



Supporting Information

for

Volatile emission and biosynthesis in endophytic fungi colonizing black poplar leaves

Christin Walther, Pamela Baumann, Katrin Luck, Beate Rothe, Peter H. W. Biedermann, Jonathan Gershenzon, Tobias G. Köllner and Sybille B. Unsicker

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Sequences of isolated endophytic fungi and identification according to NCBI database, primer used in this study, representative total ion chromatograms of single endophytic volatile blend, mass spectra of unknown volatile organic compounds, and BUSCO analysis of *Cladosporium* sp. de novo assembly

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Figure S2: Mass spectra of unknown volatile organic compounds

Figure S3: BUSCO analysis of *Cladosporium* sp. de novo assembly

Table S1: Endophytes were identified to genus level via sequencing of ribosomal DNA (ITS1F/ ITS4). The obtained sequences were compared to the NCBI sequence database and the identity (%) of best hits with their accession number is given in the main document.

Species	Sequence
<i>Alternaria infectoria</i>	TGCTTTTTGCGTACTTCTTGTTTTCTGGGTGGGCTCGCCCGCCCTCAGGACCAAC CACAAACCTTTTGCAATAGCAATCAGCGTCAGTAACAACGTAATTAATTACAACCTT TCAACAACGGATCTCTTGTTCTGGCATCGATGAAGAACGCAGCGAAATGCGATA CGTAGTGTGAATTGCAGAATTCAGTGAATCATCGAATCTTTGAACGCACATTGCG CCCTTTGGTATTCCAAAGGGCATGCCTGTTGAGCGTCATTTGTACCCTCAAGCT TTGCTTGGTGTGGGCGTCTTTTGTCTCCAGTTCGCTGGAGACTCGCCTTAAAGT CATTGGCAGCCGGCCTACTGGTTTCGGAGCGCAGCACAAGTCGCGCTCTTCGCC AGCCAAGGTCAGCGTCCAGCAAGCCTTTTTTTCAACCTTTGACCTCGGATCAGGT AGGGATACCCG
<i>Alternaria</i> sp. 1	TTCTTGTTTCCTTGTTGGTGGTTCGCCACCCTAGGACAAACATAAACCTTTTGTA TTGCAATCAGTGTGAGTAACAATAATAATTACAACCTTTCAACAACGGATCTCTT GGTCTGGCATCGATGAAGAACGCAGCGAAATGCGATAAGTAGTGTGAATTGCA GAATTCAGTGAATCATCGAATCTTTGAACGCACATTGCGCCCTTTGGTATTCCAAA GGGCATGCCTGTTGAGCGTCATTTGTACCCTCAAGCTTTGCTTGGTGTGGGC GTCTTGTCTCTAGCTTTGCTGGAGACTCGCCTTAAAGTAATTGGCAGCCGGCCTA CTGGTTTCGGAGCGCAGCACAAGTCGCACTCTCTATCAGCAAAGGTCTAGCATC CATTAAAGCCTTTTTTTCAACTTTTGACCTCGGATCAGGTAGGGATACCCGCTGAAC TTAAGCATATCAATAAGCGGAGGA
<i>Stemphylium</i> sp.	AAAAATGTGGTCTTGATGGATGCTCAACCAAGGCCGATTCAAAGTGCAAGAATTG TGCTGCGCTCCGAAACCAGTAGGTGGGCTGCCAATCATTTTAAGGCGAGTCTCG TGAGAGACAAAGACGCCAACACCAAGCAAAGCTTGAGGGTACAAATGACGCTC GAACAGGCATGCCCTTTGGAATACCAAGGGCGCAATGTGCGTTCAAAGATTG ATGATTCACTGAATTCTGCAATTCACACTACGTATCGCATTTCGCTGCGTTCTTCA TCGATGCCAGAACCAAGAGATCCGTTGTTGAAAGTTGAATAATTACATTGTTTAC TGACGCTGATTGCAATTACAAAAAGGTTTATGGTTTGGTCTGGTGGCGGGCGAA CCCGCCAGGAAACAAGAAGTGCGCAAAAGACATGGGTGAATAATTAGACAAG CTGGAGCCCTCACCGAGGTGAGGTCCCAACCCGCTTTCATATTGTGAATGATCC CTCCGCAGGTTACCC
<i>Aureobasidium</i> sp. 1	GTCCCAGGCGAGCGCCCGCCAGAGTTAAACCAAACCTCTTGTTATTTAACCGGTC GTCTGAGTTAAAAATTTGAATAAATCAAACTTTCAACAACGGATCTCTTGGTTCTC GCATCGATGAAGAACGCAGCGAAATGCGATAAGTAATGTGAATTGCAGAATTCAG TGAATCATCGAATCTTTGAACGCACATTGCGCCCTTGGTATTCCGAGGGGCATG CCTGTTGAGCGTCATTACACCACTCAAGCTATGCTTGGTATTGGGCGTCGTCCT TAGTTGGGCGCGCCTTAAAGACCTCGGCGAGGCCACTCCGGCTTTAGGCGTAGT AGAATTTATTGCAACGTCTGTCAAAGGAGAGGAACTCTGCCGACTGAAACCTTTA TTTTCTAGGTTGACCTCGGATCAGGTAGGGATACCC

Species	Sequence
<i>Aureobasidium</i> sp. 2	ATAAAGGTTTCAGTCGGCAGAGTTCCTCTCCTTTGACAGACGTTTCAATAAATTCT ACTACGCCTAAAGCCGGAGTGGCCTCGCCGAGGTCTTTAAGGCGCGCCAACTA AGGACGACGCCCAATACCAAGCATAGCTTGAGTGGTGTAAATGACGCTCGAACAG GCATGCCCTCGGAATACCAAGGGGCGCAATGTGCGTTCAAAGATTCGATGATT CACTGAATTCTGCAATTCACATTACTTATCGCATTTCGCTGCGTTCTTCATCGATG CGAGAACCAAGAGATCCGTTGTTGAAAGTTTTGATTATTCAAATTTAACTCAG ACGACCGGTTAAATAACAAGAGTTTGGTTTAACTCTGGCGGGGCGCTCGCCTGGG ACGAATCCCCAGCGGCTCGAGACCGAGCGGTCCCGCCAAAGCAACAAGGTAGTT TTAACAACAAAGGGTTGGAGGTGGGCGCTGAGCACCTTACTCTTTAATGATCC TTCCGCAGGTTACCTACGGAAGNGGATNATTAAGAGTAAGGGTGTCTGAGCGC CCGACCTCCAACCTTTGTTGTTAAAACCTACCTTGTGCTTTGGCGGGACCGCTC GGTCTCGAGCCGCTGGGATTCTCCAGGCGAGCGCCCGCCAGAGTTAAACCA AACTCTTGTATTTAACCAGGTCGTCTGAGTTAAAATTTNGAATAAATNAAAACCTTN ACAACGGANCTCTTGGTTCTCGCATCGA
<i>Didymella glomerata</i>	CCGCCGATTGGNCAATTTAAACNATTTGCAGTTGCAATCAGCGTCTGAAAAAACT TAATAGTTACAACCTTTCAACAACGGATCTCTTGGTTCTGGCATCGATGAAGAACGC AGCGAAATGCGATAAGTAGTGTGAATTGCAGAATTCAGTGAATCATCGAATCTTT GAACGCACATTGCGCCCCCTTGGTATTCCATGGGGCATGCCTGTTGAGCGTCAT TTGTACCTTTCAAGCTCTGCTTGGTGTGGGTGTTTGTCTCGCCTCTGCGTGTAGA CTCGCCTCAAACAATTGGCAGCCGGCGTATTGATTTGAGAGCGCAGTACATCTC GCGCTTGGCACTCATAACGACGACGTCCAAAAGTACATTTTTACTCTTGACCTC GGATCAGGTAGGGATACCCGCTGAACCTAAGCATATCAATAAGCGG
<i>Didymella</i> sp. 1	CCGCCGATTGGACAATTTAAACCATTTGCAGTTGCAATCAGCGTCTGAAAAAACTT AATAGTTACAACCTTTCAACAACGGATCTCTTGGTTCTGGCATCGATGAAGAACGC AGCGAAATGCGATAAGTAGTGTGAATTGCAGAATTCAGTGAATCATCGAATCTTT GAACGCACATTGCGCCCCCTTGGTATTCCATGGGGCATGCCTGTTGAGCGTCAT TTGTACCTTTCAAGCTCTGCTTGGTGTGGGTGTTTGTCTCGCCTCTGCGTGTAGA CTCGCCTCAAACAATTGGCAGCCGGCGTATTGATTTGAGAGCGCAGTACATCTC G
<i>Didymella</i> sp. 2	CTTTTAAGTACCTTACGTTTCCTCGGCGGGTCCGCCCGCGATTGGACAATTTAA ACCATTTGCAGTTGCAATCAGCGTCTGAAAAAACTTAATAGTTACAACCTTTCAACA ACGGATCTCTTGGTTCTGGCATCGATGAAGAACGCAGCGAAATGCGATAAGTAGT GTGAATTGCAGAATTCAGTGAATCATCGAATCTTTGAACGCACATTGCGCCCCCT GGTATTCCATGGGGCATGCTGTTGAGCGTCATTTGTACCTTTCAAGCTCTGCTT GGTGTGGGTGTTTGTCTCGCCTCTGCGTGTAGACTCGCCTCAAACAATTGGCA GCCGGCGTATTGATTTGAGAGCGCAGTACATCTCGCGCTTGGCACTCATAACGAC GACGTCCAAAAGTACATT
<i>Cladosporium</i> sp.	TCGGGCGGGGCTCCGGGTGGACACTTCAAACCTTTGCGTAACCTTTGCAGTCTG AGTAAACTTAATTAATAAATTAACACTTTTAAACAACGGATCTCTTGGTTCTGGCATC GATGAAGAACGCAGCGAAATGCGATAAGTAATGTGAATTGCAGAATTCAGTGAAT CATCGAATCTTTGAACGCACATTGCGCCCCCTGGTATTCCGGGGGGCATGCCTG TTCGAGCGTCATTTACCACTCAAGCCTCGCTTGGTATTGGGCAACCGG
<i>Fusarium</i> sp.	GGGACGGCCCGCCGAGGAAACCCTAAACTCTGTTTTAGTGGAACTTCTGAGT ATAAAAAACAAATAAATCAAACCTTTCAACAACGGATCTCTTGGTTCTGGCATCGA TGAAGAACGCAGCAAATGCGATAAGTAATGTGAATTGCAGAATTCAGTGAATCA TCGAATCTTTGAACGCACATTGCGCCCGCCAGTATTCTGGCGGGCATGCCTGTTG GAGCGTCAATTTCAACCCTCAAGCCAGCTTGGTGTGGGATCTGTGTGAAACAC AGTCCCCAAATTGATTGGCGGTACGTCGAGCTTCCATAGCGTAGTA
<i>Sordaria</i> sp.	CGGGCCCCGGATCCTCGGGTCTCCCGCTCGCGGGAGGCTGCCCGCCGGAGT GCCGAAACCAAACCTTTGATATTTTATGTCTCTCTGAGTAAACTTTTAAATAAGTCA AAACTTTCAACAACGGATCTCTTGGTTCTGGCATCGATGAAGAACGCAGCGAAAT GCGATAAGTAATGTGAATTGCAGAATTCAGTGAATCATCGAATCTTTGAACGCACA TTGCGCTCGCCAGTATTCTGGCGAGCATGCCTGTTGAGCGTCATTTCAACCATC AAGCTCTGCTTGGGTTGGGATCCGCGTCTGACGCGGTCCCTCAAACAACAGTGG CGGGCTCGTAGTCACACCGAGCGTAGTAACTCTACATCGCTATGGTCTGCGG CGGGTTCTTGGCGTAAAACCCCAATTTCTAAGGTTGACCTCGGATCAGGTAGGA ATACCCGCTGAACCTAAGCATATCAATAAGCGGAGGA

Species	Sequence
<i>Arthrinium</i> sp.	AAAAATGTGGTCTTGATGGATGCTCAACCAAGGCCGATTCAAAGTGCAAGAATTG TGCTGCGCTCCGAAACCAGTAGGTCGGCTGCCAATCATTTAAGGCGAGTCTCG TGAGAGACAAAGACGCCAACACCAAGCAAAGCTTGAGGGTACAAATGACGCTC GAACAGGCATGCCCTTTGGAATACCAAAGGGCGCAATGTGCGTTCAAAGATTG ATGATTCACTGAATTCTGCAATTCACACTACGTATCGCATTTTCGCTGCGTTCTTCA TCGATGCCAGAACCAAGAGATCCGTTGTTGAAAGTTGTAATAATTACATTGTTTAC TGACGCTGATTGCAATTACAAAAGGTTTATGGTTTGGTCCCTGGTGGCGGGCGAA CCCGCCAGGAAACAAGAAGTGCGCAAAAGACATGGGTGAATAATTCAGACAAG CTGGAGCCCTCACCGAGGTGAGGTCCCAACCCGCTTTCATATTGTGTAATGATCC CTCCGCAGGTTCCACC

Table S2: Primers used in this study.

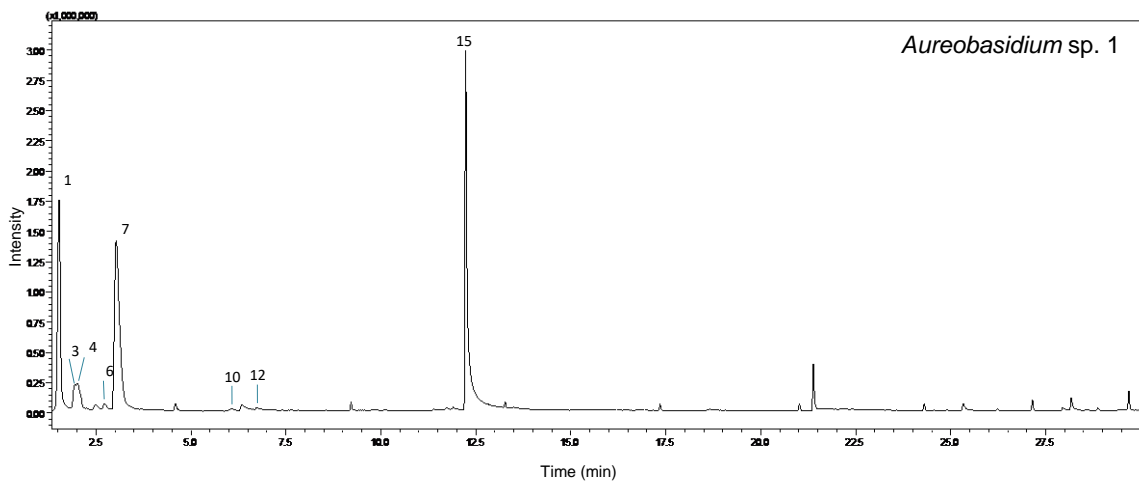
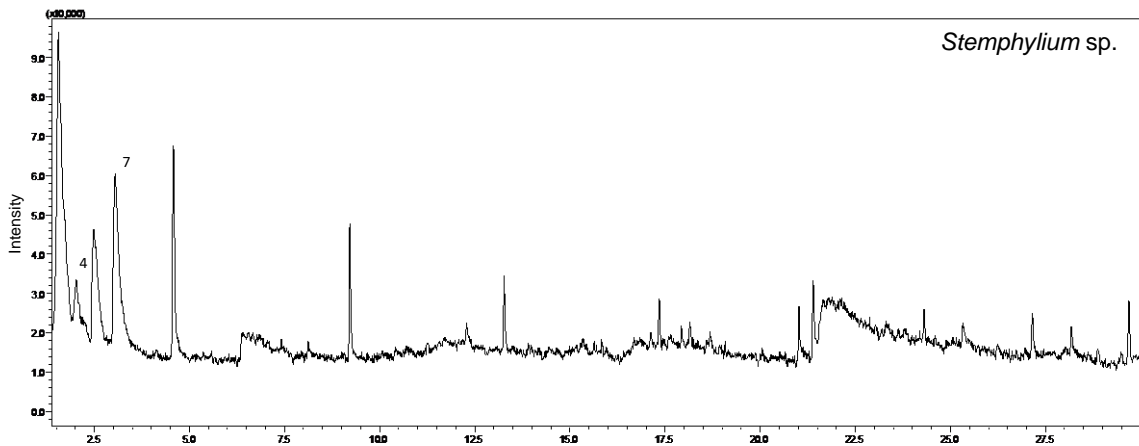
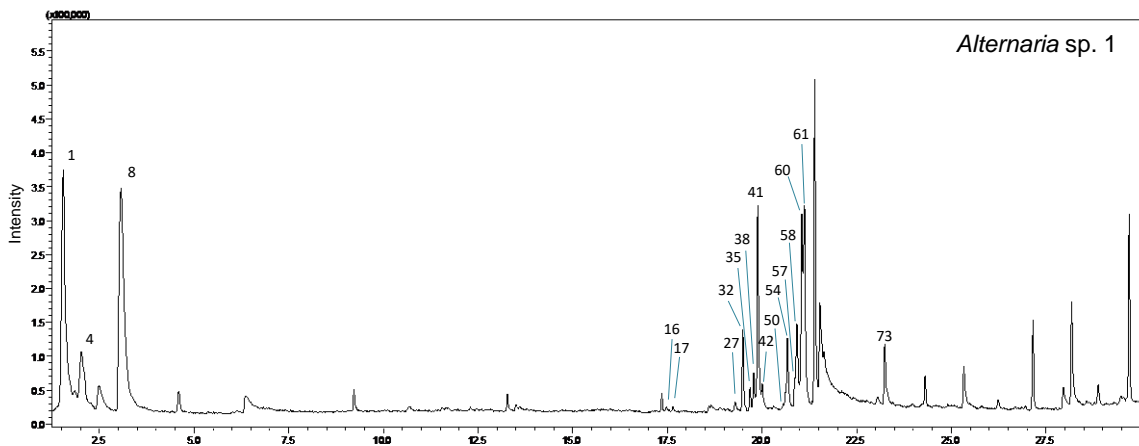
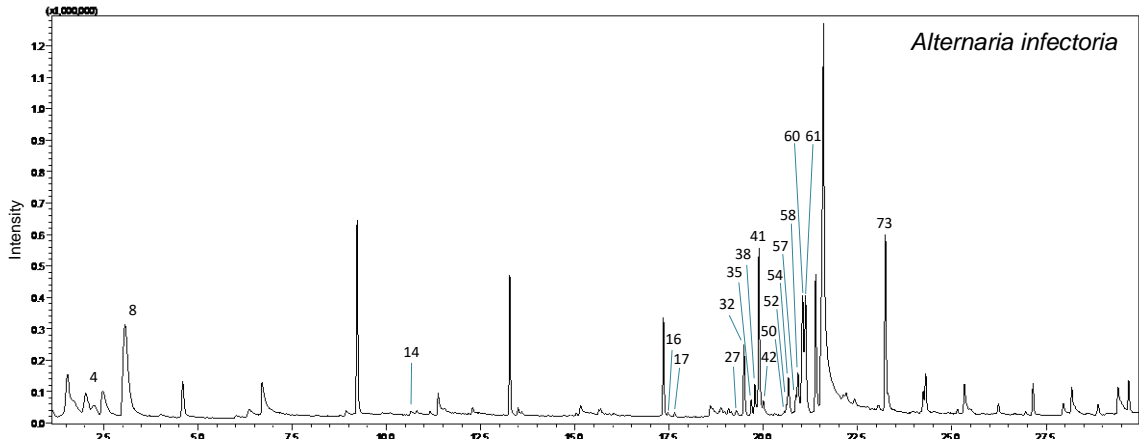
Name	Sequence	Purpose
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CxTPS1_Rev	TCACGACGCCCTCG	cloning
CxTPS2_Fwd	CACCATGTGACACCCTACTCGCC	cloning
CxTPS2_Rev	TCAGCAAACTCCAGATAGCTAGG	cloning
ITS1F	CTTGGTCATTTAGAGGAAGTAA	amplify fungal rRNA ITS
ITS4	TCCTCCGCTTATTGATATGC	amplify fungal rRNA ITS

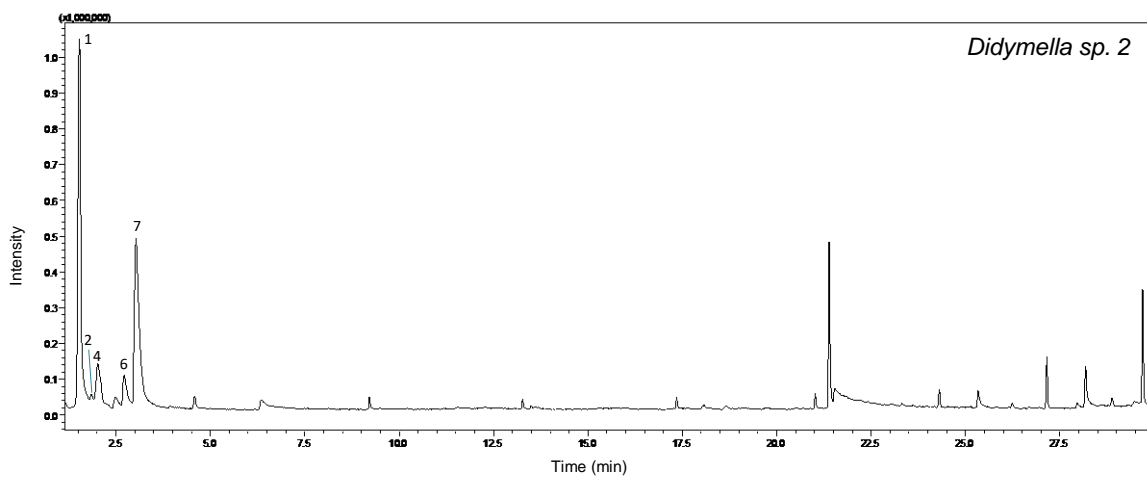
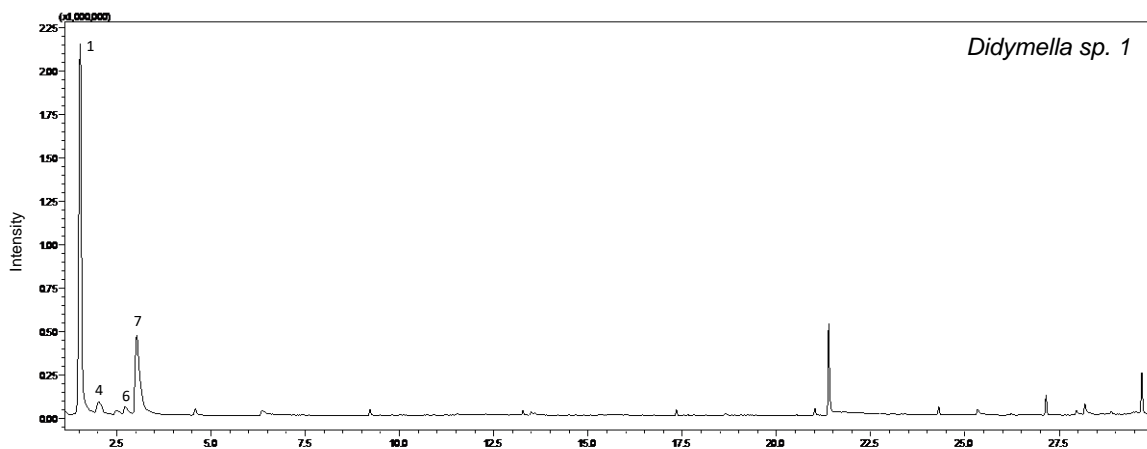
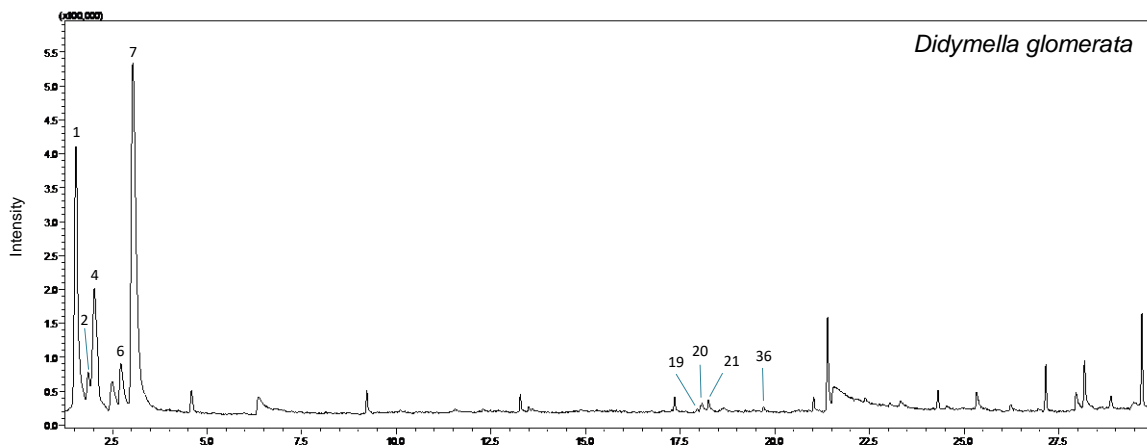
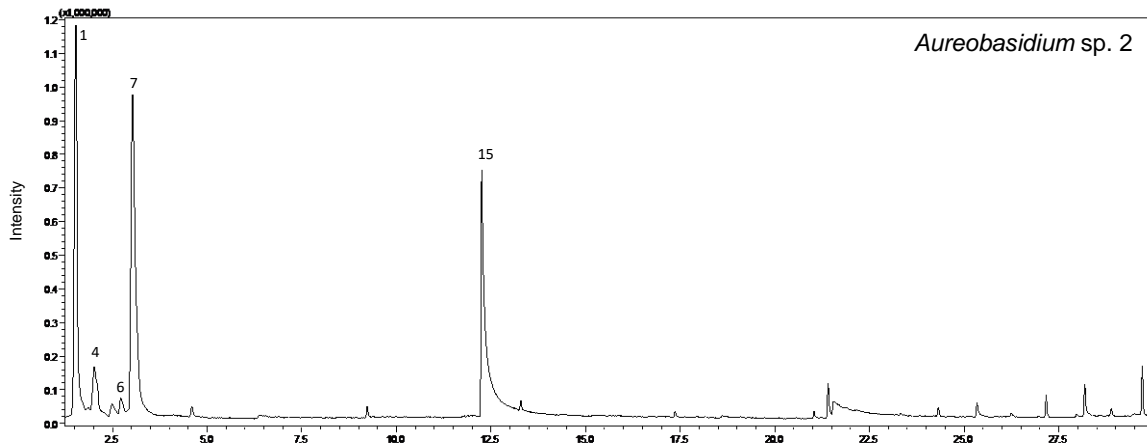
Table S3: Numbers for volatile organic compounds, which are shown in the total ion chromatograms of the volatile bouquet for each endophytic fungus in Figure S1.

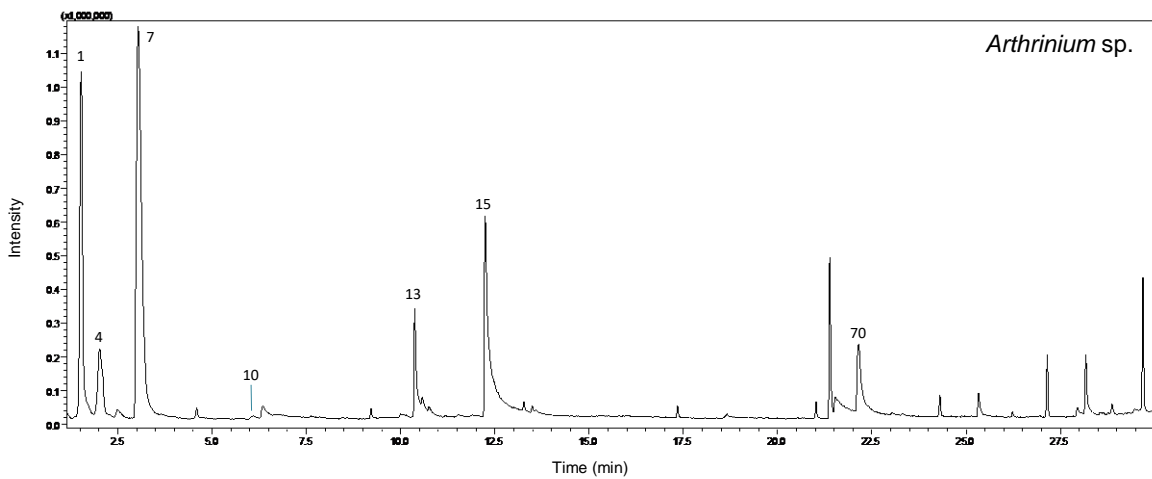
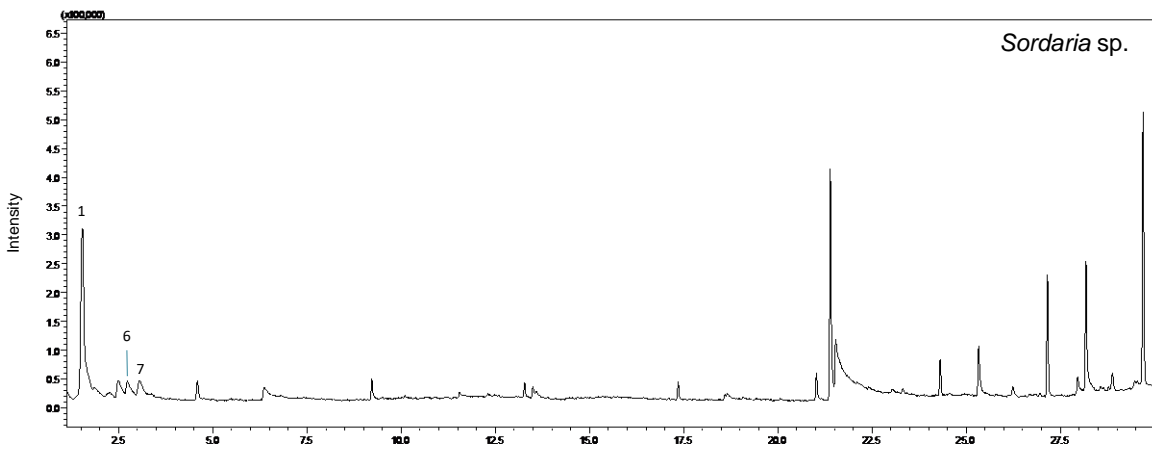
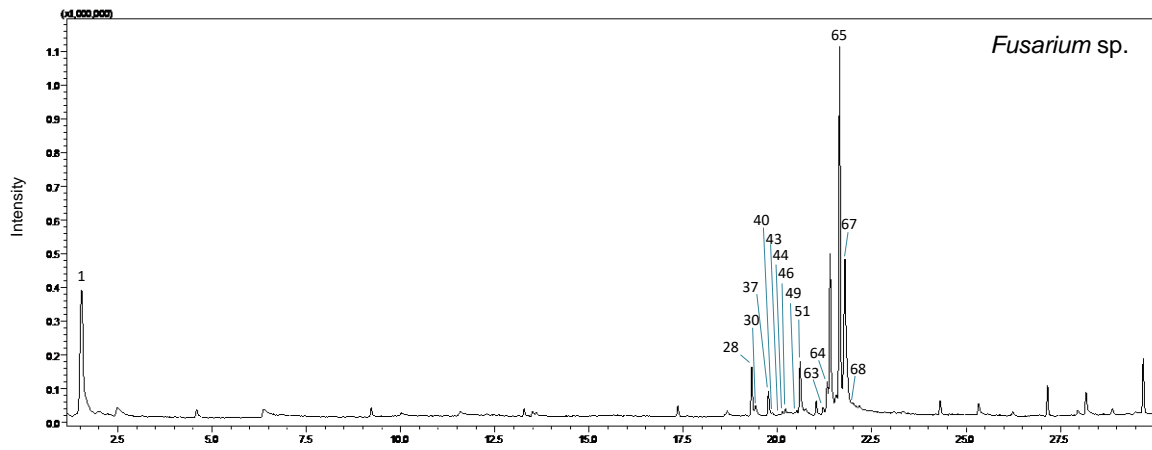
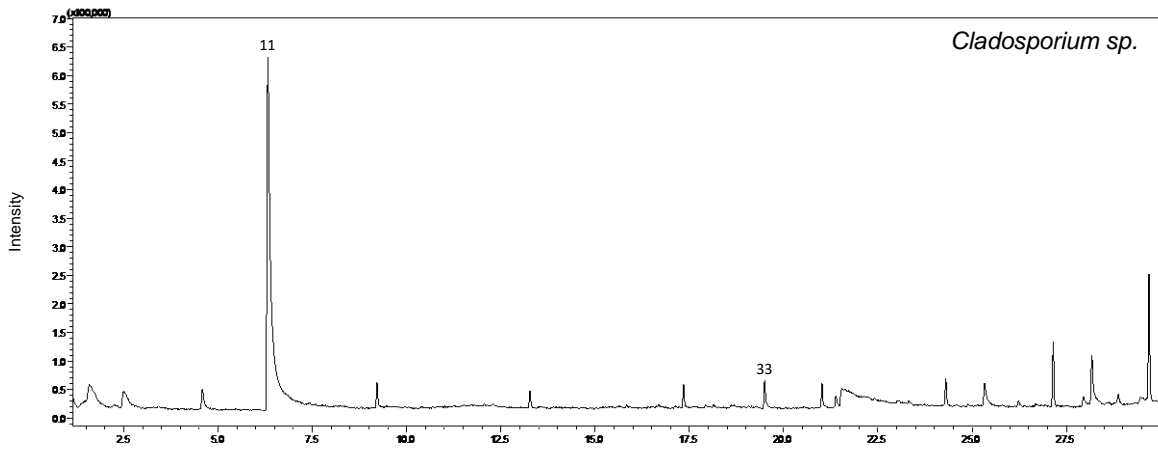
#	Volatile organic compound	R.T. (min)
1	Ethanol	1.525
2	2-Butanone	1.855
3	Ethyl Acetate	1.940
4	2-Methyl-1-propanol	2.020
5	unknown 1	2.265
6	3-Hydroxy-2-Butanone	2.720
7	3-Methyl-1-butanol	3.035
8	2-Methyl-1-butanol	3.070
9	unknown 2	3.780
10	3-Methylbutyl acetate	6.100
11	Ethenylbenzene	6.325
12	unknown 3	6.730
13	unknown 4	10.380
14	unknown 5	10.655
15	2-Phenylethanol	12.240
16	unknown 6	17.465
17	unknown 7	17.650
18	α -Cubebene	17.920

19	unknown 8	17.950
20	unknown 9	18.075
21	unknown 10	18.245
22	unknown 11	18.330
23	α -Copaene	18.525
24	unknown 12	18.765
25	unknown 13	18.850
26	Sativene	18.985
27	α -Gurjunene	19.280
28	unknown 14	19.300
29	unknown 15	19.380
30	unknown 16	19.400
31	unknown 17	19.460
32	Aristolene	19.485
33	(<i>E</i>)- β -Caryophyllene	19.500
34	unknown 18	19.535
35	unknown 19	19.675
36	unknown 20	19.695
37	Bicyclossequiphellandrene	19.745
38	β -Gurjunene	19.775
39	unknown 21	19.790
40	unknown 22	19.845
41	unknown 23	19.880
42	α -Guaiene	20.000
43	unknown 24	20.015
44	unknown 25	20.120
45	unknown 26	20.140
46	(<i>E</i>)- β -Farnesene	20.195
47	unknown 27	20.325
48	unknown 28	20.440
49	unknown 29	20.490
50	unknown 30	20.555
51	β -Chamigrene	20.585
52	unknown 31	20.610
53	unknown 32	20.610
54	α -Selinene	20.665
55	γ -Muurolene	20.685
56	unknown 33	20.800
57	unknown 34	20.860
58	β -Selinene	20.910
59	unknown 35	20.930
60	(+)-Valencene	21.040
61	unknown 36	21.115
62	α -Muurolene	21.170
63	β -Himachalene	21.195
64	β -Bisabolene	21.305
65	unknown 37	21.630

66	unknown 38	21.635
67	unknown 39	21.775
68	unknown 40	21.985
69	unknown 41	22.075
70	unknown 42	22.150
71	unknown 43	22.355
72	unknown 44	22.720
73	unknown 45	23.235
74	unknown 46	23.875
75	unknown 47	24.485
76	unknown 48	24.675
77	unknown 49	25.980







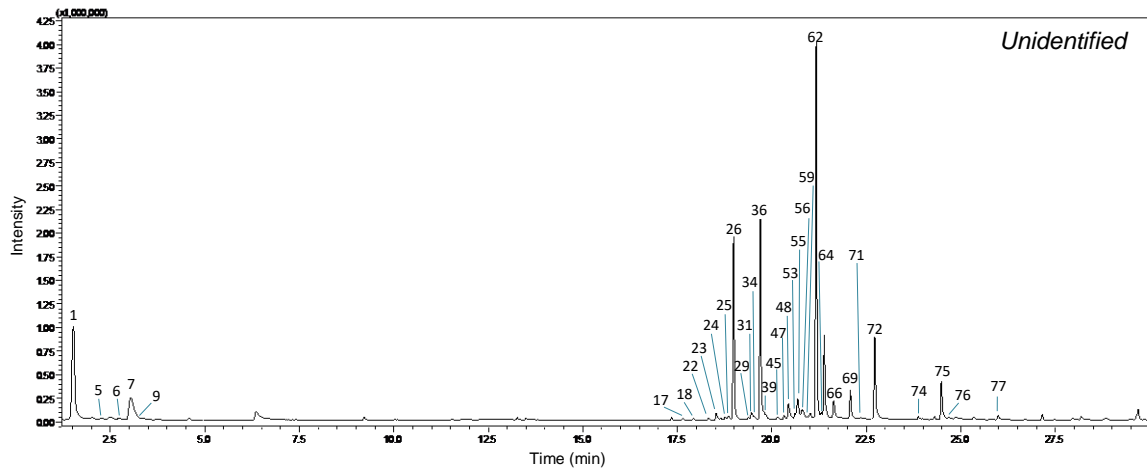
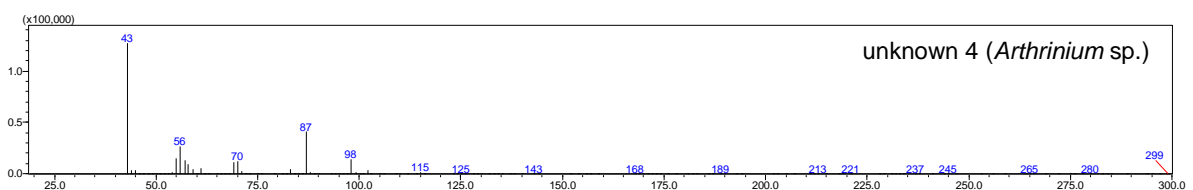
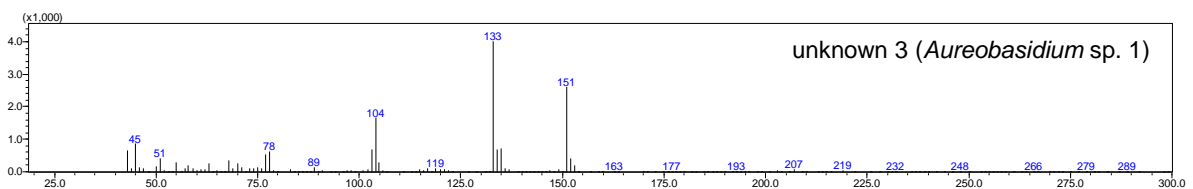
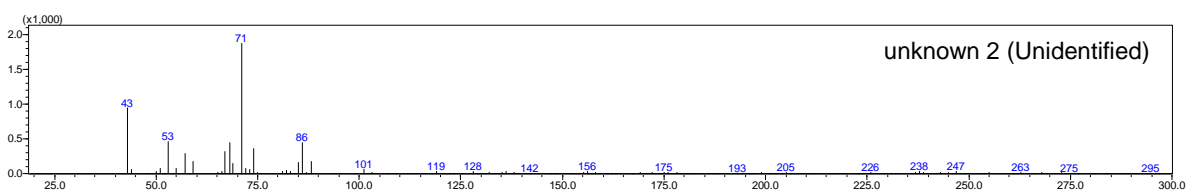
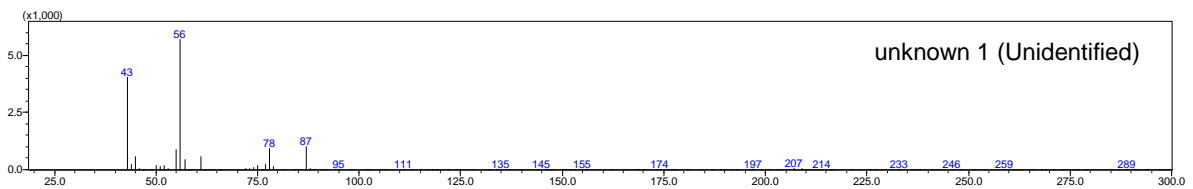
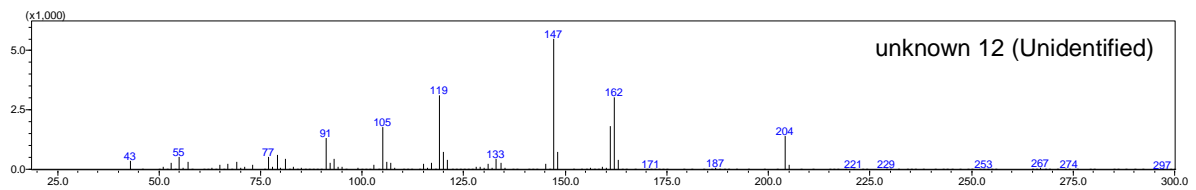
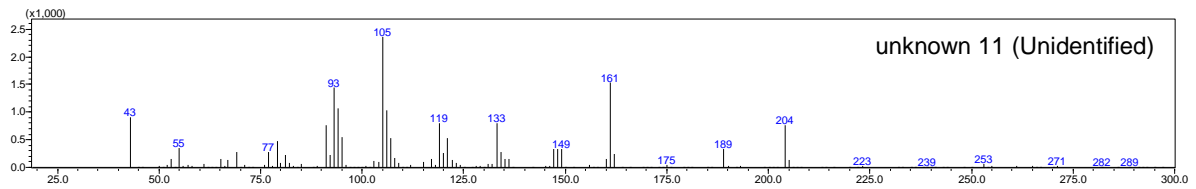
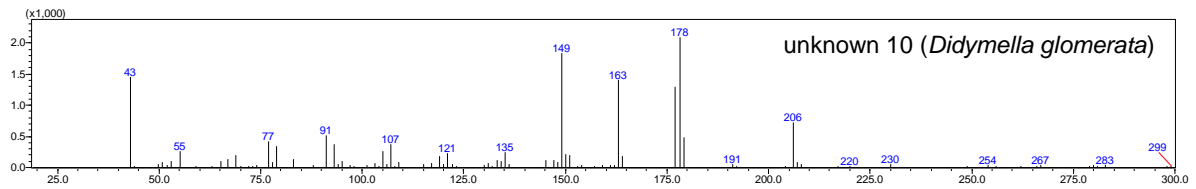
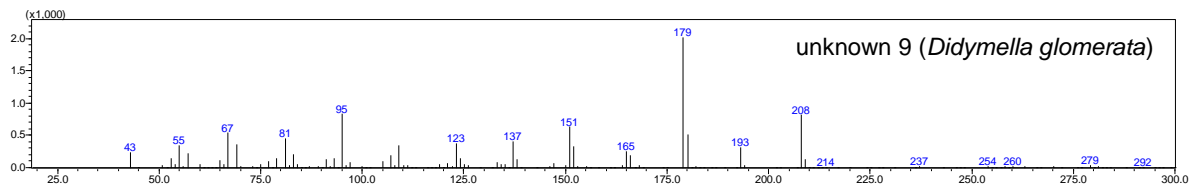
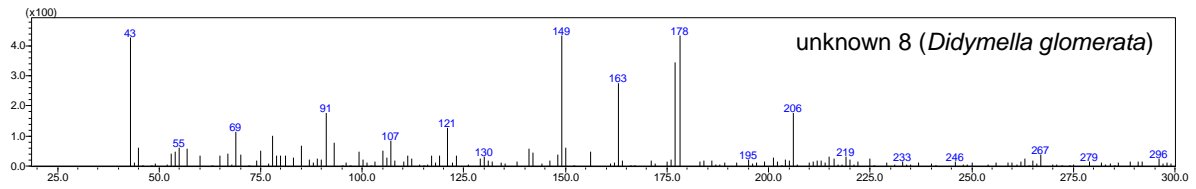
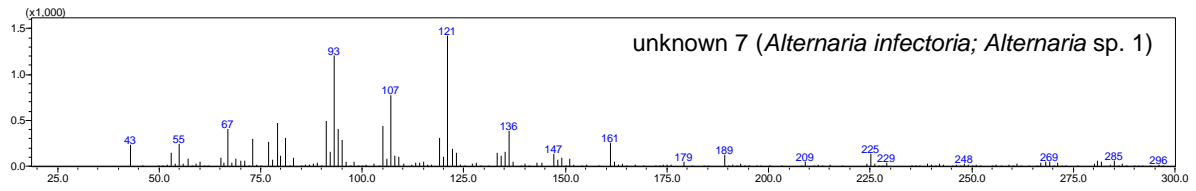
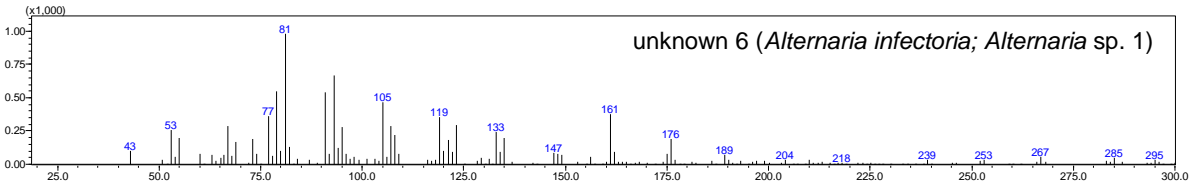
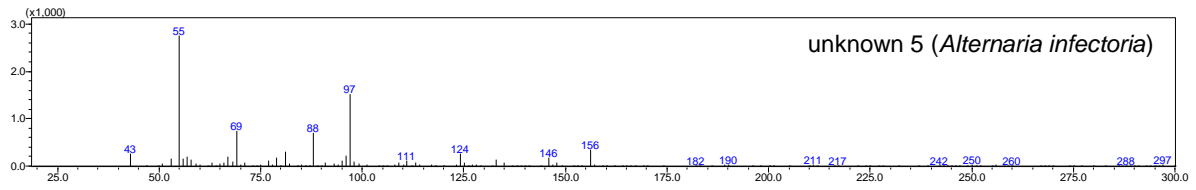
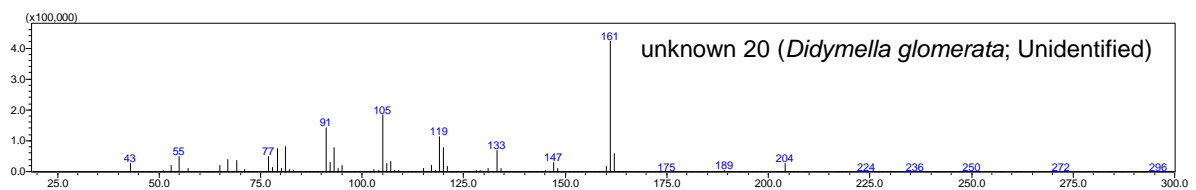
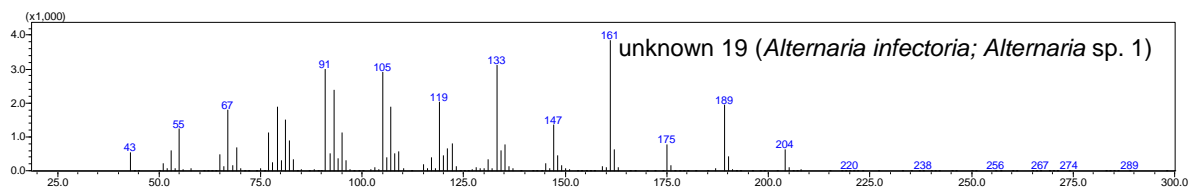
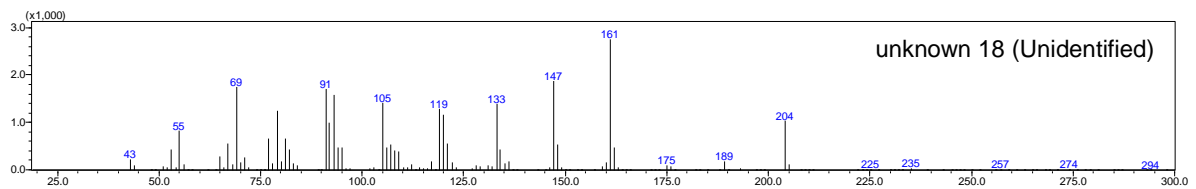
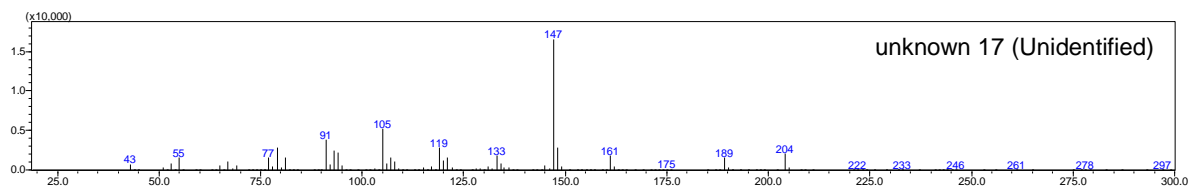
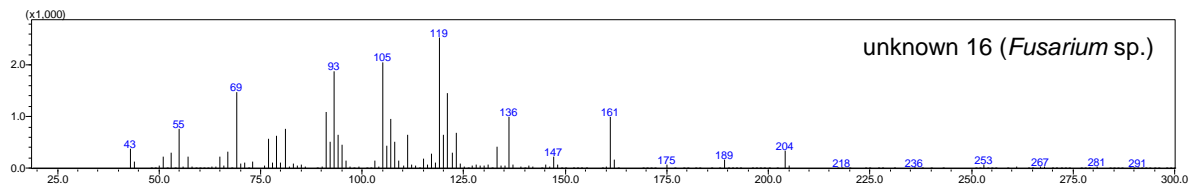
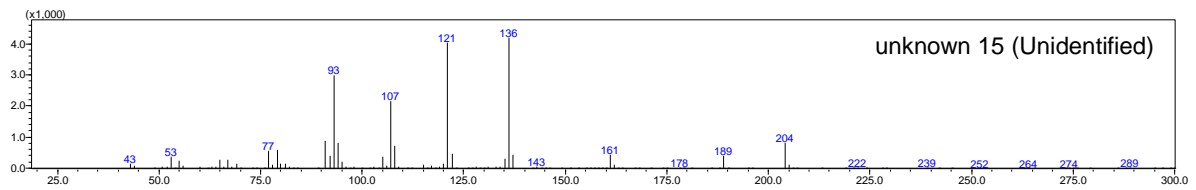
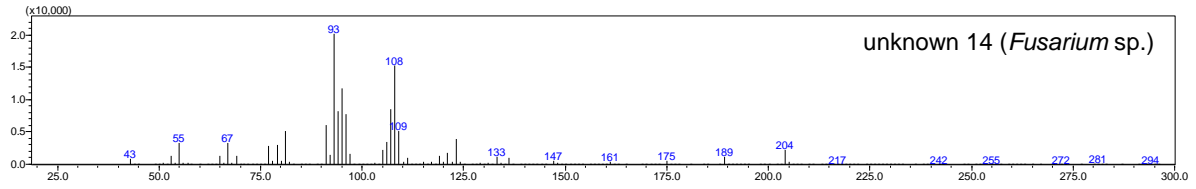
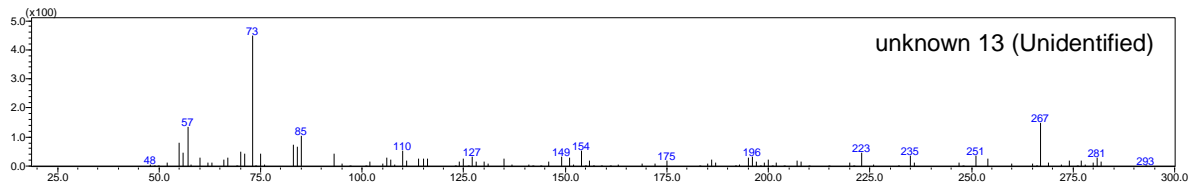
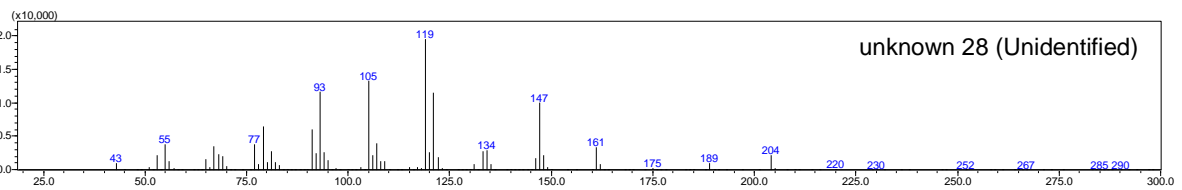
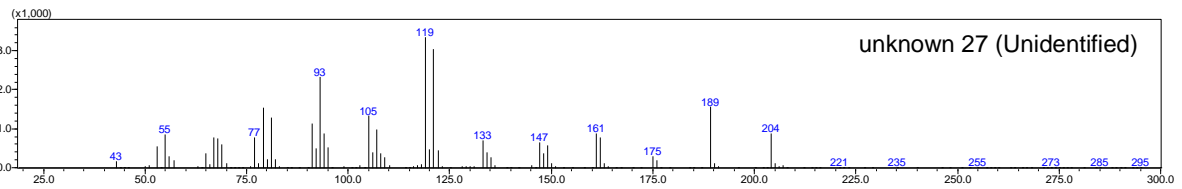
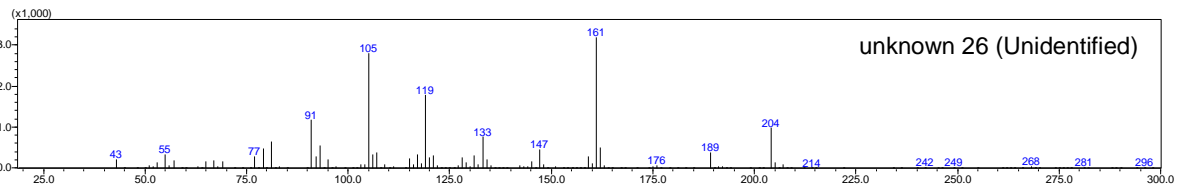
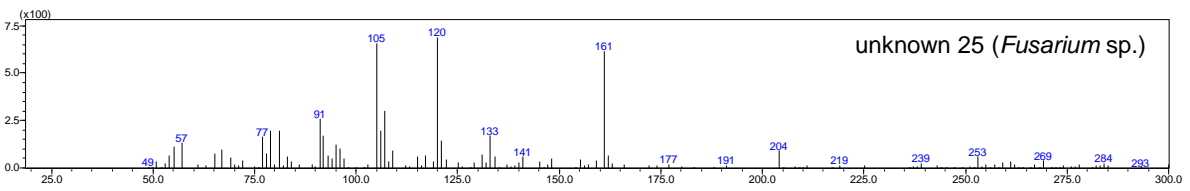
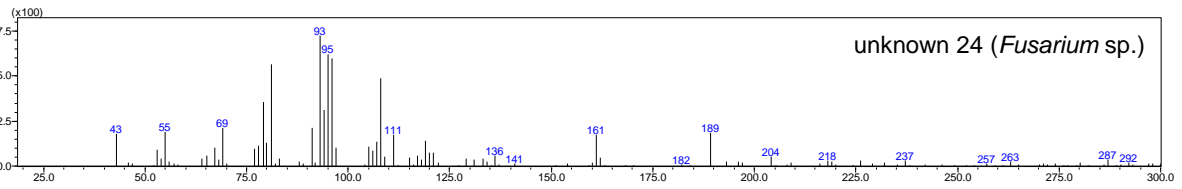
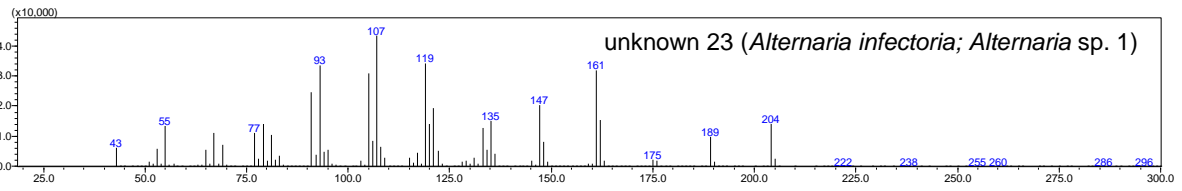
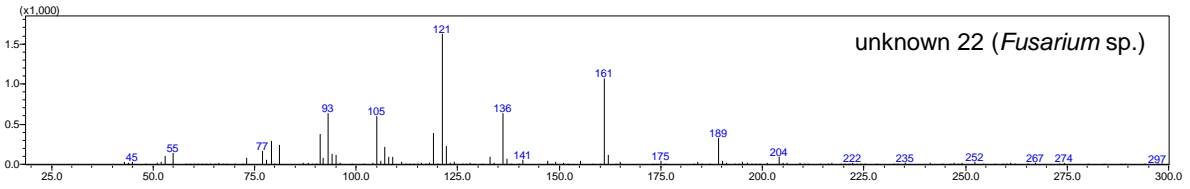
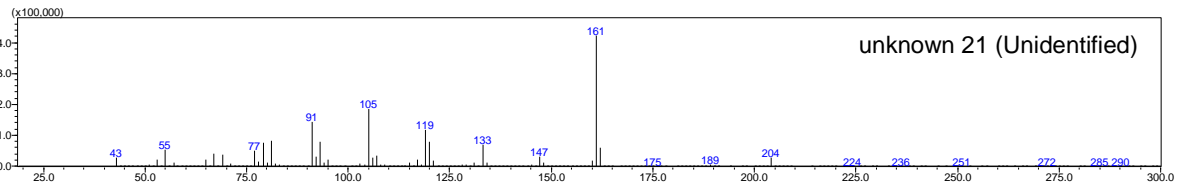


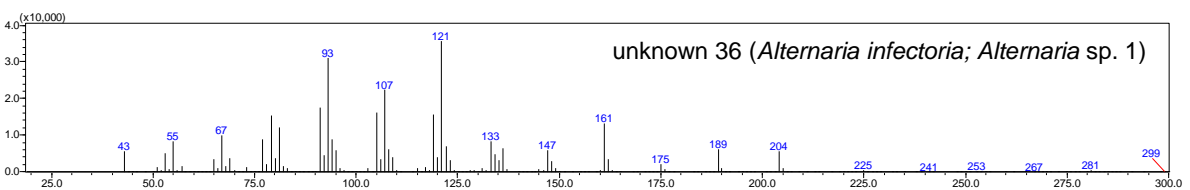
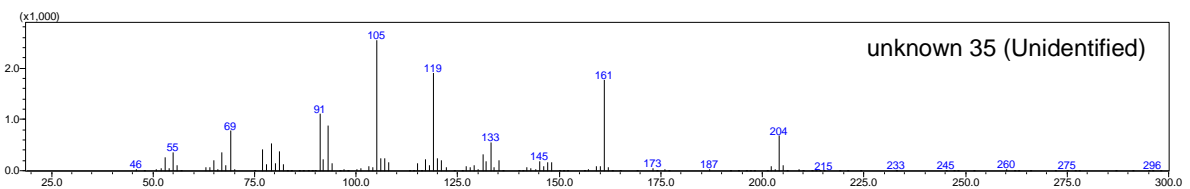
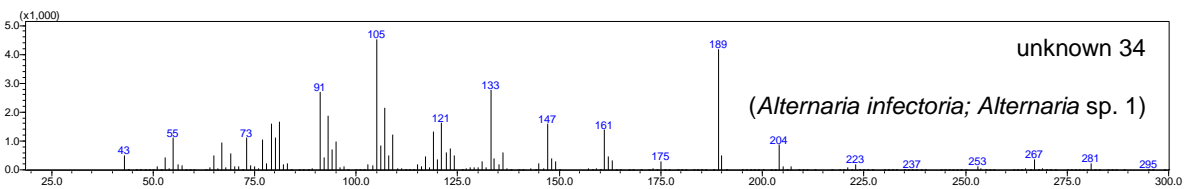
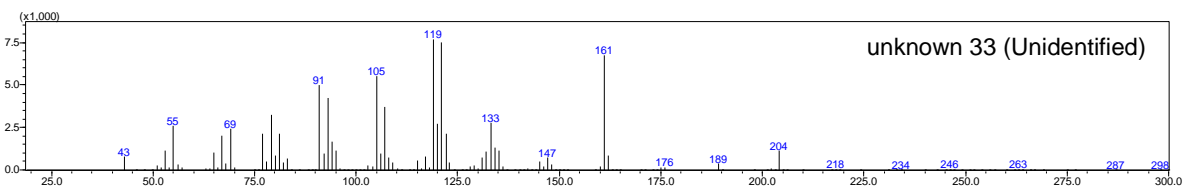
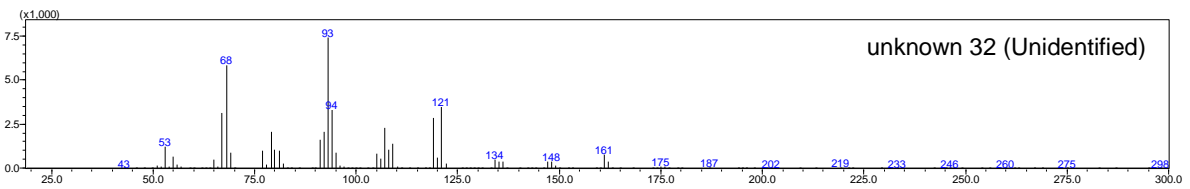
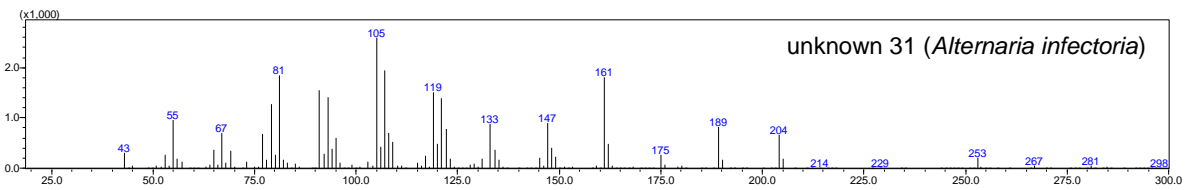
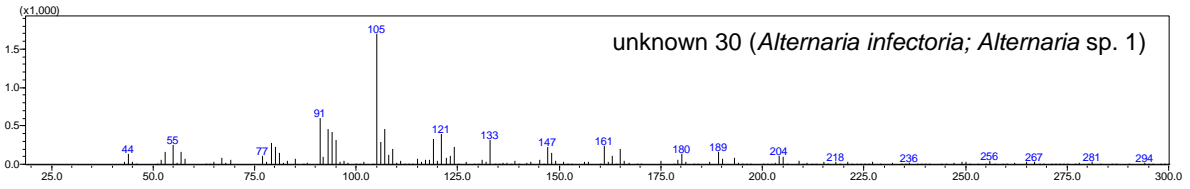
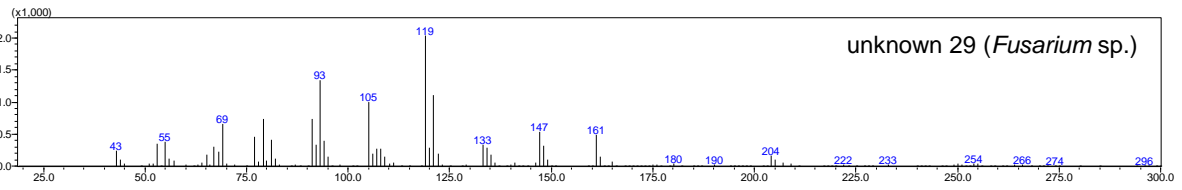
Figure S1: Representative total ion chromatogram of volatiles measured from different endophytes used in this study. Numbers indicate different volatile organic compounds, listed in Table S3. Peaks without numbers are either contamination from the PDMS tube or originate from the culture media itself.

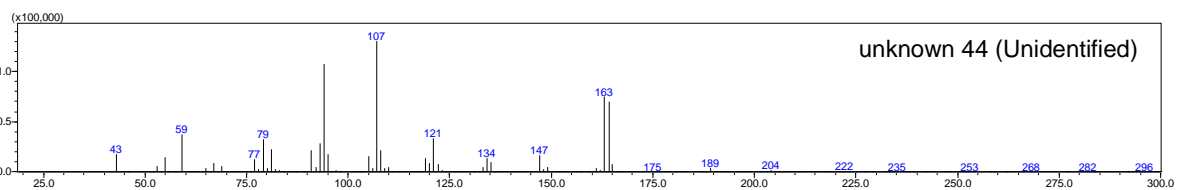
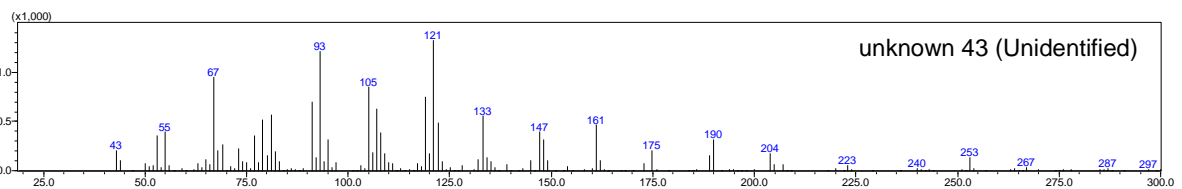
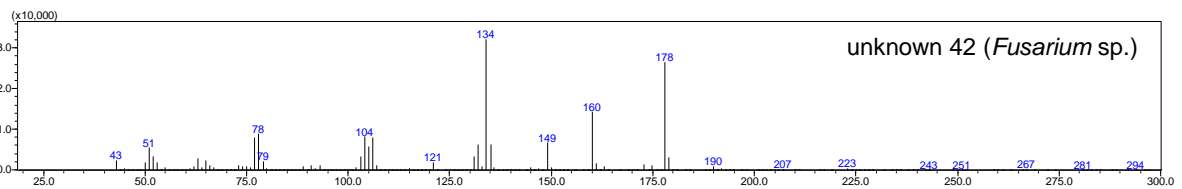
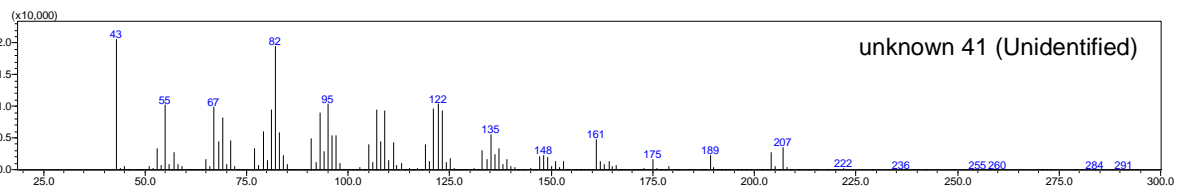
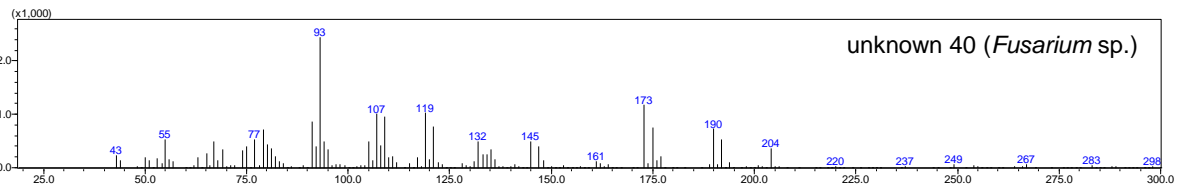
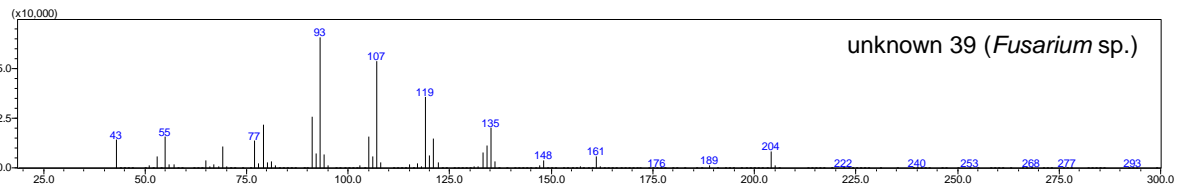
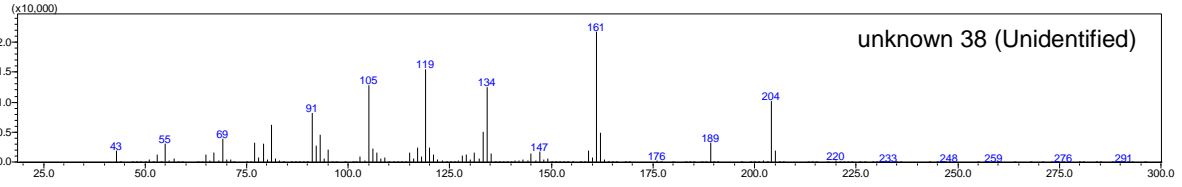
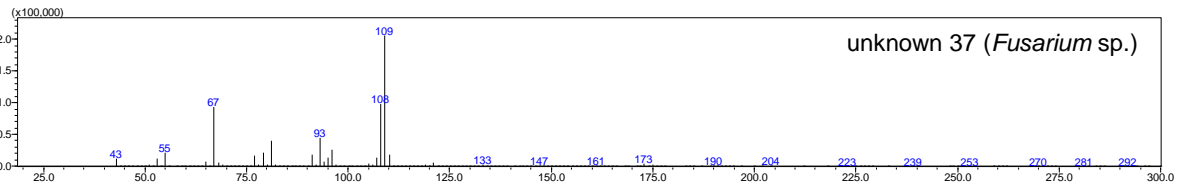












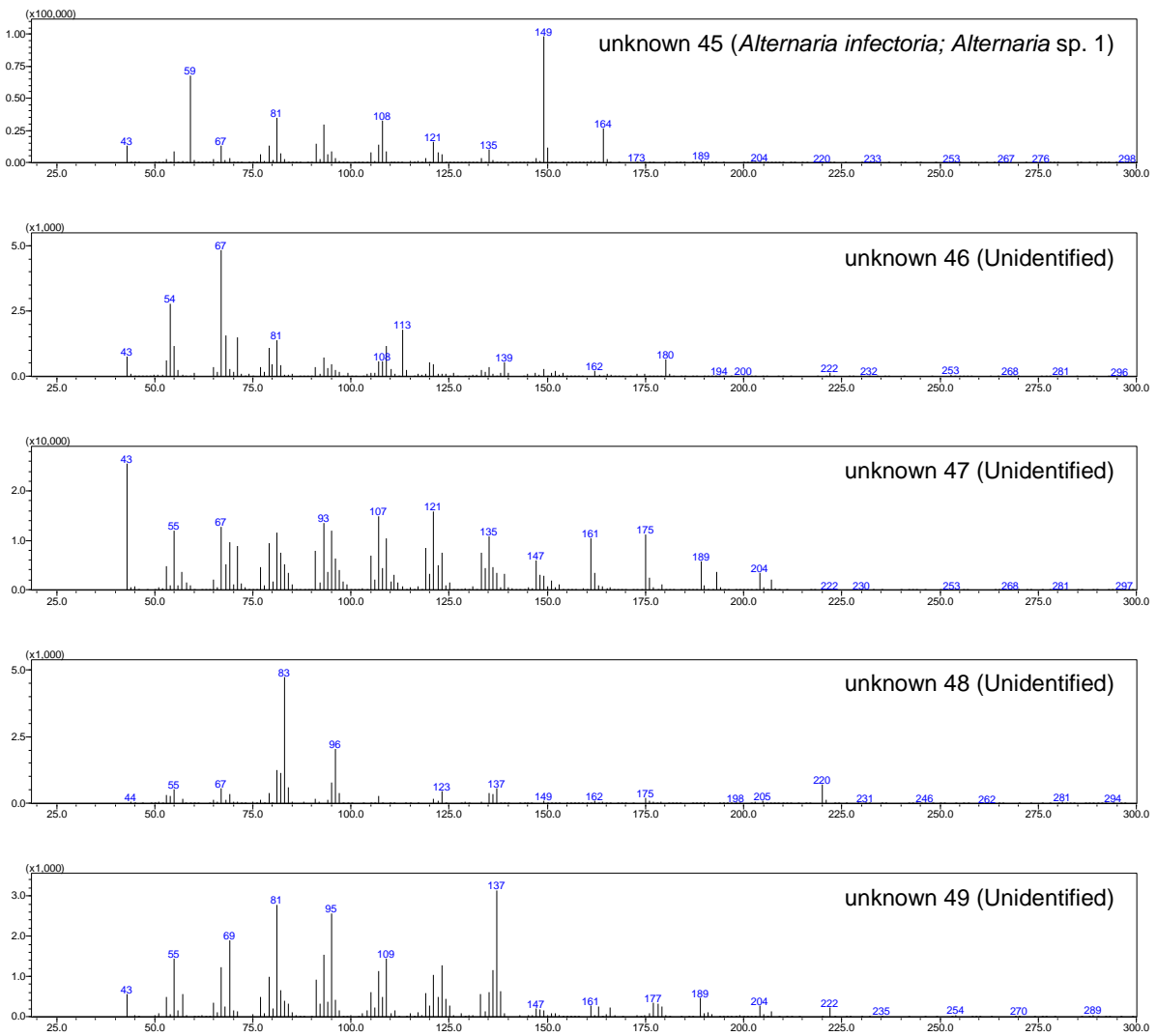


Figure S2: Mass spectra of unknown volatile organic compounds shown in Table 2. Background has been subtracted.

```
# BUSCO version is: 3.0.2
# The lineage dataset is: fungi_odb9 (Creation date: 2016-02-13, number of species: 85, number of
BUSCOs: 290)
# To reproduce this run: python /opt/software/bin/run_BUSCO.py -i /opt/software/packages/galaxy-
dist/database/files/009/dataset_9623.dat -o busco_galaxy -l
/opt/software/packages/busco/lineage/fungi_odb9/ -m transcriptome -c 11 -e 0.01 -z
#
# Summarized benchmarking in BUSCO notation for file /opt/software/packages/galaxy-
dist/database/files/009/dataset_9623.dat
# BUSCO was run in mode: transcriptome
C:98.3%[S:93.1%,D:5.2%],F:1.4%,M:0.3%,n:290
285 Complete BUSCOs (C)
270 Complete and single-copy BUSCOs (S)
15 Complete and duplicated BUSCOs (D)
4 Fragmented BUSCOs (F)
1 Missing BUSCOs (M)
290 Total BUSCO groups searched
```

Figure S3: BUSCO analysis of the *Cladosporium* sp. *de novo* assembly. The BUSCO software tool [1] was used to validate the completeness of the *de novo* assembly.

Reference

1. Afgan, E.; Baker, D.; Batut, B.; Van Den Beek, M.; Bouvier, D.; Čech, M.; Chilton, J.; Clements, D.; Coraor, N.; Grüning, B. A., *Nucleic acids research* **2018**, *46* (W1), W537-W544.