

Supplemental results

Bacterial culturing

Total counts were similar on plates incubated aerobically and anaerobically and but there was considerable variation between samples with counts ranging from lows of 10^7 bacteria/ml sample to highs of 10^{11} .

The selective culturing method indicated that more than 60% of the samples tested contained coliform bacteria, with a further 20% containing detectable *Staphylococcus*. The likelihood of culturing potentially pathogenic bacteria was the same in samples from trained and untrained producers.

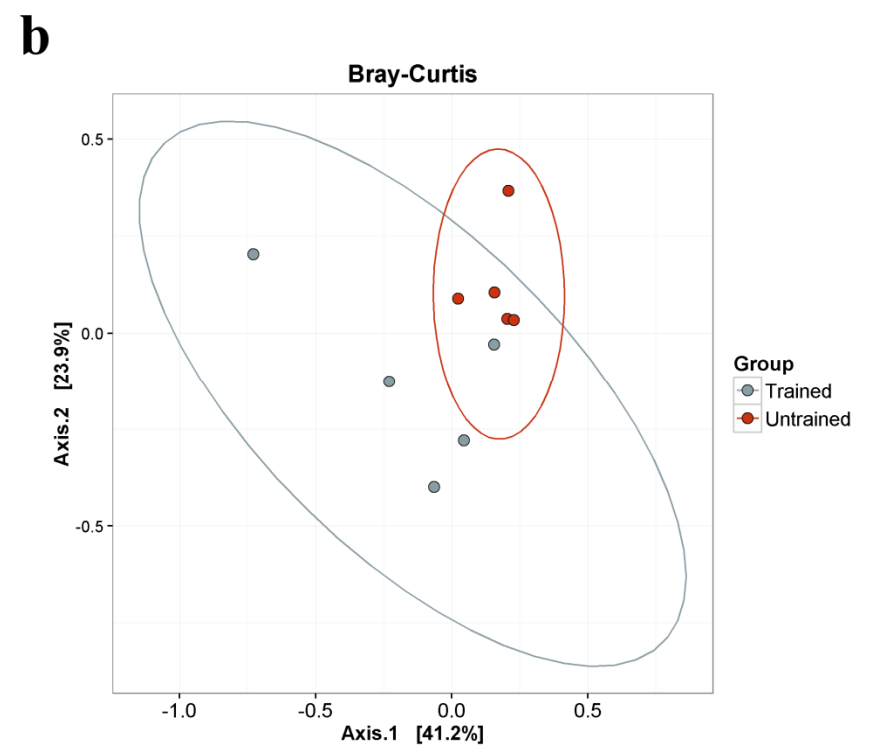
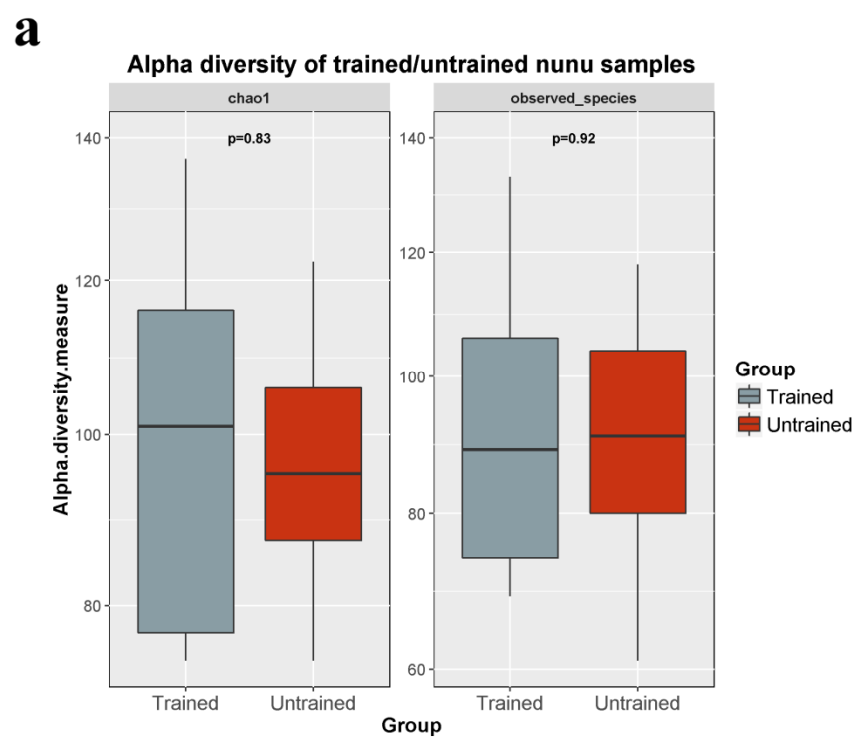


Figure S1. (A) Box plots showing the alpha diversity of nunu samples. (B) PCoA plot showing the beta diversity of nunu samples.

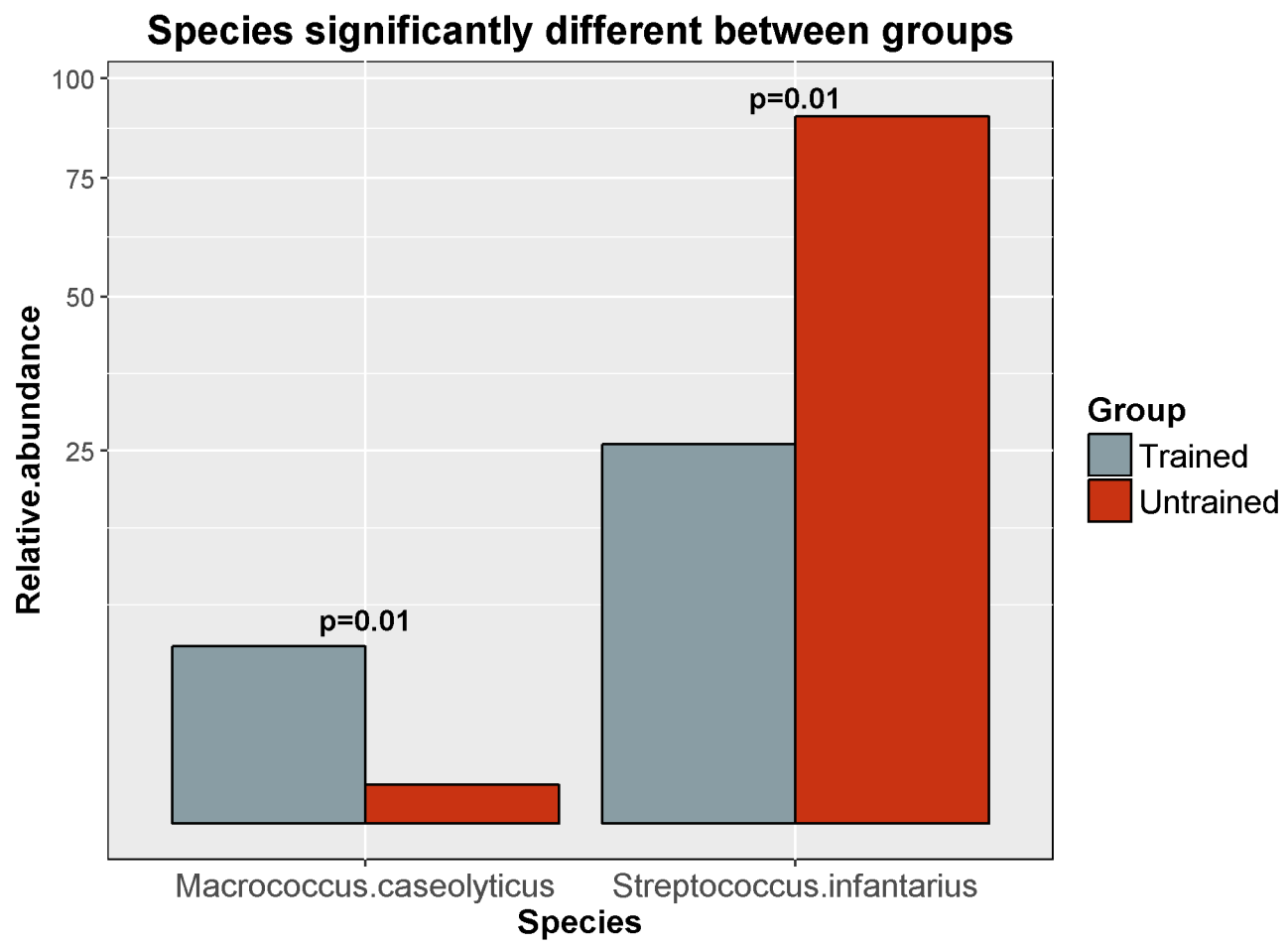


Figure S2. Bar plot showing species that were differentially abundant between nunu samples from trained producers and nunu samples from untrained producers.

Dissimilarity matrix based on HUMAnN2 output

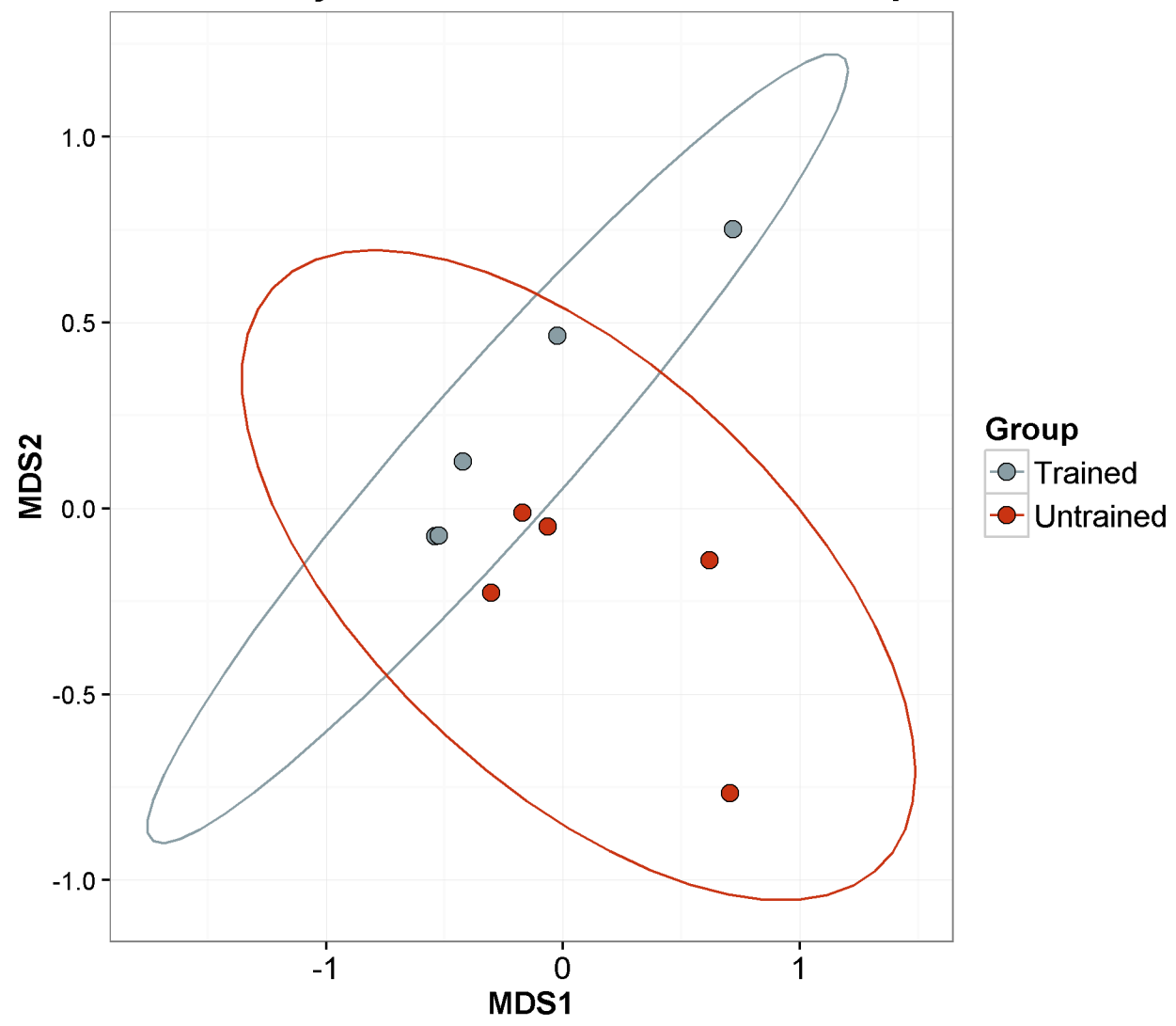


Figure S3. MDS plot showing the functional similarities between nunu samples from trained producers and nunu samples from untrained producers.

Abundances of antibiotic resistance/HGT-related functions

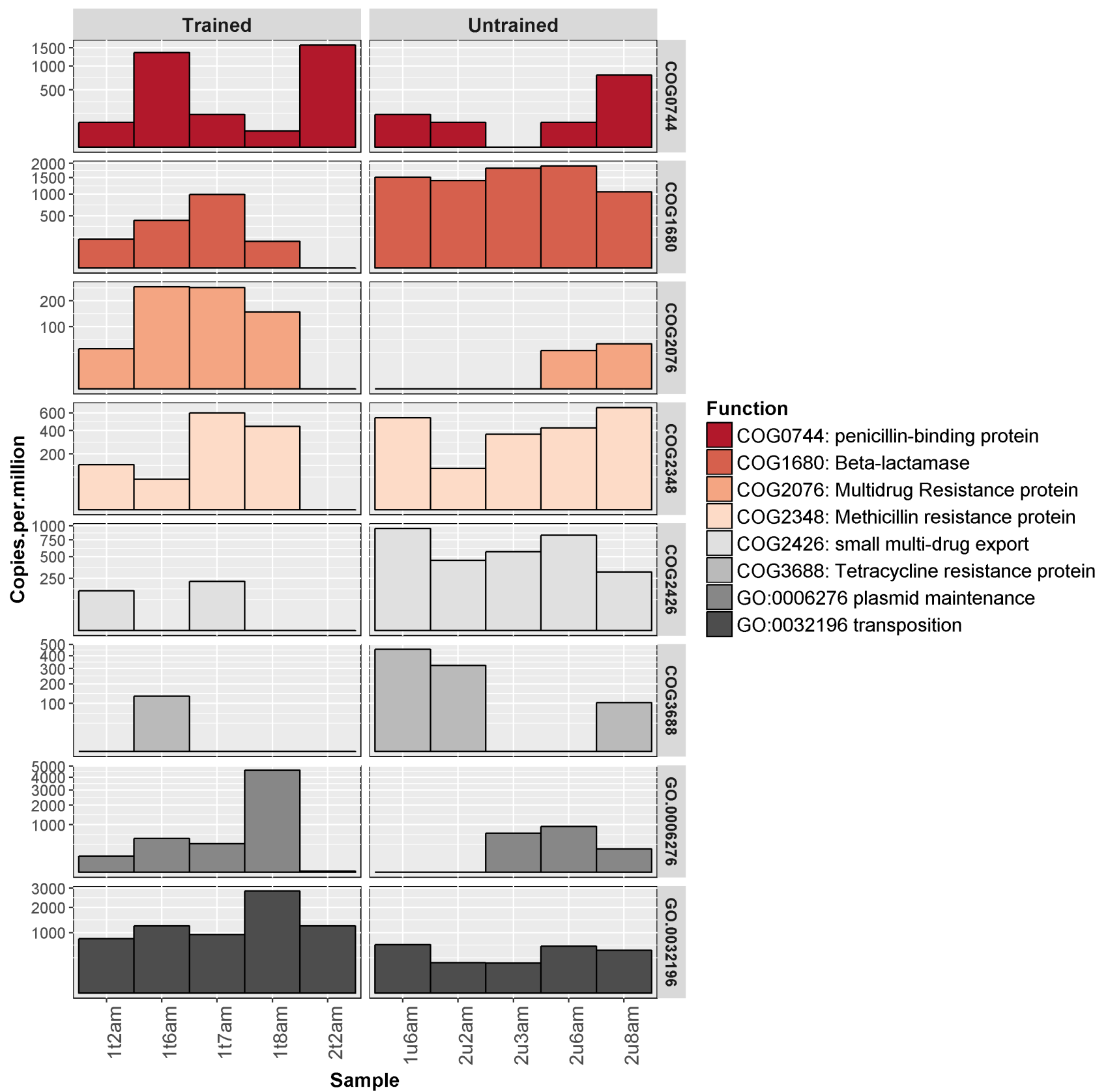
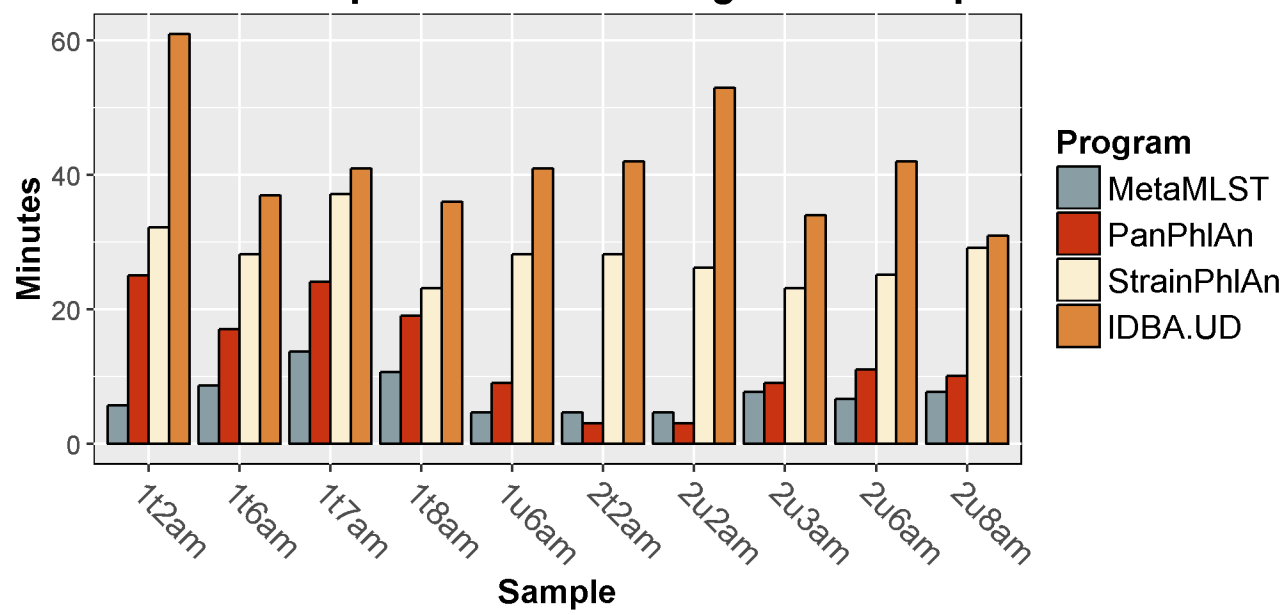


Figure S4. Bar plot showing the abundances of antibiotic resistance-associated functions and horizontal gene transfer (HGT)-associated functions in the nuNu metagenome.

A Time taken to process nunu metagenome samples



B Mean time taken to process nunu metagenome samples

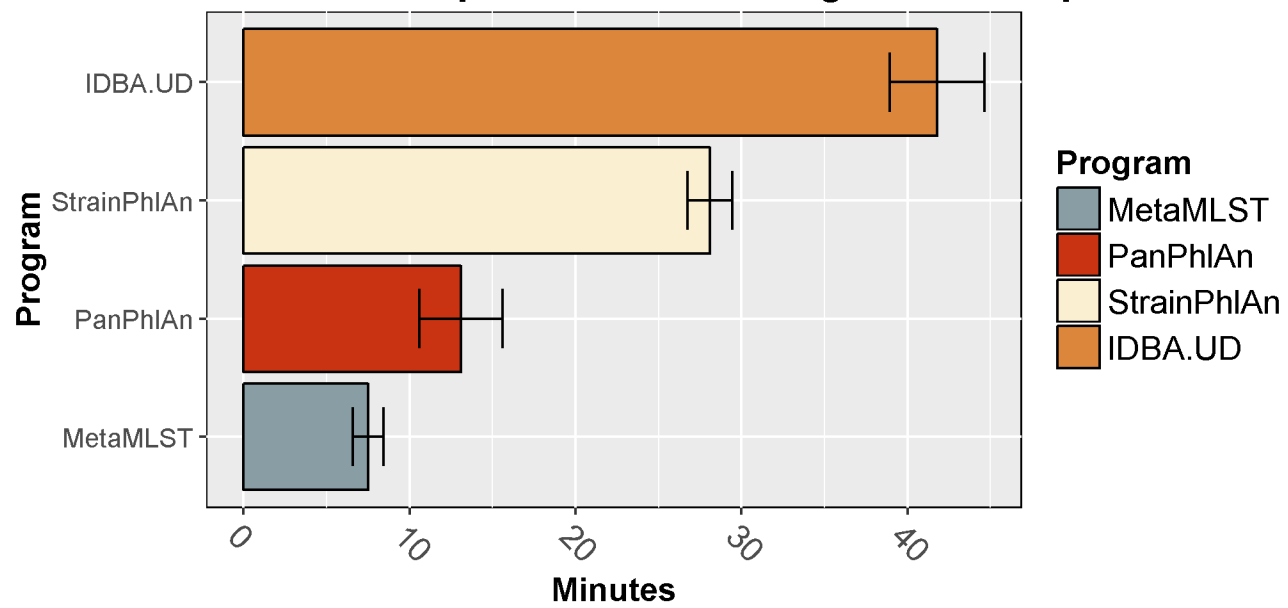


Figure S5. Bar plot showing (a) the total time taken to process nunu metagenomic samples, and (b) the mean time taken to process each nunu metagenomic sample, using IDBA-UD, MetaMLST, PanPhlAn and StrainPhlAn.

Table S1. MetaCyc pathways significantly different between groups.

MetaCyc pathway	p-value (BH adjusted)
ALLANTOINDEG.PWY.superpathway.of.allantoin.degradation.in.yeast	0.019
ARGSYNBSUB.PWY.L.arginine.biosynthesis.II.acetyl.cycle.	0.047
ARO.PWY.chorismate.biosynthesis.I	0.047
ASPASN.PWY.superpathway.of.L.aspartate.and.L.asparagine.biosynthesis	0.047
BRANCHED.CHAIN.AA.SYN.PWY.superpathway.of.branched.amino.acid.biosynthesis	0.016
CALVIN.PWY.Calvin.Benson.Bassham.cycle	0.009
COA.PWY.1.coenzyme.A.biosynthesis.II.mammalian.	0.047
COA.PWY.coenzyme.A.biosynthesis.I	0.016
COMPLETE.ARO.PWY.superpathway.of.aromatic.amino.acid.biosynthesis	0.047
DTDPRHAMSYN.PWY.dTDP.L.rhamnose.biosynthesis.I	0.047
GALACTUROCAT.PWY.D.galacturonate.degradation.I	0.009
GLUTORN.PWY.L.ornithine.biosynthesis	0.047
GLYCOGENSYNTH.PWY.glycogen.biosynthesis.I.from.ADP.D.Glucose.	0.016
HEME.BIOSYNTHESIS.II.heme.biosynthesis.I.aerobic.	0.028
HISDEG.PWY.L.histidine.degradation.I	0.047
HISTSYN.PWY.L.histidine.biosynthesis	0.028
HOMOSER.METSYN.PWY.L.methionine.biosynthesis.I	0.028
ILEUSYN.PWY.L.isoleucine.biosynthesis.I.from.threonine.	0.009
KDO.NAGLIPASYN.PWY.superpathway.of.Kdo.2.lipid.A.biosynthesis	0.034
LACTOSECAT.PWY.lactose.and.galactose.degradation.I	0.009
NONOXIPENT.PWY.pentose.phosphate.pathway.non.oxidative.branch.	0.047
P122.PWY.heterolactic.fermentation	0.028
P161.PWY.acetylene.degradation	0.009
PENTOSE.P.PWY.pentose.phosphate.pathway	0.009
PEPTIDOGLYCANSYN.PWY.peptidoglycan.biosynthesis.I.meso.diaminopimelate.containing.	0.047
POLYISOPRENSYN.PWY.polyisoprenoid.biosynthesis.E.coli.	0.047
PWY.2942.L.lysine.biosynthesis.III	0.016
PWY.3001.superpathway.of.L.isoleucine.biosynthesis.I	0.028
PWY.4242.pantothenate.and.coenzyme.A.biosynthesis.III	0.047
PWY.5097.L.lysine.biosynthesis.VI	0.016
PWY.5100.pyruvate.fermentation.to.acetate.and.lactate.II	0.028
PWY.5103.L.isoleucine.biosynthesis.III	0.016
PWY.5104.L.isoleucine.biosynthesis.IV	0.034
PWY.5173.superpathway.of.acetyl.CoA.biosynthesis	0.009
PWY.5265.peptidoglycan.biosynthesis.II.staphylococci.	0.016
PWY.5384.sucrose.degradation.IV.sucrose.phosphorylase.	0.016
PWY.5686.UMP.biosynthesis	0.028
PWY.5747.2.methylcitrate.cycle.II	0.016
PWY.5850.superpathway.of.menaquinol.6.biosynthesis.I	0.047
PWY.5860.superpathway.of.demethylmenaquinol.6.biosynthesis.I	0.047
PWY.5913.TCA.cycle.VI.obligate.autotrophs.	0.009
PWY.5973.cis.vaccenate.biosynthesis	0.009
PWY.6124.inosine.5.phosphate.biosynthesis.II	0.009
PWY.6125.superpathway.of.guanosine.nucleotides.de.novo.biosynthesis.II	0.016
PWY.6147.6.hydroxymethyl.dihydropterin.diphosphate.biosynthesis.I	0.047
PWY.6163.chorismate.biosynthesis.from.3.dehydroquinone	0.028
PWY.6168.flavin.biosynthesis.III.fungi.	0.047
PWY.621.sucrose.degradation.III.sucrose.invertase.	0.009
PWY.6282.palmitoleate.biosynthesis.I.from.5Z.dodec.5.enoate.	0.047
PWY.6385.peptidoglycan.biosynthesis.III.mycobacteria.	0.028
PWY.6386.UDP.N.acetylmuramoyl.pentapeptide.biosynthesis.II.lysine.containing.	0.028
PWY.6387.UDP.N.acetylmuramoyl.pentapeptide.biosynthesis.I.meso.diaminopimelate.containing.	0.028
PWY.6507.4.deoxy.L.threo.hex.4.enopyranuronate.degradation	0.009
PWY.6527.stachyose.degradation	0.009
PWY.6901.superpathway.of.glucose.and.xylose.degradation	0.028
PWY.6936.seleno.amino.acid.biosynthesis	0.009
PWY.7111.pyruvate.fermentation.to.isobutanol.engineered.	0.009
PWY.7115.C4.photosynthetic.carbon.assimilation.cycle.NAD.ME.type	0.047
PWY.7184.pyrimidine.deoxyribonucleotides.de.novo.biosynthesis.I	0.028
PWY.7187.pyrimidine.deoxyribonucleotides.de.novo.biosynthesis.II	0.047
PWY.7197.pyrimidine.deoxyribonucleotide.phosphorylation	0.047
PWY.7199.pyrimidine.deoxyribonucleosides.salvage	0.009
PWY.7200.superpathway.of.pyrimidine.deoxyribonucleoside.salvage	0.015
PWY.7208.superpathway.of.pyrimidine.nucleobases.salvage	0.047
PWY.7228.superpathway.of.guanosine.nucleotides.de.novo.biosynthesis.I	0.016
PWY.7242.D.fructuronate.degradation	0.047
PWY.7357.thiamin.formation.from.pyrithiamine.and.oxythiamine.yeast.	0.009
PWY.7539.6.hydroxymethyl.dihydropterin.diphosphate.biosynthesis.III.Chlamydia.	0.047

PWY.7663.gondoate.biosynthesis.anaerobic.	0.009
PWY0.1061.superpathway.of.L.alanine.biosynthesis	0.047
PWY0.1296.purine.ribonucleosides.degradation	0.009
PWY0.1297.superpathway.of.purine.deoxyribonucleosides.degradation	0.009
PWY0.1298.superpathway.of.pyrimidine.deoxyribonucleosides.degradation	0.047
PWY0.1319.CDP.diacylglycerol.biosynthesis.II	0.009
PWY0.42.2.methylcitrate.cycle.I	0.016
PWY66.409.superpathway.of.purine.nucleotide.salvage	0.009
PWY66.422.D.galactose.degradation.V.Leloir.pathway.	0.047
RHAMCAT.PWY.L.rhamnose.degradation.I	0.047
SER.GLYSYN.PWY.superpathway.of.L.serine.and.glycine.biosynthesis.I	0.047
THRESYN.PWY.superpathway.of.L.threonine.biosynthesis	0.028
TRPSYN.PWY.L.tryptophan.biosynthesis	0.009
VALSYN.PWY.L.valine.biosynthesis	0.009
X1CMET2.PWY.N10.formyl.tetrahydrofolate.biosynthesis	0.016

Table S2. The results of PanPhlAn analysis of 17 spinach samples spiked with different STEC.

Sequence accession number	Strain	<i>E. coli</i> abundance (%)	stx1A	stx1B	stx2A	stx2B
SRR4101289	<i>E. coli</i> O157:H7 str. Sakai	89.73	1	1	1	1
SRR4101293	<i>E. coli</i> O157:H7 str. TW14359	79.9	1	1	1	1
SRR4101297	<i>E. coli</i> O157:H7 str. TW14359	42.74	0	0	1	1
SRR4101299	<i>E. coli</i> O113:H21 str. CL-3	45.7	0	0	1	1
SRR4101303	<i>E. coli</i> O113:H21 str. CL-3	68.17	1	1	0	0
SRR4101307	<i>E. coli</i> serovar O145:H28	92.98	0	0	1	1
SRR4101308	<i>E. coli</i> serovar O121:H19	92.14	0	0	1	1
SRR4101310	<i>E. coli</i> EC1738	60.59	0	0	1	1
SRR4101311	<i>E. coli</i> EC1738	87.5	0	0	1	1
SRR4101312	<i>E. coli</i> O104:H4 str. 2011C-3493	80.43	0	0	1	1
SRR4101314	<i>E. coli</i> O104:H4 str. 2011C-3493	66.08	0	0	1	1
SRR4101315	<i>E. coli</i> serovar O104:H7	89.98	0	0	1	1
SRR4101317	<i>E. coli</i> serovar O145:H28	20.56	0	0	1	1
SRR4101318	<i>E. coli</i> STEC_B2F1	38.67	0	0	1	1
SRR4101319	<i>E. coli</i> STEC_B2F1	56.95	0	0	1	1
SRR4101321	<i>E. coli</i> O113:H21 str. CL-3	92.83	0	0	1	1
SRR4101323	<i>E. coli</i> O113:H21 str. CL-3	76.2	0	0	1	1

Table S3. *Escherichia coli* reference genomes used in this study.

<i>Escherichia coli</i> strain	RefSeq assembly accession
RM13514	GCF_000520035
ST540	GCF_000599625
ST2747	GCF_000599685
RM12761	GCF_000662395
RM12581	GCF_000671295
BIDMC 59	GCF_000692395
BIDMC 74	GCF_000692575
CHS 77	GCF_000692735
SE11	GCF_000010385
SE15	GCF_000010485
UTI89	GCF_000013265
536	GCF_000013305
APEC O1	GCF_000014845
E24377A	GCF_000017745
ATCC 8739	GCF_000019385
SMS-3-5	GCF_000019645
DH1	GCF_000023365
BL21-Gold(DE3)pLysS AG	GCF_000023665
IAI1	GCF_000026265
S88	GCF_000026285
UMN026	GCF_000026325
042	GCF_000027125
KO11	GCF_000147855
ABU 83972	GCF_000148365
UM146	GCF_000148605
MS 45-1	GCF_000164295
TA280	GCF_000176655
MS 145-7	GCF_000179115
W	GCF_000184185
LT-68	GCF_000188815
E1167	GCF_000190795
1.2741	GCF_000194175
3003	GCF_000194665
TW07793	GCF_000194685
UMNK88	GCF_000212715
96.0497	GCF_000215185
9.0111	GCF_000215265
UMNF18	GCF_000220005
STEC_DG131-3 clone D i14	GCF_000225125
B093	GCF_000233895
DEC2D	GCF_000242015
P12b	GCF_000249215
KO11FL	GCF_000257275
W	GCF_000258025
P4	GCF_000258145
APEC O78	GCF_000259425
KTE193	GCF_000332755
KTE233	GCF_000351025
KTE233	GCF_000351325
KTE56	GCF_000351525
KTE66	GCF_000351625
KTE67	GCF_000351645
KTE17	GCF_00035185
KTE42	GCF_000352125
KTE29	GCF_000352185
KTE79	GCF_000352245
KTE84	GCF_000352445
KTE84	GCF_000352465
KTE115	GCF_000352525
KTE135	GCF_000352585
KTE141	GCF_000352645
KTE144	GCF_000352665
KTE146	GCF_000352685
KTE147	GCF_000352705
KTE154	GCF_000352725
KTE192	GCF_000352785
KTE184	GCF_000352885
KTE183	GCF_000352905
KTE196	GCF_000352925
KTE197	GCF_000352945

KTE218	GCF_000353105
2720900	GCF_000355175
KTE114	GCF_000407765
KTE19	GCF_000407825
KTE31	GCF_000407925
KTE98	GCF_000408545
KTE102	GCF_000408585
HVH 55 (4-2646161)	GCF_000456825
HVH 58 (4-2839709)	GCF_000456865
HVH 65 (4-2262045)	GCF_000456945
HVH 111 (4-7039018)	GCF_000457555
HVH 115 (4-4465989)	GCF_000457655
HVH 139 (4-3192644)	GCF_000458035
HVH 164 (4-5953081)	GCF_000458495
HVH 188 (4-2356988)	GCF_000458825
HVH 195 (3-7155360)	GCF_000458955
KOEGE 44 (106a)	GCF_000459715
UMEA 3052-1	GCF_000460035
UMEA 3087-1	GCF_000460095
UMEA 3124-1	GCF_000460255
UMEA 3144-1	GCF_000460315
UMEA 3150-1	GCF_000460335
UMEA 3152-1	GCF_000460375
UMEA 3200-1	GCF_000460735
UMEA 3212-1	GCF_000460835
UMEA 3271-1	GCF_000461115
UMEA 3718-1	GCF_000461675
UMEA 4076-1	GCF_000461855
BIDMC 19C	GCF_000474825
JJ1886	GCF_000493755
HVH 36 (4-5675286)	GCF_000494935
K-12 substr. MG1655	GCF_000005845
12009	GCF_000010745
2009EL-2050	GCF_000299255
2009EL-2071	GCF_000299475
2011C-3493	GCF_000299455
11128	GCF_000010765
E2348/69	GCF_000026545
E24377A	GCF_000017745
EC4115	GCF_000021125
EDL933	GCF_000732965
Sakai	GCF_000008865
TW14359	GCF_000022225
Xuzhou21	GCF_000262125
11368	GCF_000091005
CB9615	GCF_000025165
RM12579	GCF_000245515
CE10	GCF_000227625
NRG 857C	GCF_000183345
55989	GCF_000026245
ETEC H10407	GCF_000210475

Table S4. *Klebsiella pneumoniae* reference genomes used in this study.

<i>Klebsiella pneumoniae</i> strain	RefSeq assembly accession
HS11286	GCF_000240185
NTUH-K2044	GCF_000009885
KCTC 2242	GCF_000220485
Kp13	GCF_000512165
KPNIH31	GCF_000785005
234-12	GCF_000981845
DHQP1002001	GCF_001704235
Kp_Goe_154414	GCF_001902335
ATCC 13884	GCF_000163455
LCT-KP214	GCF_000255975
WGLW1	GCF_000300655
WGLW2	GCF_000300675
KpQ3	GCF_000300835
WGLW5	GCF_000300955
909957	GCF_000485755
BIDMC 40	GCF_000492215
BIDMC 36	GCF_000492295
BIDMC 25	GCF_000492315
BIDMC 24	GCF_000492335
BIDMC 23	GCF_000492355
UCICRE 14	GCF_000492415
UCICRE 7	GCF_000492535
BWH 30	GCF_000492695
BWH 28	GCF_000492735
MGH 44	GCF_000492795
MGH 43	GCF_000567685
XDR	GCF_000785625
KP-7	GCF_000406385
ATCC 25955	GCF_000409715
CCBH13327	GCF_000805735
-	GCF_000821685
ATCC 11296	GCF_000826585
50531633	GCF_001462885
YMC2010/8/B2027	GCA_001901745
12-3578	GCF_000367165
1183_KPNE	GCF_001060495
570_KPNE	GCF_001063755
k414	GCF_900085035
k2254	GCF_900085435
W2-15-ERG3	GCF_900093395