Supplementary information

Identification of acyl-homoserine lactone autoinducers produced by the nitrite-oxidizing bacterium *Nitrobacter winogradskyi*.

Brett L. Mellbye^{1#}, Peter J. Bottomley², Luis A. Sayavedra-Soto^{1#}.

Department of Botany and Plant Pathology, Oregon State University, Corvallis, Oregon, USA¹; Department of Microbiology, Oregon State University, Corvallis, Oregon, USA²

*Address correspondence to Brett L. Mellbye, mellbyeb@science.oregonstate.edu, and Luis A. Sayavedra-Soto, sayavedl@science.oregonstate.edu.

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SUPPLEMENTAL MATERIALS AND METHODS

Fatty acid analysis. Stationary-phase culture samples were submitted to Microbial ID, Inc. (MIS, Microbial ID, Inc. (MIDI), Newark, Delaware, USA) for fatty acid methyl ester (FAME) generation, extraction, and identification by gas chromatography as described (1). FAME profiles were generated by Microbial ID, Inc. (MIS, Microbial ID, Inc. (MIDI), Newark, Delaware, USA) using Sherlock 6.1 as described (1).

Table S1. FAME profile of *Nitrobacter winogradskyi*.

RT^a	ECL^b	Peak Name	Percent	Comment1 ^c	Comment2 ^d
0.7395	6.6663	SOLVENT			
1.6451	12.0005	12:0	0.18	0.001	
2.7604	15.8371	Sum In Feature 3	0.38	-0.003	16:1 w7c/ 16:1 w6c
2.8108	15.9981	16:0	5.89	-0.002	
3.0667	16.8144	17:1 w8c	0.35	-0.001	
3.0880	16.8825	17:1 w6c	0.21	0.002	
3.1251	17.0008	17:0	0.20	0.001	
3.3886	17.8468	Sum In Feature 8	90.98	-0.001	18:1 w7c
3.4161	17.9349	18:1 w5c	0.23	-0.002	
3.4365	18.0006	18:0	1.40	0.001	
3.5883	18.4994	19:1 iso I	0.19	0.001	
3.6864	18.8219				
		Summed Feature 3	0.38	16:1 w7c/ 16:1 w6c	16:1 w6c/ 16:1 w7c
		Summed Feature 8	90.98	18:1 w7c	18:1 w6c

^aRetention time. ^bEquivalent chain lengths values. ^cECL deviation or unresolved peak(s) in summed features. ^dBest guess for unresolved peak(s).

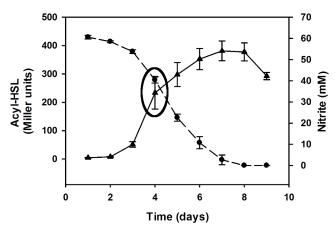


Figure S1. Acyl-HSL detection and NO₂⁻ concentration during batch culture. Total acyl-HSL level (triangles, solid line, reported as Miller units; left *y*-axis) was measured by bioassay after extraction from batch cultures. NO₂⁻ concentration (circles, dashed line, right *y*-axis) was measured by Griess assay. The circled data points highlight a statistically significant ($p \le 0.04$ by two-tailed *t*-test) spike in acyl-HSL detection (compared to previous days) that correlates with a transition to a slow growth rate ($T_d \approx 0.67$ days changes to 1.95 days). Values are the mean of three independent biological replicates. Error bars indicate the standard deviation of the mean.

SUPPLEMENTAL REFERENCES

1. **Assih EA, Ouattara AS, Thierry S, Cayol JL, Labat M, Macarie H.** 2002. *Stenotrophomonas acidaminiphila* sp. nov., a strictly aerobic bacterium isolated from an upflow anaerobic sludge blanket (UASB) reactor. Int J Syst Evol Microbiol **52:**559-568.