Table S1. Average Nucleotide Identity (ANI) between all pairs considering either all shared genes between pairs (Shared pairs) or genes shared by all strains (Shared all) or *E. coli* core genes (Core).

ANI	MG/IAI1	MG/TW1	MG/TW2	IAI1/TW1	IAI1/TW2	TW1/TW2
Shared pairs	0.985	0.927	0.927	0.923	0.914	0.913
Shared all	0.986	0.928	0.928	0.924	0.914	0.913
Core	0.988	0.934	0.933	0.929	0.92	0.919

MG: MG1655, TW1: TW11588, TW2: TW09308

Table S2. Basic growth parameters of individual strains (replicates are shown).

strain	μmax		yield/OD	
MG1655	0.72	0.72	1.1	1.09
IAI1	0.83	0.81	1.39	1.31
TW11588	0.88	0.89	1.61	1.53
TW09308	0.82	0.81	1.61	1.58

T able S3. Percentage of differentially expressed genes (DE) between individual states.

	MG1655			**IAI1**			**TW11588**			**TW09308**		
	B/C	**B/S**	**C/S**	**B/C**	**B/S**	**C/S**	**B/C**	**B/S**	**C/S**	**B/C**	**B/S**	**C/S**
DE (p<0.01)	38	53	46	48	71	64	57	77	70	38	71	61
DE (p<0.01;>= 2 fold)	12	50	44	24	57	45	29	61	49	21	61	52

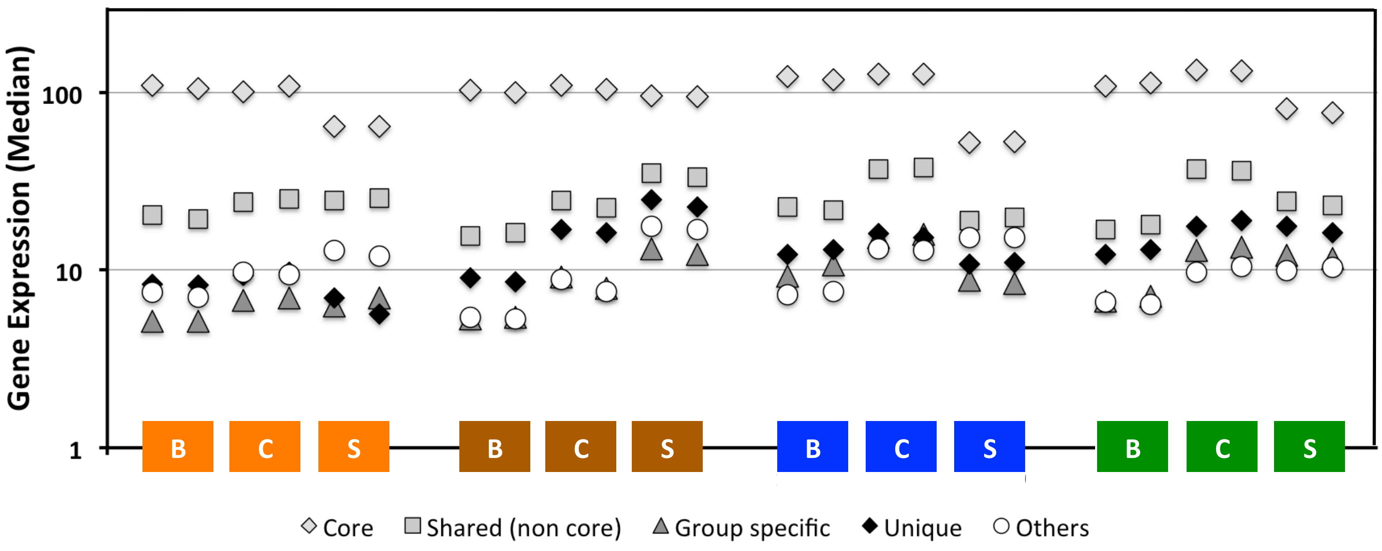


Figure S1: Median gene expression levels of individual gene sets (core: *E. coli* core genes, shared (non-core): all shard genes of isolates not included in *E. coli* core genes, group specific: genes enriched in each group, unique: genes unique to strains and others: all other genes). Strains are represented by color (orange: MG1655, brown: IAI1, blue: TW11588 and green: TW09308), whereas letters refer to individual states (B; batch, C: chemostat, S: starvation)

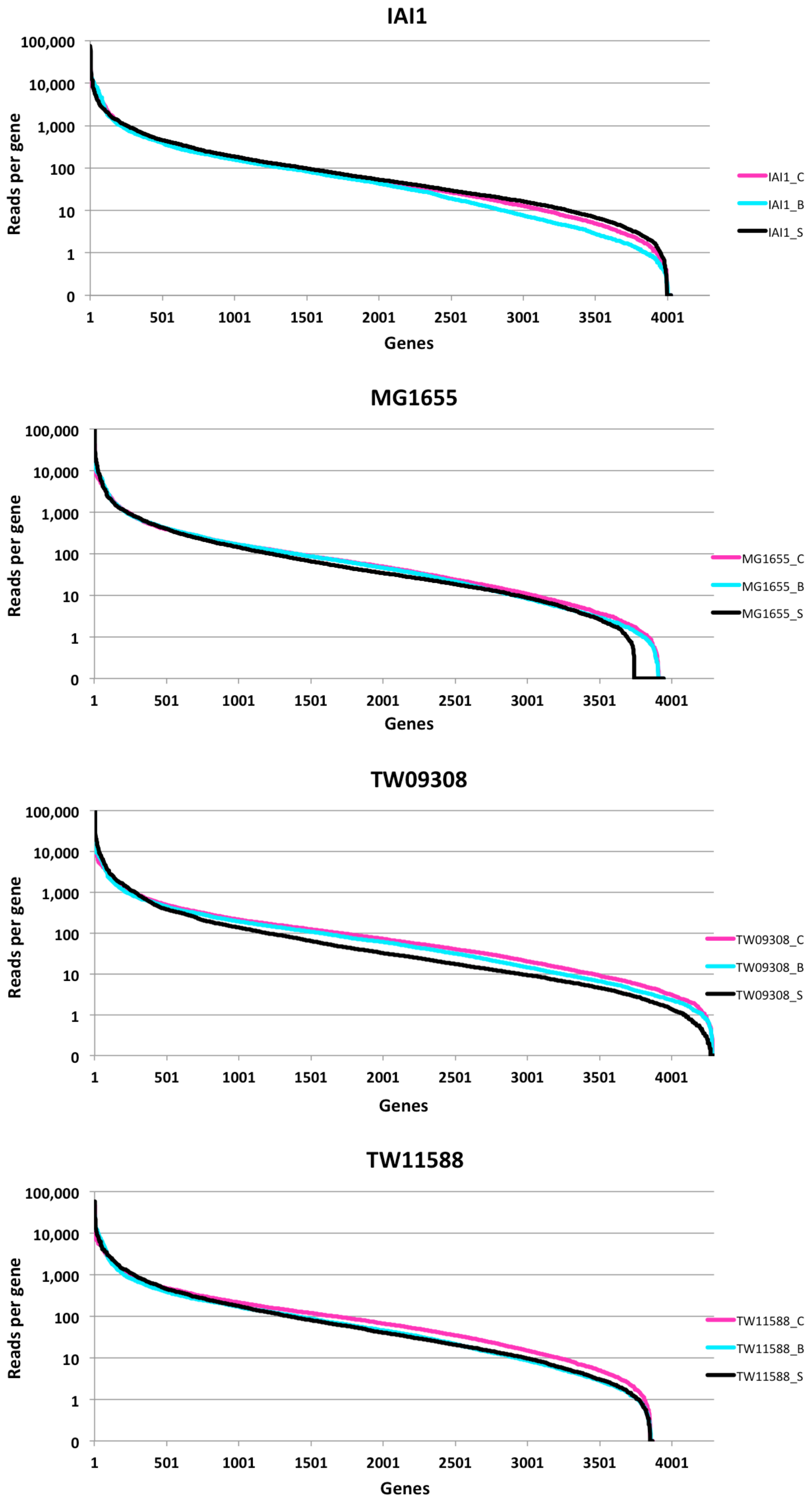


Figure S2: Overall expression level shapes of individual strains between states. B; batch, C: chemostat, S: starvation. The x-axis corresponds to all genes for individual genomes.

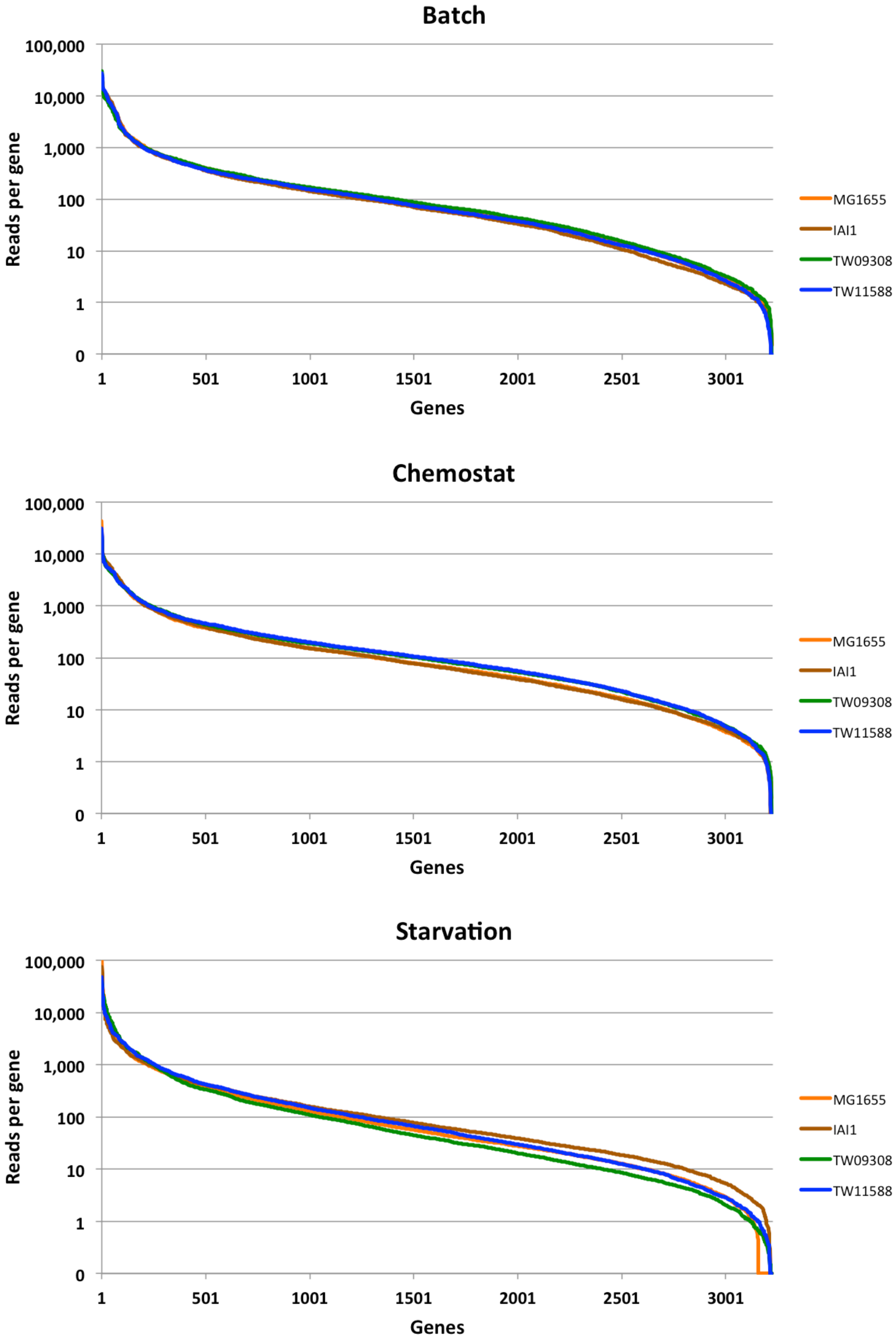
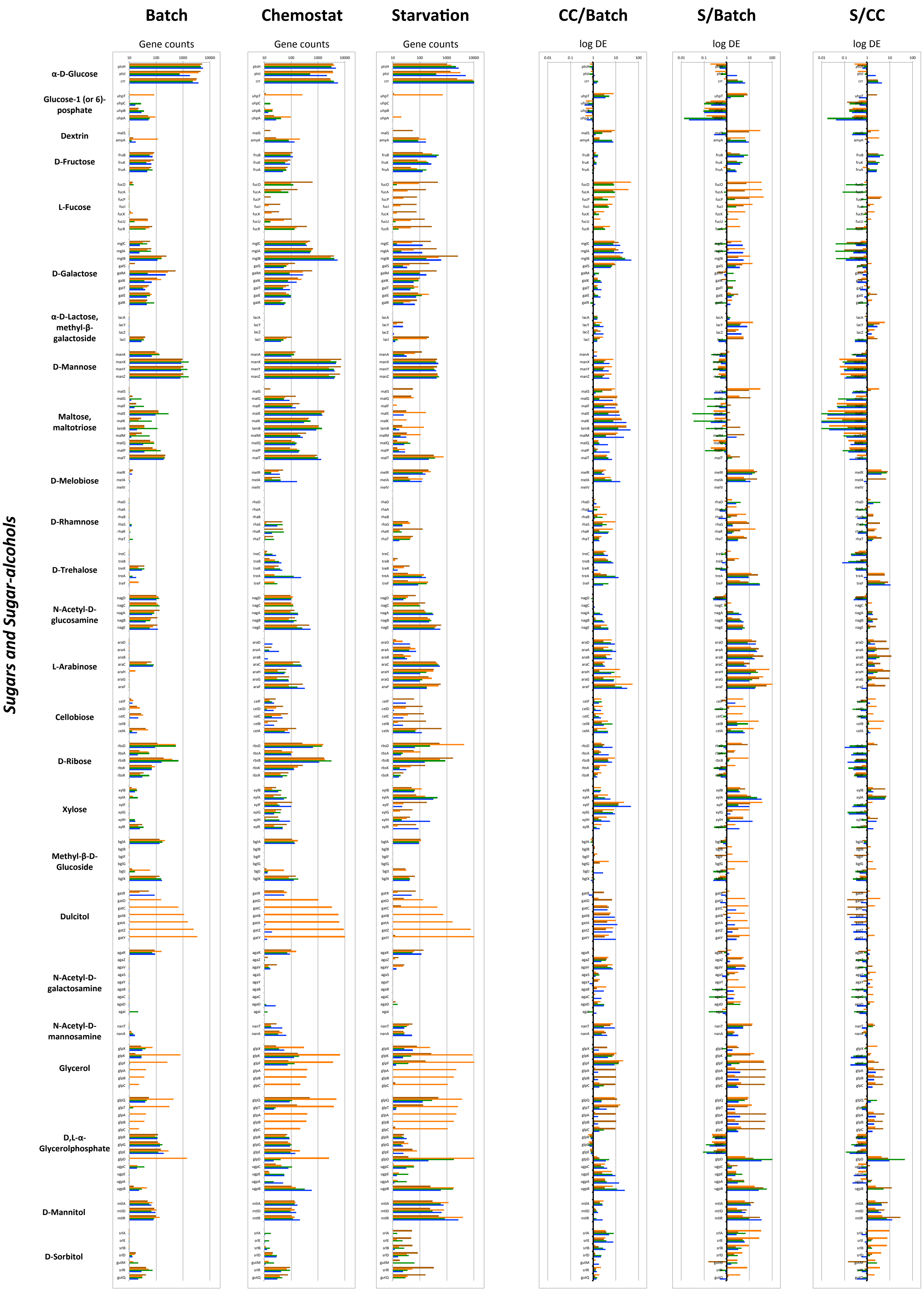


Figure S3: Overall expression level shapes of individual strains within states. Only genes shared by all four strains (x-axis) were considered for analysis. Strains are represented by color (orange: MG1655, brown: IAI1, blue: TW11588 and green: TW09308).


Batch
Chemostat
Starvation
CC/Batch
S/Batch
S/CC
Gene counts
Gene counts
Gene counts
log DE
log DE
log DE
10
100
1000
10000
0.01
0.1
1
10
100
Sugars and Sugar-alcohols
α-D-Glucose
ptsH
ptsI
crr
Glucose-1 (or 6)-posphate
uhpT
uhpC
uhpB
uhpA
Dextrin
malS
amyA
D-Fructose
fruB
fruK
fruA
L-Fucose
fucO
fucA
fucP
fucI
fucK
fucU
fucR
D-Galactose
mglC
mglA
mglB
galS
galM
galK
galT
galE
galR
α-D-Lactose, methyl-β-galactoside
lacA
lacY
lacZ
lacI
D-Mannose
manA
manX
manY
manZ
Maltose, maltotriose
malS
malG
malF
malE
malK
lamB
malM
malQ
malP
malT
D-Melobiose
melR
melA
melV
D-Rhamnose
rhaD
rhaA
rhaB
rhaS
rhaR
rhaT
D-Trehalose
treC
treB
treR
treA
treF
N-Acetyl-D-glucosamine
nagD
nagC
nagA
nagB
nagE
L-Arabinose
araD
araA
araB
araC
araH
araG
araF
Cellobiose
celF
celD
celC
celB
celA
D-Ribose
rbsD
rbsA
rbsB
rbsK
rbsR
Xylose
xylB
xylA
xylF
xylG
xylH
xylR
Methyl-β-D-Glucoside
bglA
bglB
bglF
bglG
bglJ
bglX
Dulcitol
gatR
gatD
gatC
gatB
gatA
gatZ
gatY
N-Acetyl-D-galactosamine
agaR
agaZ
agaV
agaS
agaY
agaB
agaC
agaD
agaI
N-Acetyl-D-mannosamine
nanT
nanA
Glycerol
glpX
glpK
glpF
glpA
glpB
glpC
D,L-α-Glycerolphosphate
glpQ
glpT
glpA
glpB
glpC
glpR
glpG
glpE
glpD
ugpC
ugpE
ugpA
ugpB
D-Mannitol
mtlA
mtlD
mtlR
D-Sorbitol
srlA
srlE
srlB
srlD
gutM
srlR
gutQ

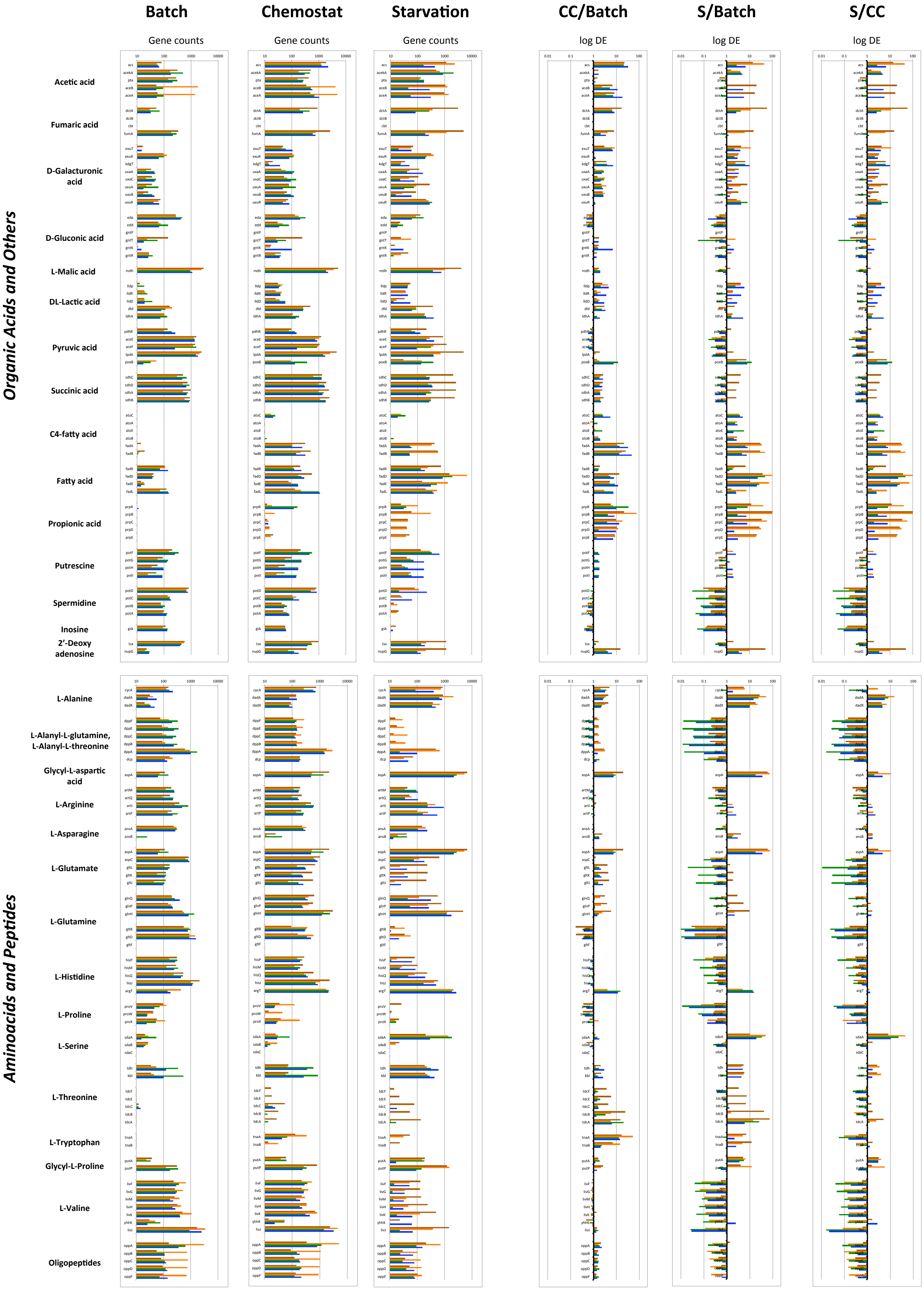

Batch
Chemostat
Starvation
CC/Batch
S/Batch
S/CC
Gene counts
Gene counts
Gene counts
log DE
log DE
log DE
10
100
1000
10000
0.01
0.1
1
10
100
Organic Acids and Others
Acetic acid
acs
aceA
pta
aceB
aceA
Fumaric acid
dctA
dctB
cbt
fumA
D-Galacturonic acid
exuT
exuR
kdgT
uxaA
uxaC
uxuA
uxuB
uxuR
D-Gluconic acid
eda
edd
gntP
gntT
gntK
gntR
L-Malic acid
mdh
DL-Lactic acid
lldp
lldR
lldD
dld
ldhA
Pyruvic acid
pdhR
aceE
aceF
lpdA
poxB
Succinic acid
sdhC
sdhD
sdhA
sdhB
C4-fatty acid
atoC
atoA
atoE
atoB
fadA
fadB
Fatty acid
fadR
fadD
fadE
fadL
Propionic acid
prpR
prpB
prpC
prpD
prpE
Putrescine
potF
potG
potH
potI
Spermidine
potD
potC
potB
potA
Inosine
gsk
2'-Deoxy adenosine
tsx
nupG
Aminoacids and Peptides
L-Alanine
cycA
dadA
dadX
L-Alanyl-L-glutamine, L-Alanyl-L-threonine
dppF
dppD
dppC
dppB
dppA
dcp
Glycyl-L-aspartic acid
aspA
L-Arginine
artM
artQ
artI
artP
L-Asparagine
ansA
ansB
L-Glutamate
aspA
aspC
gltL
gltK
gltJ
L-Glutamine
glnQ
glnP
glnH
gltB
gltD
gltF
L-Histidine
hisP
hisM
hisQ
hisJ
argT
L-Proline
proV
proW
proX
L-Serine
sdaA
sdaB
sdaC
tdh
kbl
L-Threonine
tdcF
tdcE
tdcC
tdcB
tdcA
L-Tryptophan
tnaA
tnaB
Glycyl-L-Proline
putA
putP
L-Valine
livF
livG
livM
livH
livK
yhhK
livJ
Oligopeptides
oppA
oppB
oppC
oppD
oppF

Figure S4: Gene expression (reads per gene - three columns on the left) and differential gene expression between states (three columns on the right) of genes associated to transport and catabolism of various carbon substrates (based Ihssen et al., 2007). Strains are represented by color (orange: MG1655, brown: IAI1, blue: TW11588 and green: TW09308), whereas letters refer to individual states (B; batch, C: chemostat, S: starvation).

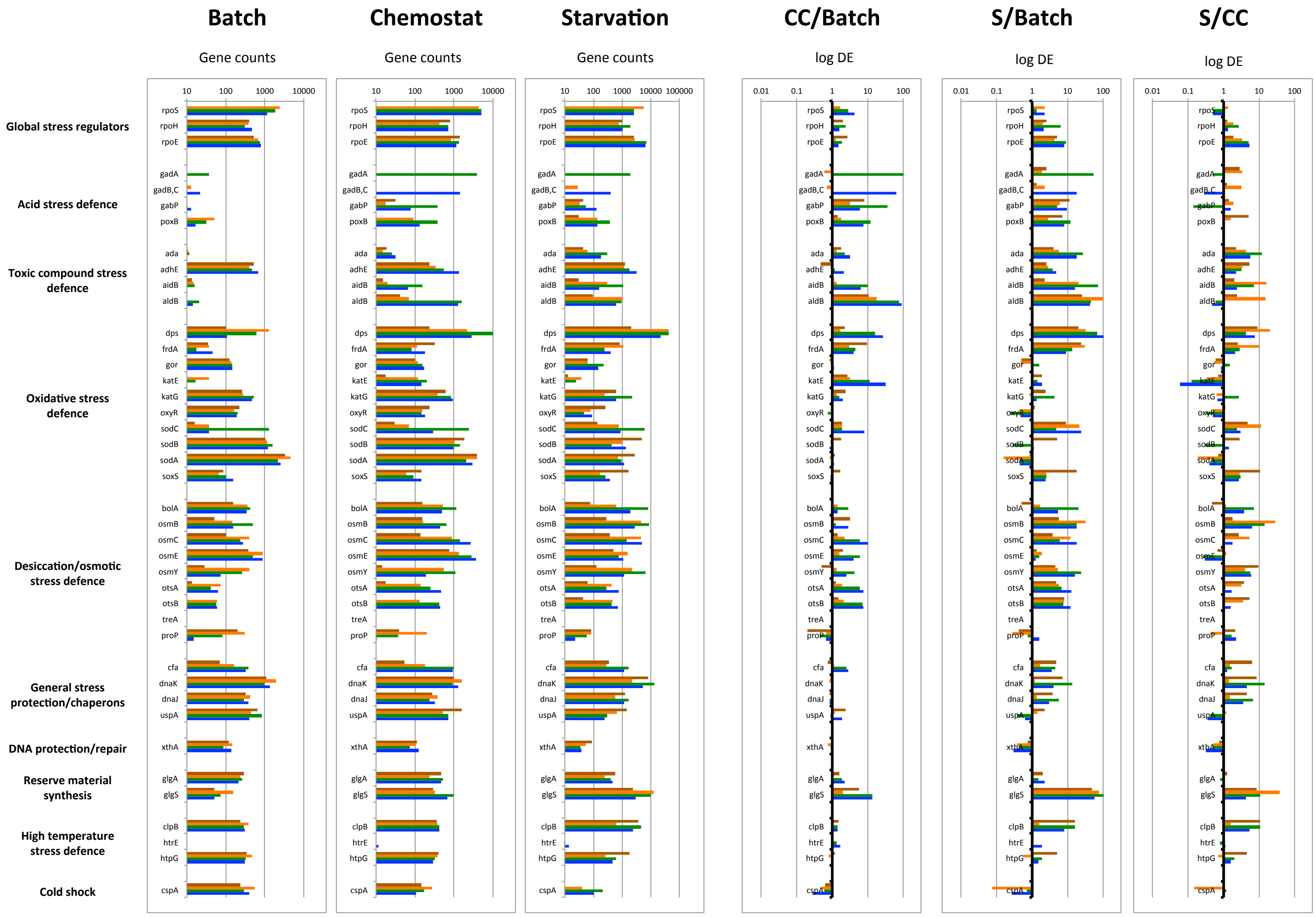


Figure S5: Gene expression (reads per gene - three columns on the left) and differential gene expression between states (three columns on the right) of genes associated to stress defense (based on Ihssen et al., 2007). Strains are represented by color (orange: MG1655, brown: IAI1, blue: TW11588 and green: TW09308), whereas letters refer to individual states (B; batch, C: chemostat, S: starvation).

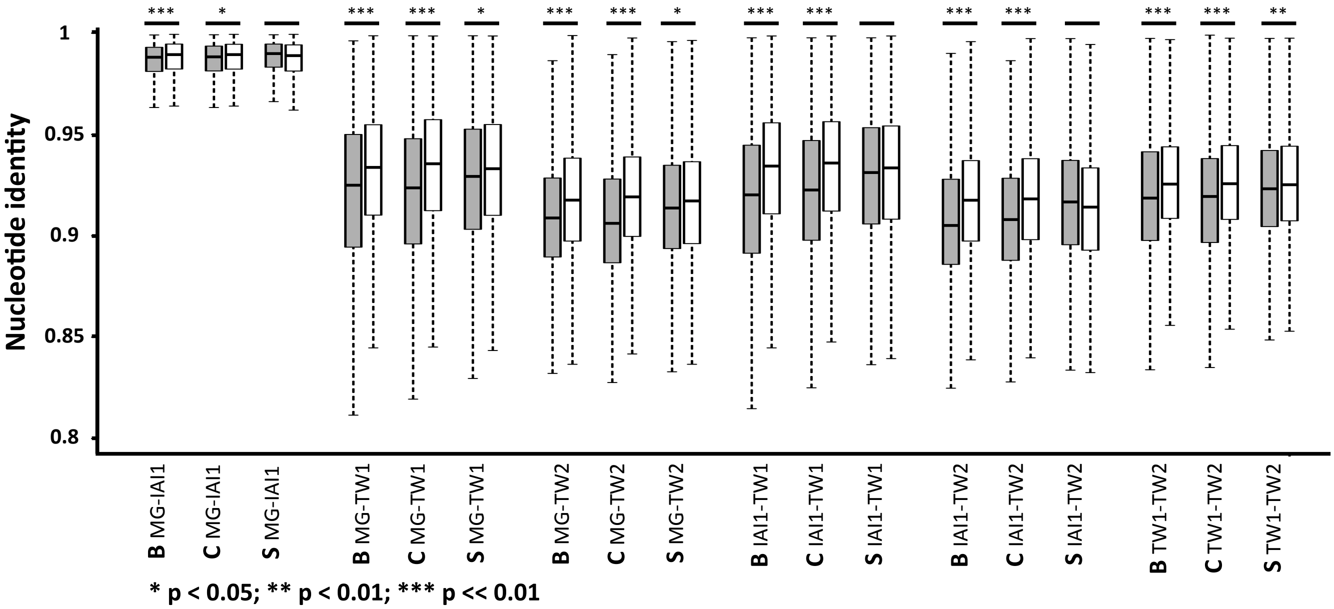


Figure S6: Nucleotide identity of highly differentially expressed genes (>= 2 fold; grey boxplots) was lower compared to nucleotide identity of all other genes (white boxplots). Individual pairwise comparisons are shown, where strains (MG: MG1655, IAI1: IAI1, TW1: TW11588 and TW2: TW09308) and individual states (B; batch, C: chemostat, S: starvation) are indicated. ***: $p << 0.01$, ** $p < 0.01$, * $p < 0.5$ (student t-test). Boxplot outliers are not indicated. Note that boxplots are given, whereas stats are based on the mean and the standard error.

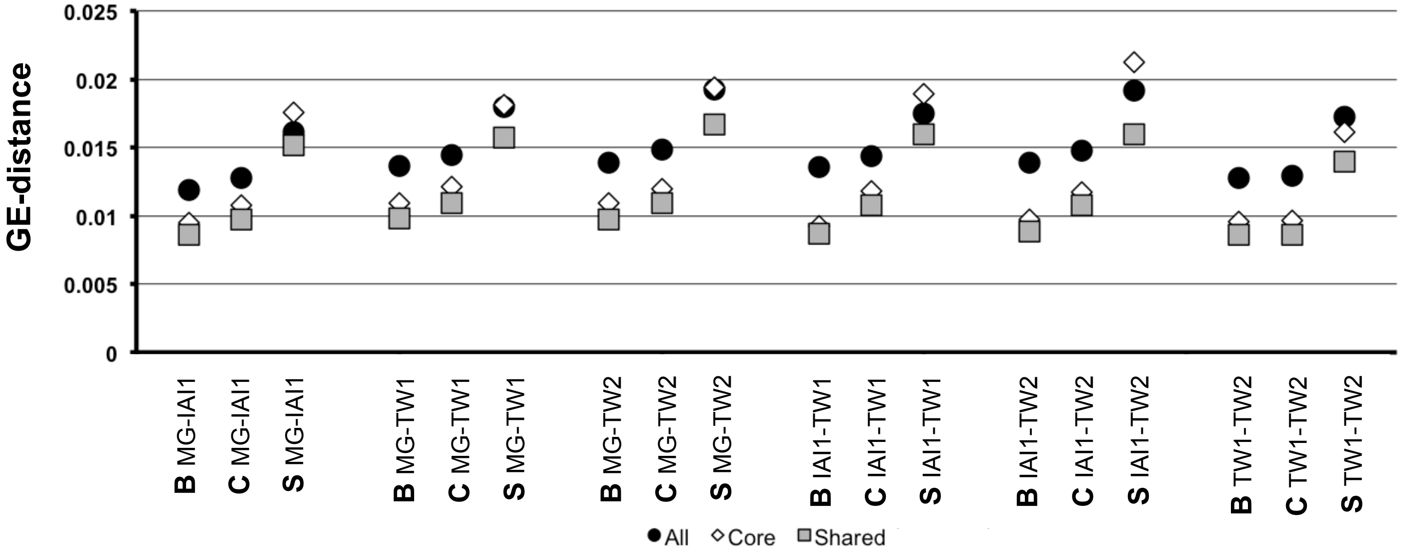


Figure S7: Gene expression profile differences (GE-distances for all pairwise comparisons for all states are shown, where strains (MG: MG1655, IAI1: IAI1, TW1: TW11588 and TW2: TW09308) and individual culture conditions (B; batch, C: chemostat, S: starvation) are indicated. Individual gene sets considered for calculations are displayed (All: all genes, Core: *E. coli* core genes, Shared: all shard genes of isolates).

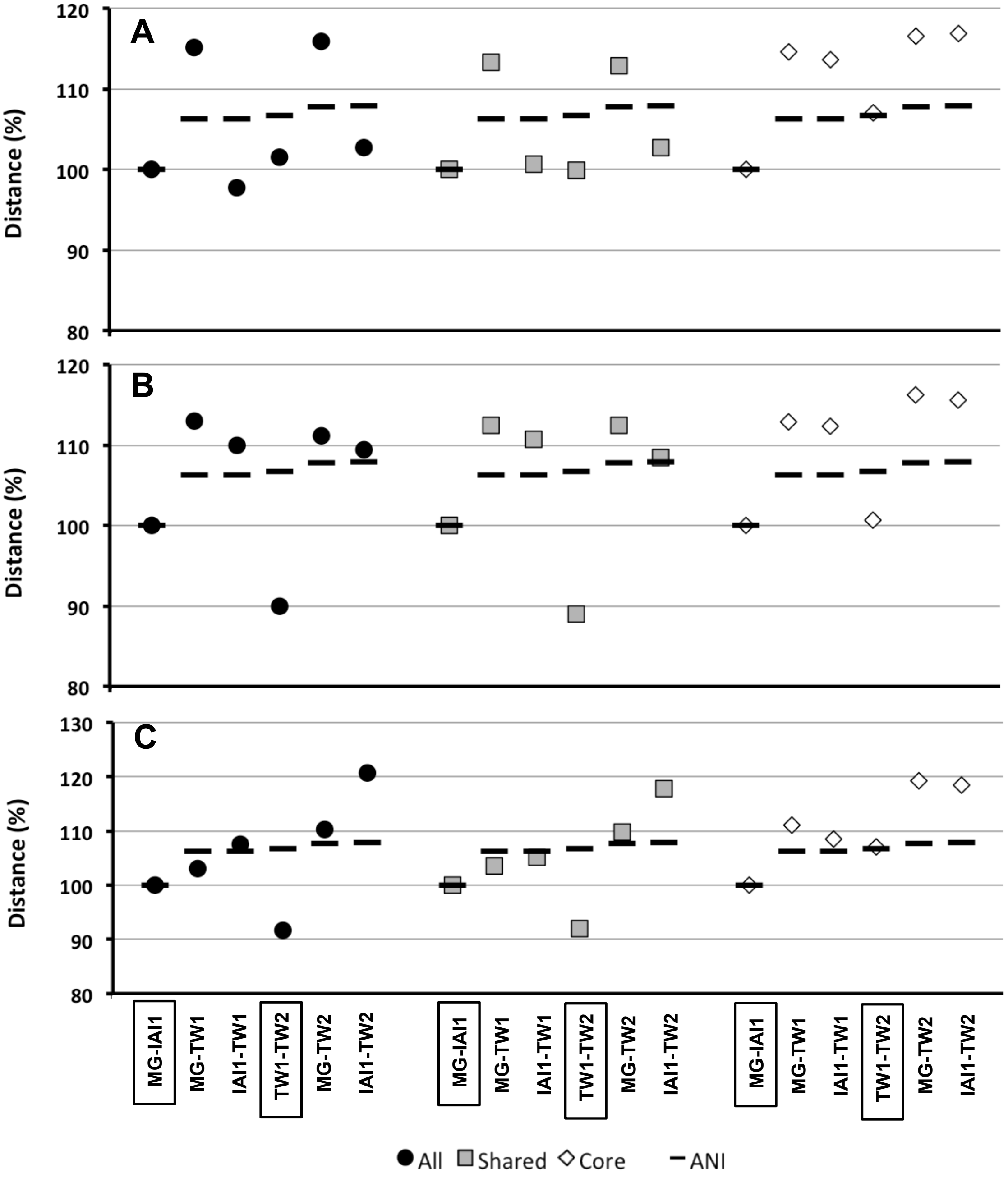


Figure S8: All pairwise comparisons for GE-distances and ANI in all states are shown (A: batch, B: chemostat, C: starvation; MG: MG1655, IAI1: IAI1, TW1: TW11588 and TW2: TW09308). Data are displayed as percentage of the distance between laboratory/commensal strains (= 100 %). Individual gene sets considered for calculations are indicated (All: all genes, Shared: all shard genes of isolates, Core: *E. coli* core genes). The percentage of the reciprocal ANI (Average Nucleotide Identity) from all shared genes between individual pairs is given. The environmental and commensal/laboratory pair, respectively, is highlighted in boxes.

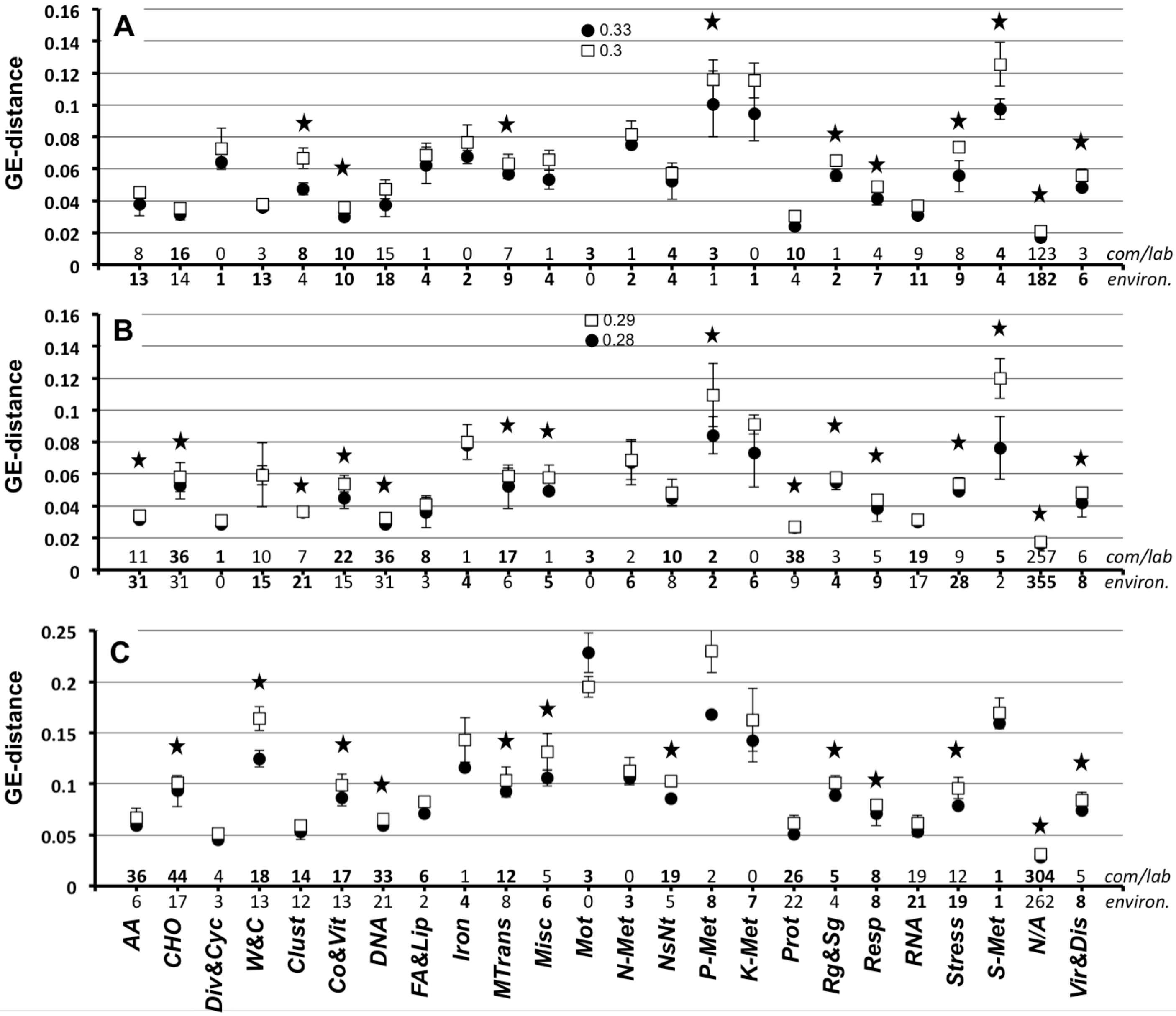


Figure S9: Average gene expression distance (GE-distance) between enteric and environmental strains (☐, n= 4 comparisons) as well as within ecological pairs (•, n= 2 comparisons) for individual RAST categories for all culture conditions (A: batch, B: chemostat and C: starvation). Genes shared by all isolates were considered for analysis. Standard deviation is indicated by error bars. Number of genes up-regulated in individual pairs are given at the bottom of each panel. A black star illustrates that pairs clustered together for that category (>= 80 % bootstrapped). For key to category abbreviations see Figure 4. Categories containing >= 20 genes were included into analysis.