Trait-based approach to bacterial growth efficiency

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Supplemental Methods

Primer Sequences

Forward (27F): AGAGTTTGATCMTGGCTCAG

Reverse (1492R): TACCTTGTTACGACTT

PCR Conditions: Concentrations (Conc.) are given per 50 µL reaction (Rxn.). Components are from the Promega Go-Taq Kit.

Component	Stock Conc.	Final Conc	µL per 50 µL Rxn.
GoTaq Reaction Buffer	5 X	1 X	10
dNTP Mix (10 mM each)	40 mM	200 µM	1
Forward Primer (10 mM)	10 µM	0.2 µM	1
Reverse Primer (10 mM)	10 µM	0.2 µM	1
GoTaq DNA Polymerase	5 U/µL	1.25 U	0.25
Molecular Grade Water			35.75
Template DNA	10 ng/L	10 ng	1

Thermal Cycler Conditions:

Temperature (ºC)	Time (sec.)	Cycles
94	180	
94	45	
50	30	30
72	90	
72	600	
4	hold	

Supplemental Tables

Table S1: Output from linear mixed-effects models describing effect of taxonomy and resource type on bacterial growth efficiency (BGE). Mixed-effects models were fit by REML using the *lme()* function in the *nlme* R package. In these models there is a main variable and a nested variable. For each analysis, the variation explained by the main variable is accounted for before the variation explained by the nested variable is determined. As such, these results indicate the relative importance of each variable when grouped together in a nested framework.

Model	Main Variable	Nested Variable	Variance Explained by Isolate	Variance Explain by Resource	AIC
Isolate Identity	Species	Resource	58.3 %	67.1 %	-48.29
Taxonomic Order	Order	Resource	19.9 %	27.6 %	-94.25
Resource Type	Resource	Species	7.99 %	63.2 %	-116.73

Table S2: Genome summary for the Huron Mountain Wildlife Foundation (HMWF) culture collection. Total number of genes is the number of annotated genes. Coverage estimate was calculated based on average contig coverage during assembly. MCR = module completion ratio from Maple analysis. For additional info see GenomeStats.xlsx or NCBI PRJNA420393.

Strain	Genome length (nucleotides)	# of Genes (total)	# of Pathways (MCR > 80)	Estimated Coverage	Scaffold N50
HMWF001	4,588,649	4,420	52	15x	6,682
HMWF003	4,460,324	4,255	47	13x	14,493
HMWF004	5,476,832	6,021	41	5x	4,613
HMWF005	5,872,459	6,493	66	7x	5,177
HMWF006	6,285,913	6,186	57	7x	14,895
HMWF007	6,051,858	6,348	63	6x	8,597
HMWF008	3,802,708	4,018	35	11x	7,977
HMWF009	3,919,360	3,860	57	14x	19,491
HMWF010	5,890,869	6,195	66	12x	35,798
HMWF011	6,024,040	5,819	69	10x	30,117
HMWF013	4,611,749	4,700	58	9x	9,462
HMWF014	4,397,736	4,521	63	9x	11,207
HMWF015	4,658,495	4,602	67	12x	22,382
HMWF016	4,523,627	4,279	74	21x	65,781
HMWF017	4,461,702	4,205	75	12x	43,441
HMWF018	5,805,605	5,820	47	18x	15,242
HMWF019	6,906,470	5,867	55	15x	19,218
HMWF021	5,954,435	5,610	66	18x	59,774
HMWF022	4,238,619	4,868	24	4x	3,647
HMWF023	4,584,820	4,363	56	14x	24,346
HMWF025	3,960,983	3,764	51	21x	44,746
HMWF026	3,392,253	3,424	50	18x	10,819
HMWF028	4,488,319	4,292	48	13x	4,397
HMWF029	4,650,641	4,374	61	20x	23,346
HMWF030	4,758,486	4,961	53	9x	4,075
HMWF031	10,014,416	12,286	82	4x	2,539
HMWF032	4,391,151	4,100	62	37x	583,475
HMWF034	6,119,028	5,738	72	26x	110,028
HMWF035	4,967,466	4,664	56	2x	69,493
HMWF036	4,455,344	4,269	65	11x	26,720

Table S3: DXS presence/absence in the Huron Mountain Wildlife Foundation (HMWF) culture collection genomes. The DXS pathway (deoxyxylulose-5-phosphate synthase) is an alternative pathway for the synthesis of vitamin B_6. BGE Group: L = Low BGE group; H = High BGE group. Results reported for all HMWF genomes but those used in the current study are indicated.

GENOME	DXS	BGE GROUP	USED IN THIS STUDY
HMWF003	TRUE	L	Yes
HMWF004	TRUE	L	Yes
HMWF005	TRUE	H	Yes
HMWF006	TRUE	H	Yes
HMWF007	TRUE	H	Yes
HMWF008	FALSE	L	Yes
HMWF009	TRUE	L	Yes
HMWF010	TRUE	H	Yes
HMWF011	TRUE	H	Yes
HMWF013	TRUE	NA	No
HMWF014	TRUE	H	Yes
HMWF015	TRUE	NA	No
HMWF017	TRUE		No
HMWF018	TRUE	H	Yes
HMWF019	TRUE	NA	No
HMWF021	TRUE	L	Yes
HMWF022	FALSE	L	Yes
HMWF023	TRUE	L	Yes
HMWF025	TRUE	L	Yes
HMWF026	TRUE	NA	No
HMWF028	TRUE	NA	No
HMWF029	FALSE	L	Yes
HMWF030	TRUE	NA	No
HMWF031	TRUE	H	Yes
HMWF032	TRUE	NA	No
HMWF034	TRUE	H	Yes
HMWF036	TRUE	L	Yes
HWMF016	TRUE	H	Yes

Supplemental Figures

Figure S1: Carbon resources used to study BGE variation in environmental isolates. **A**: Glucose – the baseline resource used to compare BGE across isolates. Glucose can be degraded by the Embden-Meyerhof-Parnas, pentose phosphate, or Entner-Doudoroff pathway. Ultimately, these pathways produce pyruvate (and then acetyl-CoA), which enters Krebs cycle and is used to produce energy and intermediates for biomass synthesis, when cells are grown aerobically. Alternatively, glucose can be fermented into organic acids (e.g., lactate), but these reactions yield less energy (Gottschalk 1986). **B**: Succinate – a simple organic acid. Succinate is an intermediate of Krebs cycle and thus it does not require previous degradation. Additionally, succinate can be directly used to produce energy via succinate dehydrogenase (Neidhardt 2007). **C**: Protocatecuate – is a complex resource with an aromatic core. Typically, it is degraded to acetyl-CoA and succinyl-CoA via the β-ketoadipate pathway (Harwood and Parales 1996). Protocatechuate is commonly used to study aromatic resource degradation in ecosystems, and the β-ketoadipate pathway is commonly found in bacteria across the phylum Proteobacteria (Buchan et al. 2000).

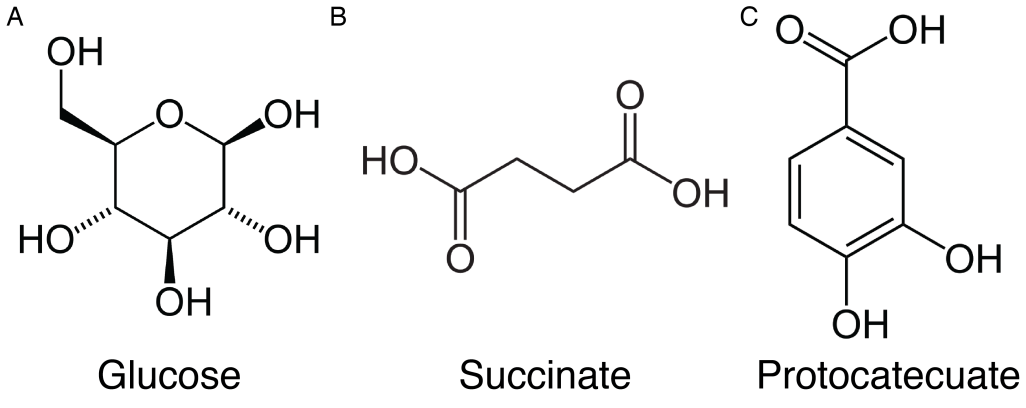

Figure S2: Maximum likelihood phylogenetic tree of lake bacterial isolates used to study BGE. Nearest relatives and other type-strains are included as a taxonomic reference. Isolates are organized by taxonomic class. The outgroup (*Aquifex*) is included as the tree root. Scale bar represents 0.01 base substitutions. The RAxML maximum likelihood tree for just the isolates used in the phylogenetic analyses can be found in Supp_tree.nwk.tre (Newick format).

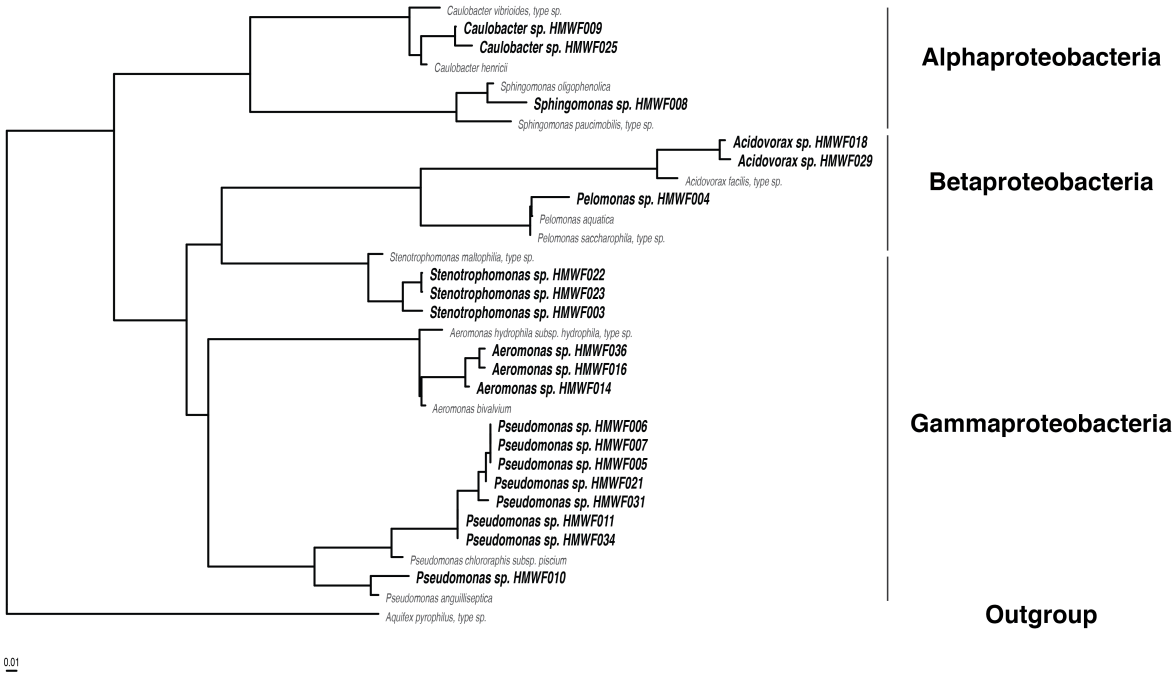

Figure S3: Kernel density of BGE when grow on glucose. Based on Hartigan's dip test, we found that there was a bimodal distribution of BGE among our isolates when supplied with glucose or succinate ($D_{glu} = 0.07$, $p = 0.58$; $D_{suc} = 0.08$, $p = 0.30$). High- and low-BGE groups were determined based on the bimodal distribution of BGE.