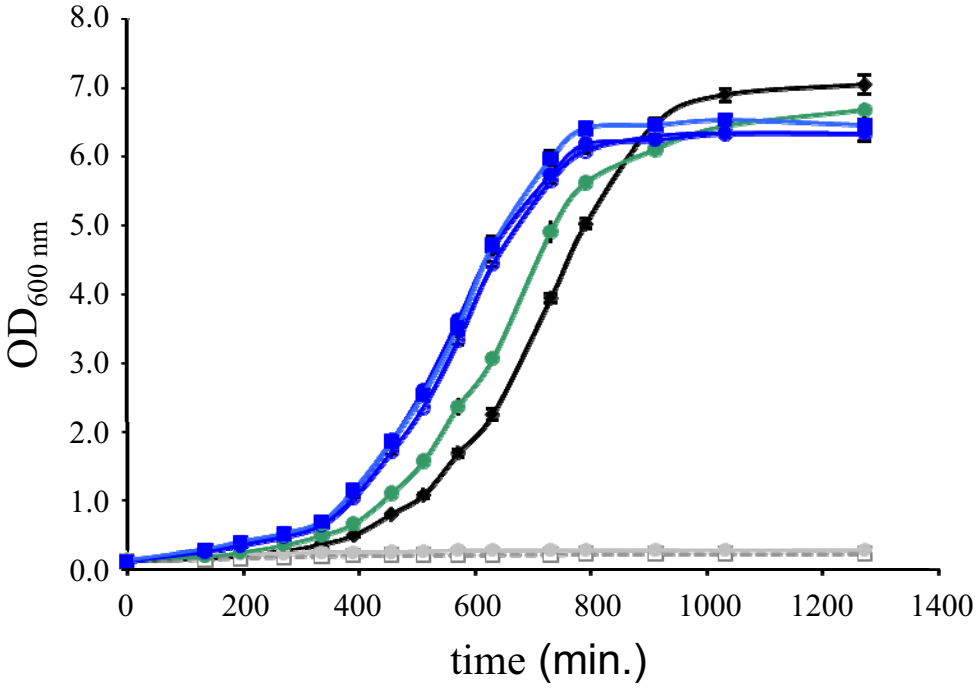
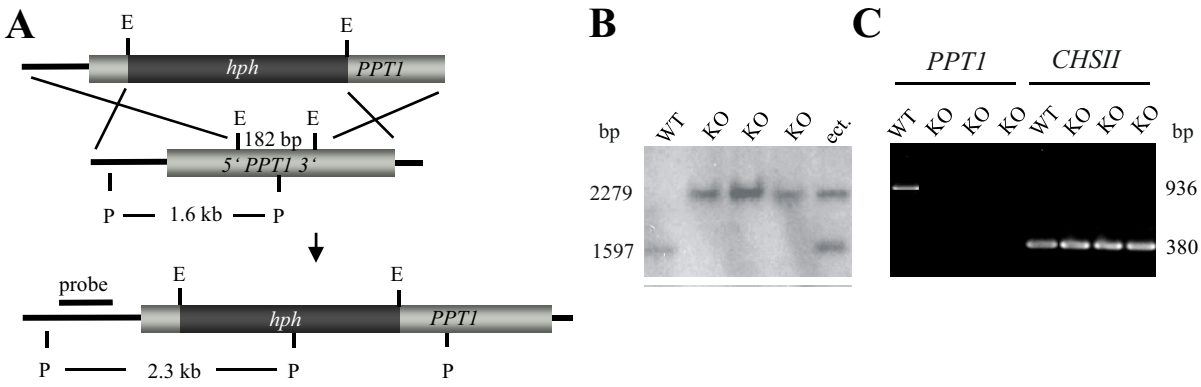
Supplemental Data. Horbach et al. (2009). Sfp-type 4'-phosphopantetheinyl transferase is indispensable for fungal pathogenicity.

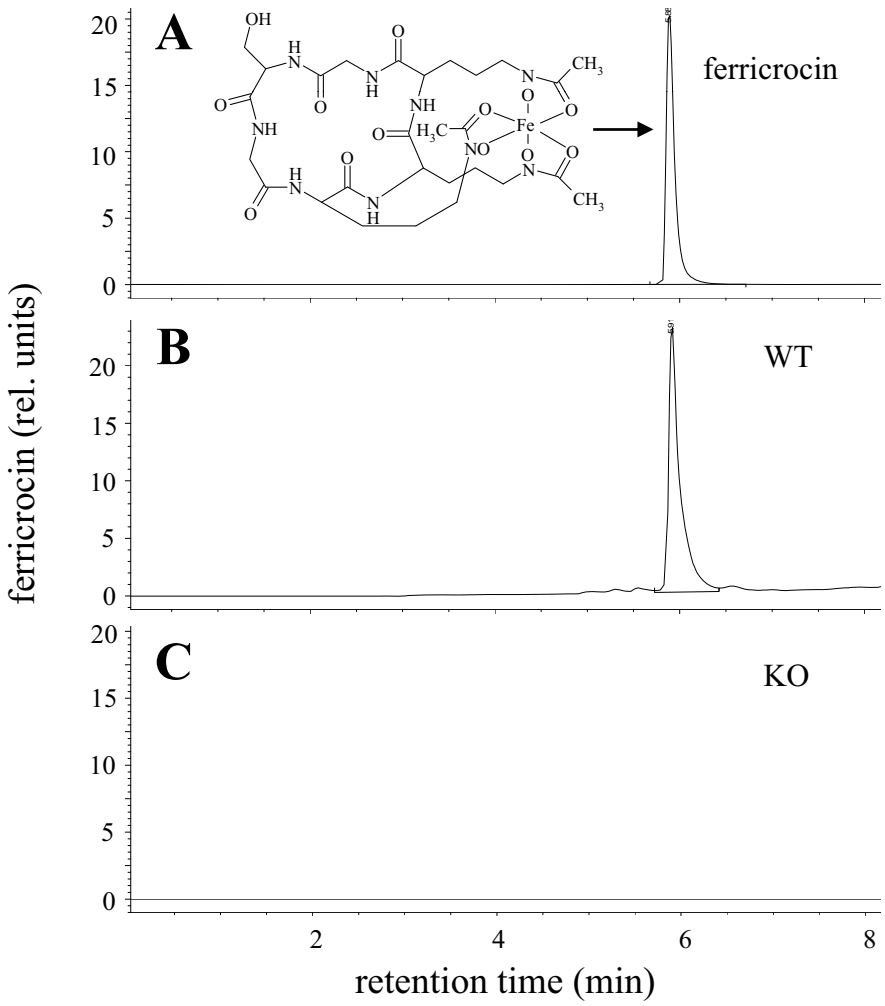


Supplemental Figure 1. Complementation of growth of the *S. cerevisiae* Δ*lys5* mutant by the PPTase genes of *A. nidulans* or *C. graminicola*. The yeast WT strain BY4741 (black line) and Δ*lys5* transformants expressing the PPTase genes *cfwA* of *A. nidulans* (green line), or *PPT1* of *C. graminicola* (blue lines), were able to grow and showed similar growth rates in liquid SD minimal medium lacking lysine. The lysine auxotrophic Δ*lys5* strain (grey line, open symbols) and the Δ*lys5* strain transformed with the empty yeast expression vector pAG300 (grey line, solid symbols) were unable to grow.



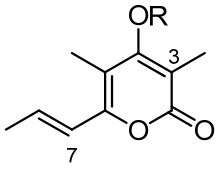
Supplemental Figure 2. Generation of Δ*ppt1* strains of *C. graminicola* by homologous recombination. (A) Scheme of targeted mutagenesis of *PPT1* by homologous recombination. A 182 bp *Eco*81I (E) fragment of the gene was replaced by a 2.6 kb fragment carrying the *E. coli* hygromycin phosphotransferase gene (*hph*) (not to scale). (B) Southern blot analysis performed with *Pst*I-digested genomic DNA from WT, ectopic (ect.), and Δ*ppt1* (KO) strains showed that the 1597 bp WT band had been replaced by the 2279 bp fragment in all independent KO strains. Transformants with ectopically integrated KO vector showed the 2279 bp band in addition to the 1597 bp WT band. (C) RT-PCR analysis performed with *PPT1*-specific primers confirmed gene inactivation at the transcript level; primers corresponding to transcripts of the constitutively expressed *CHSII* gene were used as a control. The 936 bp *PPT1* fragment was amplified only when RNA from the WT strain, but not when RNA from KO strains was used. The 380 bp *ChsII* fragment was amplified from RNA from WT and KO strains. RT-PCR products were observed after staining with EtBr.

Supplemental Figure 3. Complementation of growth of the Δ*ppt1* mutant of *C. graminicola* by the siderophore desferri-ferrichrome. On synthetic complete medium supplemented with the iron scavenger BPS and the siderophore desferri-ferrichrome not only the WT isolate CgM2 and the ectopic transformant (ect.), but also the Δ*ppt1* mutant (KO) is able to grow.



Supplemental Figure 4. Detection of the intracellular storage siderophore ferricrocin in hyphal extracts of *C. graminicola*. The HPLC profile of ferricrocin is shown in A. In contrast to the WT strain (B), the Δ*ppt1* strain (C) is unable to produce ferricrocin.

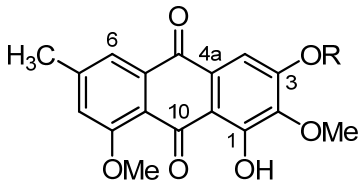
Supplemental Figure 5: Characterization of identified compounds



R = H: Colletopyrone B
R = Me: Colletopyrone C

Colletopyrone B. Beige solid, mp 188–189 °C; UV (MeCN) λ_{max} (log ε) = 221 (4.62), 325 (4.05) nm; IR (KBr) ν = 2933, 1661, 1544, 1404, 1321, 751 cm^{-1}; APCI-MS (pos.) *m/z* (%) = 181.1 (100, $[M + H]^+$); ESI-HRMS *m/z* = 203.0690 ($C_{10}H_{12}O_3 + Na^+$ requires 203.0684).

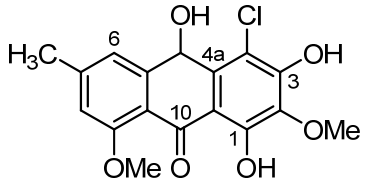
Colletopyrone C. Colourless crystals, mp 81–83 °C; UV (MeCN) λ_{max} (log ε) = 223 (4.80), 329 (4.40) nm; IR (KBr) ν = 2916, 1700, 1553, 1449, 1221, 1065, 761 cm^{-1}; APCI-MS (pos.) *m/z* (%) = 195.1 (100, $[M + H]^+$); ESI-HRMS *m/z* = 217.0846 ($C_{11}H_{14}O_3 + Na^+$ requires 217.0841).



R = H: Colletoquinone A
R = Me: Colletoquinone B

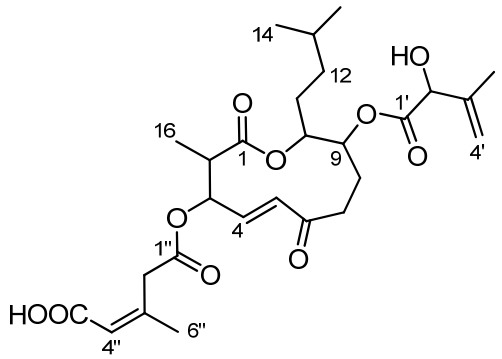
Colletoquinone A. Orange amorphous solid; UV (MeCN) λ_{max} (log ε) = 223 (4.41), 275 (4.30), 415 (3.80) nm; IR (KBr) ν = 3357, 1624, 1391, 1264, 1162, 969 cm^{-1}; APCI-MS (pos.) *m/z* (%) = 315.0 (100, $[M + H]^+$), APCI-MS (neg.) *m/z* (%) = 313.0 (100, $[M - H]^-$); FAB-HRMS *m/z* = 315.0876 ($C_{17}H_{15}O_6 + H^+$ requires 315.0869).

Colletoquinone B. Ochre-coloured solid, mp 195–196 °C (Sugawara and Strobel, 1987) (199–201 °C, NMR spectra show impurities); UV (MeCN) λ_{max} (log ε) = 214 (4.24), 274 (4.17), 416 (3.66) nm; IR (KBr) ν = 3436, 2938, 1632, 1419, 1278, 1135, 970 cm^{-1}; APCI-MS (pos.) *m/z* (%) = 329.1 (100, $[M + H]^+$), APCI-MS (neg.) *m/z* (%) = 327.1 (100, $[M - H]^-$); FAB-HRMS *m/z* = 329.1019 ($C_{18}H_{17}O_6 + H^+$ requires 329.1025).



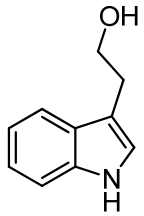
Colletoanthrone A

Colletoanthrone A. Yellow-brown powder, mp 141–145 °C (dec.); $[\alpha]_D^{26}$ = +4.0 (*c* 0.92, DMSO-d_6); UV (MeCN) λ_{max} (log ε) = 225 (sh, 4.03), 260 (3.62), 271 (3.60), 355 (3.75) nm; IR (KBr) ν = 3429, 1616, 1390, 1287, 1125, 975 cm^{-1}; APCI-MS (pos.) *m/z* (%) = 351.0 (100, $[M(^{35}Cl) + H]^+$), 353.0 (30, $[M(^{37}Cl) + H]^+$); FAB-HRMS *m/z* = 349.0488 ($[C_{17}H_{15}ClO_6 - H]^+$ requires 349.0479).



Colletolactone A

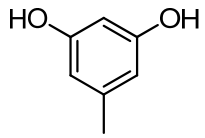
Colletolactone A. Salmon-coloured powder, mp 117–119 °C; $[\alpha]_D^{26}$ = +54.9 (*c* 0.25, $CDCl_3$); UV (MeCN) λ_{max} (log ε) = 214 (4.25) nm; IR (KBr) ν = 3436, 2957, 1737, 1166, 992 cm^{-1}; APCI-MS (neg.) *m/z* (%) = 521.1 (100, $[M - H]^-$); FAB-HRMS *m/z* = 523.2527 ($C_{27}H_{38}O_{10}$ + H^+ requires 523.2543).



Tryptophol

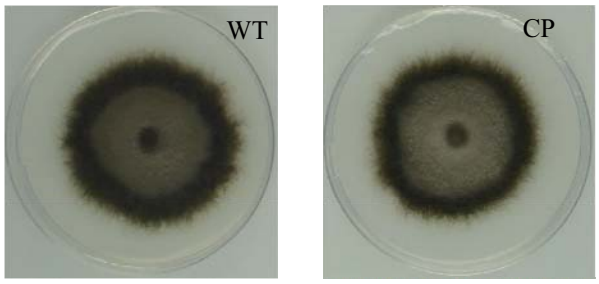
Tryptophol. Orange oil; ^1H NMR (400 MHz, $CDCl_3$) δ = 8.05 (br s, 1H, NH), 7.63 (dq, *J* = 7.9, 0.9 Hz, 1H, H-4), 7.38 (dt, *J* = 8.1, 0.9 Hz, 1H, H-7), 7.22 (ddd, *J* = 8.1, 7.0, 0.9 Hz, 1H, H-6), 7.14 (ddd, *J* = 7.9, 7.0, 0.9 Hz, 1H, H-5), 7.10 (dt, *J* = 2.4, 0.8 Hz, 1H, H-2), 3.92 (t, *J* = 6.3 Hz, 2H, H-1'), 3.05 (td, *J* = 6.3, 0.8 Hz, H-2') ppm; APCI-MS (neg.) *m/z* (%) = 160.0

(100, $[M - H]^-$); spectroscopic data is in accordance with data given in the literature (Roberge and Brassard, 1981).

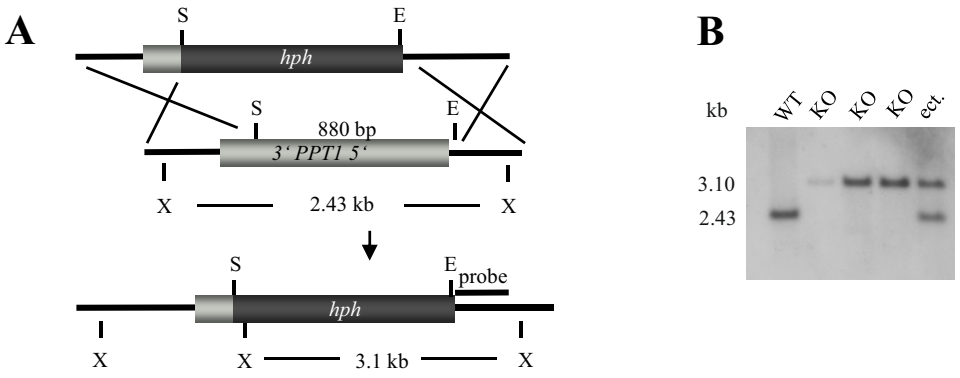


Orcinol

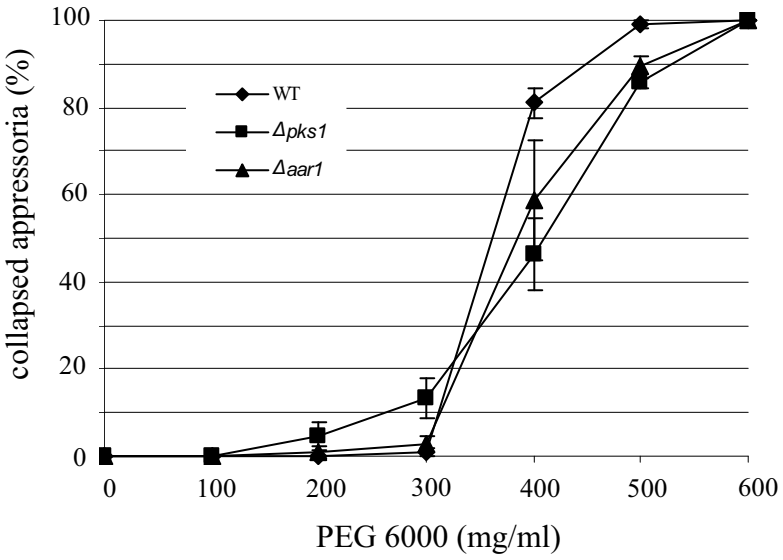
Orcinol. Tan-coloured oil; ^{1}H NMR (400 MHz, MeCN) δ = 6.14 (dq, J = 2.2, 0.8, 2H, H-2, H-6), 6.06 (tq, J = 2.2, 0.6 Hz, 1H, H-4), 2.17 (td, J = 0.8, 0.6 Hz, 3H, CH_3) ppm. APCI-MS (pos.) *m/z* (%) = 125.1 (100, $[M + H]^+$); APCI-MS (neg.) *m/z* (%) = 123.0 (100, $[M - H]^-$); spectroscopic data is in accordance with data given in the literature (Degl'Innocenti et al., 1991).



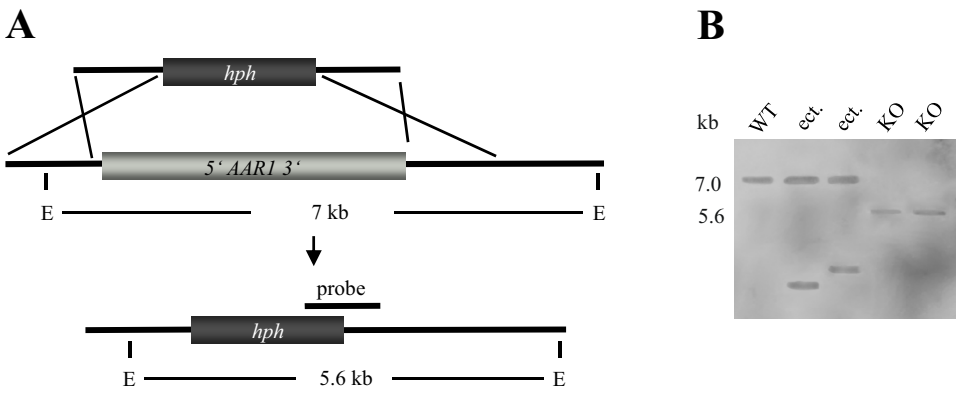
Supplemental Figure 6. Complementation of the Δ*ppt1* strains with the *PPT1* gene under control of the native promoter fully rescues growth defects on minimal medium lacking lysine. WT, wild-type strain CgM2; CP, Δ*ppt1* strain complemented with the *PPT1* gene.



Supplemental Figure 7. Generation of Δ*ppt1* strains of *M. oryzae* by homologous recombination. (A) Scheme of targeted mutagenesis of *PPT1* by homologous recombination. A 880 bp *Eco81*I/*Stu*I (E, S) fragment of the gene was replaced by a 2.6 kb fragment carrying the *E. coli* hygromycin phosphotransferase gene (*hph*) (not to scale). (B) Southern blot analyses performed with *Pst*I-digested genomic DNA from WT, an ectopic transformant (ect.), and Δ*ppt1* (KO) strains showed that the 2.43 kb WT band had been replaced by the 3.1 kb fragment in all independent KO strains. The transformant with ectopically integrated KO vector showed the 3.1 kb band in addition to the 2.43 kb WT band. This is former Fig. S4.



Supplemental Figure 8. Determination of appressorial turgor pressure by incipient-cytorrhizis with PEG 6000. One-hundred appressoria were counted 10 min. after sapplication of the PEG solution. Three replicates were counted for each PEG concentration. Appressoria of Δ*ppt1* and Δ*aar1* showed incipient-cytorrhizis at slightly higher PEG 6000 concentrations than those of the wild-type strain (WT).



Supplemental Figure 9. Generation of Δ*aar1* strains of *C. graminicola* by homologous recombination. (A) Scheme of targeted mutagenesis of *AAR1* by homologous recombination. The complete putative ORF of the gene was replaced by a 2 kb fragment carrying the *E. coli* hygromycin phosphotransferase gene (*hph*) (not to scale). (B) Southern blot analyses performed with *Eco81*I-digested genomic DNA from WT, ectopic transformants (ect.), and Δ*aar1* (KO) strains showed that the 7 kb WT band had been replaced by the 5.6 kb fragment in all independent KO strains. Transformants with ectopically integrated KO vector showed the 7 kb WT band in addition to a second band which originates from the ectopically integrated construct.

Supplemental Table 1 ^{1}H (400 MHz) and ^{13}C (101 MHz) NMR data of Colletopyrone B (CD_3OD) and Colletopyrone C ($CDCl_3$). Coupling constants (*J*) are given in Hz. ^{13}C multiplicities were determined indirectly by HSQC.

	Colletopyrone B		Colletopyrone C	
pos.	δ_H	δ_C	δ_H	δ_C
2	—	167.7 (s)	—	165.3 (s)
3	—	99.9 (s)	—	110.8 (s)
4	—	168.1 (s)	—	168.4 (s)
5	—	108.7 (s)	—	108.7 (s)
6	—	153.6 (s)	—	152.8 (s)
7	6.43 (dq, 15.4, 1.5)	121.4 (d)	6.24 (dq, 15.4, 1.7)	120.2 (d)
8	6.61 (dq, 15.4, 6.9)	134.2 (d)	6.67 (dq, 15.4, 7.0)	133.7 (d)
9	1.93 (dd, 6.9, 1.5)	18.6 (q)	1.91 (dd, 7.0, 1.7)	18.8 (q)
3-CH_3	1.93 (s)	9.0 (q)	2.04 (s)	10.5 (q)
5-CH_3	2.01 (s)	9.4 (q)	1.98 (s)	9.6 (q)
4-OCH_3	—	—	3.79 (s)	60.4 (q)

Supplemental Table 2: ^{1}H (400 MHz) and ^{13}C (101 MHz) NMR data of Colletoquinone A (DMSO-d_6 + 30% CD_3OD) and Colletoquinone B ($CDCl_3$). Coupling constants (*J*) are given in Hz. ^{13}C multiplicities were determined indirectly by HSQC.

	Colletoquinone A		Colletoquinone B	
pos.	δ_H	δ_C	δ_H	δ_C
1	—	157.3 (s)	—	156.9 (s)
2	—	140.6 (s)	—	142.2 (s)
3	—	156.6 (s)	—	157.7 (s)
4	7.15 (s)	107.9 (d)	7.40 (br s)	103.1 (d)
4a	—	128.9 (s)	—	128.5 (s)
5	—	182.4 (s)	—	182.7 (s)
5a	—	135.4 (s)	—	135.5 (s)
6	7.58 (br dd, 1.5, 0.6)	120.6 (d)	7.77 (br dd, 1.6, 0.7)	121.2 (d)
7	—	147.8 (s)	—	147.3 (s)
8	7.34 (br s)	119.9 (d)	7.14 (br s)	118.8 (d)
9	—	161.4 (s)	—	161.1 (s)
9a	—	118.4 (s)	—	118.6 (s)
10	—	187.7 (s)	—	188.2 (s)
10a	—	111.9 (s)	—	113.3 (s)
OCH_3-2	3.84 (s)	60.5 (q)	4.01 (s)	61.1 (q)
OCH_3-3	—	—	4.03 (s)	56.6 (q)
CH_3-7	2.43 (br s)	21.9 (q)	2.51 (br s)	22.5 (q)
OCH_3-9	3.92 (s)	56.7 (q)	4.06 (s)	56.8 (q)
OH-1	13.46 (s)	—	13.25 (s)	—

Supplemental Table 3: ^{1}H (400 MHz) and ^{13}C (101 MHz) NMR data of Colletoanthrone A (DMSO-d_6). Coupling constants (J) are given in Hz. ^{13}C multiplicities were determined indirectly by HSQC.

pos.	δ_H	δ_C
1	—	155.2 (s)
2	—	135.1 (s)
3	—	152.5 (s)
4	—	111.9 (s)
4a	—	135.3 (s)
5	5.57 (br s)	63.5 (d)
5a	—	145.5 (s)
6	7.06 (s)	122.9 (d)
7	—	146.5 (s)
8	7.01 (s)	113.0 (d)
9	—	160.4 (s)
9a	—	115.6 (s)
10	—	187.7 (s)
10a	—	109.9 (s)
OCH_3-2	3.78 (s)	60.3 (q)
CH_3-7	2.40 (s)	21.7 (q)
OCH_3-9	3.87 (s)	56.0 (q)
OH-1	13.61 (s)	—
OH-3	10.78 (s)	—
OH-5	5.92 (br d, 6.2)	—

Supplemental Table 4: ^{1}H (400 MHz) and ^{13}C (101 MHz) NMR data of Colletolactone A ($CDCl_3$). Coupling constants (*J*) are given in Hz. ^{13}C multiplicities were determined indirectly by HSQC.

pos.	δ_H	δ_C
1	—	172.4 (s)
2	2.86 (dq, 9.6, 6.7)	45.9 (d)
3	5.17 (ddd, 9.6, 7.2, 0.8)	76.2 (d)
4	6.33 (dd, 16.3, 7.2)	139.0 (d)
5	6.37 (d, 16.3)	131.8 (d)
6	—	202.2 (s)
7	2.60 (ddd, 14.6, 11.0, 4.3)	39.3 (t)
	2.47 (ddd, 14.6, 6.4, 3.9)	
8	2.16 (m)	29.0 (t)
	2.12 (m)	
9	4.65 (ddd, 7.1, 6.1, 3.7)	74.8 (d)
10	5.05 (td, 8.6, 3.7)	75.5 (d)
11	1.52 (m)	28.3 (t)
	1.34 (m)	
12	1.08 (m)	34.0 (t)
13	1.47 (m)	27.9 (d)
14	0.84 (d, 6.6)	22.4 (q)
15	0.84 (d, 6.6)	22.6 (q)
16	1.26 (d, 6.7)	14.0 (q)
1'	—	172.7 (s)
2'	4.56 (br s)	75.3 (d)
3'	—	141.5 (s)
4'	5.13 (br s)	116.2 (t)
	5.05 (br s)	
5'	1.72 (s)	17.6 (q)
1"	—	169.1 (s)
2"	3.79 (d, 16.2)	38.8 (t)
	3.64 (d, 16.2)	
3"	—	153.1 (s)
4"	5.91 (br q, 1.6)	118.8 (d)
5"	—	169.2 (s)
6"	2.02 (br d, 1.6)	26.5 (q)

Supplemental References

Sugawara, F., and Strobel, G.A. (1987). Tryptophol, a phytotoxin produced by Drechslera nodulosum. Phytochemistry **26,** 1349-1351.

Roberge, G., and Brassard, P. (1981). Reactions of ketene acetals. 13. Synthesis of contiguously trihydroxylated naphtho- and anthraquinones. J. Org. Chem. **46,** 4161-4166.

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