

New Pseudomonas Bacterial Strains: Biological Activity and Characteristic Properties of Metabolites

Tatiana M. Sidorova¹, **Natalia S. Tomashevich**^{1,*}, **Valeria V. Allahverdyan**¹, **Boris S. Tupertsev**^{2,3}, **Yuri I. Kostyukevich**³ and **Anzhela M. Asaturova**¹

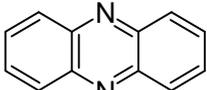
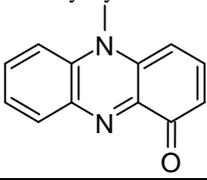
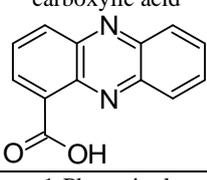
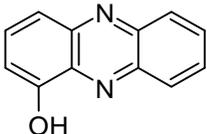
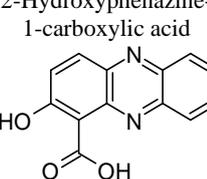
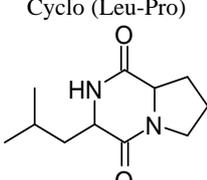
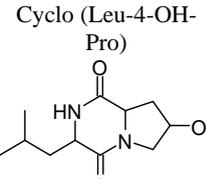
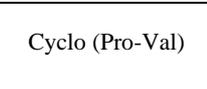
¹Federal Research Center of Biological Plant Protection, 350039 Krasnodar, Russia

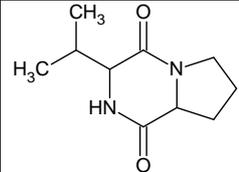
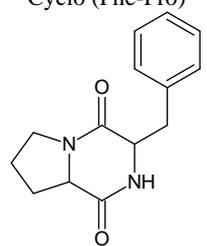
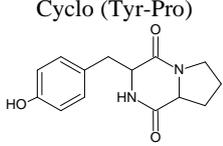
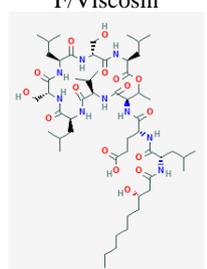
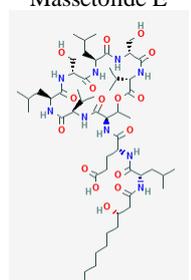
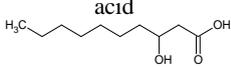
²Phystech School of Biological and Medical Physics (FBMF), Moscow Institute of Physics and Technology, 141701 Dolgoprudny, Russia

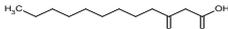
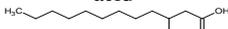
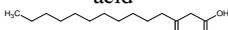
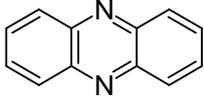
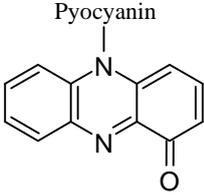
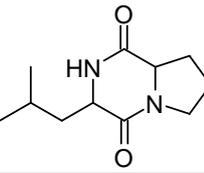
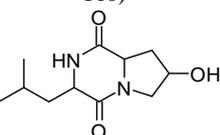
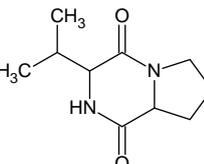
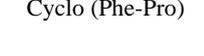
³Center of Molecular and Cellular Biology (CMCB), Skolkovo Institute of Science and Technology, 121205 Moscow, Russia

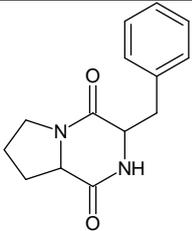
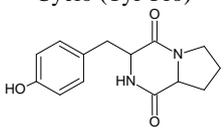
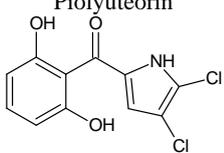
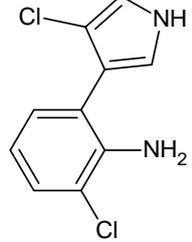
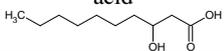
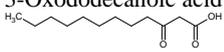
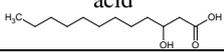
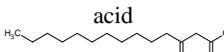
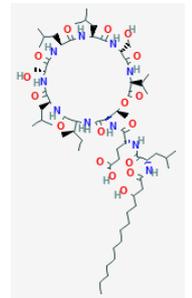
* Correspondence: tom-s2@yandex.ru; Tel.: +7-92-8038-8165

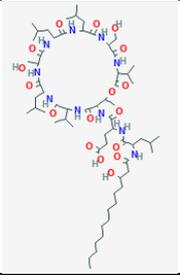
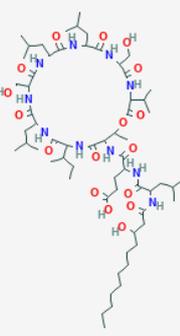
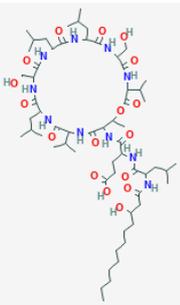
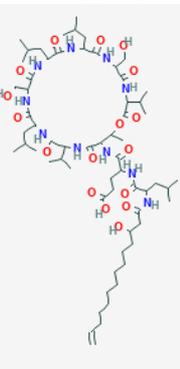
Table S1. The results of the analysis of *P. chlororaphis* BZR 245-F and *Pseudomonas* sp. BZR 523-2. C.u , are area conventional units obtained by Q Exactive Orbitrap mass spectrometer

Sample	Assignment, Structure	Molecular Formula			RT, min	Area, c.u., amount
		m/z _{ob} (Ionization Mode)	m/z _{th}	Error (ppm)		
245	Phenazine 	C ₁₂ H ₈ N ₂			5.07	2396173
		181.0760 (+)	181.0760	0		0.07 mg/g
	Pyocyanin 	C ₁₃ H ₁₀ N ₂ O			5.46	5126746
		211.0864 (+)	211.0864	0		0.16 mg/g
	Phenazine-1- carboxylic acid 	C ₁₃ H ₈ N ₂ O ₂			5.01	104904177
		225.0660 (+)	225.0659	0.4		3.22 mg/g
	1-Phenazinol 	C ₁₂ H ₈ N ₂ O			4.20	300012231
		197.0709 (+)	197.0709	0		11.27 mg/g
	2-Hydroxyphenazine- 1-carboxylic acid 	C ₁₃ H ₈ N ₂ O ₃			4.73	74811412
		241.0609 (+)	241.0608	0.4		2.26 mg/g
Phenazine-1- carboxamide 	C ₁₃ H ₉ N ₃ O			4.52	282099	
	224.0816 (+)	224.0818	-0.9		14.8 mg/g	
Cyclo (Leu-Pro) 	C ₁₁ H ₁₈ N ₂ O ₂			2.49	271658961	
	211.1441 (+)	211.1441	0			
Cyclo (Leu-4-OH- Pro) 	C ₁₁ H ₁₈ N ₂ O ₃			1.43	24334107	
	227.1389 (+)	227.1390	-0.4			
Cyclo (Pro-Val) 	C ₁₀ H ₁₆ N ₂ O ₂			1.22	18127820	

	197.1283 (+)	197.1285	-1.0		
Cyclo (Phe-Pro) 	$C_{14}H_{16}N_2O_2$			2.85	297513831
	245.1285 (+)	245.1285	0	3.04	199652217
					Sum 499190896
Cyclo (Tyr-Pro) 	$C_{14}H_{16}N_2O_3$			1.06	25762123
	261.1234 (+)	261.1234	0	1.11	28104244
				1.92	4903739
				2.13	14954527
					Sum 73724633
Massetolide F/Viscosin 	$C_{54}H_{95}N_9O_{16}$				
	1126.6975 (+)	1126.6970	0.4	11.98	2287283
Massetolide F/Viscosin 	$C_{54}H_{95}N_9O_{16}$				
	1126.6951 (+)	1126.6970	-1.7	12.09	371552
Massetolide E 	$C_{53}H_{93}N_9O_{16}$				
	1112.6814 (+)	1112.6813	0.1	11.68	344023
3-Hydroxydecanoic acid 	$C_{10}H_{20}O_3$				
	187.1338 (-)	187.1340	-1.1	6.38	24706173

	3-Oxododecanoic acid 	C ₁₂ H ₂₂ O ₃			7.49	23474555
		213.1499 (-)	213.1496	1.4		
	3-Hydroxydodecanoic acid 	C ₁₂ H ₂₄ O ₃			8.11	29080139
		215.1656 (-)	215.1653	1.4		
	3-Oxotetradecanoic acid 	C ₁₄ H ₂₆ O ₃			8.77	13172823
		241.1814 (-)	241.1809	2.1		
523	Phenazine 	C ₁₂ H ₈ N ₂			5.05	73450415 2.77 mg/g
		181.0760	181.0760	0		
	Pyocyanin 	C ₁₃ H ₁₀ N ₂ O			5.47	3482940 0.14 mg/g
		211.0864 (+)	211.0864	0		
	Cyclo (Leu-Pro) 	C ₁₁ H ₁₈ N ₂ O ₂			2.48	343763264
		211.1441 (+)	211.1441	0		
	Cyclo (Leu-4-OH-Pro) 	C ₁₁ H ₁₈ N ₂ O ₃			1.43	36169955
	227.1389 (+)	227.1388	0.4			
Cyclo (Pro-Val) 	C ₁₀ H ₁₆ N ₂ O ₂			1.22	23544170	
	197.1283	197.1285	-1.0			
Cyclo (Phe-Pro) 	C ₁₄ H ₁₆ N ₂ O ₂			2.85	374 609 480	
	245.1285 (+)	245.1285	0	3.04	246 593 042 Sum	

					621202523
Cyclo (Tyr-Pro) 	$C_{14}H_{16}N_2O_3$			1.04 1.13 1.93 2.11	34 527 281 37 763 732 6 023 925 23 816 480 Sum 102131419
Piolyuteorin 	$C_{11}H_7Cl_2NO_3$				
	261.1234	261.1234	0		
3-Chloro-4(2-amino-3-chlorophenyl)-pyrrole 	$C_{10}H_8Cl_2N_2$				
	271.9876 (+)	271.9876	0	5.05	84541796
3-hydroxydecanoic acid 	$C_{10}H_{20}O_3$				
	187.1338 (-)	187.1340	-1.1	6.38	5170154
3-Oxododecanoic acid 	$C_{12}H_{22}O_3$				
	213.1499 (-)	213.1496	1.4	7.48	15738884
3-Hydroxydodecanoic acid 	$C_{12}H_{24}O_3$				
	215.1656 (-)	215.1653	1.4	8.11	20547749
3-Oxotetradecanoic acid 	$C_{14}H_{26}O_3$				
	241.1814 (-)	241.1809	2.1	8.77	6050246
Orfamide A 	$C_{64}H_{114}N_{10}O_{17}$				
	1295.8421 (+)	1295.8436	-1.2	13.87	11559958
Orfamide B	$C_{63}H_{112}N_{10}O_{17}$				
	1281.8271 (+)	1281.8280	-0.7	13.32	11334776

					
<p data-bbox="405 427 525 454">Orfamide C</p> 	$C_{62}H_{110}N_{10}O_{17}$			13.00	897960
1267.8113 (+)	1267.8123	-0.8			
<p data-bbox="405 790 525 817">Orfamide D</p> 	$C_{61}H_{108}N_{10}O_{17}$			12.48	644091
1253.7960 (+)	1253.7967	-0.6			
<p data-bbox="405 1122 525 1149">Orfamide E</p> 	$C_{63}H_{110}N_{10}O_{17}$			12.87	221483
1279.8116 (+)	1279.8123	-0.5			

RT: 0.00 - 10.35

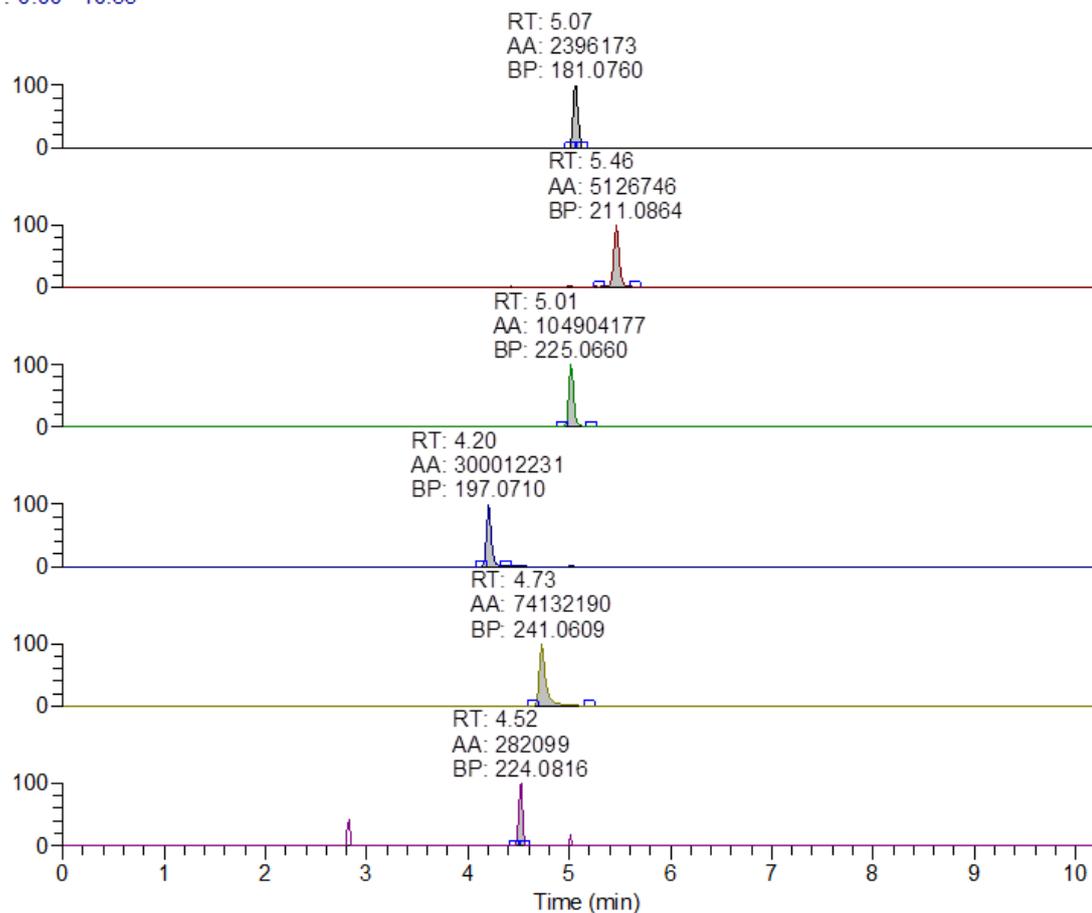


Figure S1 – Base Peak mass chromatograms of *P. chlororaphis* BZR 245-F with m/z 181.0760 (Phenazine), 211.0864 (Pyocyanin), 225.0659 (Phenazine-1-carboxylic acid), 197.0710 (1-Phenazinol), 241.0608 (2-Hydroxyphenazine-1-carboxylic acid), 224.0818 (Phenazine-1-carboxamide)

RT: 0.00 - 10.48

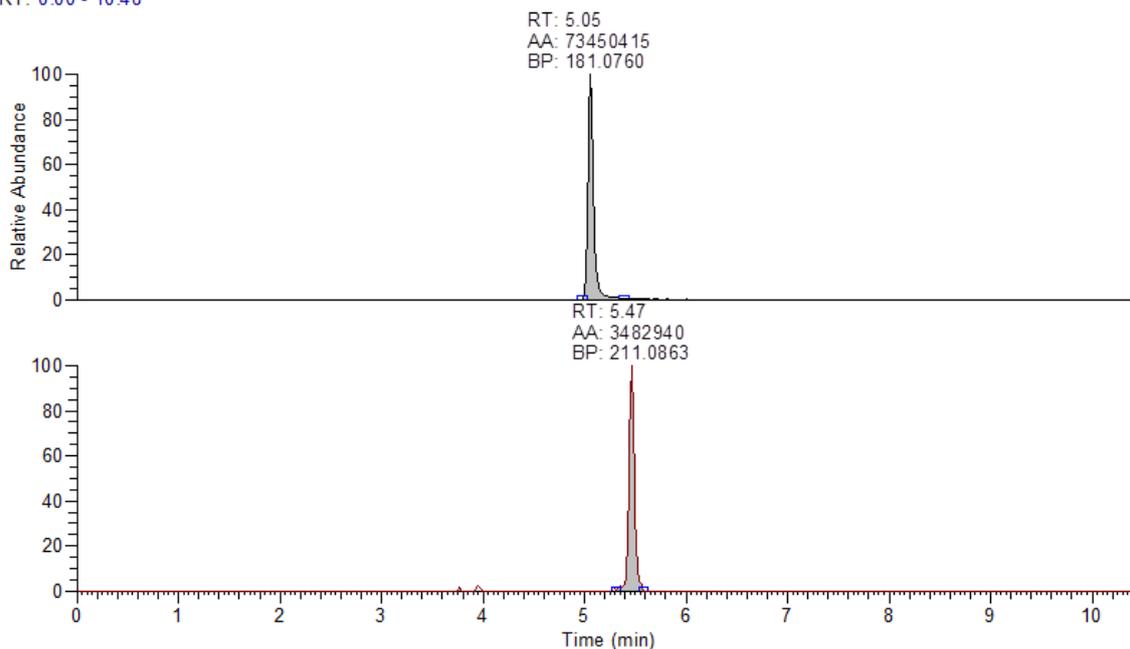


Figure S2 – Base Peak mass chromatograms of *Pseudomonas* sp. BZR 523-2 with m/z 181.0760 (Phenazine), 211.0864 (Pyocyanin)

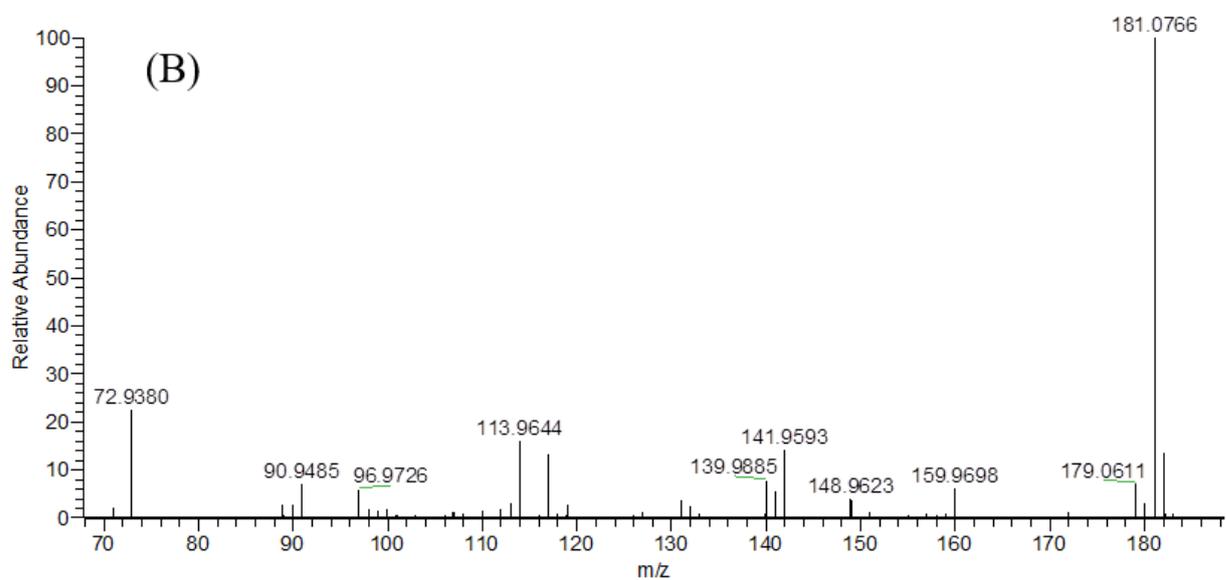
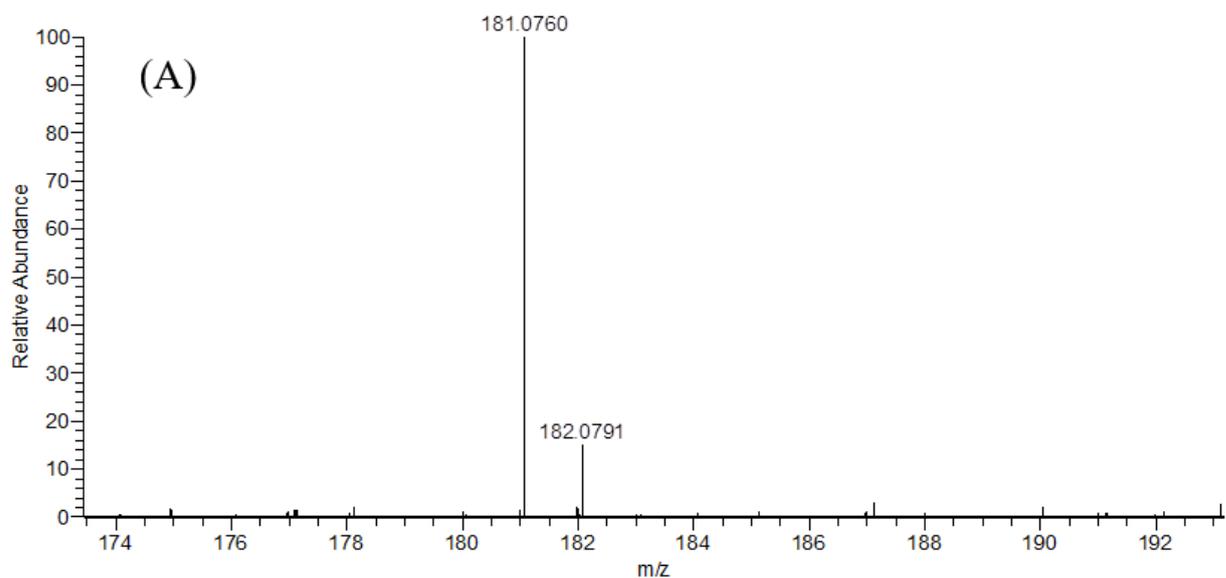


Figure S3 – MS (A) and MS/MS (B) spectra of metabolite with RT 5.05 min from *P. chlororaphis* BZR 245-F. The presented spectra are in positive-ion electrospray ionization mode and equal to those from *Pseudomonas* sp. BZR 523-2 sample

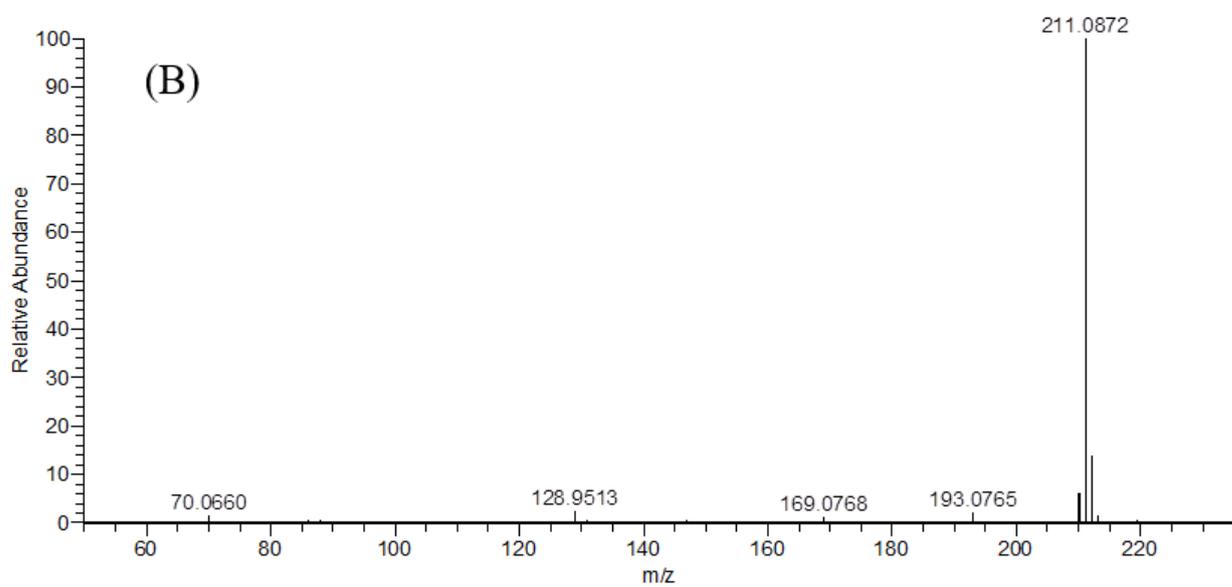
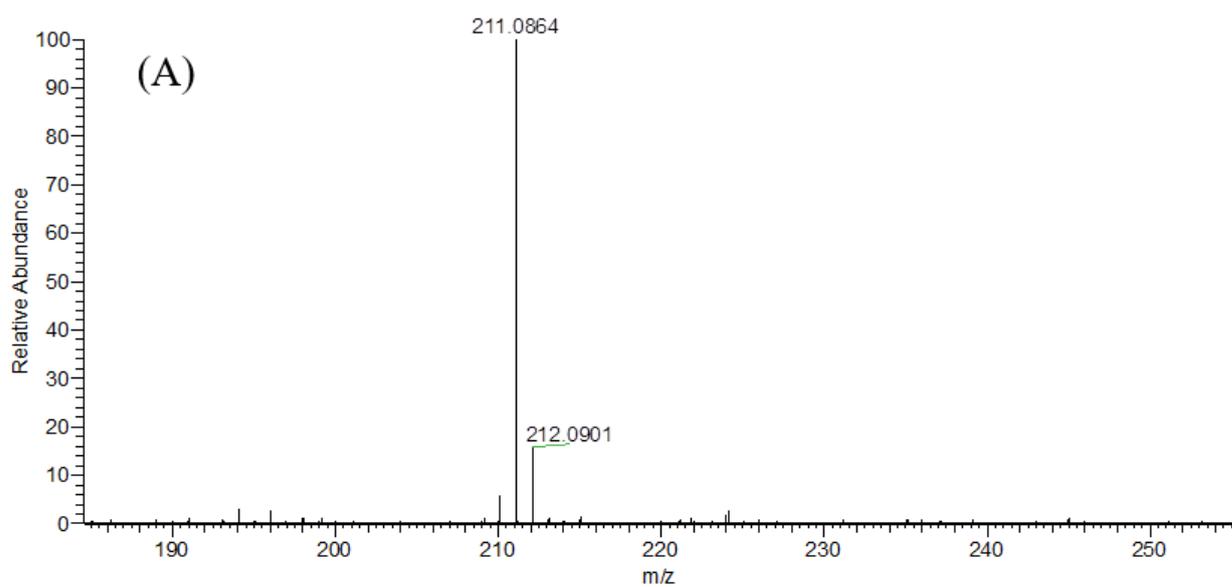


Figure S4 – MS (A) and MS/MS (B) spectra of metabolite with RT 5.45 min from *P. chlororaphis* BZR 245-F. The presented spectra are in positive-ion electrospray ionization mode and equal to those from *Pseudomonas* sp. BZR 523-2 sample

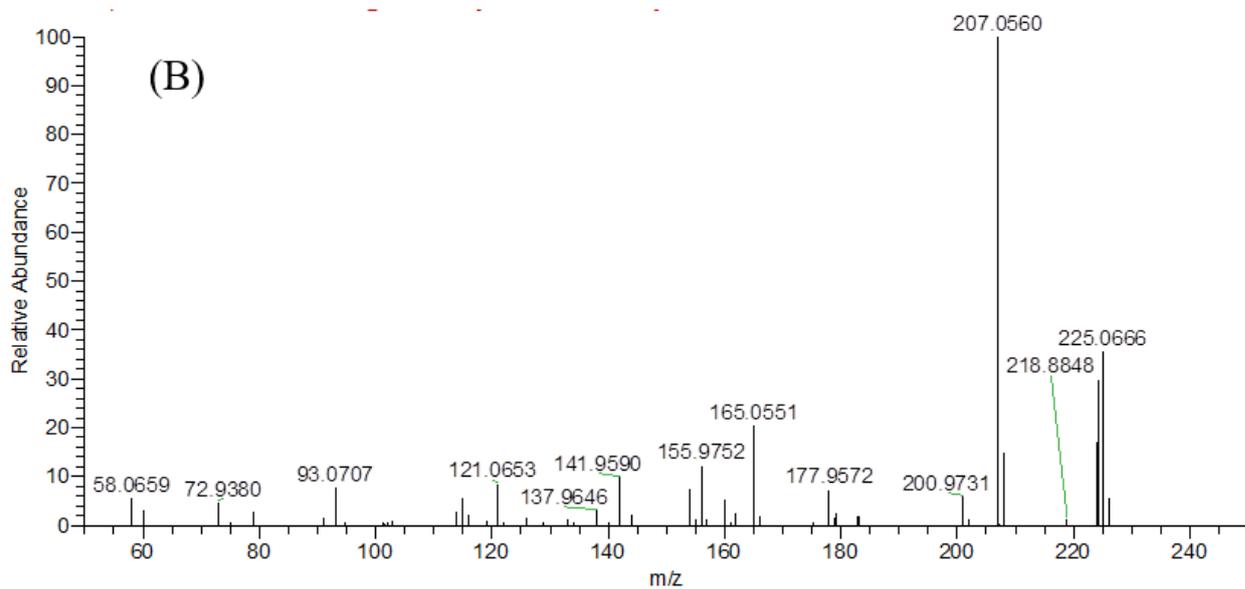
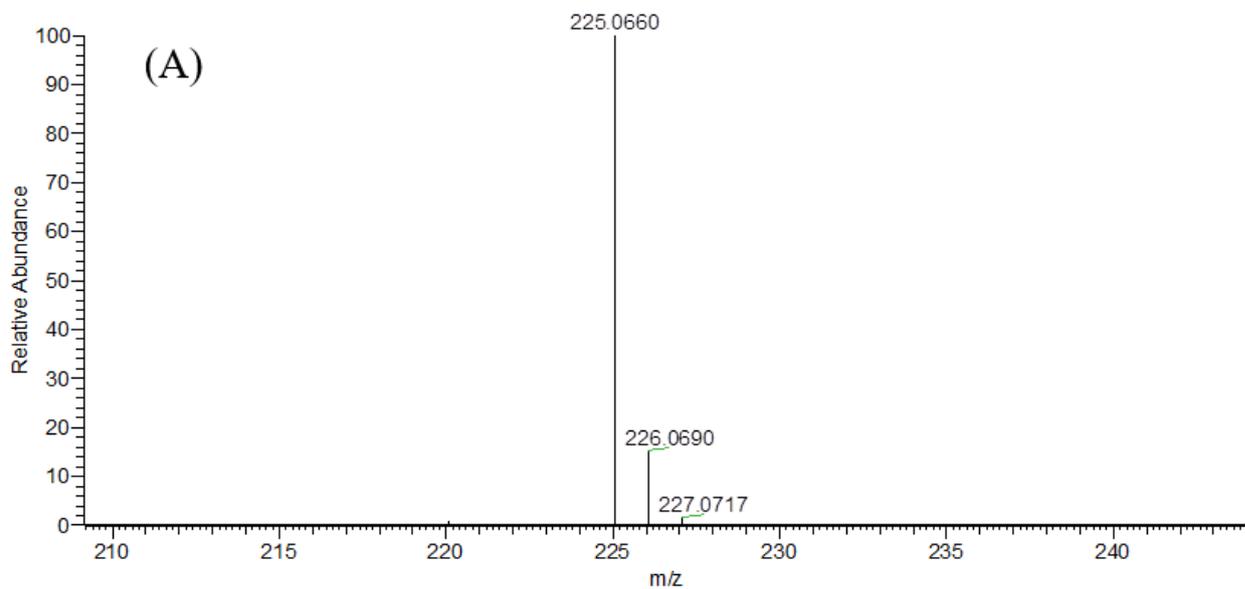


Figure S5 – MS (A) and MS/MS (B) spectra of metabolite with RT 5.01 min from *P. chlororaphis* BZR 245-F. The presented spectra are in positive-ion electrospray ionization mode

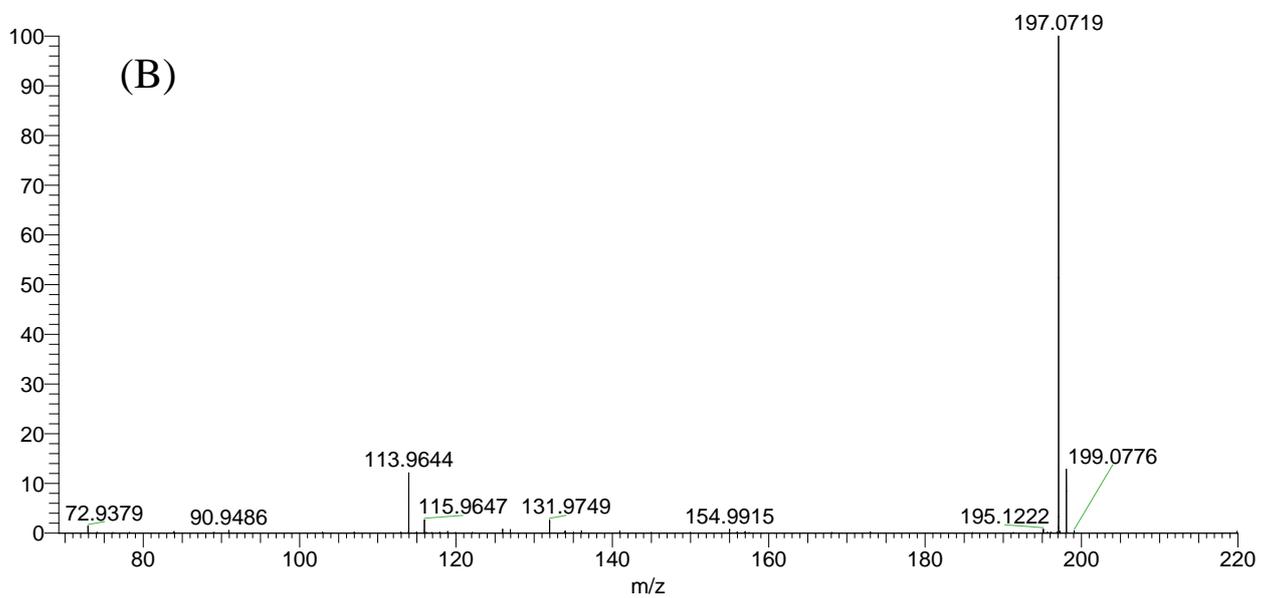
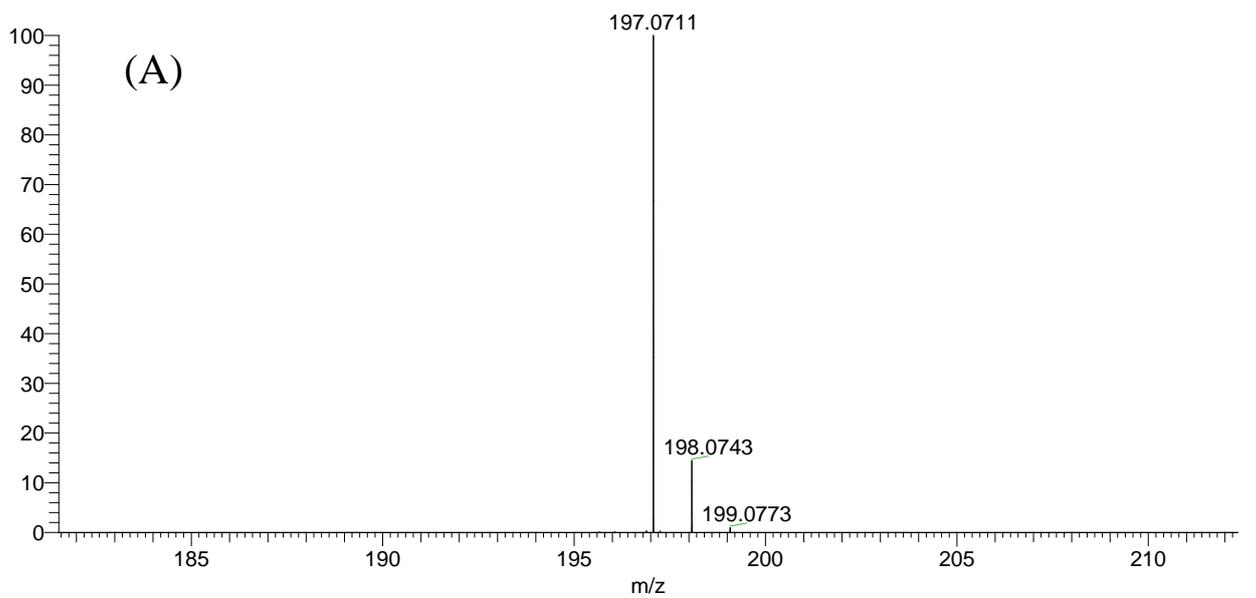


Figure S6 – MS (A) and MS/MS (B) spectra of metabolite with RT 4.20 min from *P. chlororaphis* BZR 245-F. The presented spectra are in positive-ion electrospray ionization mode

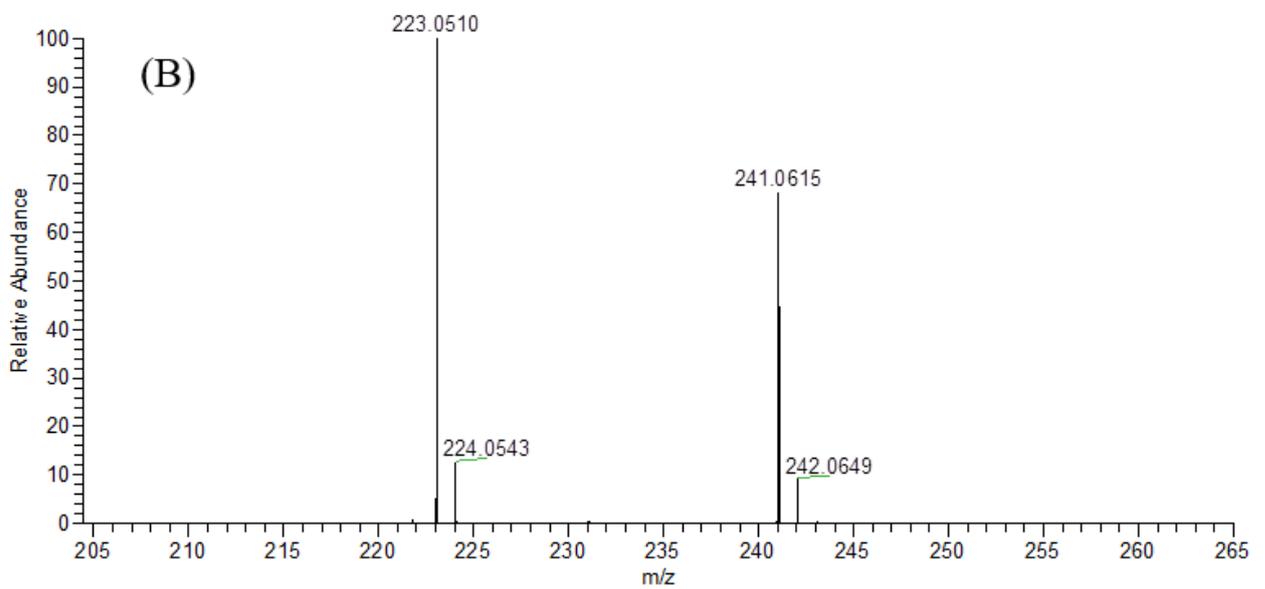
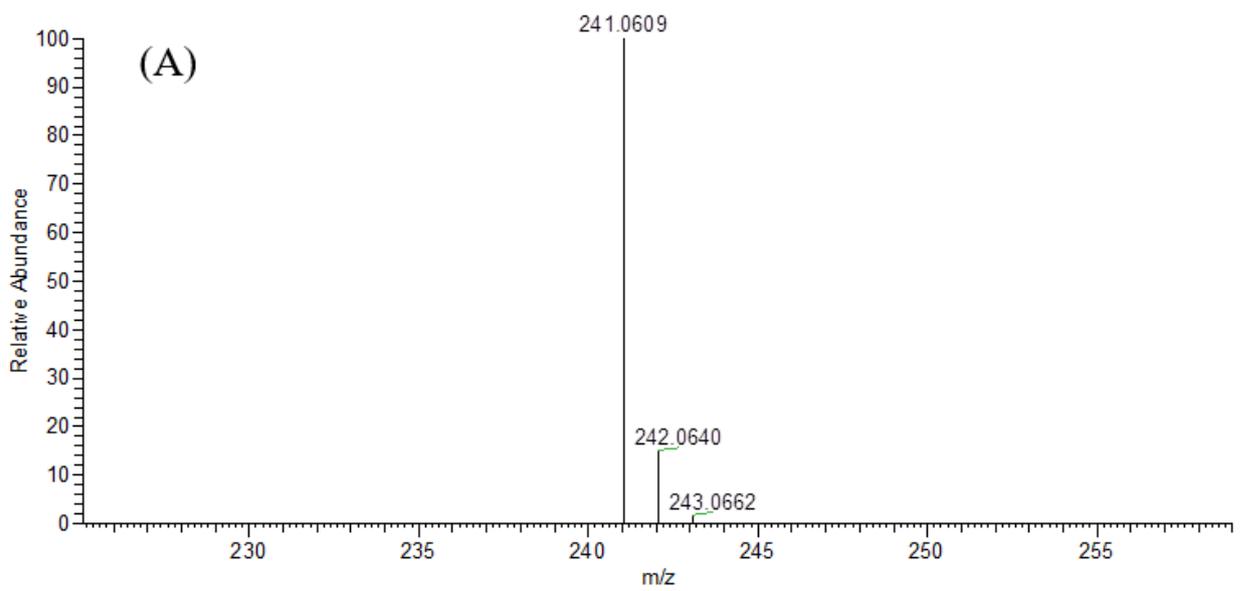


Figure S7 – MS (A) and MS/MS (B) spectra of metabolite with RT 4.73 min from *P. chlororaphis* BZR 245-F. The presented spectra are in positive-ion electrospray ionization mode

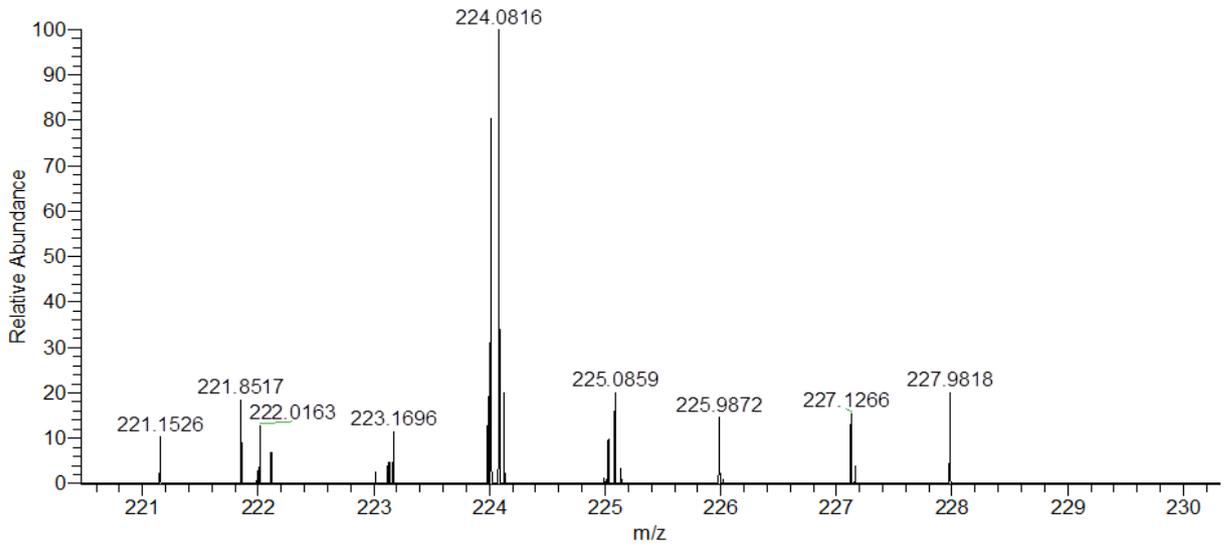


Figure S8 – MS spectrum in positive-ion electrospray ionization mode of metabolite with RT 4.52 min from *P. chlororaphis* BZR 245-F

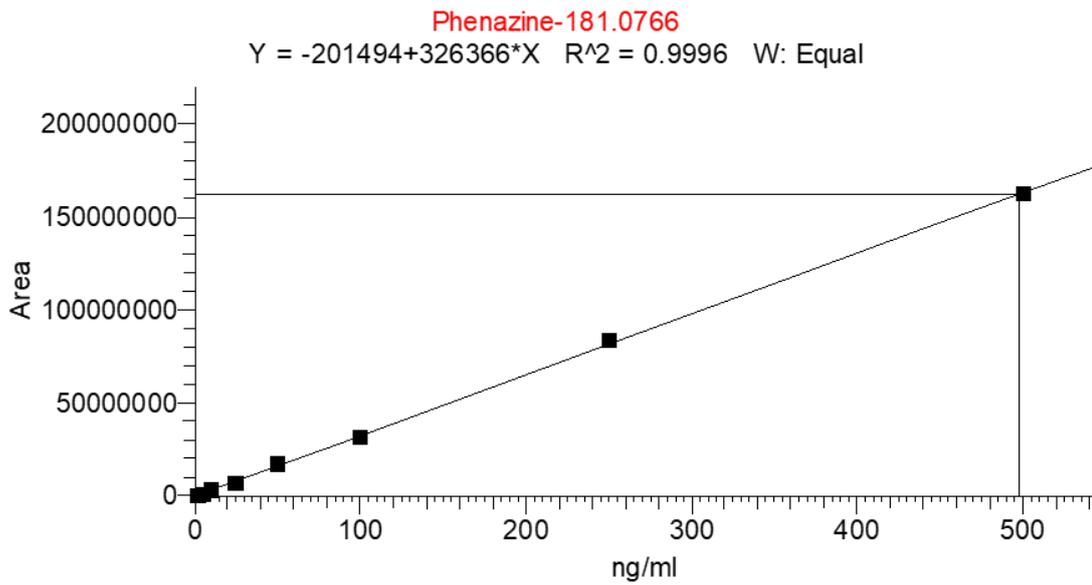


Figure S9 – Dependency graph of the signal area with m / z 181.0766 on the concentration of standard phenazine.

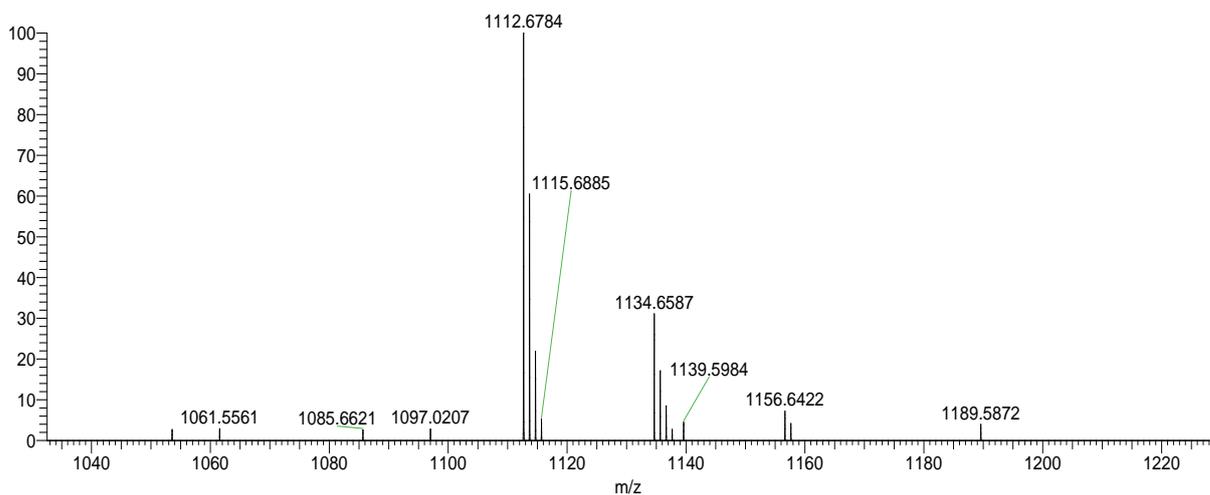


Figure S10 – MS spectrum in positive-ion electrospray ionization mode of metabolite with RT 11.68 min from *P. chlororaphis* BZR 245-F. sample

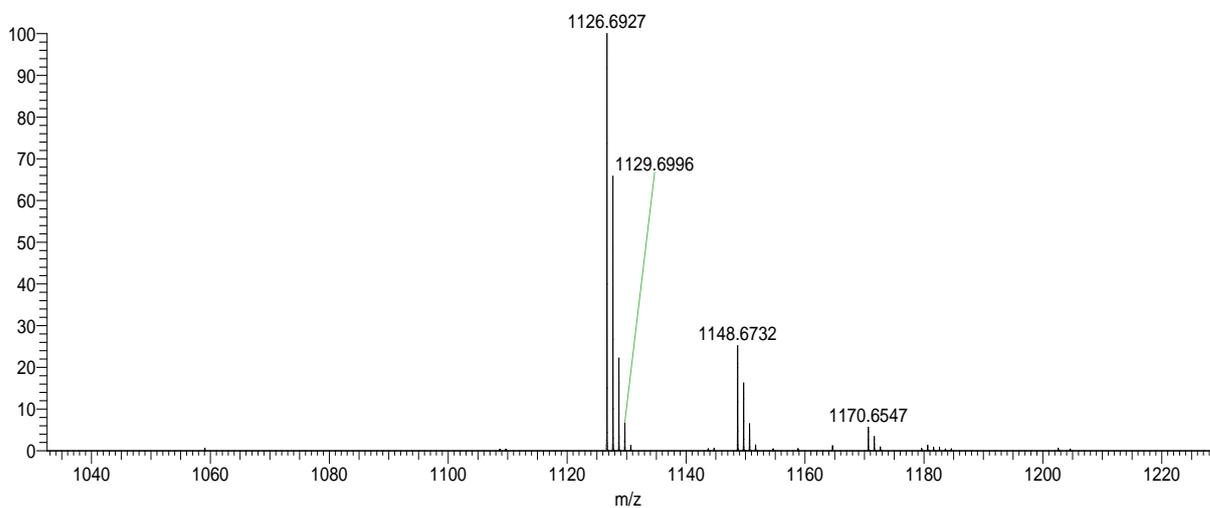


Figure S11 – MS spectrum in positive-ion electrospray ionization mode of metabolite with RT 11.98 min from *P. chlororaphis* BZR 245-F. sample

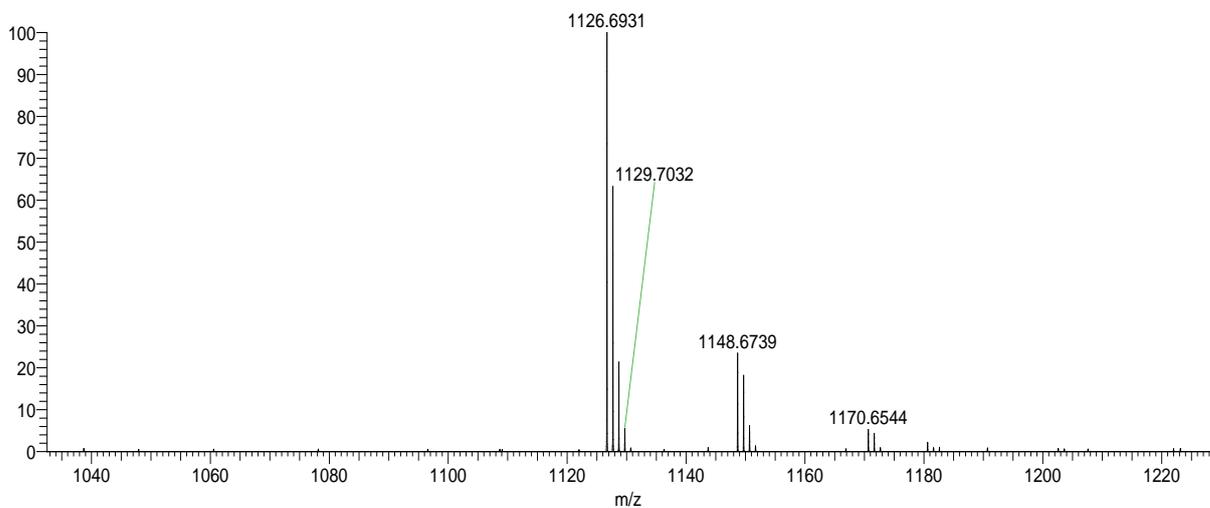


Figure S12 – MS spectrum in positive-ion electrospray ionization mode of metabolite with RT 12.09 min from *P. chlororaphis* BZR 245-F. sample

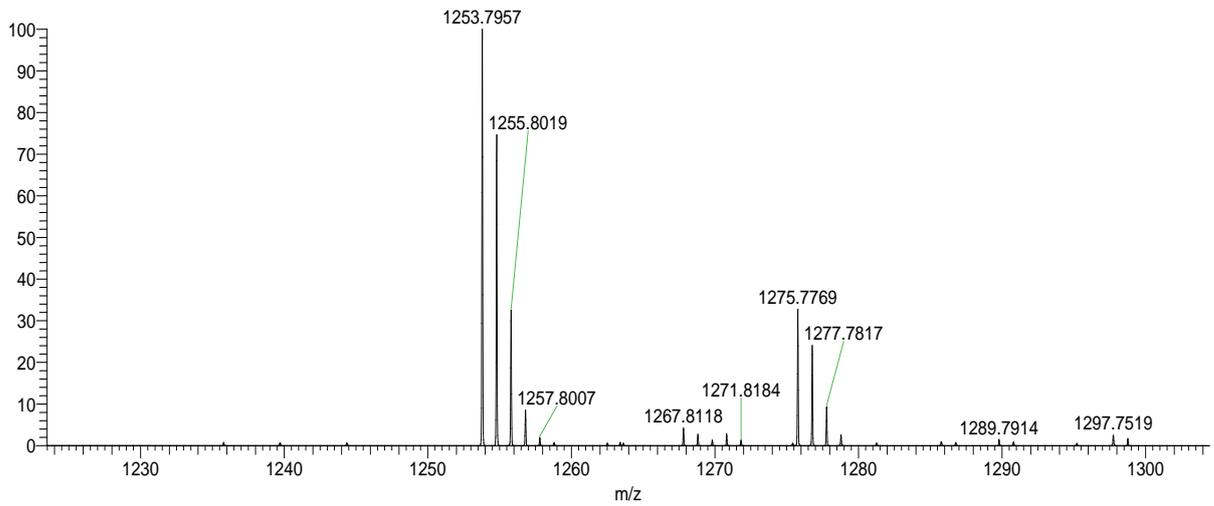


Figure S13 – MS spectrum in positive-ion electrospray ionization mode of metabolite with RT 12.48 min from *Pseudomonas* sp. BZR 523-2 sample

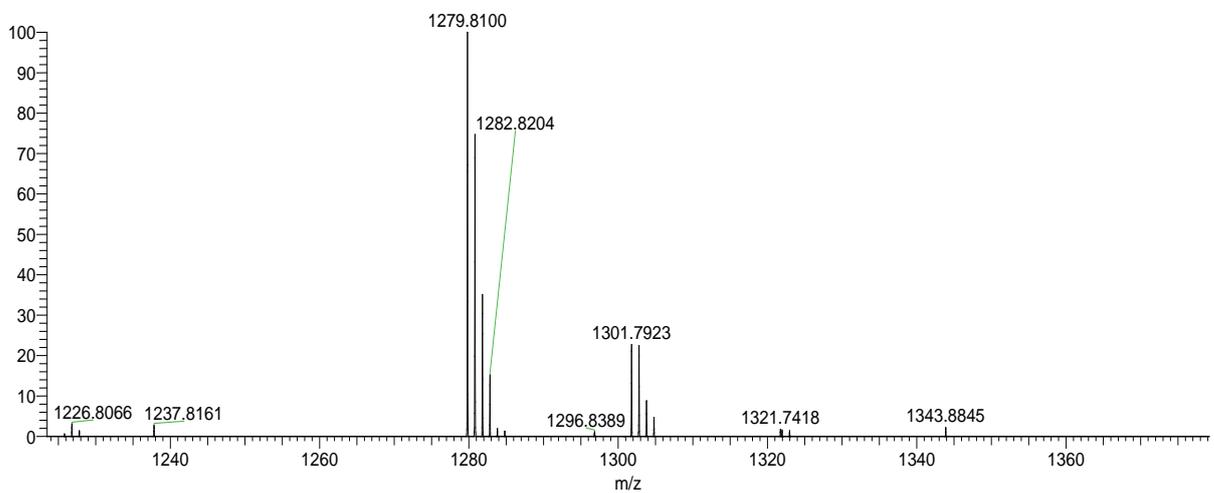


Figure S14 – MS spectrum in positive-ion electrospray ionization mode of metabolite with RT 12.82 min from *Pseudomonas* sp. BZR 523-2 sample

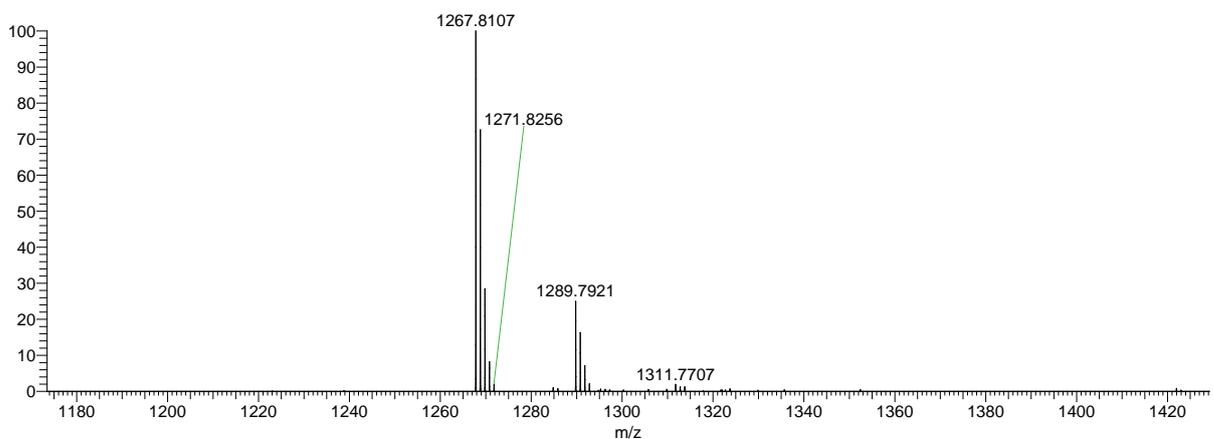


Figure S15 – MS spectrum in positive-ion electrospray ionization mode of metabolite with RT 13.00 min from *Pseudomonas* sp. BZR 523-2 sample

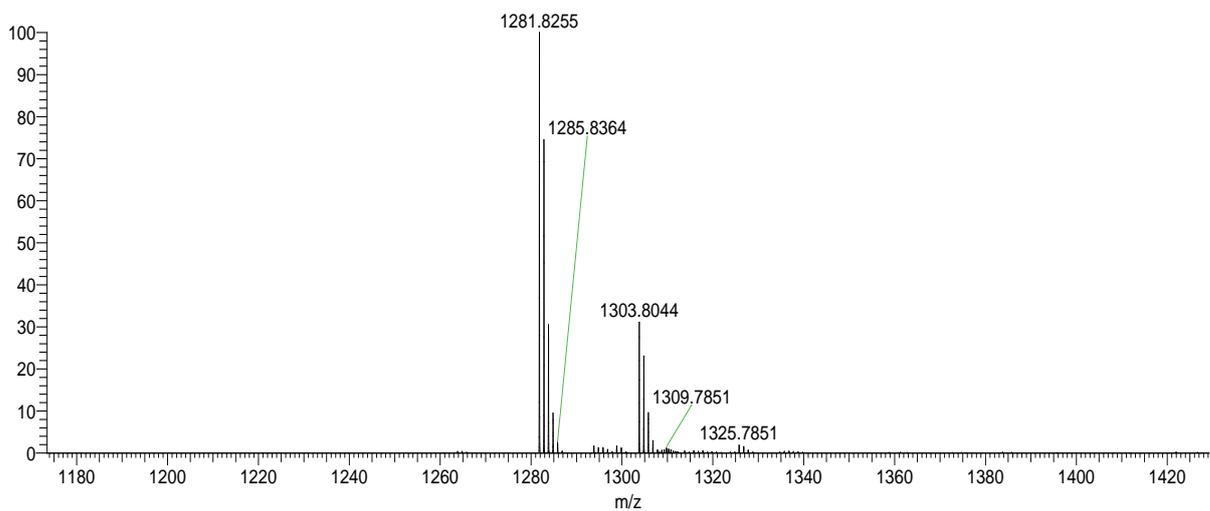


Figure S16 – MS spectrum in positive-ion electrospray ionization mode of metabolite with RT 13.32 min from *Pseudomonas* sp. BZR 523-2 sample

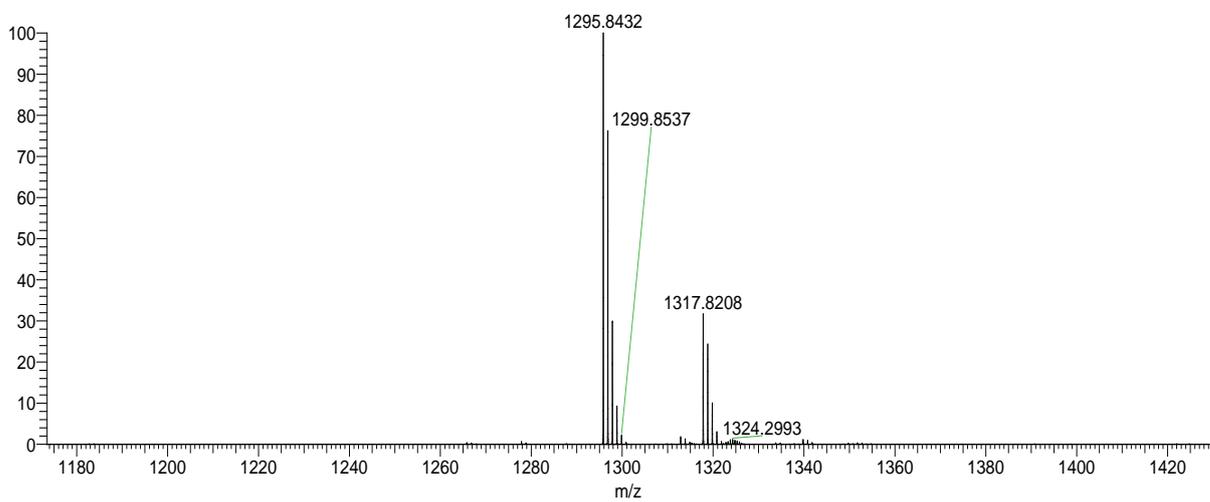


Figure S17 – MS spectrum in positive-ion electrospray ionization mode of metabolite with RT 13.80 min from *Pseudomonas* sp. BZR 523-2 sample