

Supplementary information

Table S1. Summary of chromatographic peak area of classified insect-virulence metabolites and a siderophore identified from OEpk14 in culture and *in vivo*

Compound	Observed <i>m/z</i>	Adduct	RT (min)	Peak area		
				Culture medium	3 DPI	5DPI
Enniatin A	682.4590	[M+H] ⁺	6.03	$4.35 \times 10^7 \pm 1.63 \times 10^6$	-	-
Ferricrocin	771.2446	[M+H] ⁺	1.35	$9.37 \times 10^5 \pm 1.00 \times 10^5$	-	$7.04 \times 10^5 \pm 5.10 \times 10^4$ $7.42 \times 10^5 \pm 1.84 \times 10^4$
Beauvericin	784.4120	[M+H] ⁺	5.85	$1.18 \times 10^8 \pm 1.20 \times 10^7$ $3.51 \times 10^6 \pm 6.07 \times 10^4$ $3.55 \times 10^6 \pm 3.22 \times 10^5$ $4.07 \times 10^6 \pm 9.37 \times 10^4$		
Beauvericin A/F	798.4277	[M+H] ⁺	6.00	$1.25 \times 10^8 \pm 1.63 \times 10^7$ $2.77 \times 10^5 \pm 1.87 \times 10^3$ $4.11 \times 10^5 \pm 7.83 \times 10^4$ $4.58 \times 10^5 \pm 7.71 \times 10^3$		
Beauvericin B	812.4425	[M+H] ⁺	6.15	$6.18 \times 10^7 \pm 2.47 \times 10^6$	-	-
Beauvericin C	848.4398	[M+Na] ⁺	6.42	$3.13 \times 10^6 \pm 2.91 \times 10^5$	-	-
Bassianolide	909.6098	[M+H] ⁺	6.43	$2.94 \times 10^7 \pm 9.90 \times 10^5$	-	-

Table S2. Summary of chromatographic peak area of classified insect-virulence metabolites and a siderophore identified from OEpk15 in culture and *in vivo*

Compound	Observed <i>m/z</i>	Adduct	RT (min)	Peak area			
				Culture medium	3 DPI	5DPI	7 DPI
Ferricrocin	771.2498	[M+H] ⁺	1.35	$5.75 \times 10^5 \pm 2.74 \times 10^4$	-	$1.06 \times 10^8 \pm 1.41 \times 10^6$	$2.57 \times 10^8 \pm 7.78 \times 10^6$
Beauvericin	784.4157	[M+H] ⁺	5.86	$5.03 \times 10^5 \pm 5.87 \times 10^4$	$3.34 \times 10^9 \pm 6.36 \times 10^7$	$6.24 \times 10^9 \pm 6.51 \times 10^8$	$1.90 \times 10^9 \pm 4.17 \times 10^8$
Beauvericin A/ F	798.4305	[M+H] ⁺	6.14	-	$4.80 \times 10^8 \pm 1.46 \times 10^8$	$6.71 \times 10^8 \pm 7.42 \times 10^7$	$2.15 \times 10^8 \pm 5.66 \times 10^6$
Beauvericin B	812.4464	[M+H] ⁺	6.34	-	$1.33 \times 10^7 \pm 1.48 \times 10^6$	$3.42 \times 10^7 \pm 6.79 \times 10^6$	$1.70 \times 10^7 \pm 3.25 \times 10^6$
Beauvericin C	826.4626	[M+H] ⁺	6.50	-	$7.42 \times 10^4 \pm 3.27 \times 10^4$	-	-
Beauvericin D	770.4002	[M+H] ⁺	5.85	-	$2.26 \times 10^8 \pm 2.12 \times 10^6$	$1.77 \times 10^8 \pm 2.12 \times 10^7$	$8.25 \times 10^7 \pm 3.25 \times 10^6$
Bassanolide	931.5959	[M+Na] ⁺	6.42	-	$1.55 \times 10^8 \pm 2.47 \times 10^7$	$1.89 \times 10^8 \pm 0.00$	$7.27 \times 10^7 \pm 7.99 \times 10^6$

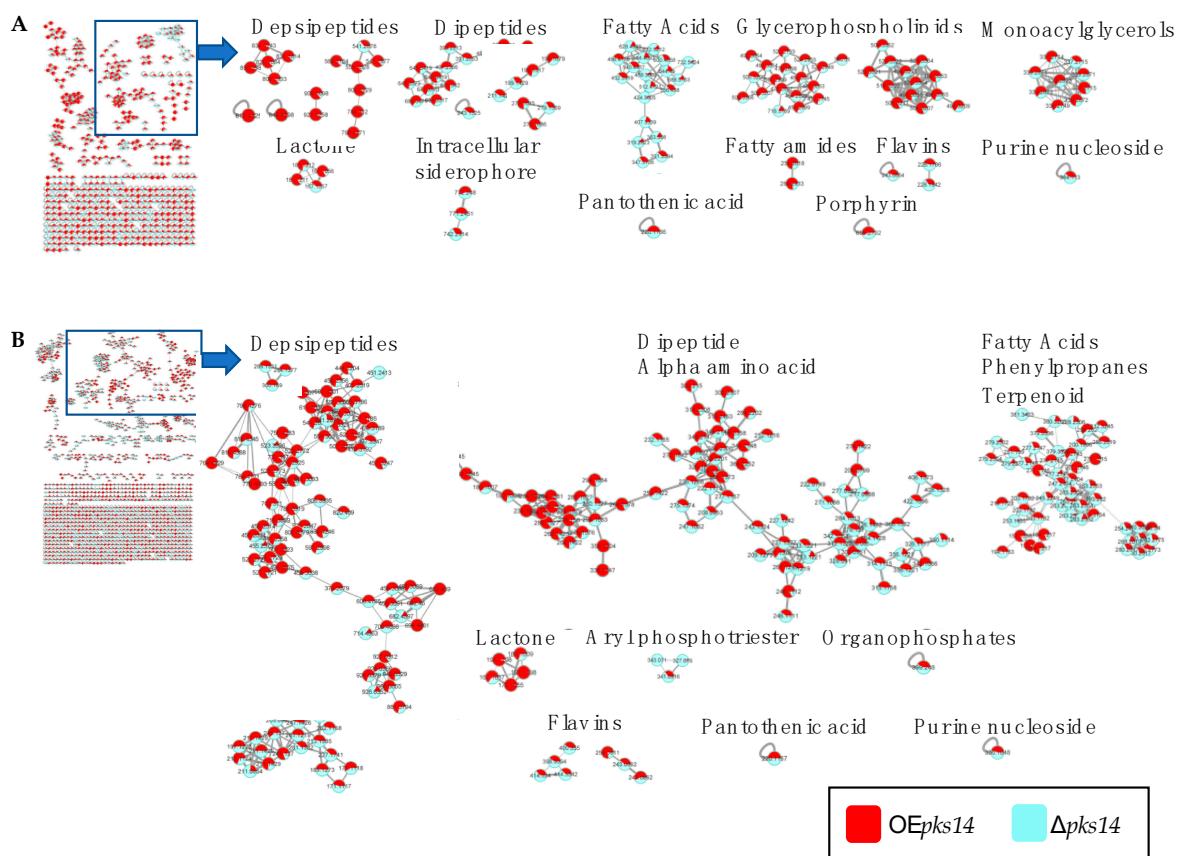


Figure S1. Molecular networking of classified metabolites for OEpk14 (red) and Δ pk14 (light blue) strains in (A) culture cells and (B) culture broth.

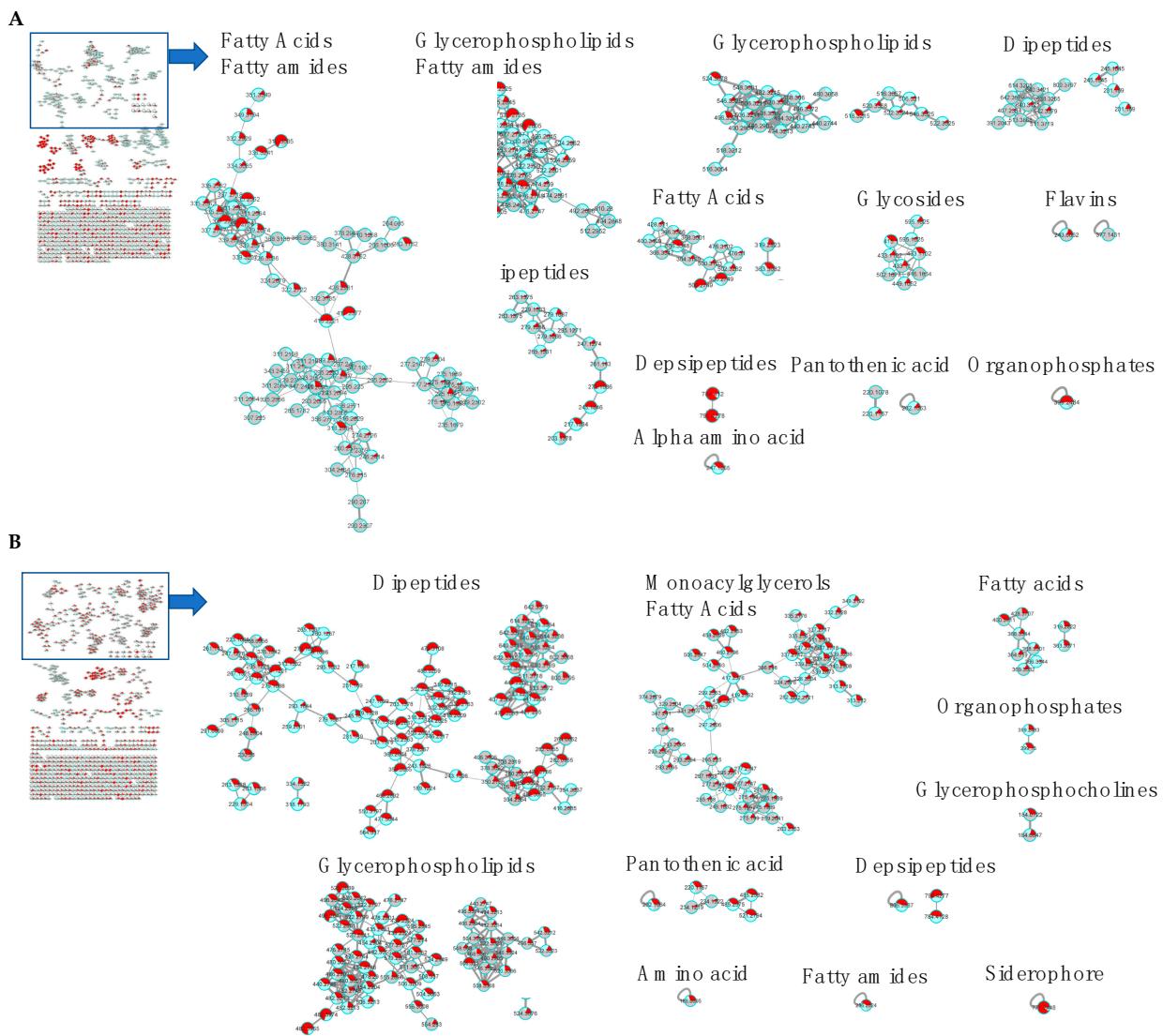


Figure S2. Molecular networking of classified metabolites in OE $pks14$ (red) and $\Delta pks14$ (light blue) strains *in vivo* at (A) early-stage infection (3 DPI) for live larvae, (B) mid-stage infection (5 DPI) for dead larvae, and (C) late-stage infection (7 DPI) for cadavers covered with fungal hyphae. Saline-injected BAWs were used as controls (gray).

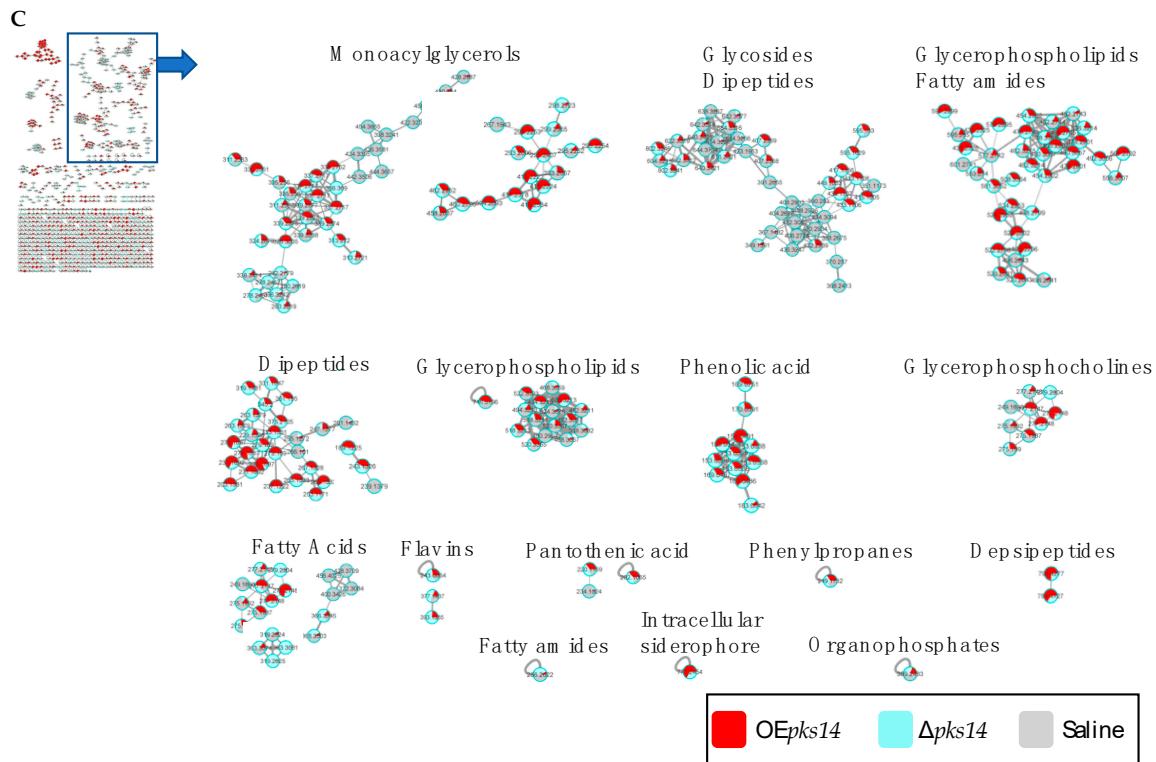


Figure S2. (continued) Molecular networking of classified metabolites in *OEpk14* (red) and $\Delta pk14$ (light blue) strains *in vivo* at (A) early-stage infection (3 DPI) for live larvae, (B) mid-stage infection (5 DPI) for dead larvae, and (C) late-stage infection (7 DPI) for cadavers covered with fungal hyphae. Saline-injected BAWs were used as controls (gray).

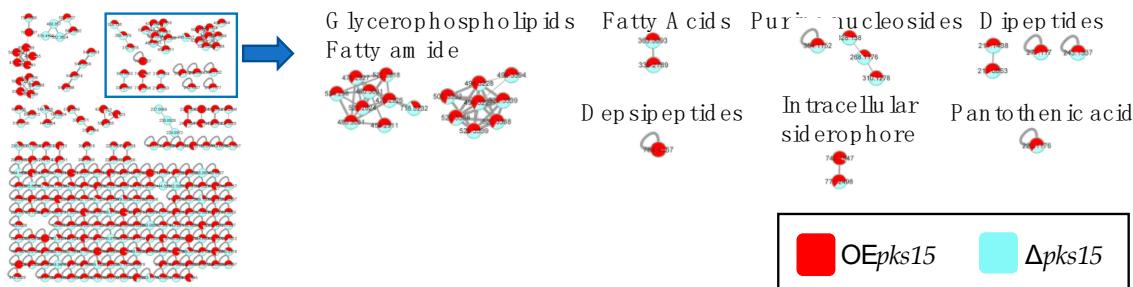


Figure S3. Molecular networking of classified metabolites for *OEpk15* (red) and $\Delta pk15$ (light blue) strains in culture.

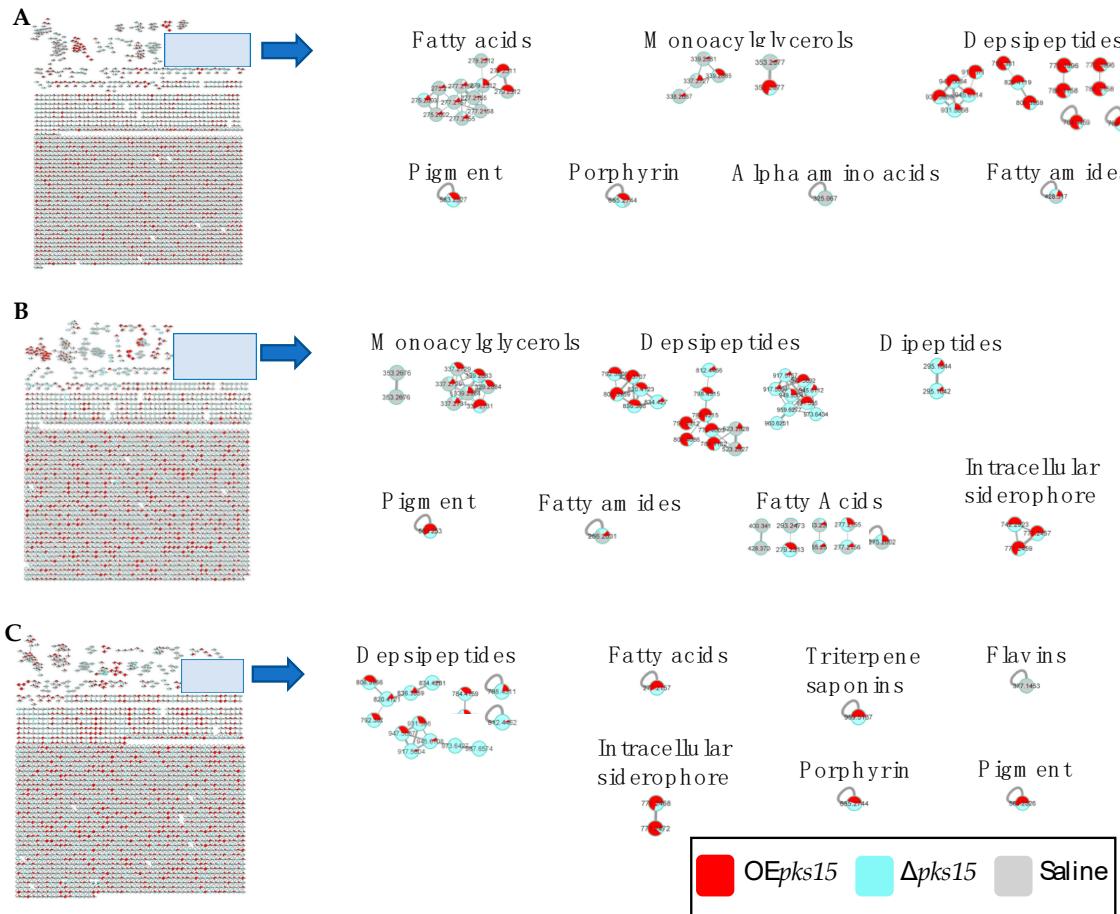


Figure S4. Molecular networking of classified metabolites for OEpk15 (red) and Apk15 (light blue) strains *in vivo* at (A) early-stage infection (3 DPI) for live larvae, (B) mid-stage infection (5 DPI) for dead larvae, and (C) late-stage infection (7 DPI) for cadavers covered with fungal hyphae. Saline-injected BAWs were used as controls (gray).

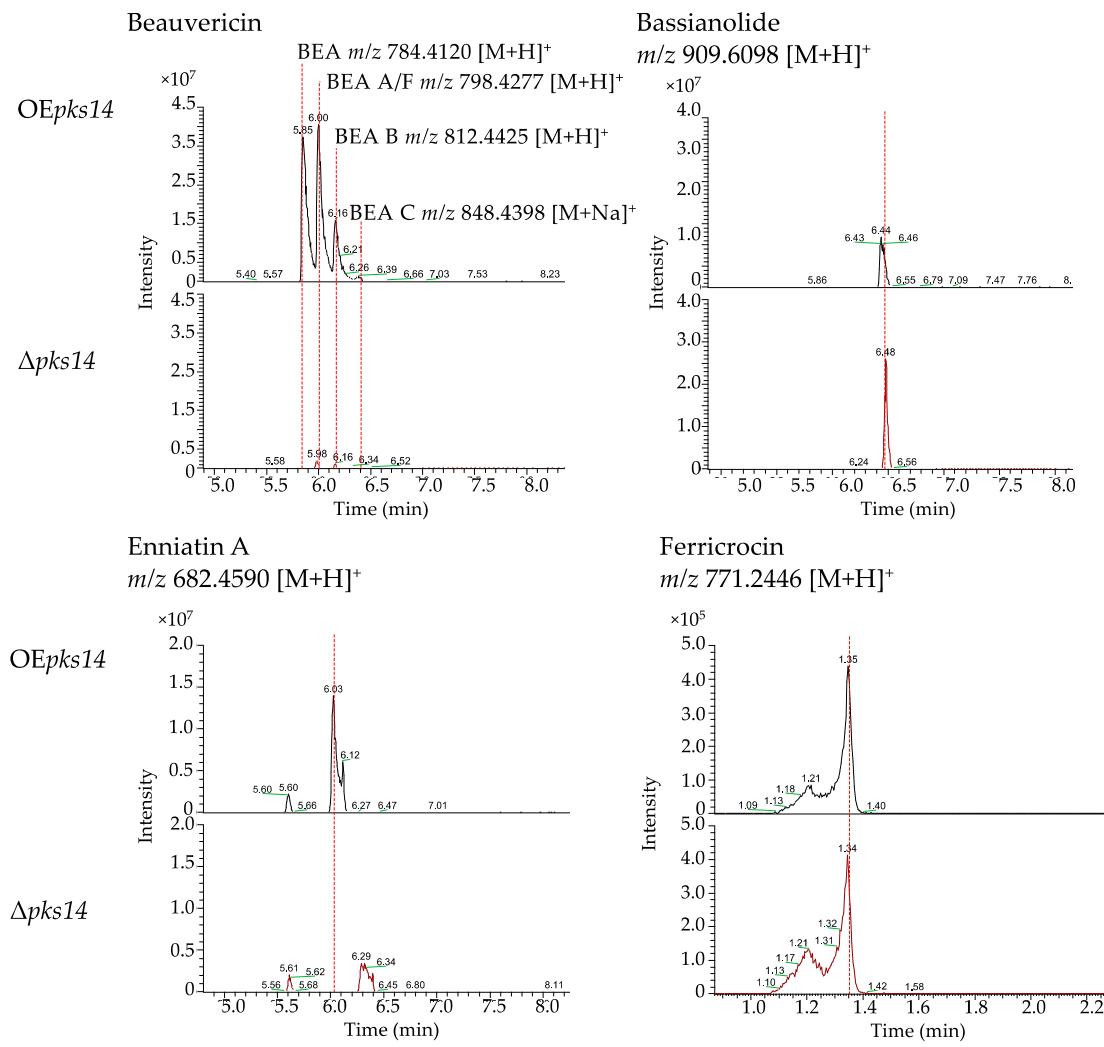


Figure S5. Comparison of full insecticides and siderophore MS profiles between OEpk14 and Δ pk14 in culture revealed up-regulation of beauvericin (BEA), beauvericin A/F (BEA A/F), beauvericin B (BEA B), beauvericin C (BEA C), bassianolide (BAS), and enniatin A (ENN A) in OEpk14 compared to Δ pk14 while no difference was seen for ferricrocin (FER).

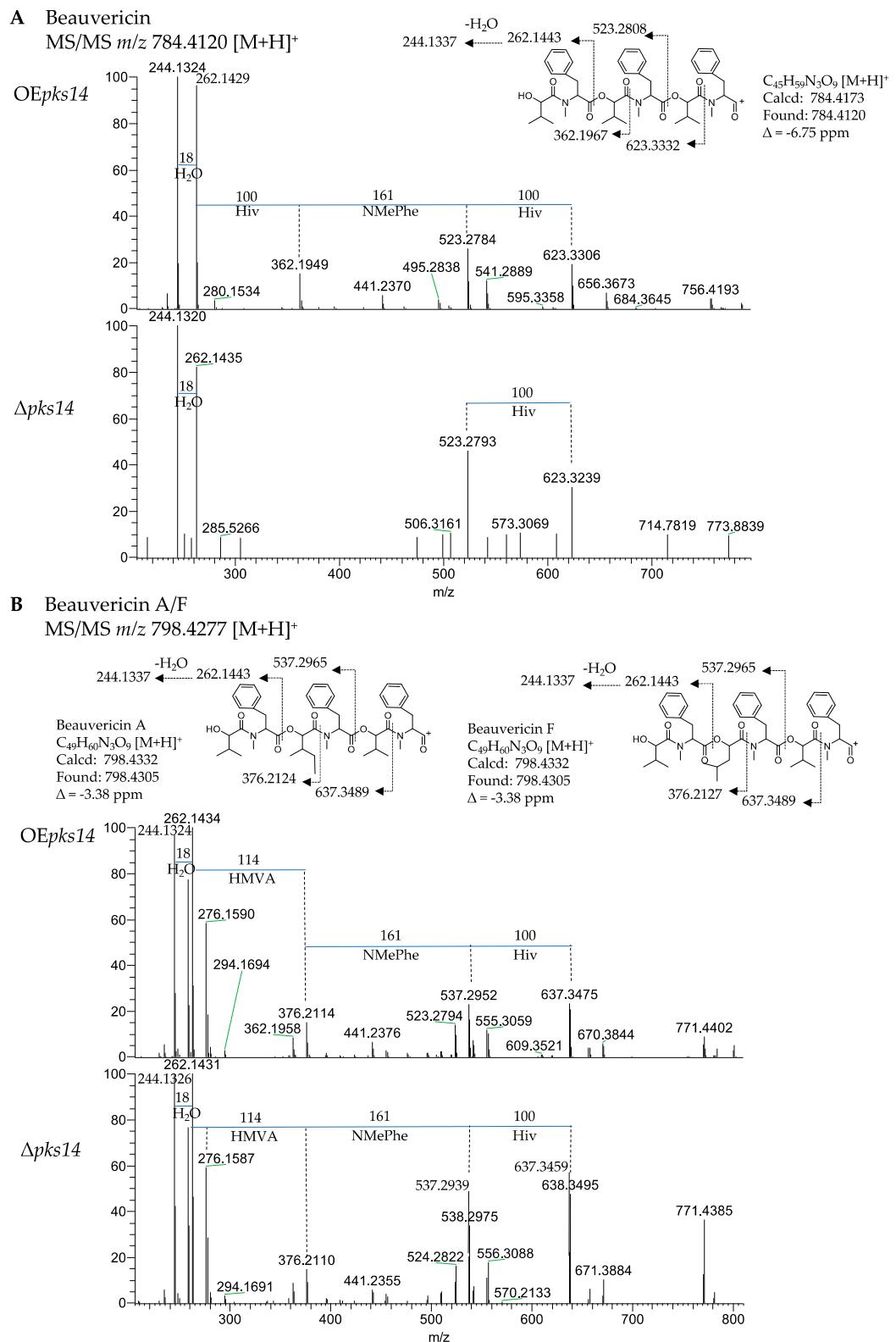


Figure S6. MS/MS spectra of (A) beauvericin (BEA), (B) beauvericin A/F (BEA A/F), (C) beauvericin B (BEA B), (D) beauvericin C (BEA C), (E) enniatin A (ENN A), (F) bassianolide (BAS), and (G) ferricrocin (FER) from OEpk14 and Δpk14 in culture. HIV = 2-hydroxyisovaleric acid, NMePhe = N-methylphenylalanine, HMVA = 2-hydroxy-3methylvaleric acid, NMelle = N-methylisoleucine, NMLeu = N-methyleucine, Gly = glycine, and L-Ser = L-serine.

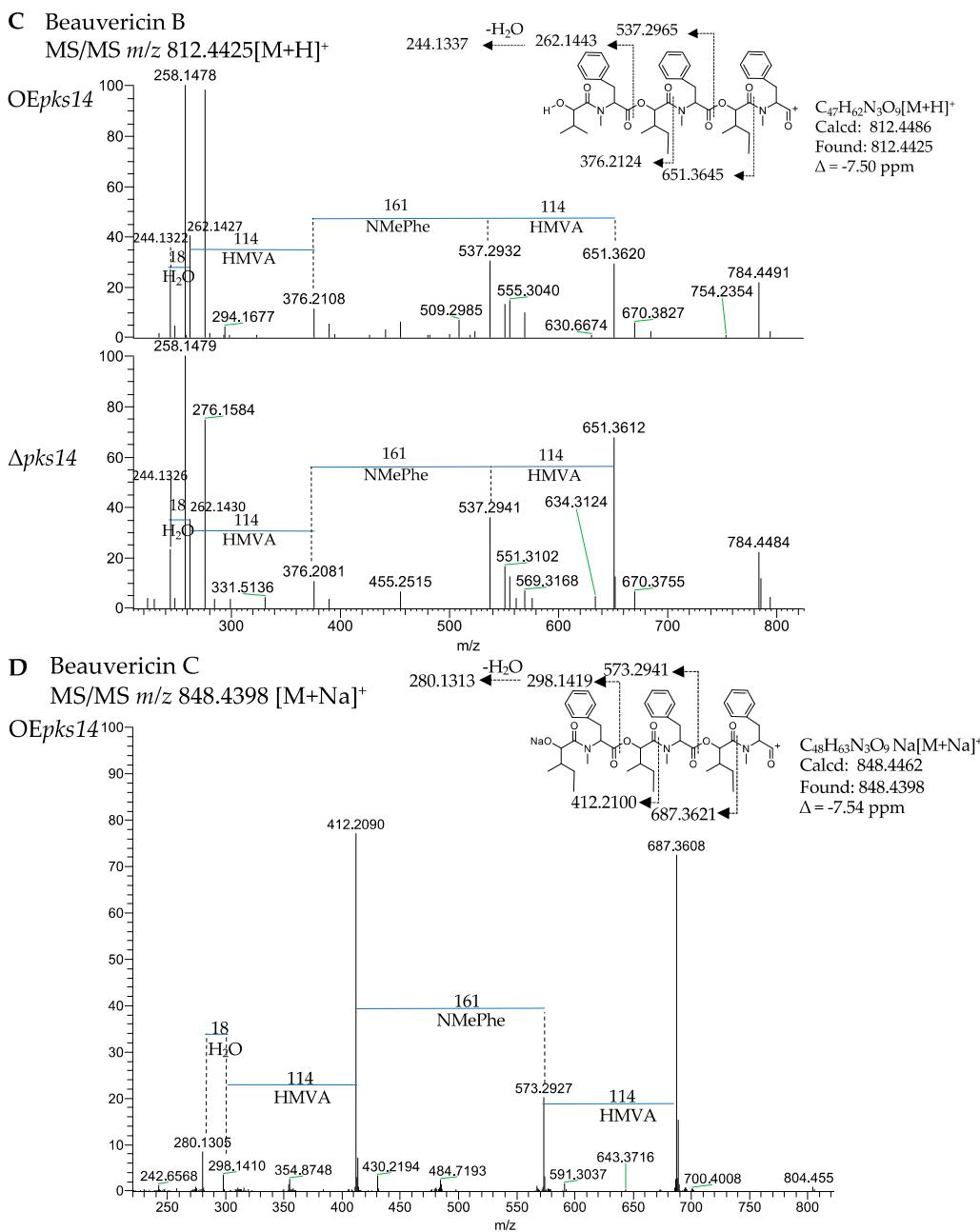


Figure S6.(continued)

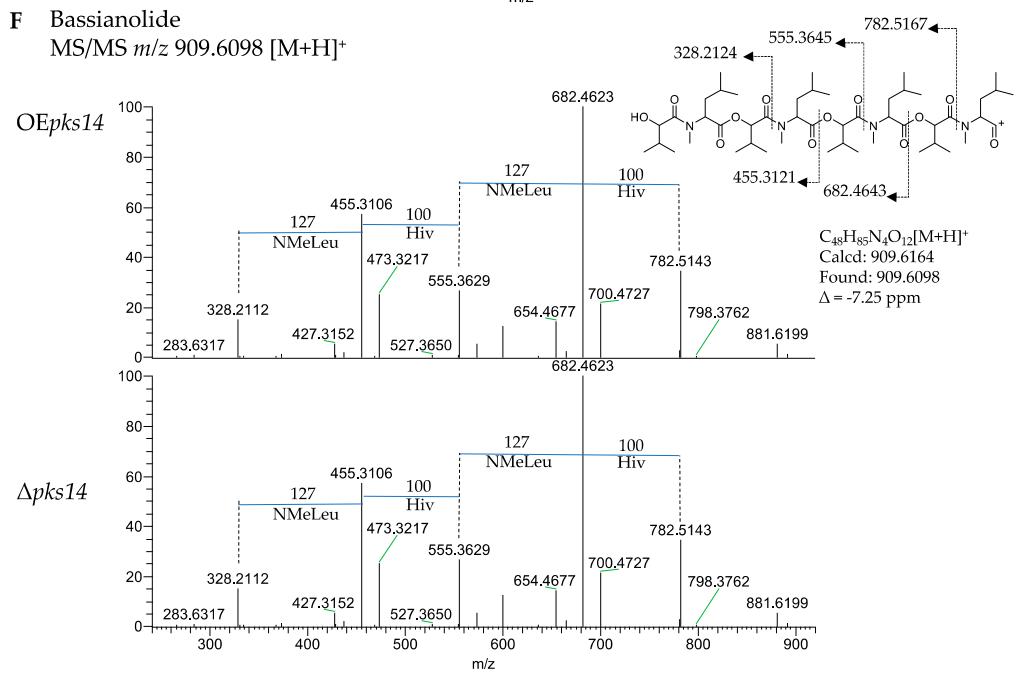
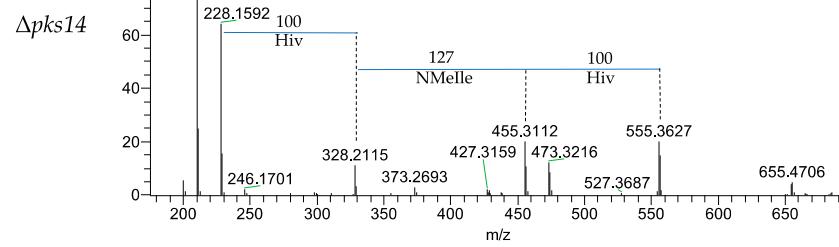
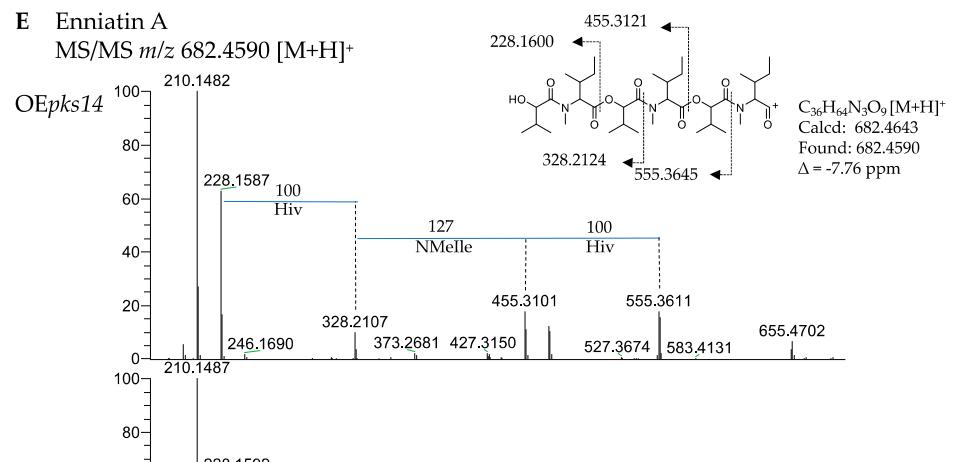


Figure S6. (continued)

G Ferricrocin
MS/MS m/z 771.2446 [M+H]⁺

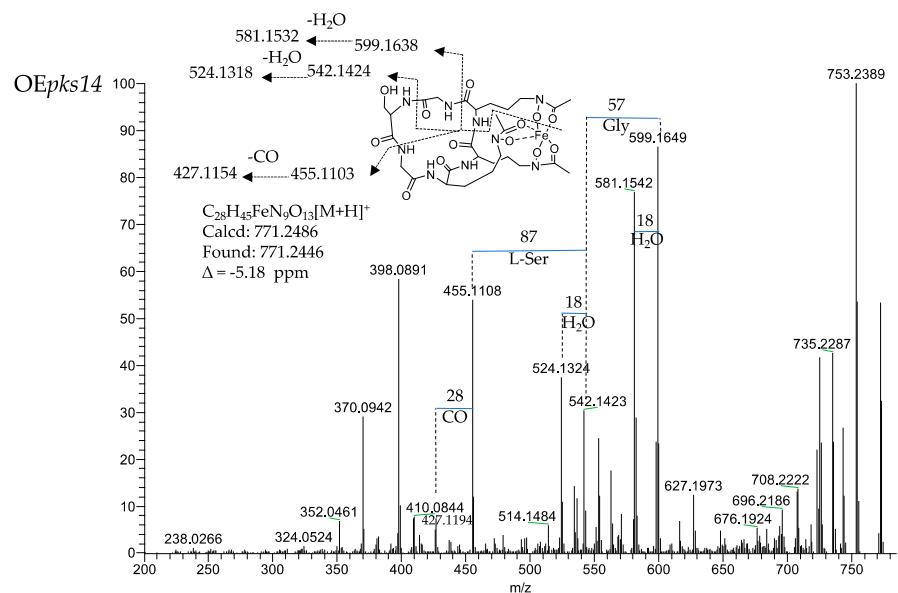


Figure S6. (continued)

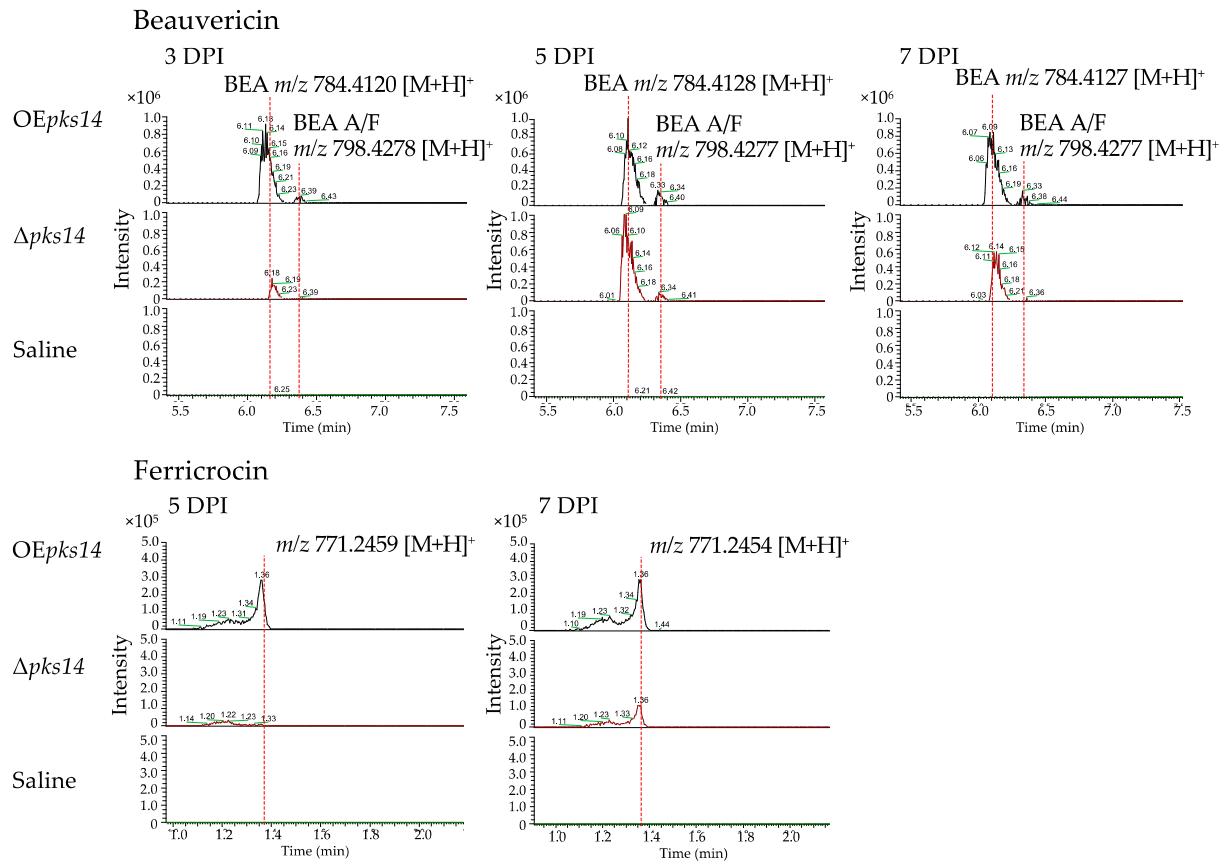


Figure S7. Comparison of full insecticides and siderophore MS profiles between OEpk14 and $\Delta pks14$ strains *in vivo* at early-stage infection (3 DPI) for live larvae, mid-stage infection (5 DPI) for dead larvae, and late-stage infection (7 DPI) for cadavers covered with fungal hyphae. Beauvericin (BEA), beauvericin A/F (BEA A/F), and ferricrocin (FER) were up-regulated in OEpk14 compared to $\Delta pks14$.

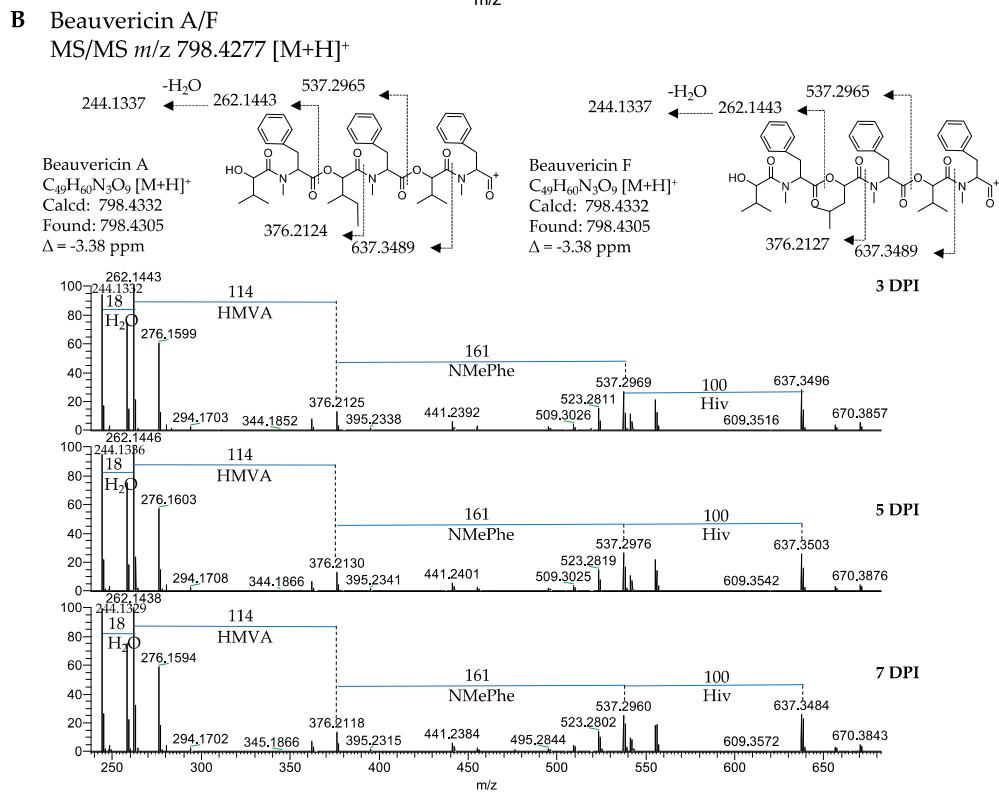
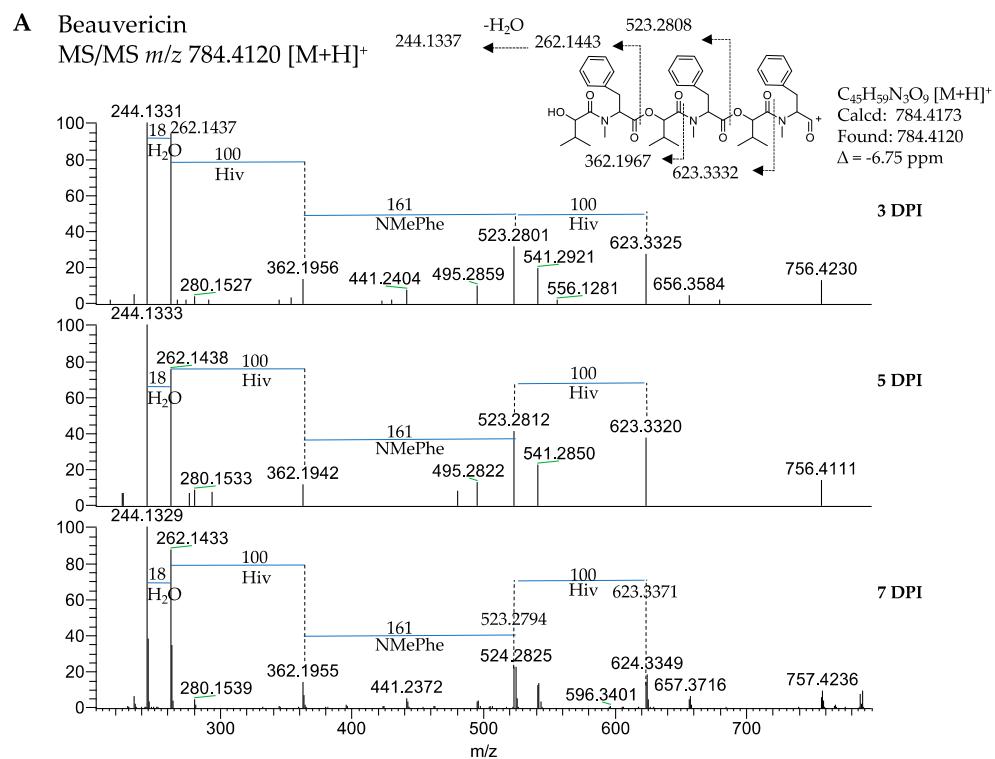


Figure S8. MS/MS spectra of (A) beauvericin (BEA), (B) beauvericin A/F (BEA A/F), and (C) ferricrocin (FER) from OEpk14 *in vivo* at early-stage infection (3 DPI) for live larvae, mid-stage infection (5 DPI) for dead larvae, and late-stage infection (7 DPI) for cadavers covered with fungal hyphae. HIV = 2-hydroxyisovaleric acid, NMePhe = N-methylphenylalanine, HMVA = 2-hydroxy-3methylvaleric acid, Gly = glycine, and L-Ser = L-serine.

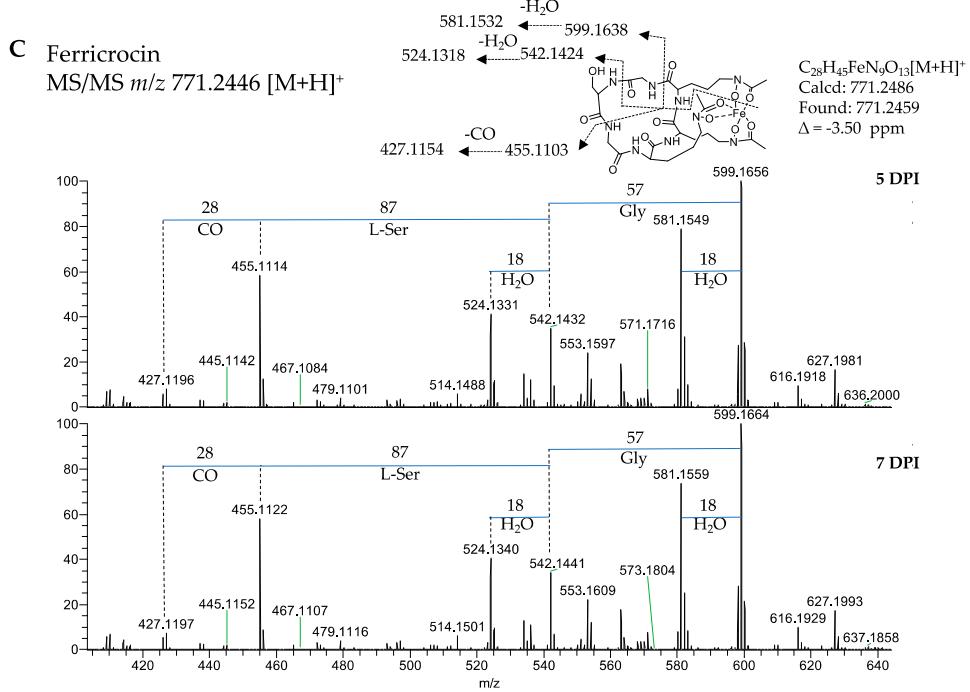


Figure S8. (continued)

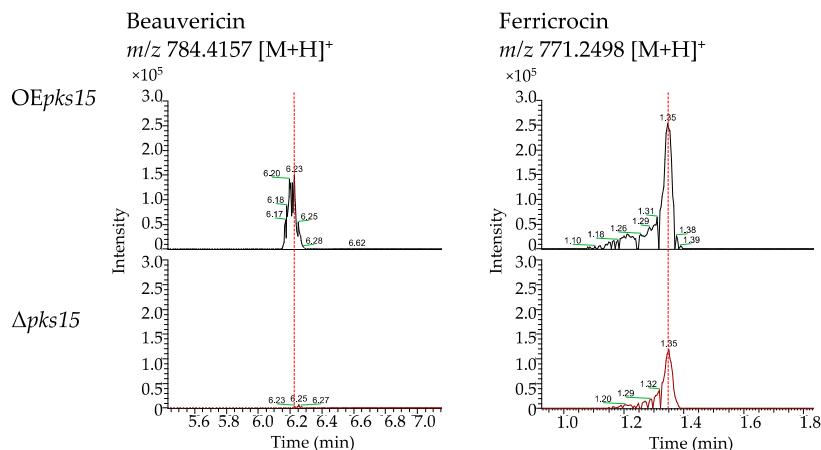


Figure S9. Comparison of insecticide and siderophore MS profiles between OEpks15 and Δ pks15 in culture revealed up-regulation of beauvericin (BEA) and ferricrocin (FER) in OEpks15 compared to Δ pks15.

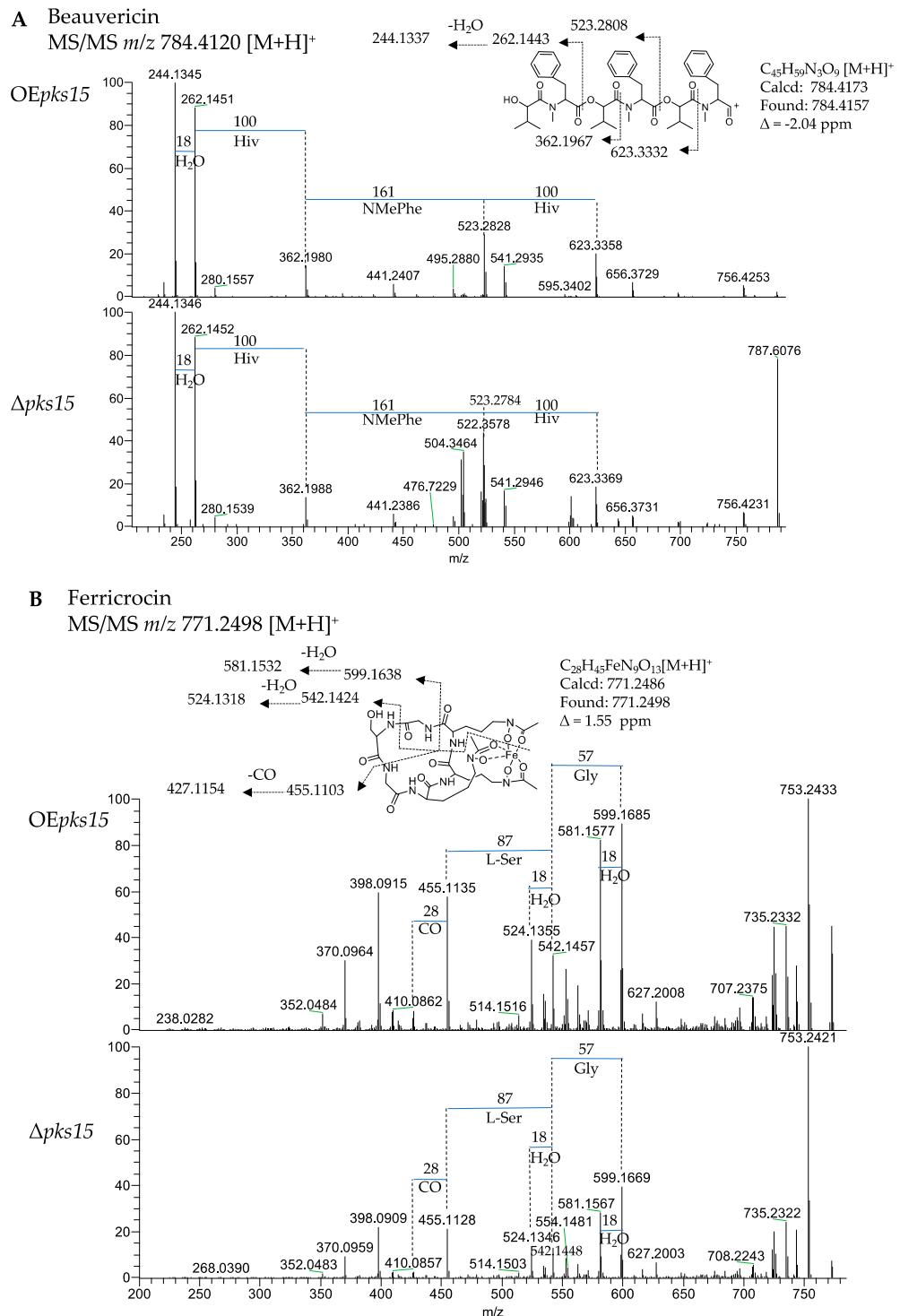


Figure S10. MS/MS spectra of (A) beauvericin (BEA) and (B) ferricrocin (FER) from OE $pks15$ and $\Delta pks15$ in culture. Hiv = 2-hydroxyisovaleric acid, NMePhe = N-methylphenylalanine, Gly = glycine, and L-Ser = L-serine.

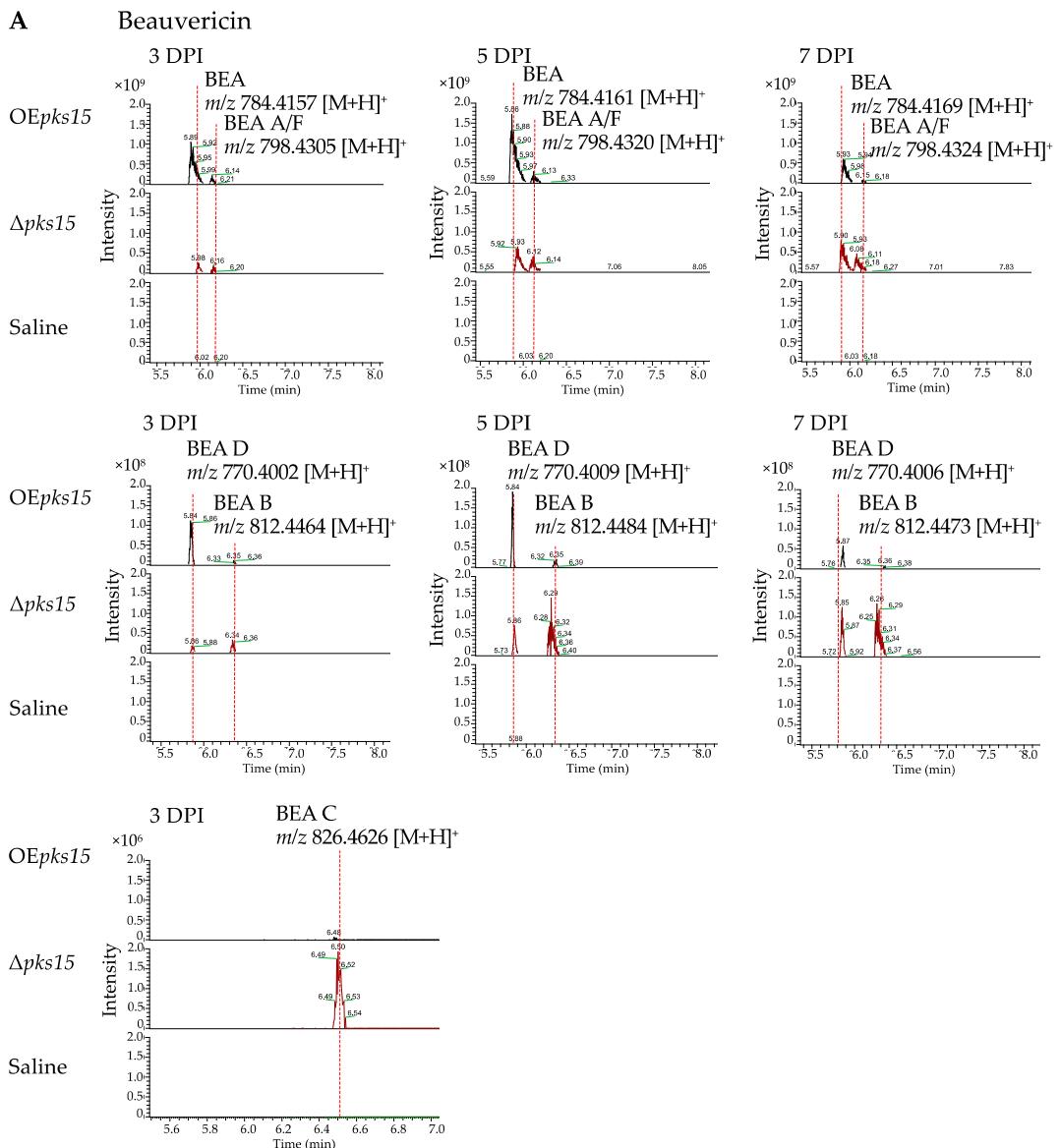


Figure S11. Comparison of insecticide and siderophore MS profiles between OE $pk15$ and $\Delta pk15$ *in vivo* at early-stage infection (3 DPI) for live larvae, mid-stage infection (5 DPI) for dead larvae, and late-stage infection (7 DPI) for cadavers covered with fungal hyphae. (A) Beauvericin (BEA), beauvericin A/F (BEA A/F), beauvericin B (BEA B), beauvericin C (BEA C), beauvericin D (BEA D), (B) bassianolide (BAS), and (C) ferricrocin (FER) were identified.

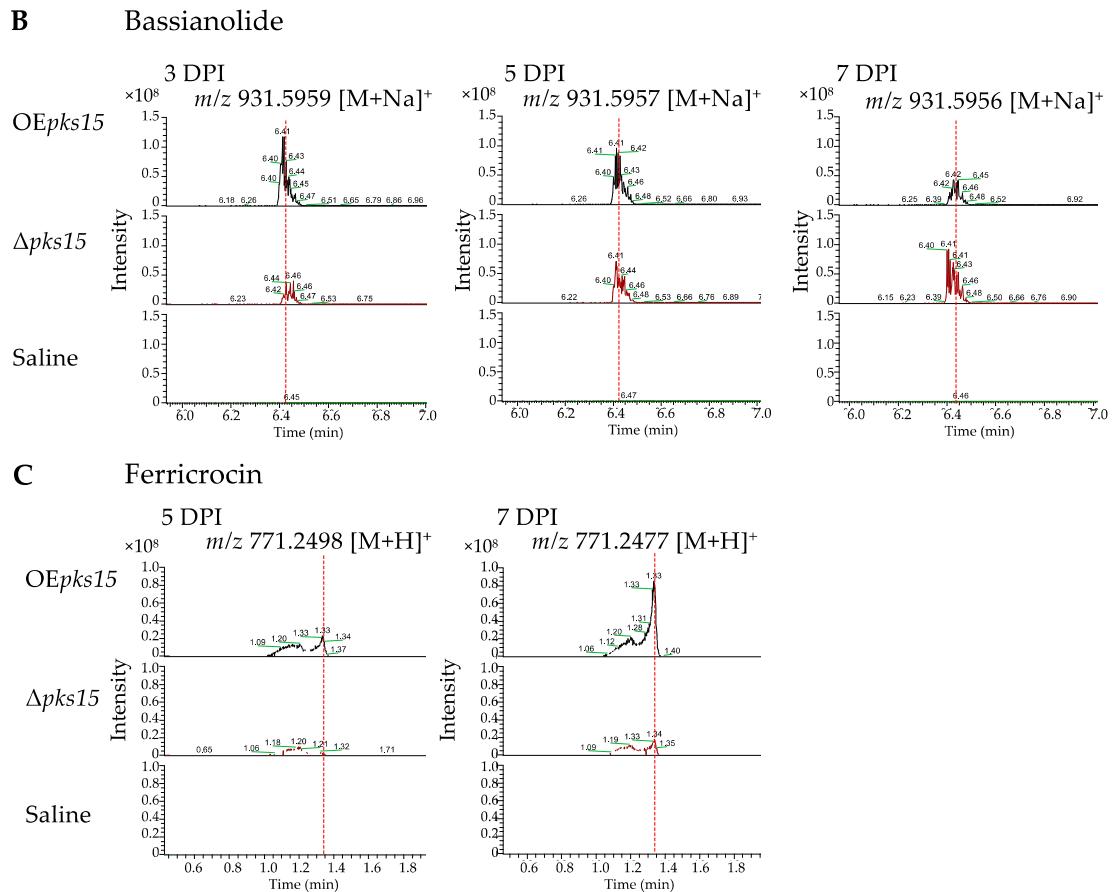


Figure S11. (continued)

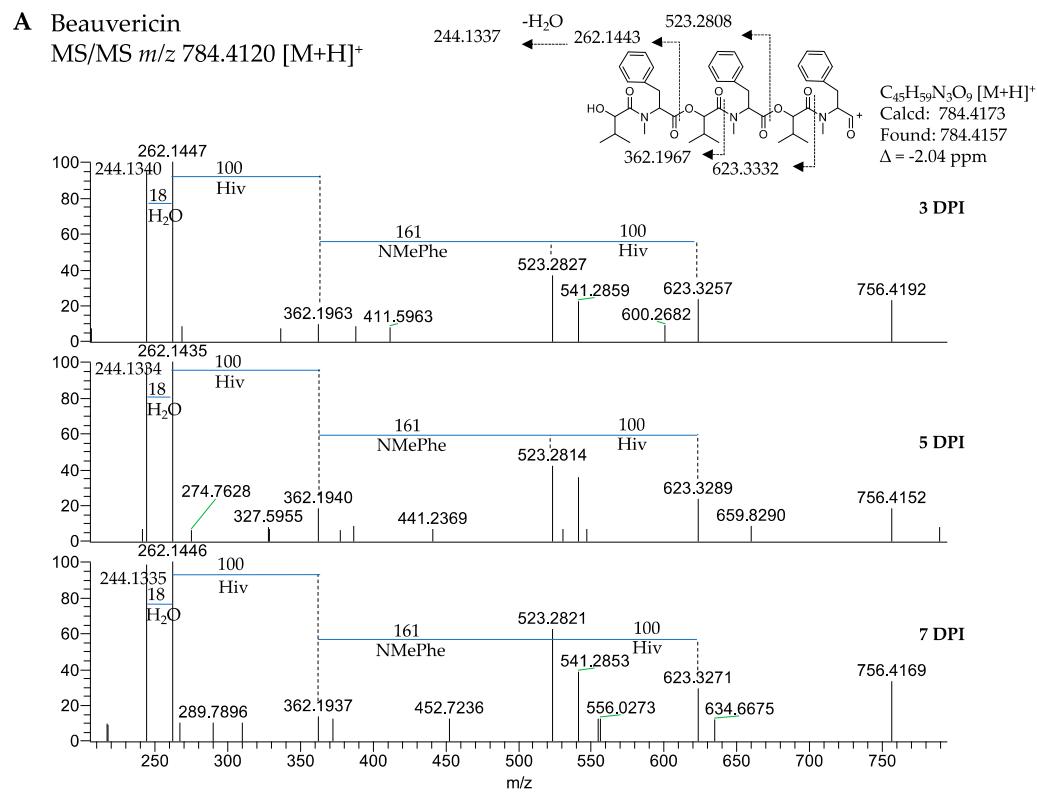


Figure S12. MS/MS spectra of (A) beauvericin (BEA), (B) beauvericin A/F (BEA A/F), (C) beauvericin B (BEA B), (E) beauvericin D (BEA D), (F) bassianolide (BAS), and (G) ferricrocin (FER) from OE^{pk15} and (D) beauvericin C (BEA D) from Δ pk15 *in vivo* at early-stage infection (3 DPI) for live larvae, mid-stage infection (5 DPI) for dead larvae, and late-stage infection (7 DPI) for cadavers covered with fungal hyphae. Hiv = 2-hydroxyisovaleric acid, NMePhe = N-methylphenylalanine, HMVA = 2-hydroxy-3methylvaleric acid, Gly = glycine, and L-Ser = L-serine.

B Beauvericin A/F
MS/MS m/z 798.4277 [M+H]⁺

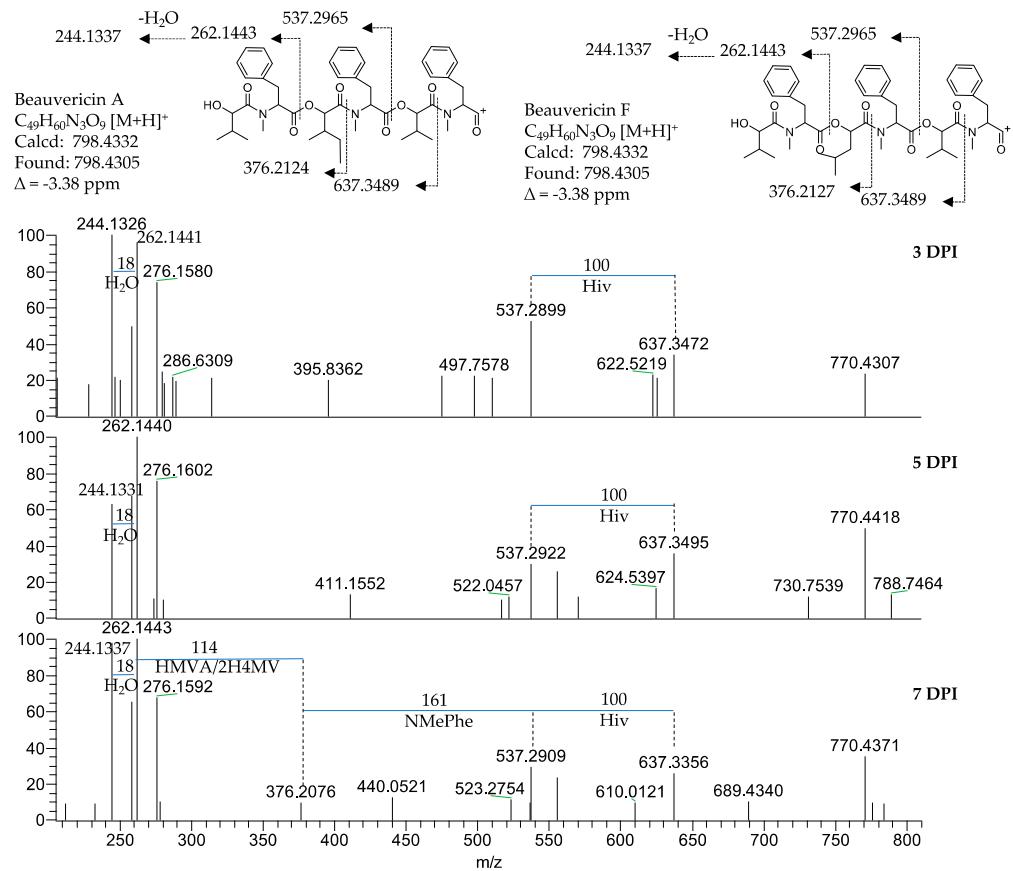


Figure S12. (continued)

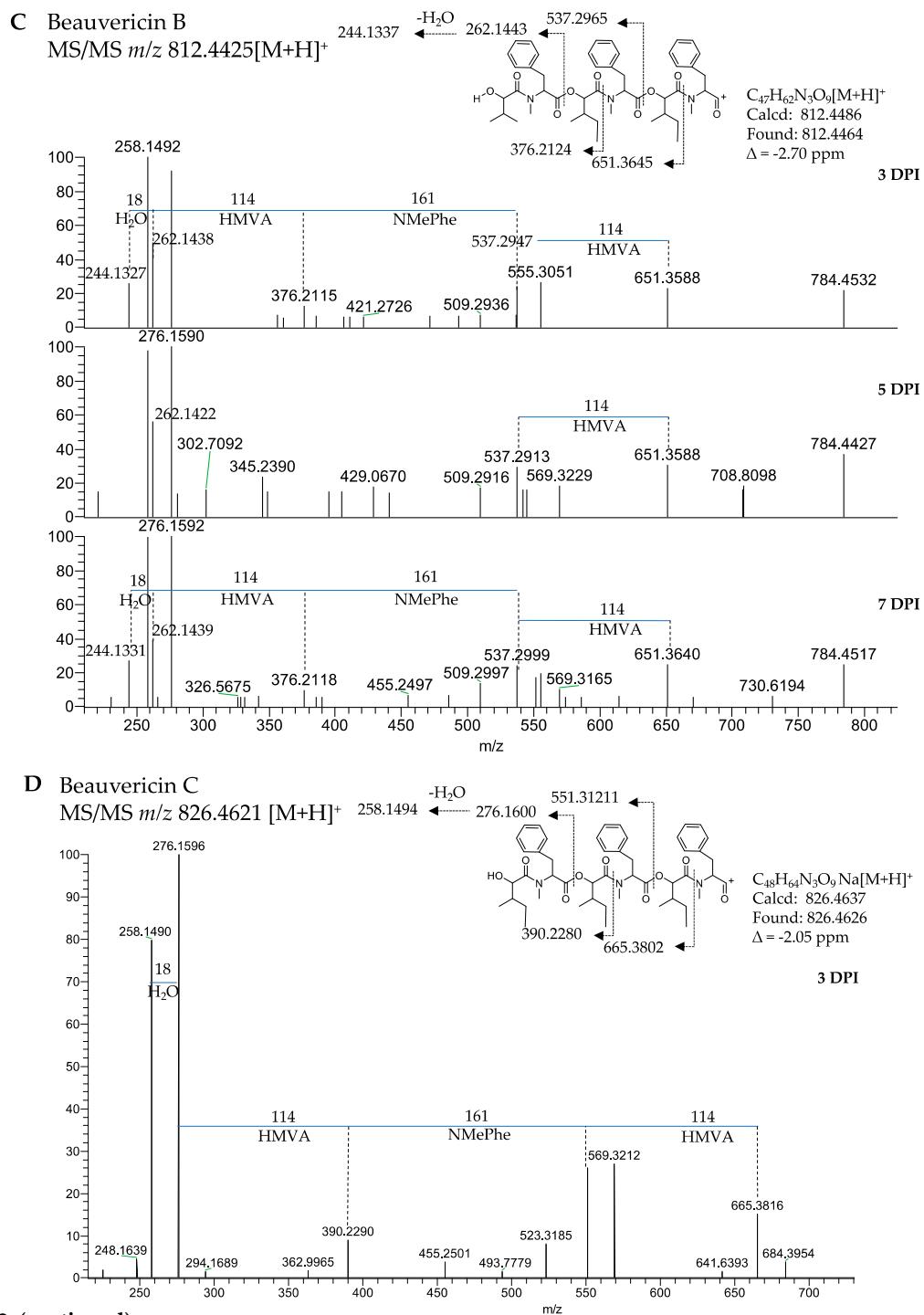


Figure S12. (continued)

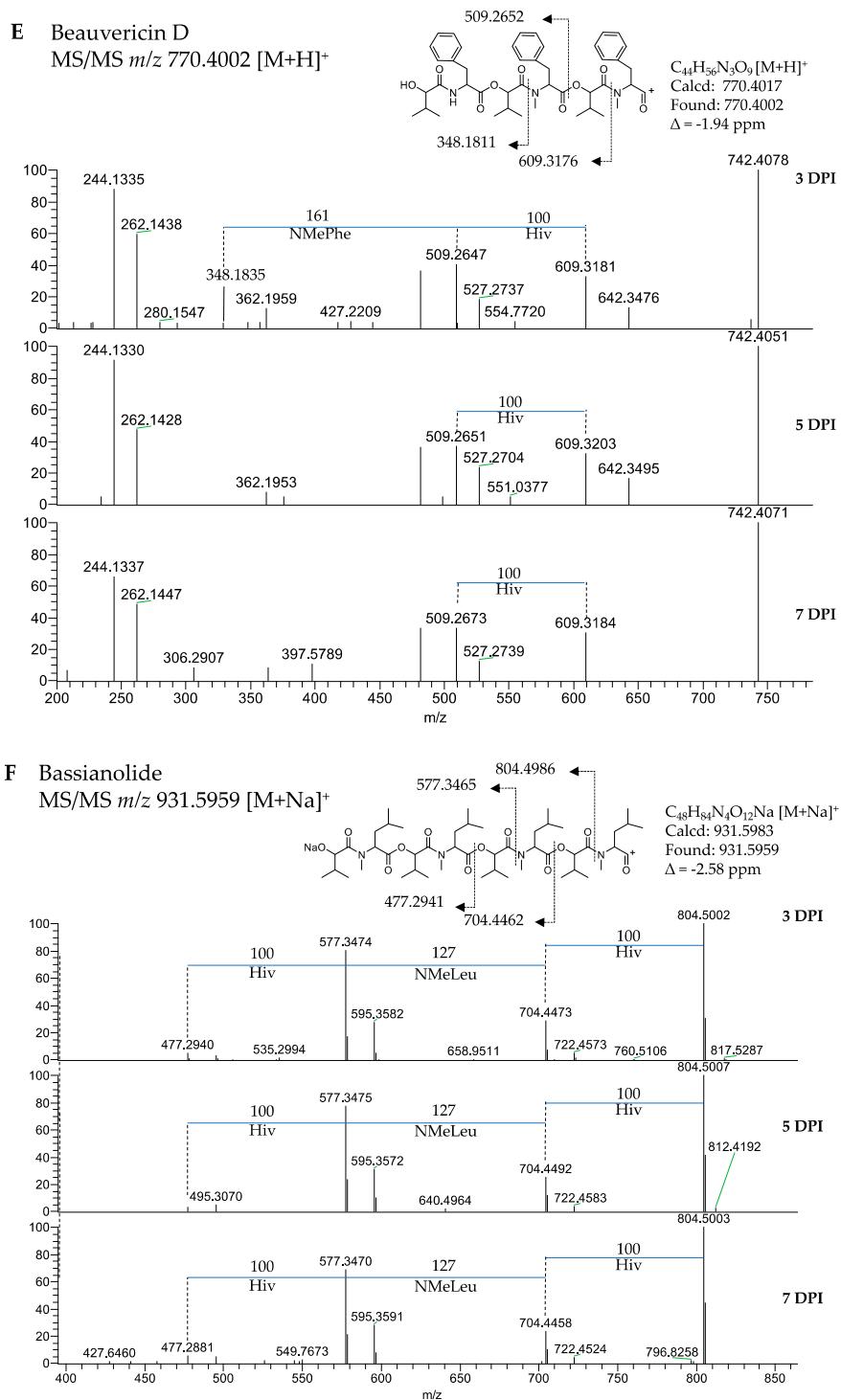


Figure S12. (continued)

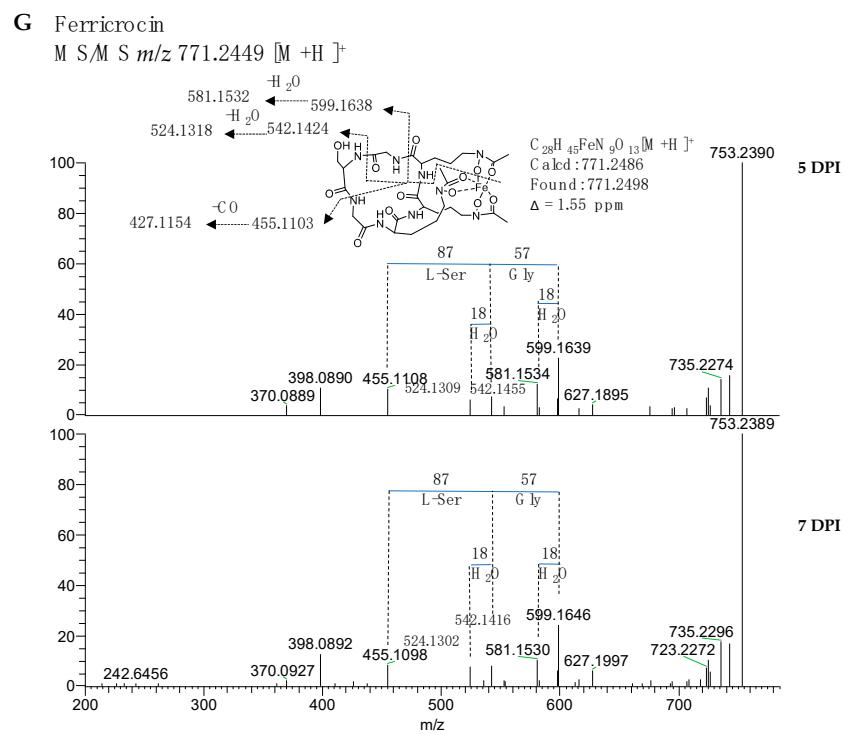


Figure S12. (continued)

A

PKS14 TCTTTATTAC TTGGCGTAAA CTCTCTTCAA GACGGGC.**CT** TGAACA.... .GAGAG
beauvericin TATTTCTGC AGC GTTTCGA GC**CTTGCCAA** CACAGGAGCT TGATCAAGCC AAGTGGG
Consensus TaTTTactTaC agcGcgTaaA cc**CTcgcCAA** c**ACaGGa**.**CT** TGAa**CA**.... .GaGaG

PKS14 GCG.....TA AAC....**TCT** CTTCAAGACGGGCC....TT GAACAGAGAG C.....
XM_008604818.1 ACGGGTGTTT TGCCCCG**TCT** CTTCGAGGTTGACCATGTTT GCGCATAATA AGTACTT
Consensus aCG.....Taa aC....**TCT** CTT**CAG**acg**GaCC**....TT GaaCAGaaaa a.....

PKS14 GATTG.CCT **AC..CGTC.G** AGTGC..**CAA** CGGCAAAAGA CTCGGTAC... .CACGC
bassianolide AGTACATACT GCAGCGTCTG CGTGC^{GG}GT CTGCGAGAGC GTAATATCTT CACATGC
Consensus aaTaCa.**act** aC..**CGTC.G** a**GTGC..caa** CgGCaAaAGa ctaagaaC... .CACGC

B

PKS15 TTGCATA TCATCATCAT TCATT^CATCC ATT^CATT^CAT TCATA^CATA. CATA^CATAAT
Beauvericin TCGCAGA CCAA. TTCTT TCTCTTGGCT GCACATTCT TCTTACCTCG CAATCATCAC
Consensus Tc**GCAGA** c**CAA.aTCaT** TCacTcagC^c aca**CATTcaT** TCa**TACaTa**. CAAa**CATAaC**

PKS15 T TGCATATCAT .CATCATTCA TTCATCCATT CATTCAT.TC ATACATACAT ACATA
XM_008604825 T TCCACATCCC GTAACGCTCA TACATTGTT ATATCAAGCC ATT^CGCTGCT TCGCT
Consensus T Tc**CACATCac** .ca**AcaCacTCA** Ta**CATCcATT** aaa**TCAA.cc** ATa**CacacaT** aCaca

PKS15 CATATCA**TCA** TCATT^CATTC ATCCATT^CAT TCATT^CATAC ATACATACAT AATTAGA
Bassianolide AAGAA. TTCA TCAATC.TAC ATC AGTCGT .CTTGCTTC ACGTCTCCTC CAGCCAT
Consensus aAgAa. a**TCA** T**CAaTC**.**TaC** ATC AgTCat .CaTT^Ca**TaC** AcacaTaCac aAgcaaa

PKS15 TGCA**TATCAT** CATCATT^CAT TCATT^CATTC ATT^CATT^CAT ACATACATAC ATAATTAA
XM_008597724.1 ACAC**TATTTT** CACCT^CTGC.T TCTTC^CACTTC ACTTC^CTCCT CTTGACA... GTACCCA
Consensus aca**TATcaT** CaCCaTgC.T TCa**TCAaTTC** AtC^Tca^TTCat acag**ACA**... aTAaccA

Figure S13. Similarities between the promoters of genes (A) *pk14* or (B) *pk15* with those of genes in the beauvericin and bassianolide biosynthetic clusters.