SUPPLEMENTAL MATERIAL

belonging to the manuscript

Response and defense of Streptomycetaceae to the plant stress hormone jasmonic acid

Anne van der Meij^{1,*}, Somayah S. M. A. Elsayed^{1,*}, Chao Du¹, Joost Willemse, Thomas M. Wood¹, Nathaniel I. Martin¹, Jos M. Raaijmakers² and Gilles P. van Wezel^{1,#}

¹ Molecular Biotechnology, Institute of Biology, Leiden University, Sylviusweg 72, 2333 BE, Leiden, The Netherlands.

² Department of Microbial Ecology, Netherlands Institute of Ecology (NIOO-KNAW), Wageningen, The Netherlands.

* these authors contributed equally. # Author for correspondence: tel. +31 71 5274310; email: g.wezel@biology.leidenuniv.nl.

Supplemental Tables

Position	$oldsymbol{\delta}_{C}$, type	$\delta_{ extsf{H}}$, mult. (J in Hz)
1	169.7 <i>,</i> C	
2	40.5, CH ₂	a: 2.41, m
		b: 2.15, m
3	38.0 <i>,</i> CH	2.15 <i>,</i> m
4	26.4, CH ₂	a: 2.00, m
		b: 1.46 <i>,</i> m
5	37.3, CH ₂	a: 2.22, m
		b: 2.00, m
6	219.1 <i>,</i> C	
7	53.5 <i>,</i> CH	1.89, dd (9.9, 5.0)
8	24.8, CH ₂	2.23 <i>,</i> m
9	125.8 <i>,</i> CH	5.23, dt (10.7, 7.4)
10	132.8 <i>,</i> CH	5.36, dt (10.7, 7.4)
11	20.1, CH ₂	2.01, m
12	14.0, CH₃	0.90, t (7.5)
1′	53.8 <i>,</i> CH	3.82 <i>,</i> m
2′	29.2, CH ₂	a: 1.84, m
		b: 1.70, m
3′	32.2 <i>,</i> CH	2.01, m
4′	174.7 <i>,</i> C	
5′	173.4 <i>,</i> C	
1'-NH		7.33, d (7.0)
4'-NH2		a: 7.44, s
		b: 6.60, s

Table S1: 1H and 13C NMR data of 1 in DMSO-d6 at 298 K

Table S2: Streptomycetaceae used in this study

reference or source
John Innes Centre John Innes Centre
(Hoskisson & Van Wezel, 2019) DSMZ culture collection
ATCC culture collection
ATCC culture collection
Reindert Nijland (WUR* strain collection)
Reindert Nijland (WUR* strain collection) Reindert Nijland (WUR*
strain collection)
Leiden MBT strain collection
van der Meij et al., 2018
van der Meij et al., 2018
Wietse de Boer (NIOO**)
Wietse de Boer (NIOO**)
Wietse de Boer (NIOO**)

*WUR: Wageningen University & Research **NIOO: Netherlands Institute of Ecology

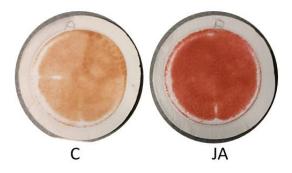


Figure S1: Pigmentation of liquid-grown mycelium of *S. coelicolor* **grown with JA.** Cultures were grown in for 24 h in liquid MM with JA (JA) or without (C) and filtered over a glass microfiber. JA induces production of red-pigmented prodigionines in the mycelia.

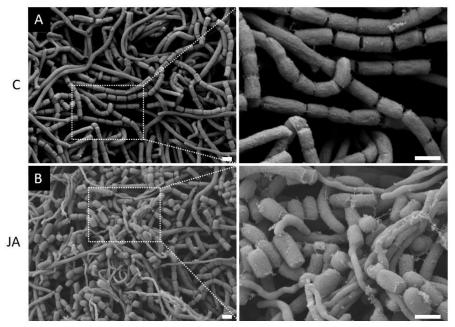


Figure S2: Scanning electron micrographs of *S. coelicolor* M145 grown in the presence of JA. (A) Aerial hyphae and spores were produced after 3 days of growth on MM. (B) On MM supplemented with 0.5 mM JA spores were produced already after 2 days of growth. Higher magnification shows JA-induced premature germination of the spore chains. Scalebars: 1 μ m.

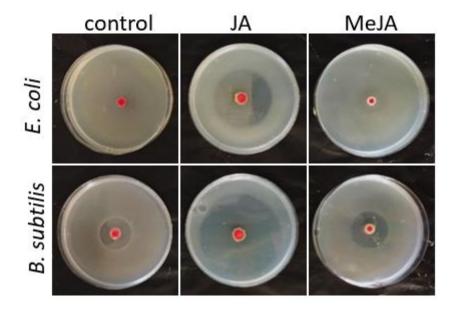


Figure S3: Antimicrobial activity of *S. roseifaciens* is strongly increased when exposed to JA.

S. roseifaciens has weak and moderate activity against *E. coli* ASD19 and *B. subtilis* 168 when grown on MM. When the medium was supplemented with 0.5 mM (0.01%) JA the antimicrobial activity strongly increased, whereas there was no visible effect of 0.5 mM (0.01%) MeJA.

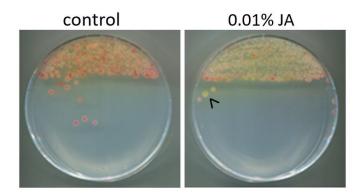


Figure S4: JA alters pigmentation of *S. roseifaciens*. When grown on MM the bacterium is red. When the medium is supplemented with JA the dominant color of the biomass is yellow, due to the absence of the red pigment.

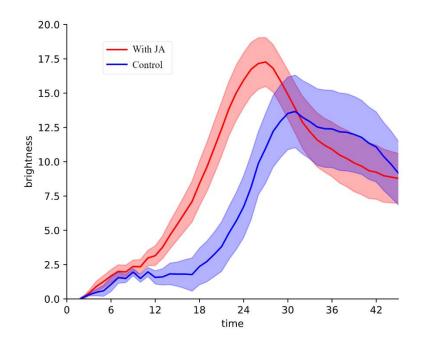


Figure S5: Growth of *S. roseifaciens* **on MM with or without JA.** Plotted is the brightness of the mycelium as a marker of growth of *S. roseifaciens* spots grown on minimal agar media measured from scanner-generated images (see materials and methods). The red line represents *S. roseifaciens* spots grown on medium supplemented with 0.5 mM JA and the blue line represents *S. roseifaciens* grown under control conditions. The solid line in the middle is the average value of 20 spots. The filled area shows the standard error of mean for each time point.

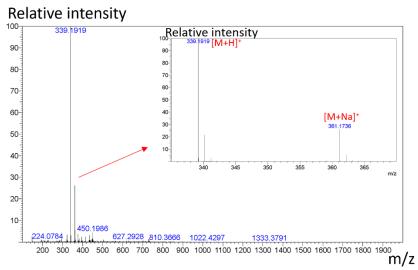


Figure S6: (+)-HRESIMS spectrum of 1

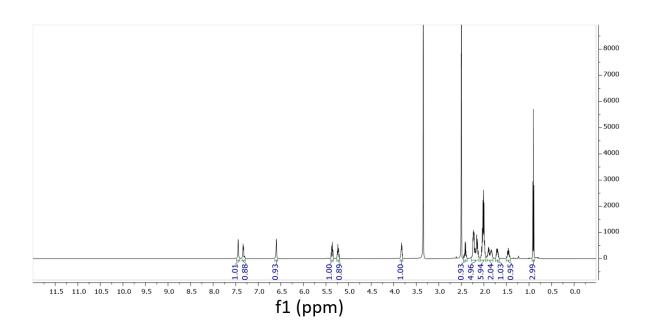


Figure S7: ¹H NMR spectrum of 1 (major isomer) in DMSO-*d6*

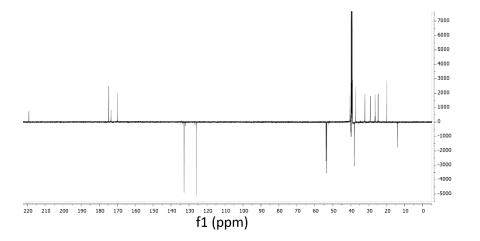


Figure S8: ¹³C APT spectrum of 1 (major isomer) in DMSO-*d6*.

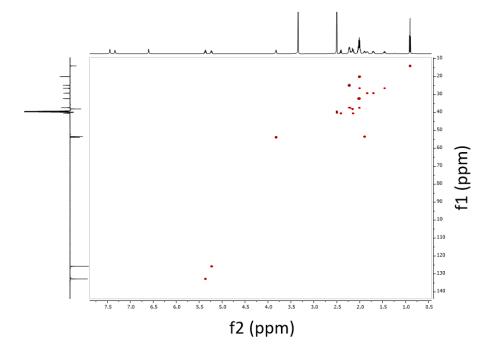


Figure S9: HSQC spectrum of 1 (major isomer) in DMSO-d6.

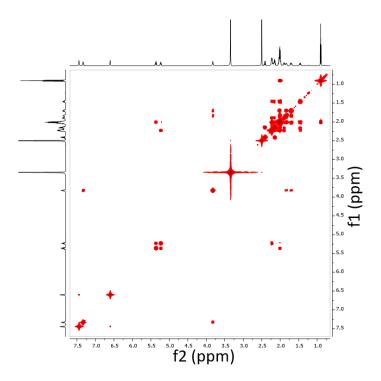


Figure S10: ¹H-¹H COSY spectrum of 1 (major isomer) in DMSO-*d6*.

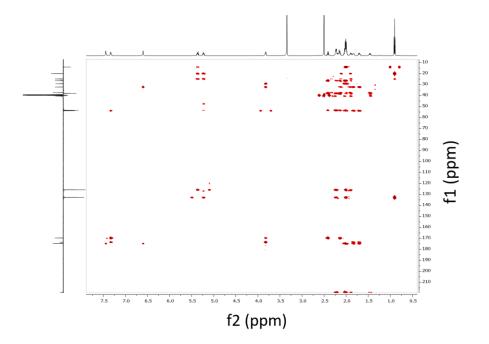


Figure S11: HMBC spectrum of 1 (major isomer) in DMSO-d6.

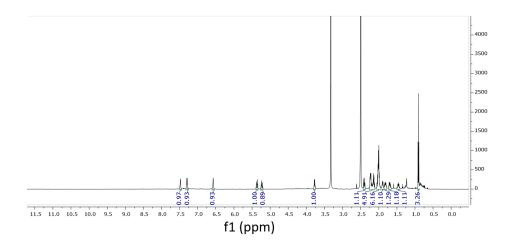


Figure S12: ¹H NMR spectrum of 1 (minor isomer) in DMSO-d6.

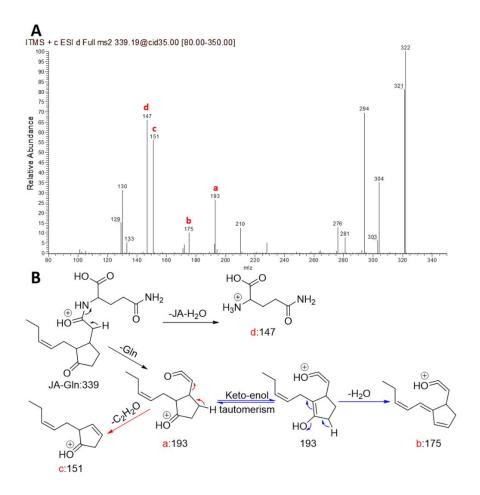


Figure S13: A: MS² spectrum of JA-Gln. The most characteristic fragments a – d are marked. **B: The likely fragmentation pathway of JA-Gln.** Fragmentation resulted in the observed fragments a – d, which are labelled using their nominal masses. The position of the positive charge is arbitrary.

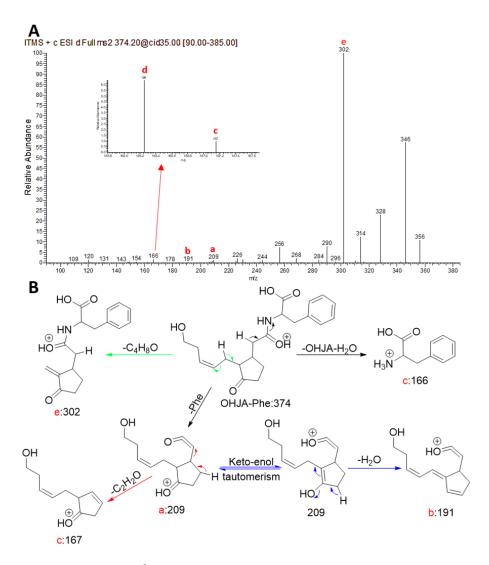


Figure S14: A: MS² spectrum of OHJA-Phe. The most characteristic fragments a – e are marked. **B: The likely fragmentation pathway of OHJA-Phe.** Fragmentation resulted in the observed fragments a – e, which are labelled using their nominal masses. The position of the positive charge is arbitrary.

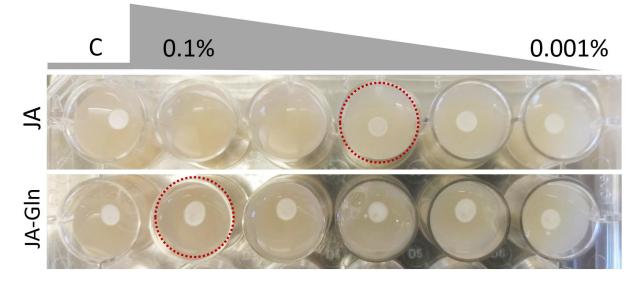


Figure S15: Glutamine-conjugated JA is not toxic to *Streptacidiphilus* **P18-A5a.** Shown are spots of *Streptacidiphilus* P18-A5a on ½ PDA with decreasing concentrations of JA or JA-Gln after 5 days of growth. From left to right, every well represents a 3-fold concentration decrease, resulting in 0.1%, 0.03%, 0.01%, 0.003% and 0.001% w/v addition of the compound indicated on the left (JA-Gln or JA). P18-A5a grows up to 0.01% (0.5mM) JA whereas it still grows 0.1% (3mM) of the glutamine conjugated version of JA (JA-Gln). C: control.

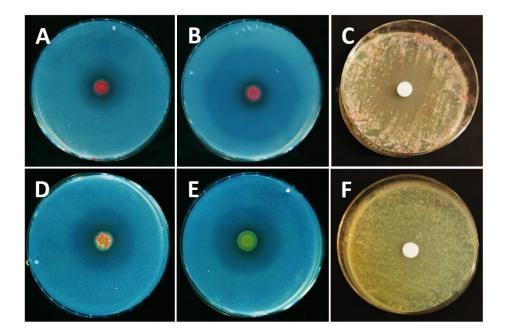


Figure S16. JA-conditioned *S. roseifaciens* is desensitized to JA. *S. roseifaciens* grown on MM with JA (B) shows increased antibiotic activity against indicator strain *E. coli* as compared to the control without JA (A). Conversely, *S. roseifaciens* that has been pre-grown for 8 generations on MM with JA (D) shows similar antibiotic activity as the control (E). JA (1 µg) spotted on a filter disk inhibits growth of the parent *S. roseifaciens* (C) whereas this growth inhibition is not observed for JA-conditioned *S. roseifaciens* (F). Plates were grown on MM agar for 5 days.