	3.63
Actinobacteria	744	2.67
Flavobacteriia	139	0.5
Bacteroidia	109	0.39
Cytophagia	84	0.3

Order

HT-1		
Taxa	Count	%
Sphingomonadales	24349	25.17
Burkholderiales	20320	21.0
Enterobacterales	13851	14.32
Rhizobiales	13432	13.88
Rhodocyclales	8053	8.32
Chitinophagales	5789	5.98
Pseudomonadales	2372	2.45
Lactobacillales	1237	1.28
Nitrosomonadales	972	1.0
Legionellales	688	0.71
HT-2		
Taxa	Count	%

HT-2		
Taxa	Count	%
Enterobacterales	48522	29.61
Sphingomonadales	34677	21.16
Burkholderiales	23732	14.48
Rhizobiales	17203	10.5
Rhodocyclales	13481	8.23
Clostridiales	4921	3.0
Pseudomonadales	3986	2.43
Chitinophagales	3191	1.95
Legionellales	1777	1.08
Lactobacillales	1612	0.98
HT-3		
Taxa	Count	%
Enterobacterales	48522	29.61
Sphingomonadales	34677	21.16
Burkholderiales	23732	14.48
Rhizobiales	17203	10.5
Rhodocyclales	13481	8.23
Clostridiales	4921	3.0
Pseudomonadales	3986	2.43
Chitinophagales	3191	1.95
Legionellales	1777	1.08
Lactobacillales	1612	0.98
HT-4		
Taxa	Count	%
Burkholderiales	62779	21.54
Enterobacterales	57187	19.62
Sphingomonadales	46899	16.09
Rhizobiales	34189	11.73
Chitinophagales	20776	7.13
Rhodocyclales	15186	5.21
Myxococcales	14448	4.96
Pseudomonadales	6646	2.28
Rhodospirillales	4870	1.67
Legionellales	3969	1.36
HT-5		
Taxa	Count	%
Burkholderiales	8703	22.11
Sphingomonadales	7758	19.71
Enterobacterales	5706	14.49
Rhodocyclales	4329	11.0
Rhizobiales	4116	10.45
Chitinophagales	3057	7.76
Pseudomonadales	971	2.47
Clostridiales	551	1.4
Nitrosomonadales	505	1.28
Legionellales	384	0.98

HT-6		
Taxa	Count	%
Enterobacterales	43517	27.74
Burkholderiales	31693	20.2
Sphingomonadales	22602	14.41
Rhizobiales	12625	8.05
Bacillales	10058	6.41
Chitinophagales	8112	5.17
Rhodocyclales	7044	4.49
Pseudomonadales	3273	2.09
Rhodospirillales	2467	1.57
Clostridiales	2462	1.57
HT-7		
Taxa	Count	%
Bifidobacterales	24124	76.86
Enterobacterales	4294	13.68
Lactobacillales	2735	8.71
Bacillales	123	0.39
Pseudomonadales	31	0.1
Burkholderiales	21	0.07
Rhizobiales	9	0.03
Sphingobacterales	9	0.03
Rhodospirillales	5	0.02
Propionibacterales	4	0.01
HT-8		
Taxa	Count	%
Burkholderiales	19079	27.98
Sphingomonadales	9410	13.8
Pseudomonadales	8686	12.74
Enterobacterales	8021	11.77
Rhizobiales	6562	9.63
Rhodocyclales	2555	3.75
Chitinophagales	2023	2.97
Lactobacillales	1961	2.88
Legionellales	1958	2.87
Nitrosomonadales	1098	1.61
HT-10		
Taxa	Count	%
Sphingomonadales	5886	19.98
Enterobacterales	5768	19.58
Burkholderiales	5092	17.29
Rhizobiales	2337	7.93
Chitinophagales	1744	5.92
Rhodocyclales	1597	5.42
Clostridiales	903	3.07
Pseudomonadales	613	2.08
Lactobacillales	588	2.0
Propionibacterales	478	1.62

HT-11		
Taxa	Count	%
Sphingomonadales	49639	20.78
Burkholderiales	41315	17.3
Enterobacterales	31329	13.12
Rhizobiales	21193	8.87
Chitinophagales	19203	8.04
Clostridiales	18324	7.67
Rhodocyclales	10723	4.49
Pseudomonadales	5962	2.5
Rhodospirillales	4845	2.03
Propionibacteriales	4801	2.01

R2_GT9		
Taxa	Count	%
Sphingomonadales	29923	24.0
Enterobacterales	21827	17.5
Burkholderiales	20680	16.58
Rhizobiales	11171	8.96
Chitinophagales	7843	6.29
Rhodocyclales	7243	5.81
Pseudomonadales	3998	3.21
Clostridiales	3224	2.59
Lactobacillales	3120	2.5
Rhodospirillales	1941	1.56

R2_GT12		
Taxa	Count	%
Burkholderiales	5757	25.61
Enterobacterales	2619	11.65
Sphingomonadales	2563	11.4
Rhizobiales	1788	7.95
Chitinophagales	1629	7.25
Clostridiales	1350	6.01
Rhodocyclales	861	3.83
Lactobacillales	662	2.95
Pseudomonadales	653	2.91
Legionellales	615	2.74

Family

HT-1		
Taxa	Count	%
Sphingomonadaceae	19880	25.93
Comamonadaceae	9176	11.97
Bradyrhizobiaceae	7458	9.73
Burkholderiaceae	6340	8.27
Enterobacteriaceae	6182	8.06
Chitinophagaceae	5789	7.55
Rhodocyclaceae	4388	5.72
Azonexaceae	2766	3.61

HT-1		
Taxa	Count	%
Pseudomonadaceae	2312	3.02
Streptococcaceae	1131	1.48
HT-2		
Taxa	Count	%
Sphingomonadaceae	26725	21.66
Enterobacteriaceae	23241	18.84
Burkholderiaceae	13070	10.6
Rhodocyclaceae	9530	7.73
Bradyrhizobiaceae	7752	6.28
Comamonadaceae	6735	5.46
Pseudomonadaceae	3909	3.17
Chitinophagaceae	3191	2.59
Clostridiaceae	3116	2.53
Azonexaceae	2911	2.36
HT-3		
Taxa	Count	%
Sphingomonadaceae	26725	21.66
Enterobacteriaceae	23241	18.84
Burkholderiaceae	13070	10.6
Rhodocyclaceae	9530	7.73
Bradyrhizobiaceae	7752	6.28
Comamonadaceae	6735	5.46
Pseudomonadaceae	3909	3.17
Chitinophagaceae	3191	2.59
Clostridiaceae	3116	2.53
Azonexaceae	2911	2.36
HT-4		
Taxa	Count	%
Sphingomonadaceae	37994	16.71
Comamonadaceae	28750	12.64
Enterobacteriaceae	26767	11.77
Chitinophagaceae	20776	9.14
Burkholderiaceae	20636	9.07
Bradyrhizobiaceae	16162	7.11
Rhodocyclaceae	8694	3.82
Pseudomonadaceae	6389	2.81
Azonexaceae	5011	2.2
Polyangiaceae	4848	2.13
HT-5		
Taxa	Count	%
Sphingomonadaceae	6510	20.77
Comamonadaceae	3569	11.39
Burkholderiaceae	3169	10.11
Chitinophagaceae	3057	9.75
Rhodocyclaceae	2410	7.69
Enterobacteriaceae	2217	7.07

HT-5		
Taxa	Count	%
Bradyrhizobiaceae	2065	6.59
Azonexaceae	1440	4.59
Pseudomonadaceae	838	2.67
Phyllobacteriaceae	452	1.44

HT-6

Taxa	Count	%
Sphingomonadaceae	18207	15.74
Burkholderiaceae	16431	14.2
Enterobacteriaceae	15143	13.09
Comamonadaceae	8985	7.77
Paenibacillaceae	8685	7.51
Chitinophagaceae	8112	7.01
Phyllobacteriaceae	5130	4.43
Rhodocyclaceae	4428	3.83
Bradyrhizobiaceae	3383	2.92
Pseudomonadaceae	2786	2.41

HT-7

Taxa	Count	%
Bifidobacteriaceae	24124	77.19
Enterobacteriaceae	2758	8.82
Streptococcaceae	2734	8.75
Erwiniaceae	1304	4.17
Bacillaceae	86	0.28
Morganellaceae	45	0.14
Staphylococcaceae	37	0.12
Yersiniaceae	33	0.11
Moraxellaceae	22	0.07
Burkholderiaceae	21	0.07

HT-8

Taxa	Count	%
Burkholderiaceae	13652	23.88
Pseudomonadaceae	8351	14.61
Sphingomonadaceae	7836	13.71
Enterobacteriaceae	3325	5.82
Bradyrhizobiaceae	2745	4.8
Comamonadaceae	2685	4.7
Chitinophagaceae	2023	3.54
Legionellaceae	1919	3.36
Streptococcaceae	1522	2.66
Rhodocyclaceae	1438	2.52

HT-10

Taxa	Count	%
Sphingomonadaceae	5061	21.15
Burkholderiaceae	3161	13.21
Enterobacteriaceae	2862	11.96
Chitinophagaceae	1744	7.29

HT-10		
Taxa	Count	%
Rhodocyclaceae	945	3.95
Comamonadaceae	899	3.76
Bradyrhizobiaceae	611	2.55
Peptococcaceae	581	2.43
Pseudomonadaceae	524	2.19
Orbaceae	468	1.96

HT-11		
Taxa	Count	%
Sphingomonadaceae	40969	21.07
Burkholderiaceae	22594	11.62
Chitinophagaceae	19203	9.88
Peptococcaceae	14305	7.36
Enterobacteriaceae	13200	6.79
Comamonadaceae	9952	5.12
Rhodocyclaceae	6742	3.47
Pseudomonadaceae	5431	2.79
Bradyrhizobiaceae	5336	2.74
Rhodospirillaceae	3955	2.03

R2_GT9		
Taxa	Count	%
Sphingomonadaceae	24959	25.44
Burkholderiaceae	10741	10.95
Enterobacteriaceae	8996	9.17
Chitinophagaceae	7843	8.0
Comamonadaceae	5549	5.66
Rhodocyclaceae	4420	4.51
Pseudomonadaceae	3473	3.54
Bradyrhizobiaceae	2907	2.96
Peptococcaceae	2035	2.07
Azonexaceae	1709	1.74

R2_GT12		
Taxa	Count	%
Burkholderiaceae	2445	13.65
Sphingomonadaceae	2145	11.97
Chitinophagaceae	1629	9.09
Enterobacteriaceae	1402	7.82
Comamonadaceae	1100	6.14
Peptococcaceae	1073	5.99
Pseudomonadaceae	599	3.34
Legionellaceae	596	3.33
Bradyrhizobiaceae	584	3.26
Orbaceae	483	2.7

Genus

HT-1		
Taxa	Count	%

HT-1		
Taxa	Count	%
Sphingomonas	7971	15.61
Cupriavidus	5340	10.46
Novosphingobium	4191	8.21
Azospira	3747	7.34
Dechloromonas	2766	5.42
Variovorax	2329	4.56
Pseudomonas	2184	4.28
Comamonas	1303	2.55
Lactococcus	1004	1.97
Flavihumibacter	1003	1.96
HT-2		
Taxa	Count	%
Cupriavidus	10987	13.68
Azospira	8652	10.77
Sphingomonas	8474	10.55
Novosphingobium	4988	6.21
Pseudomonas	3721	4.63
Escherichia	3276	4.08
Dechloromonas	2911	3.62
Clostridium	2787	3.47
Variovorax	1858	2.31
Methylobacterium	1611	2.01
HT-3		
Taxa	Count	%
Cupriavidus	10987	13.68
Azospira	8652	10.77
Sphingomonas	8474	10.55
Novosphingobium	4988	6.21
Pseudomonas	3721	4.63
Escherichia	3276	4.08
Dechloromonas	2911	3.62
Clostridium	2787	3.47
Variovorax	1858	2.31
Methylobacterium	1611	2.01
HT-4		
Taxa	Count	%
Cupriavidus	18624	12.15
Sphingomonas	15721	10.26
Variovorax	9002	5.87
Novosphingobium	7657	5.0
Azospira	7621	4.97
Pseudomonas	6062	3.96
Dechloromonas	5011	3.27
Sorangium	4776	3.12
Sandaracinus	4688	3.06
Magnetospirillum	4179	2.73

HT-5		
Taxa	Count	%
Cupriavidus	2681	12.13
Sphingomonas	2419	10.95
Azospira	2053	9.29
Novosphingobium	1509	6.83
Dechloromonas	1440	6.52
Variovorax	894	4.05
Pseudomonas	798	3.61
Comamonas	667	3.02
Niastella	435	1.97
Escherichia	394	1.78

HT-6

Taxa	Count	%
Cupriavidus	13940	16.13
Paenibacillus	7971	9.23
Sphingomonas	6352	7.35
Mesorhizobium	4601	5.33
Novosphingobium	4438	5.14
Azospira	3966	4.59
Variovorax	2744	3.18
Pseudomonas	2647	3.06
Escherichia	2559	2.96
Magnetospirillum	1895	2.19

HT-7

Taxa	Count	%
Bifidobacterium	24124	78.35
Streptococcus	2734	8.88
Escherichia	1841	5.98
Pantoea	1300	4.22
Klebsiella	251	0.82
Bacillus	86	0.28
Salmonella	58	0.19
Enterobacter	43	0.14
Staphylococcus	37	0.12
Shigella	33	0.11

HT-8

Taxa	Count	%
Cupriavidus	12248	27.49
Pseudomonas	8015	17.99
Sphingomonas	2987	6.7
Novosphingobium	1415	3.18
Lactococcus	1286	2.89
Azospira	1263	2.83
Legionella	1067	2.39
Escherichia	941	2.11
Ralstonia	927	2.08
Dechloromonas	734	1.65

HT-10		
Taxa	Count	%
Cupriavidus	2625	14.62
Sphingomonas	2203	12.27
Novosphingobium	896	4.99
Azospira	808	4.5
Escherichia	787	4.38
Desulfosporosinus	496	2.76
Pseudomonas	475	2.65
Salmonella	414	2.31
Dechloromonas	396	2.21
Gilliamella	382	2.13

HT-11

Taxa	Count	%
Cupriavidus	19136	13.27
Sphingomonas	13820	9.59
Novosphingobium	10737	7.45
Desulfosporosinus	10487	7.27
Azospira	5784	4.01
Pseudomonas	5005	3.47
Desulfotomaculum	3225	2.24
Escherichia	3106	2.15
Niastella	2502	1.74
Dechloromonas	2269	1.57

R2_GT9

Taxa	Count	%
Cupriavidus	9020	12.6
Sphingomonas	8034	11.22
Novosphingobium	6690	9.35
Azospira	3627	5.07
Pseudomonas	3298	4.61
Desulfosporosinus	1856	2.59
Escherichia	1816	2.54
Dechloromonas	1709	2.39
Variovorax	1455	2.03
Klebsiella	1248	1.74

R2_GT12

Taxa	Count	%
Cupriavidus	1159	8.4
Desulfosporosinus	942	6.83
Sphingomonas	845	6.13
Mycoavidus	659	4.78
Pseudomonas	563	4.08
Escherichia	492	3.57
Novosphingobium	426	3.09
Gilliamella	399	2.89
Azospira	351	2.54
Mycetohabitans	326	2.36

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- Langmead B. and Salzberg SL. (2012). Fast gapped-read alignment with Bowtie 2. *Nature methods*, 9(4), 357-9.