Metabolomic profiles of stony coral species from the Dry Tortugas National Park display inter- and intraspecies variation

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mSystems

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Supporting Information

Metabolomic profiles of stony coral species from the Dry Tortugas National Park display inter- and intraspecific variation

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Running title: Intra- and interspecies variation in coral metabolomes

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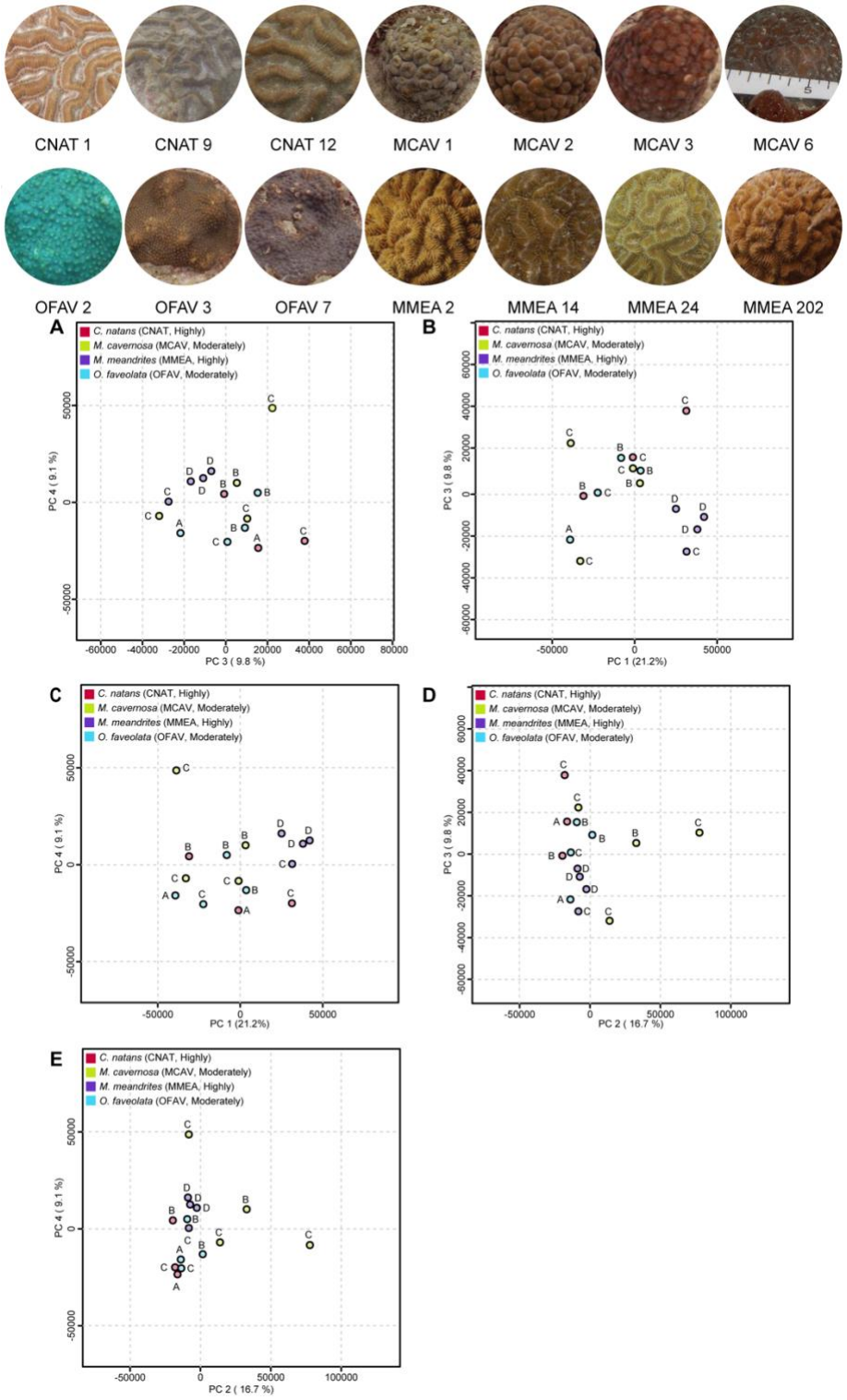


Figure S1. **Images of coral colonies collected and analyzed in this study (top) and unsupervised multivariate statistical analyses of coral extracts**. **(bottom A-E)** The first four principal components from the PCA model. The variation explained by the principal components is included on the corresponding axes. The metabolome profiles of *M. meandrites* show tight clustering across all components, implying low intraspecies variation. For the other species, the principal components shown capture intraspecies variation. The SCTLD susceptibility categorization is included in the key ('Highly', 'Moderately').

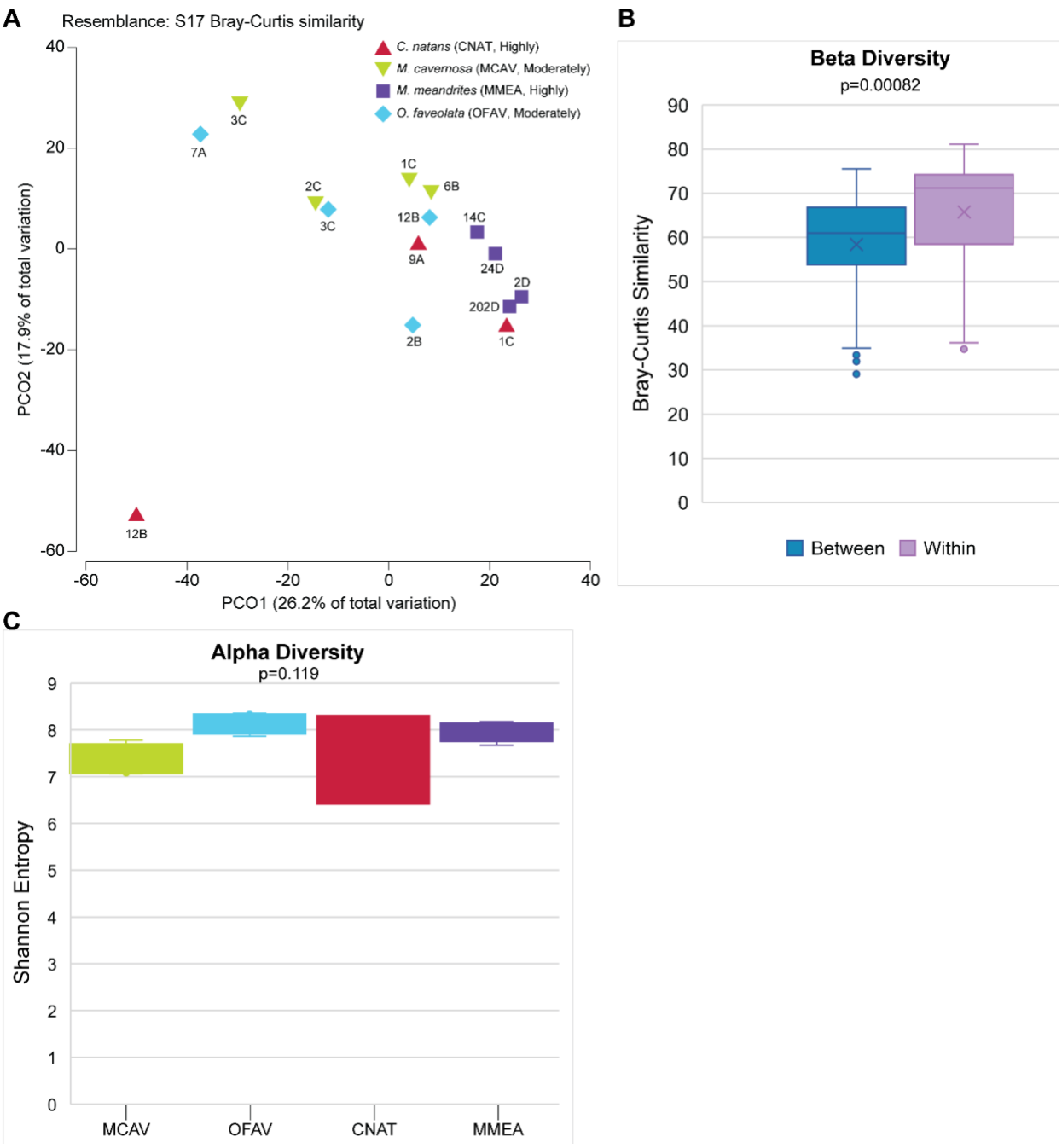


Figure S2. Alpha and beta diversity metrics. (A) Principal coordinates analysis constructed on the Bray-Curtis similarity matrix of the metabolome data. **(B)** Plot of the Bray-Curtis similarity scores between and within species. The Bray-Curtis similarity scores within species is significantly larger (p=0.00082) than the scores between species. **(C)** Plot of the alpha diversity metrics by coral species. No significant comparison was found using a Kruskal-Wallis test. The tests were conducted on a small sample size.

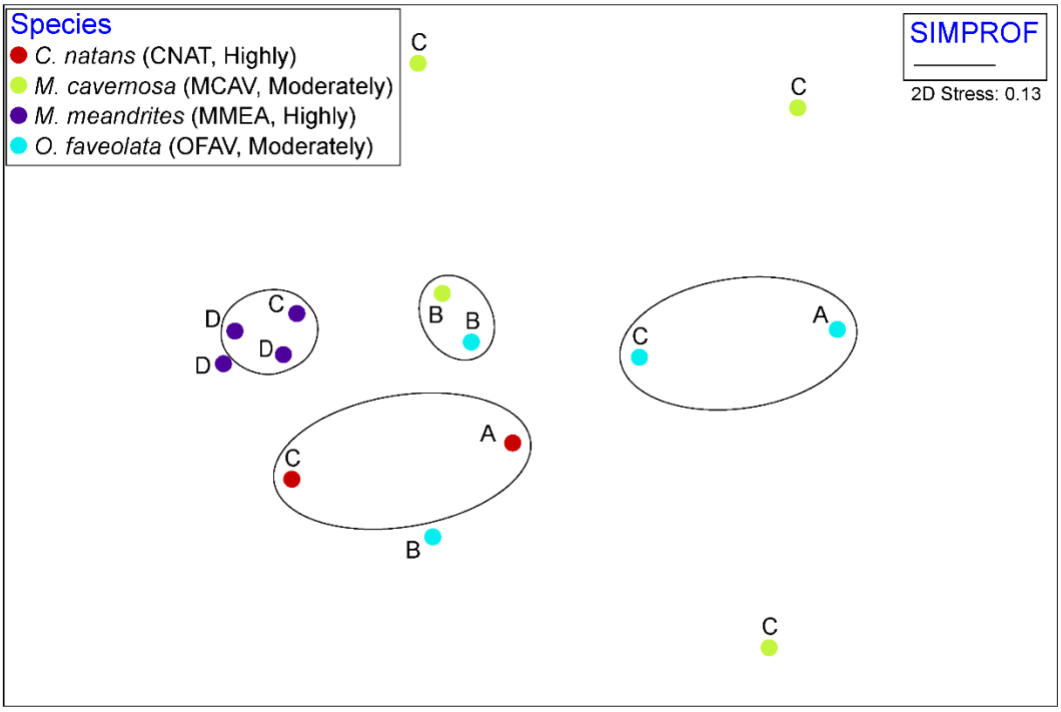


Figure S3. **Non-metric multi-dimensional scaling analysis.** n-MDS plot shows clustering of data into four groups with one group consisting of all *M. meandrites* samples. The SCTLD susceptibility categorization is included in the key ('Highly', 'Moderately'). The largest variation is observed for *M. cavernosa*. Initial analysis using SIMPROF cluster and nMDS revealed CNAT12 as an outlier; thus it was removed from this analysis. Permutational analysis of variance found significance between species (p=0.005) but not sample site.

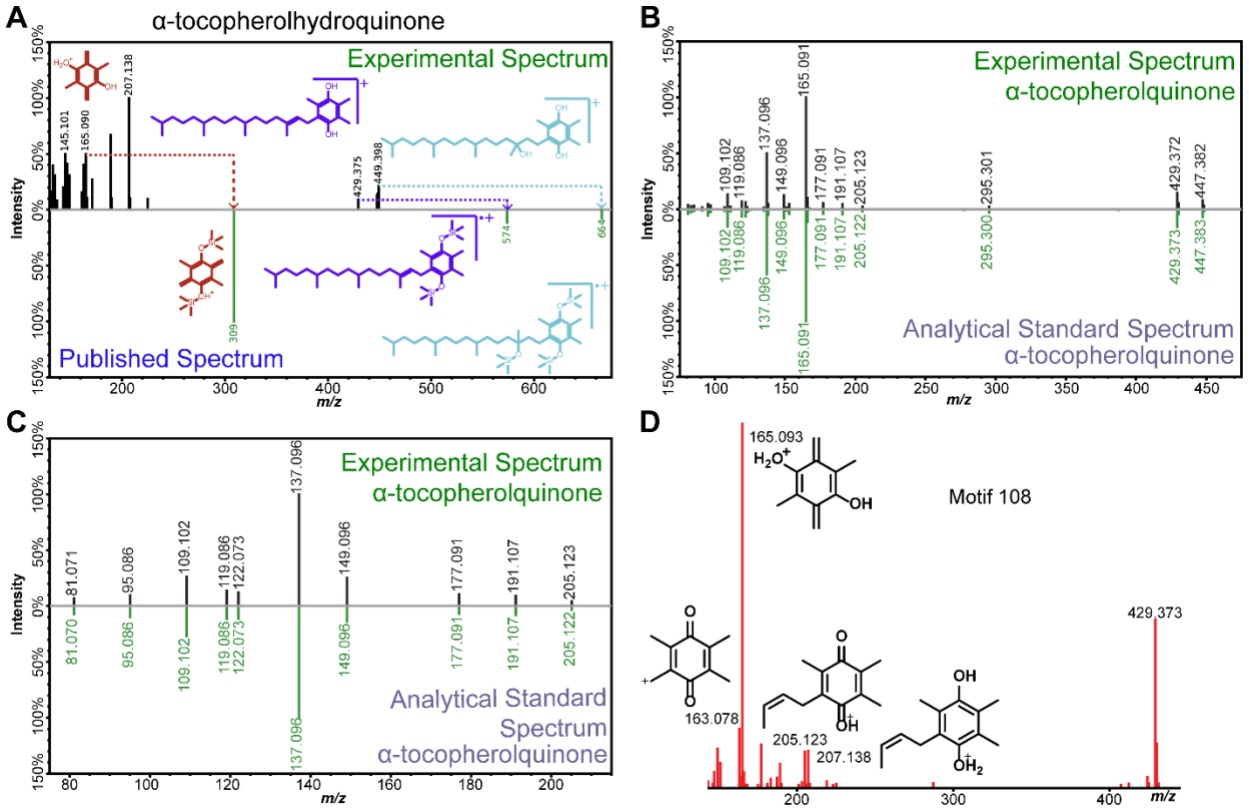


Figure S4. MS² spectra analyses of α-tocopherolhydroquinone and α-tocopherolquinone. (A) MS² mirror plot comparing the experimental spectrum of α-tocopherolquinone (top) compared to a published(1) spectrum of silicated α-tocopherolquinone (bottom). Key chemical substructures supporting the annotation are included, where substructures of the same color correspond. **(B)** MS² mirror plot comparing the experimental spectrum of *m/z*_RT 447.383_21.3 (top) with the spectrum acquired on an analytical standard of α-tocopherolquinone (bottom). **(C)** MS² mirror plot of (B), zoomed in to the *m/z* 80-215 range where the fragment peak at *m/z* 165 has been removed to show matching fragment peaks detected at lower intensity. **(D)** MS2LDA motif 108 used to aid annotations. Chemical substructures for characteristic fragment peaks are included.

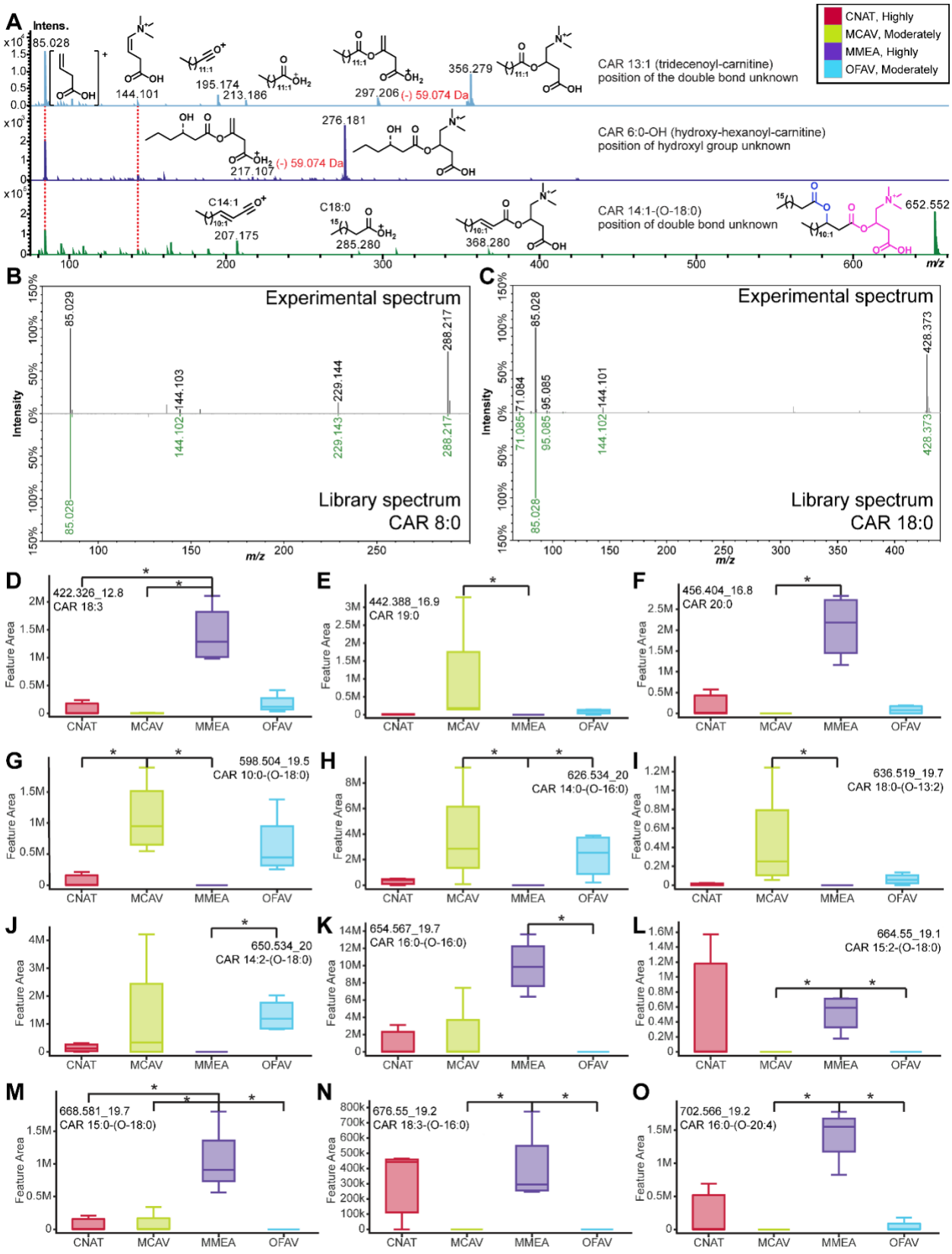
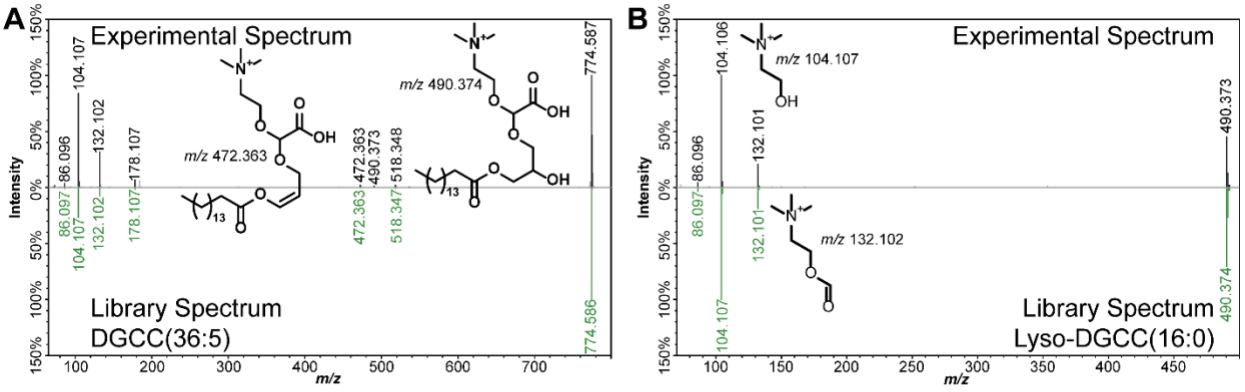

A
Intens.
85.028
144.101
195.174
213.186
297.206
(-) 59.074 Da
356.279
CAR 13:1 (tridecenoyl-carnitine)
position of the double bond unknown
276.181
217.107
(-) 59.074 Da
CAR 6:0-OH (hydroxy-hexanoyl-carnitine)
position of hydroxyl group unknown
C14:1
207.175
C18:0
285.280
368.280
652.552
CAR 14:1-(O-18:0)
position of double bond unknown
m/z
CNAT, Highly
MCAV, Moderately
MMEA, Highly
OFAV, Moderately
B
Experimental spectrum
Library spectrum
CAR 8:0
Intensity
m/z
C
Experimental spectrum
Library spectrum
CAR 18:0
D
422.326_12.8
CAR 18:3
E
442.388_16.9
CAR 19:0
F
456.404_16.8
CAR 20:0
G
598.504_19.5
CAR 10:0-(O-18:0)
H
626.534_20
CAR 14:0-(O-16:0)
I
636.519_19.7
CAR 18:0-(O-13:2)
J
650.534_20
CAR 14:2-(O-18:0)
K
654.567_19.7
CAR 16:0-(O-16:0)
L
664.55_19.1
CAR 15:2-(O-18:0)
M
668.581_19.7
CAR 15:0-(O-18:0)
N
676.55_19.2
CAR 18:3-(O-16:0)
O
702.566_19.2
CAR 16:0-(O-20:4)
Feature Area
CNAT
MCAV
MMEA
OFAV

Figure S5. **Acylcarnitine annotation and differential detection. (A)** The MS^2 spectra of representative acylcarnitine analogues are shown. Conserved fragment peaks, with the chemical substructure, are indicated between spectra. Key fragment peaks used to support the acylcarnitine annotation include *m/z* 85.028 and 144.101.(2) A representative MS^2 spectra of an acylcarnitine analogue containing a conjugated fatty acyl ester of hydroxy fatty acid (FAHFA) is also shown (bottom spectrum) The acylcarnitine headgroup is shown in pink, and the novel linkage of the fatty acyl ester is in blue. The fragment observed at *m/z* 285.280 is representative of C18:0 and supports this annotation. **(B)** and **(C)** MS^2 mirror plots for the experimental spectrum (top) and the library spectrum in GNPS (bottom). **(D-O)** The box plots of the relative abundance of features annotated as acylcarnitines. Asterisks indicate significant differences between the compared groups as determined by a Kruskal Wallis test with Dunn's post-hoc test (adjusted $p<0.05$). Each box plot is labeled with the *m/z*_RT and proposed annotation. The SCTLD susceptibility categorization is included in the key at the top of the figure ('Highly', 'Moderately').



C

```
QUERY scaninfo(MS2DATA) WHERE
MS2PROD=104.1070:TOLERANCEPPM=5
AND MS2PROD=132.1019:TOLERANCEPPM=5
AND MS2PROD= 472.3633:TOLERANCEPPM=5
AND MS2PROD=490.3738:TOLERANCEPPM=5
```

Figure S6. Annotation of DGCC betaine lipids. (A) and **(B)** MS^2 mirror plots for the experimental spectra (top) and the library spectra in GNPS (bottom). Key fragment peaks at *m/z* 490.373 and 472.373 enabled (A) to be further annotated as DGCC(16:0_20:5) (Figure S6). The corresponding chemical substructures for these fragment peaks are included, including substructures for *m/z* 132.102 and 104.107. **(C)** Shows the query submitted to MassQL to search for lyso-DGCC(16:0) analogues containing the 16:0 fatty acid tail.

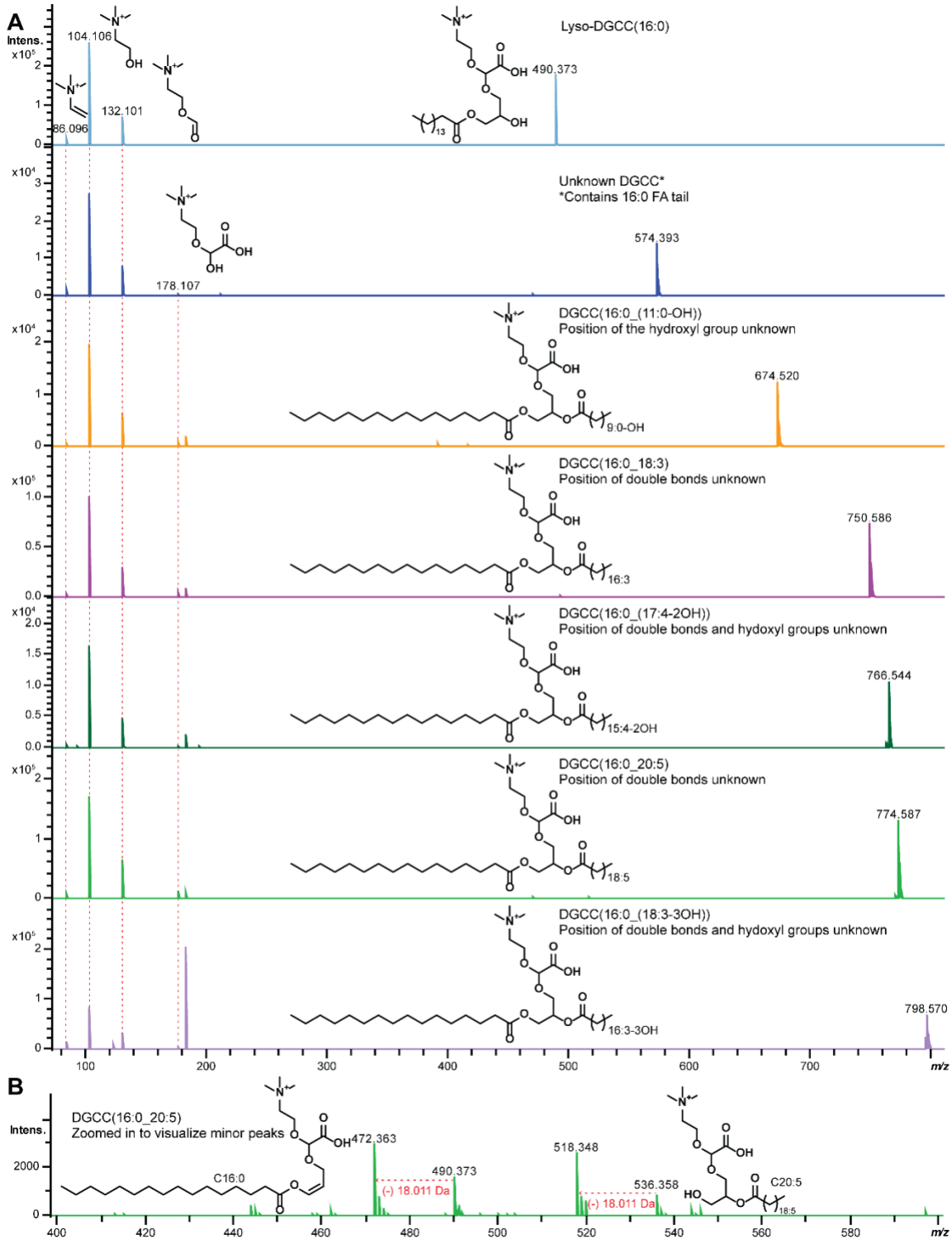
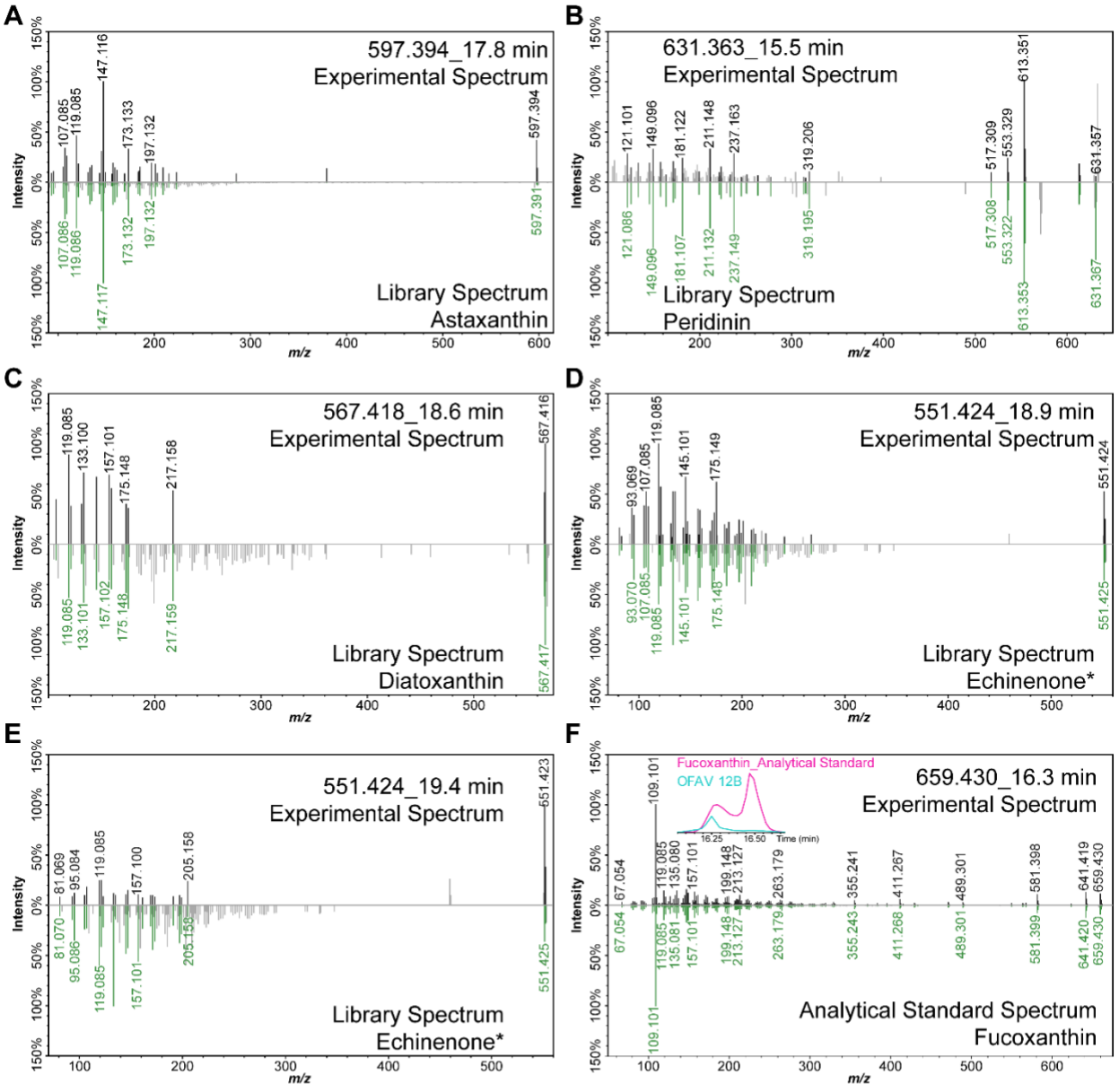


Figure S7. MS^2 spectral analysis of lyso-DGCC(16:0) and analogues. (A) The MS^2 spectra of lyso-DGCC (monoacylated; 16:0) and diacylated DGCCs are shown. Conserved fragment peaks, including the chemical substructure, are indicated between spectra. Fragment peaks *m/z* 86.096, 104.107, 132.102, and 178.107 support the DGCC annotation. Mass shifts between key fragment peaks that aided in annotation are indicated. **(B)** A zoomed in of the spectrum of DGCC (16:0_20:5) in the *m/z* range 400-600 is shown. The fragment peaks at *m/z* 490.373 and 472.363 support the 16:0 fatty acyl tail.



G

*m/z*_RT	Putative Annotation	*Breviolum*	*Durusdinium*	*Symbiodinium*
551.424_19.4	Echinenone or isomer	A	A	A
551.425_18.9	Echinenone or isomer	A	A	A
567.418_18.6	Diatoxanthin	A	A	A
583.415_18.1	Diadinoxanthin or Diadinochrome*	A	A	A
583.415_20.5	Diadinoxanthin or Diadinochrome*	P	P	A
585.43_17	Antheraxanthin	A	A	A
597.394_17.8	Astaxanthin	A	A	A
601.423_15.1	Neoxanthin	A	A	A
613.352_17.7	Pyrroxanthin	P	P	A
631.363_15.5	Peridinin	P	P	P
659.430_16.3	Fucoxanthin	A	P	A
583.416_18.4	Diadinoxanthin or Diadinochrome*	P	A	P
		P: present	A: absent	

Figure S8. MS^2 mirror plots of features annotated as pigments. (A-E) MS^2 mirror plots for the experimental spectra (top) and the library spectra in GNPS (bottom) are shown, along with the pigment name, *m/z* and retention time. The * indicates features that have identical MS^2 spectra, but different

retention times representing isomeric species. **(F)** MS^2 mirror plot for the experimental spectrum of the feature annotated as fucoxanthin compared with the spectrum acquired on a fucoxanthin analytical standard. The extracted ion chromatogram for *m/z* 659.430 is shown as an inset for an *O. faveolata* extract and the analytical standard. **(G)** The list of annotated pigments, including their detection pattern in the zooxanthellae genera included in this study.

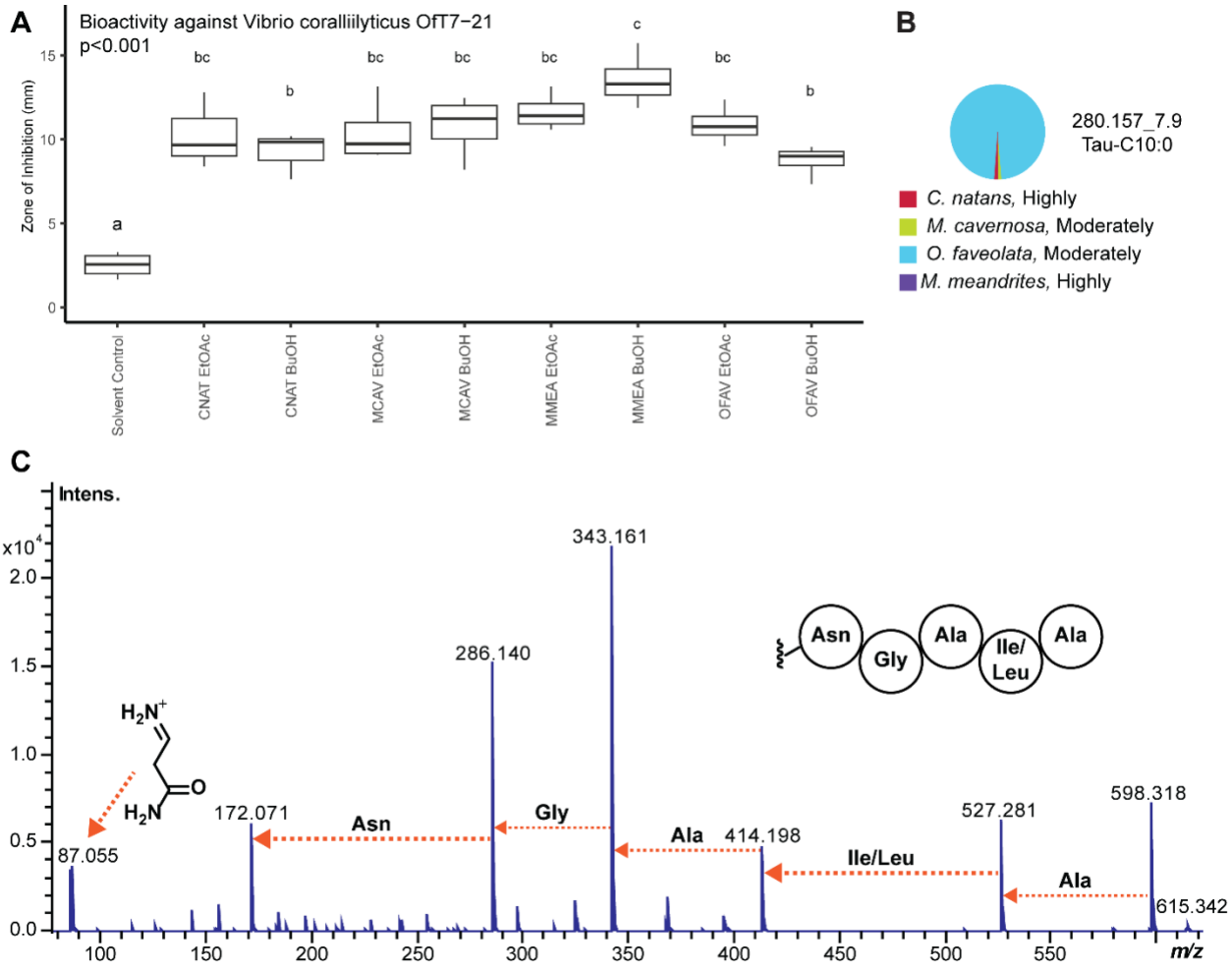


Figure S9. Bioactivity of BuOH partitions and annotations of compounds detected. (A) Bioactivity of EtOAc and BuOH partitions of crude extracts determined using agar diffusion growth inhibition assay against the coral pathogen *V. coralliilyticus* OfT7-21. **(B)** The feature annotated as Tau-C10:0 showed highest detected relative abundance in *O. faveolata*. The SCTLD susceptibility categorization is included in the key ('Highly', 'Moderately'). **(C)** Partial annotation of a polypeptide. Feature 615.3461_5.9 min was proposed as an analogue of tunicyclin G by DEREPLICATOR. The experimental MS^2 spectrum supports the annotation of a polypeptide containing amino acid residues NGAI/LA. The detection of the Asn immonium ion (*m/z* 87.055) aided the annotation and refuted the annotation of a tunicyclin G analogue.

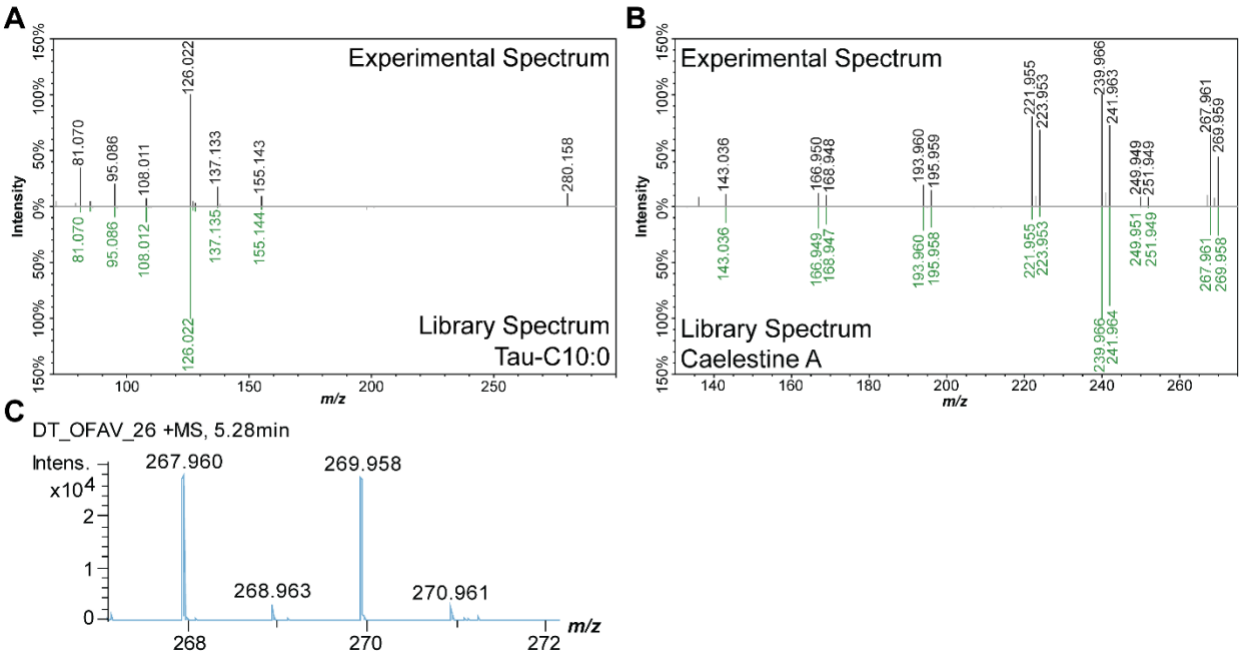


Figure S10. MS^2 Mirror Plots for annotations of compounds uniquely detected in BuOH partitions. **(A)** Tau-C10:0 was annotated through the GNPS library where the experimental spectrum (top) is compared to the GNPS library spectrum (bottom). **(B)** MS^2 mirror plot of the feature annotated as Caelestine A, where the experimental spectrum (top) is compared to the GNPS library spectrum (bottom). **(C)** Isotopic pattern representative of bromination.

Links to MassQL Query Output:

Acylcarnitine analogues:
https://gnps.ucsd.edu/ProteoSAFe/status.jsp?task=a1029fbb7b2b49e787febf1942032b2f

Lyso-DGCC(16:0) analogues:
https://gnps.ucsd.edu/ProteoSAFe/status.jsp?task=8737be5237d445889bba22013b2cfd70

Table S1. Sample and collection site information.

Species	Identification Number & Site	Depth (m)	Fragment Surface Area (cm^2 or cmxcm)	Date Collected
M. meandrites	14, C	14.6	7.98	1/28/2020
M. meandrites	24, D	16.8	4	1/28/2020
M. meandrites	2, D	13.4	1	1/28/2020
M. meandrites	202, D	13.7	5.75	1/28/2020
C. natans	1, C	13.4	2.1	1/28/2020
C. natans	9, A	18.3	13.6	1/29/2020
C. natans	12, B	17.4	5.6	1/28/2020
M. cavernosa	2, C	14.3	9	1/28/2020
M. cavernosa	3, C	14.3	11.2	1/28/2020
M. cavernosa	1, C	14.3	5.4	1/28/2020
M. cavernosa	6, B	16.8	3.4	1/28/2020
O. faveolata	2, B	15.8	5x4	1/28/2020
O. faveolata	3, C	14	6x3	1/28/2020
O. faveolata	7, A	18	5x4	1/29/2020
O. faveolata	12, B	16.5	5x5	1/28/2020

Site	A	B	C	D
Coordinates	24°38.564 N, 82°58.161 W	24°35.608 N, 82°58.862 W	24°35.275 N, 82°58.767 W	24°35.000 N, 82°58.236 W

Approximate Distance Between Sites (km)	A	B	C	D
B	6			
C	6	1		
D	7	2	1	

Table S2. Metabolite features annotated in this study.

*m/z*_RT	Putative Annotation	Annotation Method
Tocopherol Analogues		
429.373_20.5	α-tocomonoenol*	Previous study[1]
429.373_20.7	α-tocomonoenol*	Previous study[1]
429.373_21.7	α-tocomonoenol*	Previous study[1]
429.373_22.3	α-tocomonoenol*	Previous study[1]
447.383_21.1	α-Tocopherolquinone*	SIRIUS with CSI:Finger ID
447.383_21.3	α-Tocopherolquinone*	FBMN Propagation, Analytical Standard[3]
449.398_21.3	α-Tocopherolhydroquinone	MolDiscovery and SIRIUS with CSI:Finger ID
441.337_19.0	Hydroxy-α-tocotrienol	SIRIUS with CSI:Finger ID
424.333_19.7	α-tocotrienol	Previous study[1]
Acylcarnitines		
276.18_2.8	CAR 6:0-OH	MS2LDA
288.217_7.4	CAR 8:0	GNPS Library Match
344.279_11.1	CAR 12:0	MS2LDA
356.279_11.2	CAR 13:1	SIRIUS with CSI:Finger ID
372.311_12.6	CAR 14:0	SIRIUS with CSI:Finger ID
396.311_12.2	CAR 16:2	MS2LDA
398.326_13.1	CAR 16:1	SIRIUS with CSI:Finger ID
400.342_13.9	CAR 16:0	MassQL
422.326_12.8	CAR 18:3	MolDiscovery
426.357_14.3	CAR 18:1	MassQL
428.373_16.6	CAR 18:0	GNPS Library Match
442.388_16.9	CAR 19:0	MS2LDA
446.327_12.7	CAR 20:5	MassQL
456.404_16.8	CAR 20:0	MS2LDA
476.373_14.4	CAR 22:4	MolDiscovery
484.436_18.1	CAR 22:0	MS2LDA
510.451_18.2	CAR 24:1	MS2LDA
512.467_19.1	CAR 24:0	MS2LDA
536.467_18.7	CAR 26:2	MS2LDA
598.504_19.5	CAR 10:0-(O-18:0)	MassQL
626.534_20	CAR 14:0-(O-16:0)	MS2LDA
636.519_19.7	CAR 18:0-(O-13:2)	MassQL
640.55_19.9	CAR 13:0-(O-18:0)	MassQL
648.519_19.7	CAR 16:3-(O-16:0)	MassQL
650.534_20	CAR 14:2-(O-18:0)	MassQL
652.551_20.1	CAR 14:1-(O-18:0)	MassQL
654.567_19.7	CAR 16:0-(O-16:0)	MassQL
664.55_19.1	CAR 15:2-(O-18:0)	MS2LDA
666.566_20.0	CAR 13:0-(O-20:1)	MassQL

668.581_19.7	CAR 15:0-(O-18:0)	MS2LDA
676.55_19.2	CAR 18:3-(O-16:0)	MassQL
702.566_19.2	CAR 16:0-(O-20:4)	MS2LDA
728.581_19.9	CAR 20:4-(O-18:1)	MassQL
DGCC		
798.567_18.3	DGCC(16:0_(18:3-3OH))	MassQL
750.587_20.3	DGCC(16:0_18:3)	MassQL
574.395_14	Unknown DGCC[#]	Manual MS^2 Annotation
674.52_16.1	DGCC(16:0_(11:0-OH))	Manual MS^2 Annotation
766.545_18.6	DGCC(16:0_(17:4-2OH))	Manual MS^2 Annotation
774.584_19.6	DGCC(16:0_20:5)	GNPS Library Match
490.373_13.4	Lyso-DGCC(16:0)	GNPS Library Match
Pigments		
551.424_19.4	Echinenone*	GNPS Library Search
551.425_18.9	Echinenone*	GNPS Library Search
567.418_18.6	Diatoxanthin	GNPS Library Search
583.415_18.1	Diadinoxanthin or Diadinochrome*	Literature Search[2]
583.415_20.5	Diadinoxanthin or Diadinochrome*	Literature Search[2]
585.43_17	Antheraxanthin	Literature Search[2]
597.394_17.8	Astaxanthin	GNPS Library Hit
601.423_15.1	Neoxanthin	Literature Search[2]
613.352_17.7	Pyrroxanthin	Literature Search[2]
631.363_15.5	Peridinin	GNPS Library Search
659.430_16.3	Fucoxanthin	Analytical Standard[3]
583.416_18.4	Diadinoxanthin or Diadinochrome*	Literature Search[2]

[1]Deutsch *et al.* 2021 (3)

[2]Wakahama *et al.* 2012 (4)

[3]Level 1 Annotation, all other reported annotations are Level 2 (5)

[#]Contains 16:0 FA tail

*isomeric compounds with identical MS^2 spectra but different retention times

Table S3. MASST results for features annotated as acylated acylcarnitines and *N*-acyl taurine, where "X" indicates that feature contained a match from the dataset (massIVE ID indicated in the rightmost column).

Putative Annotation	*m/z*_RT	*Organism/Sample Type* *Montipora capitata*	*Porites*	*Sarcophyton* spp.	*Montastraea cavernosa*	*Lobophytum* spp.	Tunicata	Marine sponges, algae and tunicates	Coral	MassIVE IDs (MSV0000…)
Car 14:0-(O-16:0)	626.534_20	X								85925
Car 13:0-(O-18:0)	640.55_19.9		X	X	X	X	X			87449, 80632, 80597, 84587, 87471, 84430
Car 14:2-(O-18:0)	650.534_20						X			84143
Car 14:1-(O-18:0)	652.551_20.1		X						X	80597, 80632, 79808
Car 16:0-(O-16:0)	654.567_19.8						X	X		87449
Car 15:0-(O-18:0)	668.581_19.7						X			87449
Car 18:3-(O-16:0)	676.55_19.2						X			87449, 84143
Car 16:0-(O-20:4)	702.566_19.3						X			84143
		Bacillariophyta	*Lingulodinium*	*Pseudonitzschia*	*Synechococcus*	Coral Reef Seawater	Dissolved Organic Matter and Algal Extracts			
Tau-C10:0	280.157_7.9	X	X	X	X		X			85555, 87692, 81731, 88187, 87702, 87935, 88062, 87995, 88008, 87608, 87322, 87861, 86926, 85779, 85843, 83632, 83889, 85480, 86236, 87863, 88823, 85852, 88734, 88723, 82082, 82952, 86993, 87747

Table S4. Predicted chemical classes of features uniquely detected in BuOH partitions.

*m/z*_RT	Most Specific Class (CANOPUS)
230.248_12.4	1,2-aminoalcohols
258.279_14.3	1,2-aminoalcohols
286.31_17.3	1,2-aminoalcohols
428.445_20.6	1,2-aminoalcohols
523.472_17.4	1,2-diacylglycerols
353.232_5.7	1,2-diacylglycerols
304.176_4.8	1,3-substituted cyclopentyl purine nucleosides
512.278_11.7	1-acyl-sn-glycero-3-phosphocholines
492.272_10.1	1-acyl-sn-glycero-3-phosphocholines
508.304_12	1-acyl-sn-glycero-3-phosphocholines
496.339_23.9	1-acyl-sn-glycero-3-phosphocholines
511.257_17.5	1-acyl-sn-glycero-3-phosphocholines
564.365_11.2	1-acyl-sn-glycero-3-phosphocholines
468.307_9.6	1-acyl-sn-glycero-3-phosphocholines
552.401_11.9	1-acyl-sn-glycero-3-phosphocholines

550.349_10.9	1-acyl-sn-glycero-3-phosphocholines
542.323_10.5	1-acyl-sn-glycero-3-phosphocholines
468.309_9.9	1-acyl-sn-glycero-3-phosphocholines
572.334_10.9	1-acyl-sn-glycero-3-phosphocholines
542.324_10.7	1-acyl-sn-glycero-3-phosphocholines
742.574_18.4	1-alkyl,2-acylglycero-3-phosphocholines
738.543_23.8	1-alkyl,2-acylglycero-3-phosphocholines
524.37_23.9	1-alkyl,2-acylglycero-3-phosphocholines
564.402_16	1-alkyl,2-acylglycero-3-phosphocholines
812.585_20	1-alkyl,2-acylglycero-3-phosphocholines
742.575_20.1	1-alkyl,2-acylglycero-3-phosphocholines
469.342_17.9	3-alkylindoles
344.279_12.4	Acylcarnitines
266.139_4.1	Acylcarnitines
484.436_20	Acylcarnitines
374.29_9.1	Acylcarnitines
422.326_13.7	Acylcarnitines
400.341_17	Acylcarnitines
428.372_18.1	Acylcarnitines
456.405_19.2	Acylcarnitines
476.374_17.3	Acylcarnitines
506.42_19.2	Acylcarnitines
504.404_18.6	Acylcarnitines
442.389_18.7	Acylcarnitines
428.372_18.4	Acylcarnitines
510.451_20.1	Acylcarnitines
484.435_20.1	Acylcarnitines
414.355_17.9	Acylcarnitines
398.326_16.1	Acylcarnitines
448.342_16	Acylcarnitines
193.195_14.8	Alkatrienes
295.172_14.6	Alkyl glycosides
357.213_7.3	Alkyl glycosides
760.536_19.6	Alpha amino acid amides
718.547_19.7	Alpha amino acid amides
324.167_5.7	Alpha amino acid esters
516.403_12.1	Alpha amino acid esters
504.389_16.1	Alpha amino acids
462.342_12.9	Alpha amino acids
520.346_10.9	Alpha amino acids
542.288_11.3	Alpha amino acids
504.39_17.5	Alpha amino acids
528.353_11.2	Alpha amino acids

514.373_13.8	Alpha amino acids
361.21_7.9	Alpha amino acids
472.363_17.8	Alpha amino acids
486.342_12.6	Alpha amino acids
722.555_21	Alpha amino acids
504.389_17.3	Alpha amino acids
472.363_17.8	Alpha amino acids
504.389_16.8	Alpha amino acids
414.356_17.6	Alpha amino acids
434.312_15.3	Alpha amino acids
412.377_17.7	Alpha amino acids
529.276_10.2	Alpha amino acids and derivatives
451.236_16.4	Alpha amino acids and derivatives
412.378_17.4	Alpha amino acids and derivatives
485.337_16	Alpha amino acids and derivatives
289.153_6	Alpha amino acids and derivatives
345.993_7	Alpha amino acids and derivatives
518.405_17.5	Alpha amino acids and derivatives
540.426_17.6	Alpha amino acids and derivatives
540.426_17.6	Alpha amino acids and derivatives
434.312_14.4	Alpha amino acids and derivatives
504.389_16.4	Alpha amino acids and derivatives
311.244_8	Alpha amino acids and derivatives
414.353_18.3	Alpha amino acids and derivatives
708.309_16.1	Alpha amino acids and derivatives
519.355_24	Amino acids
332.149_9	Amino acids
343.2_7.9	Amino acids
346.274_15.9	Amino acids and derivatives
282.059_9.5	Amino acids and derivatives
463.338_16	Amino acids and derivatives
285.29_11.5	Amino acids and derivatives
271.274_11.5	Amino acids and derivatives
313.321_12.8	Amino acids and derivatives
546.436_19	Amino acids and derivatives
369.384_18.1	Amino acids and derivatives
394.342_17.8	Amino acids and derivatives
402.323_17.1	Amino acids and derivatives
369.383_17.7	Amino acids and derivatives
500.394_18.5	Amino acids and derivatives
498.452_20.5	Amino acids and derivatives
309.178_7.1	Amino acids and derivatives
340.323_17.6	Amino acids and derivatives

426.394_20	Amino acids and derivatives
452.373_17.5	Amino acids and derivatives
418.317_16.9	Amino acids and derivatives
286.201_9.3	Amino acids and derivatives
294.157_6.5	Amino acids, peptides, and analogues
482.284_18.4	Aminocyclitol glycosides
368.153_19.4	Aminotriazines
296.237_17.5	Aniline and substituted anilines
248.143_9.1	Aniline and substituted anilines
527.217_11.1	Anisoles
246.128_8.6	Aralkylamines
307.161_6.8	Aralkylamines
380.354_19.6	Azacyclic compounds
165.091_20.3	Benzaldehydes
455.328_19.6	Benzene and substituted derivatives
345.278_16	Benzene and substituted derivatives
329.211_10	Benzene and substituted derivatives
340.214_8.7	Benzene and substituted derivatives
345.278_15.9	Benzene and substituted derivatives
340.248_7.1	Benzene and substituted derivatives
305.139_21.8	Benzoic acids and derivatives
327.122_5.3	Benzoic acids and derivatives
453.021_7.4	Benzothiazoles
339.936_5.7	Bisphosphonates
314.305_18.2	Carboxylic acid amides
480.384_19	Carboxylic acid derivatives
414.393_19.3	Ceramides
540.499_22.4	Ceramides
476.358_13.7	Cholines
506.404_14.3	Cholines
520.345_10.6	Cholines
506.404_14.1	Cholines
725.58_12.7	Cyclic depsipeptides
330.336_19.1	Dialkyl ethers
594.339_8.2	Dicarboxylic acids and derivatives
601.36_12.4	Dicarboxylic acids and derivatives
378.222_16.5	Diphenylmethanes
378.222_16.7	Diphenylmethanes
331.226_12.4	Eicosanoids
459.488_12.6	Ethers
259.19_12	Fatty acid esters
375.252_10.8	Fatty acid esters
447.332_15.8	Fatty acid esters

514.373_13.8	Fatty acid esters
444.404_17.3	Fatty acid esters
392.205_16.4	Fatty acid esters
512.467_21	Fatty acid esters
538.483_21	Fatty acid esters
530.368_12.3	Fatty acid esters
510.429_19.7	Fatty acid esters
521.492_20.4	Fatty acid esters
568.457_18.5	Fatty acid esters
339.23_12.1	Fatty acid esters
430.388_18.6	Fatty acid esters
390.284_8.2	Fatty acids and conjugates
547.331_19.7	Fatty acyl glycosides
836.602_20.6	Fatty acyl glycosides of mono- and disaccharides
407.352_20.1	Fatty Acyls
525.498_12.6	Fatty Acyls
400.378_18.5	Fatty Acyls
364.321_19.7	Fatty Acyls
398.363_19.1	Fatty Acyls
374.362_17.8	Fatty Acyls
365.231_7.3	Fatty alcohols
825.584_20.5	Fatty alcohols
588.512_15.9	Fatty amides
268.263_13.3	Fatty amides
314.305_17.6	Fatty amides
268.263_12.8	Fatty amides
426.373_20.7	Fatty amides
698.591_21.1	Fatty amides
784.557_18.8	Glycerophosphocholines
848.564_16.1	Glycerophosphocholines
800.588_23.9	Glycerophosphocholines
784.56_18.9	Glycerophosphocholines
838.576_19.3	Glycerophosphocholines
568.287_8.1	Glycerophosphocholines
654.427_17.9	Glycerophosphocholines
840.617_20.2	Glycerophosphocholines
612.386_10.7	Glycerophosphocholines
848.565_16.2	Glycerophosphocholines
824.649_12.8	Glycerophosphocholines
558.318_11.3	Glycerophosphocholines
772.572_21	Glycerophosphocholines
762.586_21.4	Glycerophosphocholines

574.314_10.2	Glycerophosphocholines
483.254_11.6	Glycerophospholipids
332.208_8.7	Glycosylamines
379.379_19.9	Guanidines
379.378_19.6	Guanidines
215.082_3.3	Harmala alkaloids
487.411_24.1	Heteroaromatic compounds
371.221_9.6	Kaurane diterpenoids
530.287_23.8	L-alpha-amino acids
260.222_11.5	Leucine and derivatives
507.294_10.2	Lipids and lipid-like molecules
540.427_16.8	Lipids and lipid-like molecules
318.226_7.3	Long-chain fatty acids
329.21_8.6	Long-chain fatty acids
528.272_11.2	Lysophosphatidylcholines
524.277_11.3	Lysophosphatidylcholines
558.319_12.1	Lysophosphatidylcholines
522.355_9.4	Lysophosphatidylcholines
506.287_10.2	Lysophosphatidylethanolamines
506.288_10.1	Lysophosphatidylethanolamines
588.446_15.6	Macrolides and analogues
286.201_9.4	Methyl-branched fatty acids
516.984_7.4	Monoalkyl phosphates
383.242_9.5	Monosaccharides
365.363_19.2	Monoterpenoids
329.316_13.3	N-acyl amines
301.285_11.6	N-acyl amines
240.232_11.6	N-acyl amines
546.436_18.8	N-acyl amines
574.467_20	N-acyl amines
329.316_12.9	N-acyl amines
329.316_12.8	N-acyl amines
396.31_13	N-acyl amines
518.405_17.8	N-acyl amines
500.394_18.8	N-acyl amines
468.441_19	N-acyl amines
440.409_17.6	N-acyl amines
568.457_18.9	N-acyl amines
440.409_16.9	N-acyl amines
708.577_23	N-acyl amines
468.441_19.1	N-acyl amines
574.467_19.9	N-acyl amines
383.373_17.2	N-acyl amines

380.217_9	N-acyl amines
554.55_20.4	N-acyl amines
301.286_12.2	N-acyl amines
440.409_17.7	N-acyl amines
398.363_19	N-acyl-alpha amino acids
346.258_8.2	N-acyl-alpha amino acids
344.242_8.8	N-acyl-alpha amino acids
300.216_6.9	N-acyl-alpha amino acids
546.4_17.6	N-acyl-alpha amino acids
532.421_18.2	N-acyl-alpha amino acids
604.406_16.1	N-acyl-alpha amino acids
571.431_14.8	N-acyl-alpha amino acids
546.399_17	N-acyl-alpha amino acids
344.242_9.3	N-acyl-alpha amino acids
358.258_9.6	N-acyl-alpha amino acids and derivatives
388.249_9.8	N-acyl-alpha amino acids and derivatives
345.203_5.9	N-acyl-alpha amino acids and derivatives
520.347_7.2	N-acyl-alpha amino acids and derivatives
570.34_7.7	N-acyl-alpha amino acids and derivatives
604.406_16.7	N-acyl-alpha amino acids and derivatives
518.405_18.3	N-acyl-alpha amino acids and derivatives
544.384_17.2	N-acyl-L-alpha-amino acids
544.384_16.9	N-acyl-L-alpha-amino acids
299.198_6.9	N-acyl-L-alpha-amino acids
590.391_16.3	N-acyl-L-alpha-amino acids
437.025_7	Naphthalenes
399.237_7.9	Naphthalenes
410.218_6.6	N-carbamoyl-alpha amino acids and derivatives
686.358_16.5	Oligopeptides
469.313_15.8	Oligopeptides
457.313_14.8	Oligopeptides
568.287_9.1	Oligopeptides
729.468_10.1	Oligopeptides
680.473_18.2	Oligopeptides
562.361_13.4	Oligopeptides
402.284_9.4	Oligopeptides
574.395_16.5	Oligopeptides
522.362_8.4	Oligopeptides
405.219_16.8	Oligopeptides
594.339_8.3	Oligopeptides
574.395_17	Oligopeptides
314.232_9.5	Organic phosphonic acids
326.199_8.1	Organic phosphoramides

438.503_21	Organonitrogen compounds
360.29_17.8	Oxosteroids
373.31_16.7	Oxosteroids
396.214_7.8	Peptides
574.358_10.2	Peptides
500.394_18.9	Peptides
616.442_17.4	Peptides
532.385_13.7	Peptides
496.333_9.4	Peptides
682.488_18.2	Peptides
506.404_16	Peptides
468.42_23	Phenanthrenes and derivatives
450.394_18	Phenol ethers
467.203_8.3	Phenoxy compounds
512.299_9.1	Phenylalanine and derivatives
618.421_17.4	Phenylalanine and derivatives
404.313_16.8	Phenylalanine and derivatives
388.393_19.1	Phenylmethylamines
360.362_18	Phenylmethylamines
332.331_17.1	Phenylmethylamines
332.331_15.7	Phenylmethylamines
332.331_16.4	Phenylmethylamines
360.362_18.4	Phenylmethylamines
416.425_20	Phenylmethylamines
846.606_20.1	Phosphatidylcholines
800.581_23.8	Phosphatidylcholines
826.594_23.8	Phosphatidylcholines
732.554_16.7	Phosphatidylcholines
732.554_17.2	Phosphatidylcholines
754.537_23.8	Phosphatidylcholines
828.609_23.8	Phosphatidylcholines
818.589_23.8	Phosphatidylcholines
764.573_23.9	Phosphatidylcholines
732.553_19.1	Phosphatidylcholines
732.553_19.1	Phosphatidylcholines
652.455_16.6	Phosphatidylcholines
688.497_17.9	Phosphatidylcholines
620.392_18.1	Phosphatidylcholines
786.601_17.2	Phosphatidylcholines
552.329_9.3	Phosphatidylcholines
580.397_23.9	Phosphatidylcholines
700.455_14.9	Phosphatidylcholines
612.386_11.4	Phosphatidylcholines

812.613_15.3	Phosphatidylcholines
732.554_18.6	Phosphatidylcholines
756.553_15.5	Phosphatidylcholines
756.553_15.8	Phosphatidylcholines
794.592_15.7	Phosphatidylcholines
806.568_14.5	Phosphatidylcholines
774.564_23.8	Phosphatidylcholines
554.344_8.8	Phosphatidylcholines
596.357_10.7	Phosphatidylcholines
580.397_15.4	Phosphatidylcholines
735.522_16.3	Phosphatidylcholines
808.584_20.8	Phosphatidylcholines
600.329_9	Phosphatidylcholines
822.564_19.8	Phosphatidylcholines
830.613_20.8	Phosphatidylcholines
846.617_18.3	Phosphatidylcholines
776.577_20.1	Phosphatidylcholines
800.578_21.1	Phosphatidylcholines
842.589_23.7	Phosphatidylcholines
582.34_12.8	Phosphatidylethanolamines
594.377_14	Phosphatidylethanolamines
702.473_16.4	Phosphatidylethanolamines
706.501_19.6	Phosphatidylethanolamines
528.272_10.3	Phosphatidylethanolamines
666.397_11.5	Phosphatidylserines
355.216_11.4	Phosphinic acid esters
542.344_9.7	Phosphocholines
531.366_16.6	Phosphocholines
610.334_7.3	Phosphocholines
674.519_18.6	Phosphosphingolipids
671.512_19.3	Phosphosphingolipids
701.559_23.9	Phosphosphingolipids
585.366_13	Phosphosphingolipids
456.275_8.5	p-Hydroxybenzoic acid alkyl esters
277.201_9.6	Polyethylene glycols
363.31_11.7	Polyethylene glycols
581.462_16.5	Polyethylene glycols
553.43_14.9	Polyethylene glycols
306.243_11.2	Prenol lipids
351.348_18.9	Prenol lipids
379.173_3	Purine nucleosides
423.01_6.7	Pyrimidine nucleotides
539.289_17.1	Saccharolipids

307.227_10.2	Steroid acids
464.374_19.7	Steroid acids
432.347_19.2	Steroid esters
424.341_16.2	Steroidal alkaloids
496.415_19.4	Steroidal alkaloids
456.42_22.9	Steroidal alkaloids
416.336_13.4	Steroidal alkaloids
373.31_13.6	Steroids and steroid derivatives
428.388_21.5	Steroids and steroid derivatives
442.404_22.1	Steroids and steroid derivatives
440.389_20.8	Steroids and steroid derivatives
246.128_8.6	Styrenes
403.209_10.2	Styrenes
767.497_17.2	Sulfoquinovosyldiacylglycerols
765.481_16.5	Sulfoquinovosyldiacylglycerols
274.159_14.3	Triarylamines
553.268_10.5	Tricarboxylic acids and derivatives
437.341_10.2	Triterpenoids
674.519_18.3	Very long-chain fatty acids
505.497_20.1	Wax monoesters
341.305_17.1	-
318.128_16.2	-
886.617_20.5	-
395.783_24.1	-
258.279_13.8	-
501.339_14.6	-
629.366_13.3	-
300.992_13.2	-
373.274_10.1	-
357.347_14.1	-
270.315_17.4	-
308.15_18.3	-
417.3_19.2	-
376.313_24	-
160.17_4.8	-
371.28_23.9	-
272.222_12.3	-
228.268_13.5	-
258.279_14	-
200.237_12.1	-
268.001_6.8	-
286.31_17	-
221.014_6.2	-

348.144_9	-
285.16_5.9	-
366.155_9	-
220.015_6.2	-
385.378_18.1	-
260.003_6.2	-
248.143_9.2	-
313.321_12.9	-
222.013_6.2	-
429.318_11.8	-
229.202_5.1	-
241.203_9.6	-
252.996_6	-
164.645_5.2	-
268.001_6.3	-
244.008_6.5	-
213.016_5.5	-
270.315_15.6	-
286.31_16.8	-
182.106_5.3	-
362.253_8	-
381.225_5.3	-
473.344_11.9	-
228.268_13.9	-
382.2_6.9	-
241.203_9.8	-
221.014_6.3	-
242.284_14	-
395.783_24.1	-
246.243_9.4	-
427.389_14.8	-
598.263_8.4	-
355.262_10.1	-
815.482_16.1	-
402.386_19.8	-
270.315_17	-
499.396_17.9	-
541.277_12.3	-
575.503_23.8	-
213.016_5.3	-
206.009_5.2	-
212.017_5.2	-
242.248_16.8	-

260.003_5.9	-
205.665_7.7	-
164.644_5	-
244.008_6.1	-
374.291_13.8	-
252.996_5.8	-
313.273_23.8	-
480.331_11.7	-
319.263_15.5	-
383.79_23.9	-
271.263_24.9	-
212.017_5.6	-
214.015_5.7	-
236.01_6	-
221.007_5.2	-
206.009_5.5	-
245.007_6.5	-
206.037_5.1	-
191.003_4.6	-
214.253_12.2	-
241.203_10.1	-
176.036_4.5	-
258.279_16.2	-
206.037_5.4	-
157.092_6	-
281.009_4.8	-
498.342_9.3	-
286.31_17.9	-
314.232_9.5	-
344.352_20.5	-
366.265_11.4	-
316.248_10.4	-
267.027_10.3	-
353.744_15.1	-
280.3_17.7	-
411.769_24.2	-
250.642_10.4	-
371.722_15.8	-
268.001_6.6	-
298.346_18.9	-
201.16_4.7	-
314.341_19	-
360.237_7.1	-

200.237_12	-
380.211_7.3	-
404.786_24.1	-
260.258_10.4	-
304.284_12.4	-
268.671_10.9	-
333.349_21.4	-
292.192_24	-
341.187_8.6	-
999.756_14.6	-
289.153_5.2	-
239.225_12.2	-
320.699_24	-
425.287_24.2	-
263.684_24.1	-
389.254_24.1	-
291.302_19.2	-
484.762_5.9	-
268.176_24.1	-
354.844_24.3	-
268.263_13	-
526.483_21.2	-
396.456_21.5	-
206.009_5.9	-
244.999_5.9	-
765.749_7.1	-
342.278_23.9	-
209.14_4.1	-
414.191_7.1	-
382.199_6.7	-
372.383_23.3	-
372.383_23.4	-
371.722_15.2	-
244.21_24	-
929.577_21	-
1593.361_10.8	-
1199.27_10.8	-
389.321_23.7	-
332.165_3.6	-
1191.271_11	-
252.207_24.1	-
258.279_15.8	-
420.757_24.1	-

238.191_24.1	-
296.258_17.4	-
1199.269_10.7	-
323.158_4.5	-
248.193_7.3	-
426.393_20.6	-
433.763_24.1	-
850.594_23.8	-
584.282_6.4	-
382.331_23.9	-
383.79_20.3	-
244.21_21.4	-
286.31_17.5	-
239.225_16.7	-
296.684_24.1	-
858.582_15.6	-
422.399_22.5	-
852.608_23.8	-
389.321_22.6	-
358.367_21.9	-
358.368_22.6	-
1109.723_9.9	-
536.755_6.5	-
255.182_6.1	-
441.198_7.4	-
403.336_24	-
1082.454_10.6	-
860.6_23.8	-
454.404_22.2	-
402.328_22.6	-

Table S5. Symbiodiniaceae strains cultured in this study (strains were originally sourced from the BURR collection and kindly provided by Mary Alice Coffroth).

Genera	Strain ID from BURR Collection
Breviolum	*Breviolum* MF 1.05b.01 SCI 07-205
Breviolum	*Breviolum* MF 1.05b.01 SCI 07-209
Breviolum	*Breviolum* Mf 10.14.02 SCI
Durusdinium	*Durusdinium* Mf 2.2b-2
Durusdinium	*Durusdinium* Mf 2.2b-2
Symbiodinium	*Symbiodinium* Mf 10.02a
Symbiodinium	*Symbiodinium* 04-503 SCI.01

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