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# **Supporting Information**

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#### 1. General Methods and Materials

**NMR measurements** were performed on a Bruker Avance II 300 MHz, Bruker AVANCE III 500 MHz and a Bruker AVANCE III 600 MHz spectrometer, equipped with a Bruker Cryoplatform. The chemical shifts are reported in parts per million (ppm) relative to the solvent residual peak of CDCl<sub>3</sub> (<sup>1</sup>H: 7.26 ppm, singlet; <sup>13</sup>C: 77.16 ppm, triplett), C<sub>6</sub>D<sub>6</sub> (<sup>1</sup>H: 7.16 ppm, singlet; <sup>13</sup>C: 128.06 ppm, triplett), H<sub>3</sub>PO<sub>4</sub> (<sup>31</sup>P 0.00 ppm), DMSO-*d*<sub>6</sub> (<sup>1</sup>H: 2.50 ppm, quintet; <sup>13</sup>C: 39.52 ppm, heptet) and MeOD-*d*<sub>4</sub> (<sup>1</sup>H: 3.31 ppm, quintet; <sup>13</sup>C: 49.0 ppm, heptet). The following abbreviations were used to explain the multiplicities: s = singlet, d = doublet, t = triplet, q = quartet, hept = heptet, m = multiplet, br = broad.

**UHPLC-ESI-HRMS** measurements were carried out on a Dionex Ultimate3000 system (Thermo Scientific) combined with a Q-Exactive Plus mass spectrometer (Thermo Scientific) equipped with an elecrospray ion (ESI) source. Metabolite separation was carried out by reverse phase liquid chromatography at 40 °C using a Luna Omega C18 column (100 x 2.1 mm, particle size 1.6 µm, 100 Å, Phenomenex) preceded by a SecurityGuard<sup>TM</sup> ULTRA guard cartridge (2 x 2.1 mm, Phenomenex). Mobile phases were consisted of H<sub>2</sub>O (A) and acetonitrile (B). 5 µl of sample were injected into a gradient as follows: 0–1 min, 10% B; 1–3 min, 22% B; 3–7 min, 27%; 7–20 min, 95% B; 20–22.5 min, 95% B; 22.5–22.6 min, 10% B; 22.6–27 min, 10% B at a constant flow rate of 0.3 mL/min. Metabolites were detected in negtive (MS<sup>1</sup>) ionization mode within a range of m/z 150 – 1800 with a resolving power of 70,000 at m/z 200. MS<sup>2</sup> measurements were performed using combined methods of data-dependent MS<sup>2</sup> analysis and Top10 experiments. The resolving power was set to 70,000 at m/z 200 for MS<sup>1</sup> and 17500 for MS<sup>2</sup>, with an isolation window of 1.0 m/z and a stepped normalized collision energy (NCE) of 20/30/40.

**UHPLC-MS measurements** were performed on a Shimadzu LCMS-2020 system equipped with single quadrupole mass spectrometer using a Kinetex C18 column (50 x 2.1 mm, particle size 1.7  $\mu$ m, pore diameter 100 Å, Phenomenex). Column oven was set to 40 °C; scan range of MS was set to m/z 150 to 2,000 with a scan speed of 10,000 u/s and event time of 0.25 s under positive and negative mode. DL temperature was set to 250 °C with an interface temperature of 350 °C and a heat block of 400 °C. The nebulizing gas flow was set to 1.5 L/min and dry gas flow to 15 L/min.

**Semi-preparative HPLC** was performed on a Shimadzu HPLC system using a Gemini C18(2) 250 x 10 mm column (particle size 5  $\mu$ m, pore diameter 100 Å, Phenomenex) and a Luna Phenyl-Hexyl 250 x 10 mm column (particle size 5  $\mu$ m, pore diameter 100 Å, Phenomenex). **Preparative HPLC** was performed on a Shimadzu HPLC system using a Luna Phenyl-Hexyl 250 x 21.2 mm column (particle size 5  $\mu$ m, pore diameter 100 Å, Phenomenex).

**Solid phase extraction** was carried out using Chromabond  $C_{18}$ ec cartridges filled with 2 g of octadecylmodified silica gel (Macherey-Nagel, Germany).

**Chemicals**: Methanol (VWR, Germany); water for analytical and preparative HPLC (Millipore, Germany), formic acid (Carl Roth, Germany); acetonitrile (VWR and Th. Geyer as LC-MS grade), ammonium hydroxide solution 50% (Alfa Aesar, Germany), chloroform (VWR, Germany), Tropic Marine Sea Salt (catalog # 10134), Bacto Peptone (catalog # 211677) and Bacto Yeast extract (catalog # 210933) were purchased as indicated. All other reagents and solvents for synthesis were purchased from Acros Organics, Alfa Aesar, Carbolution Chemicals, Carl Roth, Fluorochem, Sigma Aldrich, TCI, Th. Geyer and VWR and used without further purification.

All reactions were performed in flame-dried glassware under argon atmosphere, unless otherwise stated.

#### 2. HRMS-based analysis of lipid extracts

Bacterial strains were grown in 50 mL marine broth media (Carl Roth) with 180 rpm at 30 °C until OD<sub>600</sub> reached around 0.8. The cell culture was pelleted under 4°C and the cell pellets were lyophilized for 2 days. The 50 ml supernatant was extracted with resin (30 g/L, HP21:XAD8 1:1) and stirring in a 50 ml falcon tube for overnight. The resin was filtered and extracted with 10 ml MeOH and transferred into a new vial. The 10 ml MeOH extract (from cell supernatant) was dried under a GeneVac. The dried cell pellets were further resuspened in 10 mL freshly prepared MeOH (0.1% NH4OH) and extracted exhaustively with the help of sonication for 30 min. 10 mL of the supernatant (from cell supernatant or pellets) was dissovled into a new vial and evaporated under a GeneVac. The dry extract (from cell supernatant or pellets) was dissovled into a concentration of 50 µg/mL freshly prepared MeOH (0.1%NH4OH) following with sonication for 30 min. After pelleting the extract, 5 µL of the supernatant (from cell supernatant or pellets) was transferred into a HPLC vial for UPLC-HRMS<sup>2</sup> metabolomic-based analysis.

Extracts of bacterial pellets were analyzed for natural products based on UPLC-HRMS measurements and MS/MS fragmentation based network analysis carried out on the global natural products social molecular networking platform (GNPS).<sup>1</sup> A molecular network was created using the online workflow at GNPS. The data was filtered by removing all MS/MS peaks within +/- 17 Da of the precursor *m/z*. MS/MS spectra were window filtered by choosing only the top 10 peaks in the +/- 50 Da window throughout the spectrum. The data was then clustered with MS-Cluster with a parent mass tolerance of 0.02 Da and a MS/MS fragment ion tolerance of 0.02 Da to create consensus spectra. Further, concensus spectra that contained less than 2 spectra were discarded. A network was then created where edges were filtered to have a cosine score above 0.7 for general networks and more than 6 matched peaks. Further edges between two nodes were kept in the network if and only if each of the nodes appeared in each other's respective top 5 most similar nodes. Finally, the maximum size of a molecular family was set to 25, and the lowest scoring edges were removed from molecular families until the molecular family size was below this threshold. The spectra in the network were then searched against GNPS' spectral libraries. The library spectra were filtered in the same manner as the input data. All matches kept between network spectra and library spectra were required to have a score above 0.7 and at least 6 matched peaks.



**Figure S1.** GNPS network analysis of HR-MS/MS measurement in negative mode for relative abundance and composition of cell membrane crude extract from marine broth (brown), *A. machipongonensis* PR1 (black), *D. fermentans* DSM 18053 (green), *Z. uliginosa* ATCC 14397 (red), *E. pacifica* KMM 6172 (purple) and *B. baltica* BA134 (blue). Precursor ion mass tolerance 0.02 Da, fragment ion mass tolerance 0.02 Da, cosine score 0.7. Red circle showing molecular ion clusters assigned to sulfonosphingolipids.



**Figure S2.** GNPS network analysis of HR-MS/MS measurement in negative mode for relative abundance and composition of cell membrane crude extract from background signal, MeOH (brown), marine broth (blue), *Z. galactonovorans* Dsij (black) and *Z. uliginosa* ATCC 14397 (red). Precursor ion mass tolerance 0.02 Da, fragment ion mass tolerance 0.02 Da, cosine score 0.7. Red circle showing molecular ion clusters assigned to sulfonosphingolipids.



**Figure S3.** GNPS network analysis of HR-MS/MS measurement in negative mode for relative abundance and composition of crude extract from culture supernatant after cultivation in marine broth (brown), *A. machipongonensis* PR1 (black), *D. fermentans* DSM 18053 (green), *Z. uliginosa* ATCC 14397 (red), *E. pacifica* KMM 6172 (purple) and *B. baltica* BA134 (blue). Precursor ion mass tolerance 0.02 Da, fragment ion mass tolerance 0.02 Da, cosine score 0.7.

#### 3. Isolation and structural analysis of sulfonolipds

All pilot-scale cultivations were conducted in batch mode in 75 L stirred tank reactor (bbi, Germany). The filling volume was 50 L, inoculated by 0.5 L pre-culture (shake culture, marine broth, 48 hours). The strirring rate was adjusted to 200 rpm. The aeration rate was kept constant as well with a value of 15 L/min. These process parameters ensured aerobic conditions during the cultivations, proven by an online measured dissolved oxygen tension, which was always higher that 50%. The cell pellets were harvested by centrifugation and extracted twice with 1000 mL CHCl<sub>3</sub>/MeOH (2/1) and twice with 1000 mL CHCl<sub>3</sub>/MeOH (1/1). The combined organic extract was filtered, evaporated and concentrated to give approximately 35 g crude lipid extract. The crude lipid extract was sequentially dissolved first in 50% and then in 80% MeOH (0.1% NH<sub>4</sub>OH) to yield a sulfonosphingolipid rich 80% MeOH fraction. The enriched sulfonosphingolipid fraction was first purified using a Biotage Isolera system with a reverse-phase gradient (solvents A (0.1% NH<sub>4</sub>OH in water) and B (0.1% NH<sub>4</sub>OH in acetonitrile): 50% B for 3 CV; 50-100% B for 15 CV, 100% B for 8 CV at a constant flow rate of 25 mL/min. Sulfonosphingolipid enriched fractions were further purified by preparative HPLC using a gradient of solvents A (5% methanol in water) and B (methanol): 0–6 min, 40% B; 6-35 min, 100% B; 35-45 min, 100% B at a constant flow rate of 20 mL/min. If necessary, semipreparative reversed phase HPLC was repeated using a gradient of solvents A (0.1% NH<sub>4</sub>OH in water) and B (0.1% NH4OH in ACN): 0-4 min, 35% B; 4-30 min, 90% B; 30-35 min, 100% B; 35-45 min, 35% B at a constant flow rate of 2 mL/min until analytical pure compounds were obtained. The obtained isolates were repeatedly dissolved in MeOH and dried under vacuum or lyophilized. For NMR assignment, see Table S2 and Table S3.



**Figure S4.** Semi-prep HPLC purification (from top to bottom:  $\lambda = 190, 254, 210 \text{ nm}$ ) of ZU-590 (fraction 10), ZU-602 (fractions 11-12), ZU-572 (fractions 14-15) and ZU-616 (fractions 16-17).



Figure S5. LC-MS/MS profile (5 ppm range) of sulfonosphingolipids isolated from Z. uliginosa based on ESI-HRMS(-) analysis.



**Figure S6.** Zoom in of LC-MS/MS profile (5 ppm range) of sulfonosphingolipids isolated from Z. uliginosa based on ESI-HRMS(-) analysis and the proposed fragments of the isolated sulfonosphingolipids.

#### 4. Choanoflagellate husbandry and bioassays

For bioassay, the *S. rosetta* cell line co-cultured with the non-rosette–inducing prey bacterium *Echinocola pacifica*, called "SrEpac" (for *S. rosetta* + *E. pacifica*), was used. The SrEpac cell line reproducibly yields high percentages of cells in rosettes (>80%) in response to live *Algoriphagus*, outer membrane vesicles (OMVs) isolated from *Algoriphagus*-conditioned medium and bulk lipids extracted from *Algoriphagus*.<sup>2</sup> SrEpac was propagated in 5% Sea Water Complete media (5% vol/vol in artificial sea water). Artificial sea water was made by adding 32.9 g Tropic Marine sea salts (Wartenberg, Germany) to 1L distilled water to a salinity of 32-27 ppm. SrEpac was passaged 1:10 into 9 mL fresh 4% SWC once a day to stimulate rapid growth (cells were grown in 25 cm<sup>2</sup> Corning cell culture flask). For all rosette development bioassays, cultures of single cells were induced shortly after passaging at a density of approximately 10<sup>5</sup> cells/mL.

**Testing of rosettes-activity:** For all assays, rosette development was quantified approximately 22-24 h postinduction. To quantify rosette development, 100  $\mu$ L of treated SrEpac was first pipetted vigorously to disrupt clusters of cells that were not in rosettes, then cell were fixed in 1% formaldehyde immediately before counting (Bright-Line hemacytometer, Hausser Scientific). To determine the fraction of cells in rosettes, single cells and cells within rosettes were scored until 400 total cells had been counted (per technical replicate). To quantify rosette size, the number of cells in each rosette were counted. A group of four or more cells were qualified as a rosette if the cells maintained an organized polarity relative to a central focus (with each cell oriented with the apical flagellum pointing outward) after vigorous physical perturbation, in this case from pipetting up and down or vortexing. Three biological replicates were performed for each assay.

Assay with bacteria extracts and single sulfonosphingolipid: SrEpac was cultured as described above, and aliquoted into 96-well plates (Sarstedt). Bacteria extracts were resuspended in DMSO to a concentration of 5 mg/mL (sonication of samples), and then added to SrEpac to yield the desired concentration. Each sample was tested for cell viability (constant viability) and rosette-inducing activity at final concentrations of 5, 10, 20, 30, 40 and 50  $\mu$ g/mL. Samples showing concentration-dependent induction were further investigated. Purified lipids were resuspended in DMSO with vigorous sonication to a concentration of 4 mM or 8 mM, and then added to SrEpac to yield the final concentrations of 1, 3, 5, 10, 20, 40 and 80  $\mu$ M. Outer membrane vesicles (OMVs) from A. *machipongonensis* PR1 and DMSO were used as positive and negative control, respectively. Rosette development was quantified as described above. Graphs were generated using GraphPad Prism 9 statistical software.

**Table S1.** Bacterial species used for testing and analysis (strains were obtained from the German Collection of Microorganisms and Cell Cultures GmbH (DSMZ).

Sulfonosphingolipids-producing bacterial species	Abbreviation	Induction of rosettes
Algoriphagus machipongonensis PR1	AM	+
Cyclobacterium marinum LMG 13164	СМ	+
Echinicola pacifica KMM 6172	EP	-
Belliella baltica BA134	BB	-
Dyadobacter fermentans DSM 18053	DF	+
Zobellia galactonovorans Dsij	ZG	+
Zobellia uliginosa ATCC 14397	ZU	+

### 5. NMR Assignments



Figure S7. Chemical structures and <sup>1</sup>H and <sup>13</sup>C-NMR assignments.

	<sup>1</sup> H-NMR (600 MHz)						<sup>13</sup> C-NMR (150 MHz)				
	ZU-602	ZU-616		ZU-572	ZU-590		ZU-602	ZU-616		ZU-572	ZU-590
Pos.	δ <sub>H</sub> [a]	$\delta_{\mathrm{H}}^{[a]}$	Pos.	$\delta_{H^{[a]}}$	$\delta_{\mathrm{H}^{[a]}}$	Pos.	$\delta c^{[a]}$	$\delta c^{[a]}$	Pos.	$\delta c^{[a]}$	$\delta c^{[a]}$
	multi. $(J \text{ in Hz})$	2.00		2.00	2.11		-	-		-	-
1	(14, 14, 4, 0, 2)	3.00 (dd 14.4.0.0)	1	3.06 (dd 14.4.80)	3.11	1			1		
	(uu, 14.4, 9.2) 3 12	(uu, 14.4, 9.0) 3 12		(uu, 14.4, 8.0) 3 11	3 11		51.63	51.67		51.6	51.77
	(dd 14433)	5.12 m		(dd 14435)	5.11 m						
	-	-		-	-						
1'			1'			1'	173.92	173.92	1'	175.94	175.99
2	4.33	4.33	•	4.24	4.2	2	50 75	50.70	2	50.96	52.2
2	(ddd, 9.1, 5.9, 3.4)	(ddd, 9.1, 5.9, 3.3)	2	m	(p, )	2	52.75	52.72	2	52.80	55.2
2'	2.31	2.3	21	2.19	2.22	2'	45 44	45 44	21	37 51	37 48
2	m	m	2		(td, 7.6, 3.7)	2		45.44	2	57.51	57.40
	4.18	4.17		4.21	3.98		7504	75.05	•	7504	70.00
3	m	(ddd, 9.6, 5.1, 2.1)	3	(q, 6.0)	(ddd, 9.4, 6.3, 2.2)	3	75.04	75.06	3	75.04	70.99
	2.06	2.06		16	3.2) 1.62						
3'	5.90 m	5.90	3'	1.0 m	1.02 m	3'	69.79	69.79	3'	26.91	26.87
	1 45	(1, 0.7) 1 45		111	111						
4'	m	m				4'	26.7	26.71			
	5.48	5.48		5.47	1.53						
4		(ddt, 15.4, 6.8,	4	(ddt, 15.2, 6.7,	(-, (2))	4	130.53	130.53	4	130.66	41.67
	m	1.4)		1.5)	(p, 6.3)						
	5.73	5.73		5.72	3.82						
5	(dtd 150 68 11)	(dtd, 15.1, 6.8,	5	(dtd, 15.0, 6.8,	m	5	134.85	134.88	5	134.81	69.08
	(ulu, 15.0, 0.0, 1.1)	1.1)		1.1)	–						
6	2.03	2.05	6	2.04	1.45	6	33.51	33.52	6	33.51	26.79
	m	m	<b>F</b> 12 Al	m	m				<b>F</b> 12 AI		
7-13, 5'-12'	1.38 - 1.17	1.38 - 1.17	7-13, 4'-	1.38 - 1.17	1.38 - 1.17	7-13, 5'-12'	30.0-24.7	30.0-24.7	7-13, 4'-	30.0-24.7	30.0-24.7
	m	m	12	m	m				14		
	1 38 - 1 17	1 38 - 1 17		1 16 - 1 11	1 16 - 1 11		40.27	40.27		40.27	40.27
14, 13'	m	m	14, 13'	m	m	14, 13'	10.27	40.27	14, 13'	10.27	10.27
15 14	1.52	1.52	15 14	1.52	1.52	15.14	29.17	29.17	1 - 1 4	29.17	29.17
15, 14'	m	m	15, 14' m	m	15, 14'			15, 14'			
15', 16'	0.88	0.88	15', 16'	0.88	0.88	15', 16'	22.06	22.06	15', 16'	22.06	22.06
16, 17	(d, 6.6)	(d, 6.6)	16, 17	(d, 6.6)	(d, 6.6)	16, 17	23.00	23.00	16, 17	23.00	23.00

Table S2. <sup>1</sup>H and <sup>13</sup>C-NMR (MeOD-*d*<sub>4</sub>) data for sulfonosphingolipids isolated from Z. *uliginosa*. For NMR spectra: see Figure S7 to Figure S26

<sup>[a]</sup>All values in ppm.

	<sup>1</sup> H-NMR (600 MHz)						<sup>13</sup> C-NMR (150 MHz)				
	ZU-602	ZU-616		ZU-572	ZU-590	1	ZU-602	ZU-616		ZU-572	ZU-590
Pos.	$\delta_{H}^{[a]}$	$\delta_{H}^{[a]}$	Pos.	$\delta_{H}^{[a]}$	$\delta_{H}^{[a]}$	Pos.	S [a]	8 [a]	Pos.	S [a]	S [a]
	multi. (J in Hz)						UC -	UC		UC .	UC -
1	2.51	2.50	1	2.63	2.62	1			1		
•	m	m	-	(dd, 14.0, 4.9)	m		51.37	51 41	-	51.48	51.76
	2.66	2.66		2.70	2.77		01107	01111		01110	01110
	(d, 5.5)	(t, 4.4)		(dd, 14.0, 6.4)	(dd, 14.1, 6.1)						
1'	-	-	1'	-	-	1'	170.34	170.36	1'	171.73	171.57
	- 2.05	- 2.09		- 2.01	-						
2	5.95 m	3.90 m	2	(444, 77, 38, 1.4)	(dtd 70.62.45)	2	51.27	51.25	2	51.43	51.44
	2.07	2 07		2 00	(uiu, 7.9, 0.2, 4.3) 2 02						
2'	2.07 m	2:07 m	2'	2:00 m	(t, 7, 3)	2'	44.64	44.66	2'	35.85	35.88
3'-OH	4.69	4.69	3'-OH	-	-	3'-ОН			2'-OH		
	(d, 4.2)	(d, 4.0)		-	-		-	-	_	-	-
2	3.98	3.98	2	4.07	3.78	2	72.00	75 12	2	72	60.16
3	m	m	5	(d, 6.0)	(dtd, 7.9, 6.2, 4.5)	5	73.09	75.15	3	75	09.10
3-OH	5.03	5.03	3-ОН	5.11	4.87	3-OH		_	3-ОН		
	(d, 5.2)	(d, 4.5)		(d, 5.5)	(d, 5.9)						
3'	3.73	3.72	3'	1.45	1.49	3'	67.52	67.55	3'	25.27	26.83
	m	m	_	m	1.05	-			-		
4'	1.29	1.28	4	5.33	1.25	4	130.83	130.95	4	130.94	40.92
	m 5.24	m 5 24		(aat, 15.4, 0.5, 1.4)	m 2.50						
4	3.34 (dd 15.4 5.5)	3.34 (dd 15 4 5 5)	5	5.51 m	(1,0,0)	5	130.93	130.88	5	130.76	66.41
	(uu, 15.4, 5.5) 5 51	(uu, 15.4, 5.5) 5 51			(d, 9.0) 4 17						
5	(dd, 14.2, 6.7)	(dd, 14.2, 6.7)	5-OH	-	(d. 5.5)						
	1.93	1.93	-	1.93	1.29			21.05		21.02	20.5
6	m	m	6	m	m	6	31.82	31.86	6	31.82	38.5
7-13, 5'-12'	1.30 - 1.17	1.30 - 1.17	7-13, 4'-12'	1.30 - 1.17	1.30 - 1.17	7-13, 4'-12'	30.0-24.7	30.0-24.7	7-13, 4'-12'	30.0-24.7	30.0-24.7
	m	m		m	m						
14 13'	1.30 - 1.17	1.30 - 1.17	14 13'	1.16 - 1.11	1.16 - 1.11	14 13'	38.5	38.5	14 13'	38.5	38.5
14, 15	m	m	14, 15	m	m	14,15			14, 15		
15, 14'	1.49	1.49	15, 14'	1.49	1.49	15, 14'	27.41	27.44	15, 14'	27.42	27.41
10,11	m	m		m	m						
15', 16'	0.84	0.84	15', 16'	0.84	0.84	15', 16'	22.54	22.56	15', 16'	22.53	22.54
16, 17 NU	(a, b.b)	(a, b.b)	16, 17 NH	(d, 6.6)	(d, b.b)	16, 17			16, 17		
NH	/.58	1.59	INH	/.50	1.57						
	(dd, 7.9, 2.1)	(dd, 7.9, 2.1)		(d, 7.7)	(d, 7.9)						

**Table S3.** <sup>1</sup>H and <sup>13</sup>C-NMR (DMSO-*d*<sub>6</sub>) data for sulfonosphingolipids isolated from *Z. uliginosa*. For NMR spectra: see Figure S28 to Figure S47.

<sup>[a]</sup>All values in ppm.

### 6. NMR spectra of isolated sulfonosphingolipids



<sup>6.1</sup> Sulfonosphingolipids in MeOD-d4

Figure S8. <sup>1</sup>H-NMR spectrum (MeOD-*d<sub>4</sub>*) of sulfonosphingolipid ZU-572.



Figure S9. <sup>13</sup>C-NMR spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid ZU-572.



Figure S10. HSQC spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid **ZU-572**.



Figure S11. COSY spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid ZU-572.



Figure S12. HMBC spectrum (MeOD-d4) of sulfonosphingolipid ZU-572.



Figure S13. <sup>1</sup>H-NMR spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid **ZU-590**.



Figure S14. <sup>13</sup>C-NMR spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid **ZU-590**.



Figure S15. HSQC spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid ZU-590.



Figure S16. COSY spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid **ZU-590**.



Figure S17. HMBC spectrum (MeOD-d4) of sulfonosphingolipid ZU-590.



Figure S18. <sup>1</sup>H-NMR spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid **ZU-602**.



Figure S19. <sup>13</sup>C-NMR spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid ZU-602.



Figure S20. HSQC spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid **ZU-602**.



Figure S21. COSY spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid **ZU-602**.



Figure S22. HMBC spectrum (MeOD-d<sub>4</sub>) of sulfonosphingolipid ZU-602.



**Figure** S23. <sup>1</sup>H-NMR spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid **ZU-616** (**flavocristamide A**).



Figure S24. <sup>13</sup>C-NMR spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid ZU-616 (flavocristamide A).



Figure S25. HSQC spectrum (MeOD-d<sub>4</sub>) of sulfonosphingolipid ZU-616 (flavocristamide A).



Figure S26. COSY spectrum (MeOD-*d*<sub>4</sub>) of sulfonosphingolipid **ZU-616** (flavocristamide A).



Figure S27. HMBC spectrum (MeOD-d4) of sulfonosphingolipid ZU-616 (flavocristamide A).



#### Sulfonosphingolipids in DMSO-d<sub>6</sub> 6.2

Figure S28. <sup>1</sup>H-NMR spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid **ZU-572**.

Supporting Information

**—** 



Figure S29. <sup>13</sup>C-NMR spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid ZU-572.



Figure S30. HSQC spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid ZU-572.



Figure S31. COSY spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid ZU-572.



Figure S32. HMBC spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid ZU-572.



Figure S33. <sup>1</sup>H-NMR spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid ZU-590.



Figure S34. <sup>13</sup>C-NMR spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid ZU-590.



Figure S35. HSQC spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid ZU-590.



Figure S36. COSY spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid ZU-590.



Figure S37. HMBC spectrum (DMSO-d<sub>6</sub>) of sulfonosphingolipid ZU-590.

![](_page_43_Figure_0.jpeg)

Figure S38. <sup>1</sup>H-NMR spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid **ZU-602**.

![](_page_44_Figure_0.jpeg)

Figure S39. <sup>13</sup>C-NMR spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid **ZU-602**.

![](_page_45_Figure_0.jpeg)

Figure S40. HSQC spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid **ZU-602**.

![](_page_46_Figure_0.jpeg)

Figure S41. COSY spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid **ZU-602**.

![](_page_47_Figure_0.jpeg)

Figure S42. HMBC spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid **ZU-602**.

![](_page_48_Figure_0.jpeg)

Figure S43. <sup>1</sup>H-NMR spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid **ZU-616 (flavocristamide A)**.

![](_page_49_Figure_0.jpeg)

Figure S44. <sup>13</sup>C-NMR spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid **ZU-616** (flavocristamide A).

![](_page_50_Figure_0.jpeg)

Figure S45. HSQC spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid ZU-616 (flavocristamide A).

![](_page_51_Figure_0.jpeg)

Figure S46. COSY spectrum (DMSO-d<sub>6</sub>) of sulfonosphingolipid ZU-616 (flavocristamide A).

![](_page_52_Figure_0.jpeg)

Figure S47. HMBC spectrum (DMSO-*d*<sub>6</sub>) of sulfonosphingolipid ZU-616 (flavocristamide A).