Table S1. Primers for *phqK* exon amplification and fusion

Name	**Sequence**
*phqK*_Int1_F	5′–ATGGGCTCTTTAGGTGAAGAAGTTCAAG–3′
*phqK*_Int1_R	5′–GTTGCTTTGAAGACCAATACAGTCTCCGATGGACTTCA GTATATTGCTTTTC–3′
*phqK*_Int2_F	5′–GAAAAGCAATATACTGAAGTCCATCGGAGACTGTATTG GTCTTCAAAGCAAC–3′
*phqK*_Int2_R	5′–CAGACGTCTAGGAGATTTCTTGTATCCTGATGAATGCAG AACCACGAAAAG–3′
*phqK*_Int3_F	5′–CTTTTCGTGGTTCTGCATTCATCAGGATACAAGAAATCT CCTAGACGTCTG–3′
*phqK*_Int3_R	5′–CTAGGGTGACTTGTTCTGCAATGG–3′

Table S2. Crystallographic information. Values in parentheses pertain to the outermost shell of data.

Ligand	Malbrancheamide B SeMet (**21**)	None	Malbrancheamide C (**22**)	Paraherquamide K (**14**)	Paraherquamide L (**15**)
Space Group	*P* 2_1 2_1 2_1	*P* 2_1 2_1 2_1	*P* 2_1 2_1 2_1	*P* 2_1 2_1 2_1	*P* 2_1 2_1 2_1
Cell Dimensions					
a,b,c (Å)	48.0, 82.4, 119.3	47.7, 84.2, 116.4	48.1, 82.8, 120.1	48.2, 83.2, 119.6	64.9, 79.9, 87.7
	90.0, 90.0, 90.0	90.0, 90.0, 90.0	90.0, 90.0, 90.0	90.0, 90.0, 90.0	90.0, 90.0, 90.0
X-ray Source	APS 23ID-B	APS 23ID-B	APS 23ID-B	APS 23ID-B	APS 23ID-B
Wavelength (Å)	0.979	1.033	0.7293	1.033	1.033
d_{min} (Å)	1.69 (1.75 - 1.69)	1.71 (1.77 - 1.71)	1.25 (1.30 - 1.25)	1.89 (1.96 - 1.89)	2.09 (2.17 - 2.09)
R-merge	0.082 (0.948)	0.071 (1.195)	0.069 (1.548)	0.193 (1.463)	0.155 (1.160)
Avg I/σ	21.58 (1.55)	19.97 (1.34)	17.88 (1.11)	10.93 (1.14)	12.08 (1.66)
Completeness (%)	99 (88)	99 (91)	100 (99)	99 (95)	100 (98)
Multiplicity	12.0 (8.0)	12.4 (9.1)	13.0 (10.7)	12.7 (11.8)	12.6 (11.9)
Total Reflections	635,907	630,819	1,725,902	495,499	345,452
$CC_{1/2}$	1.00 (0.72)	1.00 (0.60)	1.00 (0.53)	1.00 (0.56)	1.00 (0.54)
CC*	1 (0.91)	1 (0.87)	1 (0.83)	1 (0.85)	1 (0.84)
Refinement					
Reflections (#)	53,023 (4,655)	50,852 (4,603)	132,940 (13,092)	39,106 (3,831)	27,473 (2,646)
R_{work}/R_{free} (%)	17.0/20.7	16.2/19.2	15.4/17.5	21.5/25.6	20.8/25.6
Number of Non-Hydrogen Atoms	4,061	3,991	4,265	3,823	3,696
Macromolecules	3,506	3,538	3,570	3,517	3,454
Ligands	79	53	79	85	86
Solvent	476	400	616	221	156
Amino Acid Residues	444	445	444	444	438
Deviation From Ideality					
Bond Lengths (Å)	0.006	0.006	0.005	0.007	0.008
Bond Angles (deg)	0.9	0.86	0.85	0.94	1.01
Average B-Factor ($Å^2$)	30.7	37.4	26.0	39.9	57.2
Macromolecules	29.6	36.6	23.6	39.8	57.3
FAD	21.2	25.6	19.3	30.1	38.3
Substrate	30.8		20.8	57.3	88.8
Solvent	40.2	46.3	40.5	41.0	53.9
Ramachandran Plot					
Favored (%)	97.96	97.52	98.19	97.06	96.54
Allowed (%)	2.04	2.48	1.81	2.94	3.46
Outliers (%)	0	0	0	0	0

Table S3. ^{13}C-NMR, ^{1}H-NMR, gHMBCAD, and gCOSY, correlations for paraherquamide K (**14**) isolated from *phqK* knockout in *P. simplicissimum*. HRMS (ESI-QTOF): m/z $[M+H]^+$ calculated for $C_{27}H_{33}N_3O_2$ = 432.2651, experimental (isolated) = 432.2662.

Paraherquamide K $(CD_3)_2SO$-d_6, 800 MHz

	∂ ^{13}C	∂ ^{1}H (*J* [Hz])	gHMBCAD	gCOSY
1	NH	10.53 (s)	2, 3, 8, 9	
2	140.09			
3	103.76			
4	117.32	7.01 (8.3)	3, 6, 8	5
5	108.52	6.44 (8.3)	6, 7, 9	4
6	147.43			
7	104.78			
8	132.82			
9	121.51			
10	28.95	2.65 (d, 15.5)	2, 3, 11, 12, 20	
		2.71 (d, 15.5)	2, 3, 11, 12	
11	55.62			
12	59.53	2.05 (d, 10.2)	11, 16, 20	
		3.27 (m)		
13	64.36			
14	39.84	1.85 (dd, 16.6, 9.1)		17
15	30.18	1.70 (ddd, 21.8, 11.1, 5.4)		16
		1.91 (m)		
16	53.93	2.14 (m)		15
		3.08 (m)		
17	13.13	1.32 (d, 7.0)	13, 14	
18	172.70			
19	29.34	1.58 (dd, 12.8, 4.0)	13, 18, 21	
		2.02 (dd, 12.5)		
20	46.16	1.97 (m)		
21	33.87			
22	30.02	1.28 (s)	2, 20, 21, 23	
23	23.38	1.31 (s)	2, 20, 21, 22	
24	118.13	6.94 (d, 9.8)	6, 26	25
25	128.91	5.72 (d, 9.7)	7, 26	24
26	74.98			
27	27.08	1.36	25, 26, 28	
28	27.08	1.36	25, 26, 27	
29	NH	8.11	10, 11, 13	

Table S4. ^{13}C-NMR, ^{1}H-NMR, gHMBCAD, and gCOSY, correlations for paraherquamide L (**15**) isolated from *phqK* knockout in *P. simplicissimum*. HRMS (ESI-QTOF): m/z $[M+H]^+$ calculated for $C_{27}H_{33}N_3O_3$ = 448.2600, experimental (isolated) = 448.2612.

Paraherquamide L $(CD_3)_2SO$-d_6, 600 MHz

	∂ ^{13}C	∂ ^{1}H (*J* [Hz])	gHMBCAD	gCOSY
1	NH	10.89 (s)	2, 3, 9, 10	
2	141.57			
3	104.01			
4	112.32	6.92 (d, 8.3)	6, 8	5
5	115.81	6.64 (d, 8.3)	6, 7, 9	4
6	139.58			
7	136.82			
8	127.96			
9	124.69			
10	29.03	2.71 (m)	2, 3, 9, 11, 20	
11	55.6			
12	59.54	2.05 (m)		
		3.28 (d, 9.9)		
13	64.38			
14	39.91	1.85 (m)	18	15
15	30.19	1.68 (ddt, 15.5, 10.5, 5.0)	14, 16, 17	14, 16
		1.90 (m)		
16	53.43	2.14 (td, 10.2, 4.7)		15
		3.08 (q, 8.8)		
17	13.17	1.31 (s)	14	
18	172.72			
19	29.44	1.58 (dd, 12.4, 3.5)	13, 18, 20, 21	
		2.02 (m)		
20	46.2	1.98 (m)		
21	34.07			
22	29.83	1.29 (s)	23	
23	23.22	1.30 (s)	2, 21, 22	
24	139.25	6.51 (d, 7.6)	7, 25, 26	
25	115.35	4.97 (d, 7.6)	24, 26	
26	78.86			
27	29.5	1.35 (s)	25, 26	
28	29.5	1.35 (s)	25, 26	
29	NH	8.13 (s)	10, 11	

Table S5. ^{13}C-NMR, ^{1}H-NMR, gHMBCAD, gCOSY, and NOESY correlations for paraherquamide M (**16**) isolated from PhqK *in vitro* reactions. HRMS (ESI-QTOF): m/z $[M+H]^+$ calculated for $C_{27}H_{33}N_3O_3$ = 448.2600, experimental (isolated) = 448.2604.

Paraherquamide M $(CD_3)_2SO$-d_6, 800 MHz

Position	∂ ^{13}C	∂ ^{1}H (*J* [Hz])	gHMBCAD	gCOSY	NOESY
1	NH	10.61 (s)	8		24
2	183.17				
3	172.02				
4	125.91	7.03 (d, 8.1)	6, 7, 8, 10	5	5, 10
5	108.02	6.32 (d, 8.1)	6, 7	4	4
6	152.00				
7	104.31				
8	138.35				
9	122.13		5		
10	38.41	1.78 (d, 14.8)	2, 12, 16		
		2.27 (d, 14.8)	2, 21		29
11	61.37				
12	60.15	2.31 (d, 10.5)	16, 18		
		3.58 (d, 10.7)	13, 20		
13	66.96				
14	39.34	1.81 (m)	13		17
15	30.05	1.64 (m)		16	
		1.90 (m)			
16	52.43	2.13 (m)		15	
		3.05 (m)	13		
17	13.06	1.30 (d, 7.0)	13, 16		
18	60.74				
19	27.06	1.27 (m)	20	20	20
		1.88 (m)	3, 17		20
20	52.53	2.84 (t, 10.4)	19, 22, 23	19	19
21	45.21				
22	23.99	0.72 (s)	11, 20, 21, 23		
23	20.24	0.94 (s)	20, 21, 22		
24	116.68	6.57 (d, 9.9)	6, 7, 8, 25, 26	25	
25	130.1	5.73 (d, 9.9)	7, 27	24	
26	75.73				
27	27.52	1.36 (d, 18.5)	25, 26, 28		
28	27.52	1.36 (d, 18.5)	25, 26, 27		
29	NH	8.47 (s)			

Table S6. ^{13}C-NMR, ^{1}H-NMR, gHMBCAD, gCOSY, and NOESY correlations for paraherquamide N (**17**) isolated from PhqK *in vitro* reactions. HRMS (ESI-QTOF): m/z $[M+H]^+$ calculated for $C_{27}H_{33}N_3O_4$ = 464.2549, experimental (isolated) = 464.2550.

Paraherquamide N $(CD_3)_2SO$-d_6, 600 MHz

Position	∂ ^{13}C	∂ ^{1}H (*J* [Hz])	gHMBCAD	gCOSY	NOESY
1	NH	10.51 (s)	3, 6, 8		
2	182.45				
3	62.03				
4	120.96	6.99 (d, 8.1)	3, 8, 9	5	10, 23
5	116.17	6.60 (d, 8.0)	6, 7, 9	4	
6	126.22				
7	135.04				
8	133.41				
9	145.61				
10	38.33	1.82 (s)	2, 21, 16		
		2.28 (s)	2, 21		29
11	60.87				
12	60.14	2.31 (d, 10.7)	13, 16		
		3.61 (d, 10.8)	13, 16		
13	66.93				
14	39.46	1.80 (m)	13, 15, 16, 19		
15	30.06	1.64 (m)	14, 16, 17	16	
		1.91 (m)	13		
16	52.45	2.14 (m)	12	15	
		3.05 (m)	13, 14, 15		
17	13.03	1.30 (d, 7.0)	13, 14, 15		
18	172.00				
19	27.05	1.27 (m)	11, 18, 20, 21	20	
		1.89 (m)	13, 18		
20	52.45	2.81 (t, 20.5, 10.3)	19, 21, 22, 23	19	19, 19
21	45.21				
22	24.11	0.72 (s)	3, 20, 21, 23		
23	20.40	0.95 (s)	3, 20, 21, 23		
24	139.00	6.36 (d, 7.6)	7, 25, 26	25	25
25	115.17	4.97 (d, 7.6)	24, 26, 27, 28	24	24
26	79.28				
27	29.22	1.36 (d, 16.7)	24, 25, 26, 28		
28	29.22	1.36 (d, 16.7)	24, 25, 26, 27		
29	NH	8.50 (s)	10, 13, 11		10

Table S7. ^{1}H-NMR for malbrancheamide (**19**) isolated from *Malbranchea aurantiaca*. HRMS (ESI-QTOF): m/z $[M+H]^{+}$ calculated for $C_{21}H_{23}Cl_2N_3O$ = 404.1291, experimental (isolated) = 404.1287.

Malbrancheamide CD_3OD, 600 MHz

Position	∂ ^{1}H (*J* [Hz])
1	2.56 (m)
	1.49 (m)
2	1.87 (m)
3	2.03 (m)
	2.19 (m)
5	2.27 (dd, 10.3, 1.5)
	3.43 (d, 10.2)
6	2.84
7	7.48 (s)
10	7.40 (s)
12a	2.13 (m)
13	2.03 (d, 13.1)
	1.96 (dd, 13.1, 5.2)
16	1.32 (s)
17	1.42 (s)

Table S8. ^{1}H-NMR, gHMBCAD, gCOSY, and NOESY correlations for spiromalbramide (**20**) isolated from PhqK *in vitro* reactions. HRMS (ESI-QTOF): m/z $[M+H]^+$ calculated for $C_{27}H_{33}N_3O_4$ = 420.1246, experimental (isolated) = 420.1243.

Spiromalbramide CD_3OD, 600MHz

Position	∂ ^{13}C	∂ ^{1}H (*J* [Hz])	gHMBCAD	gCOSY	NOESY
1	28.19	2.46 (ddd, 12.8, 9.5, 3.9)	14	2	2
	28.21	1.46 (td, 11.7, 7.1)	13a	2	
2	22.99	1.89 (m)		1, 3	1, 3
3	54.08	3.11 (td, 8.9, 3.0)		2	2
		2.25 (q, 8.9)		2	
5	59.77	3.71 (d, 11.3)	3, 5a, 13a		17
	59.73	2.65 (d, 10.0)			
5a	68.62				
6	39.31	2.39 (d, 15.1)	5, 6a, 6b, 11a, 12, 12a		
	39.24	2.06 (d, 15.2)			
6a	63.75				
6b	131.82				
7	128.72	7.51 (s)	6a, 8, 9, 10a		6, 17
8	125.49				
9	132.24				
10	112.12	7.02 (s)	8, 9, 10a		
10a	143.75				
11a	184.11				
12	46.97				
12a	55.10	3.05 (t, 10.1)		13	16
13	28.81	1.86 (m)	5a, 14	12a	12a
	28.78	1.71 (dd, 12.7, 10.0)			17
13a	62.60				
14	175.61				
16	24.40	0.83 (s)	6a, 12, 12a, 17		
17	20.93	1.12 (s)	6a, 12, 12a, 16		5, 13

Table S9. ^{1}H-NMR for malbrancheamide B (**21**) isolated from *Malbranchea aurantiaca*. HRMS (ESI-QTOF): m/z $[M+H]^{+}$ calculated for $C_{21}H_{23}ClN_3O$ = 370.1681, experimental (isolated) = 370.1675.

Malbrancheamide B, $(CD_3)_2SO$-d_6, 600 MHz

Position	∂ ^{1}H (*J* [Hz])
1	2.47 (m)
	1.34 (m)
2	1.75 (m)
3	2.97 (m)
	2.43 (m)
5	2.16 (d, 9.6)
	3.29 (d, 9.9)
6	2.77 (s)
7	7.33 (d, 8.4)
8	6.96 (dd, 8.4, 1.9)
10	7.28 (s)
12a	1.89 (d, 13.0)
13	1.83 (dd, 13.0, 4.8)
	1.91 (m)
16	1.27 (s)
17	1.33 (s)

Table S10. ^{1}H-NMR, gHMBCAD, gCOSY, and NOESY correlations for spiromalbrancheamide B (**25**) isolated from PhqK *in vitro* reactions. HRMS (ESI-TOF): m/z $[M+H]^+$ calculated for $C_{21}H_{24}ClN_3O_2$ = 386.1635, experimental (isolated) = 386.1630.

Spiromalbrancheamide B CD_3OD, 600MHz

Position	∂ ^{13}C	∂ ^{1}H (*J* [Hz])	gHMBCAD	gCOSY	NOESY
1	28.38	1.45 (m)	14		
		2.46 (ddd, 13.0, 9.4, 4.0)	14	14	
2	23.09	1.89 (s)		3	1, 3
3	54.44	2.25 (q, 8.9)		2	5
		3.11 (td, 8.6, 2.7)			2
5	59.96	2.65 (dd, 11.3, 1.8)	12a, 13a		
		3.71 (d, 11.2)	3, 5a		6, 17
5a	68.89				
6	39.59	2.03 (d, 15.1)	6b, 11a, 12a, 13a		5
		2.39 (d, 15.1)	11a, 12, 13a		
6a	63.61				
6b	130.18				
7	128.25	7.31	6a, 9, 10a	8	17
8	122.49	7.01 (dd, 8.1, 2.0)	6b, 9	7	
9	135.1				
10	110.86	6.89 (s)	6b, 9		
10a	144.94				
11a	184.82				
12	46.88				
12a	55.17	3.05 (m)	12, 13a, 16	13	13, 16
13	28.91	1.71 (dd, 12.8, 9.9)	14	12a	17
		1.86 (m)	13a	12a	
13a	62.99				
14	175.86				
16	24.59	0.83 (s)	6a, 12, 12a, 17	17	12a
17	21.22	1.11 (s)	6a, 12, 12a, 16	16	5, 6, 13

Table S11. ^{1}H-NMR for malbrancheamide C (**22**) isolated from MalA bromination *in vitro* reaction. HRMS (ESI-QTOF): m/z $[M+H]^{+}$ calculated for $C_{21}H_{24}BrN_3O$ = 414.1181, experimental (*in vitro*) 414.1169.

Malbrancheamide C CD_3OD, 700 MHz

Position	∂ ^{1}H (*J* [Hz])
1	2.53 (ddd, 12.0, 9.0, 5.5)
	1.47 (m)
2	1.89 (m)
3	3.06 (m)
	2.18 (m)
5	2.26 (dd, 11.9, 1.6)
	3.45 (d, 9.8)
6	2.87 (d, 9.7)
7	7.26 (d, 10.4)
8	7.07 (dd, 11.4, 1.8)
10	7.42 (d, 1.8)
12a	2.16 (m)
13	2.01 (dd, 13.1, 11.1)
	1.96 (dd, 13.3, 5.0)
16	1.34 (s)
17	1.43 (s)

Table S12. ^{1}H-NMR, gHMBCAD, gCOSY, and NOESY correlations for spiromalbrancheamide C (**26**) isolated from PhqK *in vitro* reactions. HRMS (ESI-QTOF): m/z $[M+H]^+$ calculated for $C_{21}H_{24}BrClN_3O_2$ = 430.1130, experimental (isolated) = 430.1138.

Spiromalbrancheamide C CD_3OD, 800MHz

Position	∂ ^{13}C	∂ ^{1}H (*J* [Hz])	gHMBCAD	gCOSY	NOESY
1	28.38	1.47 (m)	13a, 14	2	
		2.46 (ddd, 13.3, 9.6, 4.0)		13	2
2	23.05	1.89 (m)			1, 3
3	54.44	2.25 (q, 9.1)	5	13	2
		3.11 (td, 8.7, 2.8)	13a	2	
5	59.97	2.65 (d, 10.8)	3, 5a		
		3.71 (d, 11.2)	13a, 14		17
5a	63.01				
6	39.56	2.03 (d, 15.1)	5, 5a, 6b, 11a, 12a		
		2.39 (d, 15.1)	5, 5a, 11a		
6a	63.68				
6b	130.70				
7	128.59	7.26 (d)	6a, 10, 10a		6, 17
8	125.48	7.17 (d)	6b, 9		
9	113.72	7.04 (s)	6b, 8, 10		
10	122.71				
10a	145.11				
11a	184.69				
12	46.86				
12a	55.19	3.05 (t, 10.4)	5, 6a, 12, 13	13	13, 16
13	28.92	1.71 (dd, 12.8, 9.8)	13a, 14	12a	17
		1.87 (m)	5a, 12a	12a	
13a	68.89				
14	175.88				
16	24.60	0.83 (s)	6a, 12, 12a, 17		
17	21.22	1.10 (s)	6a, 12, 12a, 16		7, 13

Table S13. ^{1}H-NMR for isomalbrancheamide D (**24**) isolated from MalA bromination and chlorination *in vitro* reactions. HRMS (ESI-QTOF): m/z $[M+H]^+$ calculated for $C_{21}H_{23}BrClN_3O$ = 448.0786, experimental (*in vitro*) 448.0783.

Isomalbrancheamide D CD_3OD, 700 MHz

Position	∂ ^{1}H (*J* [Hz])
1	2.53 (ddd, 12.2, 9.1, 5.6)
	1.46 (m)
2	1.88 (m)
3	3.06 (m)
	2.16 (q, 8.8)
5	2.26 (d, 10.4)
	3.43 (d, 10.3)
6	2.84 (d, 4.8)
7	7.51 (s)
10	7.57 (s)
12a	2.15 (m)
13	2.01 (m)
	1.94 (m)
16	1.33 (s)
17	1.43 (s)

Table S14. ^{1}H-NMR, gHMBCAD, gCOSY, and NOESY correlations for spiroisomalbrancheamide D (**28**) isolated from PhqK *in vitro* reactions. HRMS (ESI-QTOF): m/z $[M+H]^{+}$ calculated for $C_{21}H_{24}BrClN_3O_2$ = 464.0740, experimental (isolated) = 464.0715.

Spiroisomalbrancheamide D CD_3OD, 800MHz (600 MHz for ^{1}H-NMR)

Position	∂ ^{13}C	∂ ^{1}H (*J* [Hz])	gHMBCAD	gCOSY	NOESY
1	28.37	1.45 (m)		2	13
		2.46 (ddd, 12.8, 9.4, 3.9)			
2	23.04	1.89 (s)		1, 3	
3	54.40	2.25 (m)	13a		
		3.11 (td, 8.6, 2.7)		2	
5	59.88	2.65 (d, 11.8)			
		3.71 (d, 11.4)	13a		
5a	62.95				
6	39.34	2.06 (d, 15.2)	6b, 11a		
		2.39 (d, 15.2)	5		
6a	64.010				
6b	132.400				
7	128.73	7.53 (s)	6a, 8, 9, 10a		5, 17
8	127.82				
9	122.49				
10	115.32	7.17 (s)	6b, 8, 9		
10a	143.69				
11a	184.23				
12	47.10				
12a	55.14	3.05 (m)			13
13	28.85	1.71 (m)	13a		
		1.86 (m)			
13a	68.88				
14	170.3				
15		8.55 (s)			
16	24.50	0.83 (s)	6a, 12, 12a, 17		12a
17	21.02	1.12 (s)	6a, 12, 12a, 16		6,13

Table S15. ^{1}H-NMR, gHMBCAD, and gCOSY correlations for malbrancheamide E (**23**) isolated from MalA *in vitro* reactions. HRMS (ESI-QTOF): m/z $[M+H]^+$ calculated for $C_{21}H_{24}BrBrN_3O_2$ = 492.0286, experimental (isolated) = 492.0293.

Malbrancheamide E CD_3OD, 800MHz (600 MHz for 1H-NMR)

Position	∂ ^{13}C	∂ ^{1}H (*J* [Hz])	gHMBCAD	gCOSY	NOESY
1	28.37	1.45 (m)		2	13
		2.46 (ddd, 12.8, 9.4, 3.9)			
2	23.04	1.89 (s)		1, 3	
3	54.40	2.25 (m)	13a		
		3.11 (td, 8.6, 2.7)		2	
5	59.88	2.65 (d, 11.8)			
		3.71 (d, 11.4)	13a		
5a	62.95				
6	39.34	2.06 (d, 15.2)	6b, 11a		
		2.39 (d, 15.2)	5		
6a	64.010				
6b	132.400				
7	128.73	7.53 (s)	6a, 8, 9, 10a		5, 17
8	127.82				
9	122.49				
10	115.32	7.17 (s)	6b, 8, 9		
10a	143.69				
11a	184.23				
12	47.10				
12a	55.14	3.05 (m)			13
13	28.85	1.71 (m)	13a		
		1.86 (m)			
13a	68.88				
14	170.3				
15		8.55 (s)			
16	24.50	0.83 (s)	6a, 12, 12a, 17		12a
17	21.02	1.12 (s)	6a, 12, 12a, 16		6,13

Table S16. ^{1}H-NMR, gHMBCAD, gCOSY, and NOESY correlations for spiromalbrancheamide E (**27**) isolated from PhqK *in vitro* reactions. HRMS (ESI-QTOF): m/z $[M+H]^+$ calculated for $C_{21}H_{24}BrBrN_3O_2$ = 508.0235, experimental (isolated) = 508.0223.

Spiromalbrancheamide E CD_3OD, 800MHz (600 MHz for ^{1}H-NMR)

Position	**∂ ^{13}C**	**∂ ^{1}H (*J* [Hz])**	**gHMBCAD**	**gCOSY**	**NOESY**
1	28.37	1.46 (td, 11.8, 11.4, 7.2)	13a, 14	2	2
		2.46 (ddd, 13.0, 9.4, 3.9)			
2	23.04	1.89 (m)			1, 3
3	54.40	2.25 (q, 9.0)	2	2	5
		3.11 (td, 8.9, 3.2)	1, 13a		
5	59.88	2.65 (d, 11.6)	5a		3
		3.71 (d, 11.3)			6, 17
5a	62.94				
6	39.31	2.06 (d, 15.2)	5a, 6b, 11a, 12, 12a		
		2.39 (d, 15.1)	5, 5a		
6a	63.91				
6b	133.02				
7	131.82	7.65 (s)	6a, 8, 9, 10a		6, 17
8	117.06				
9	124.82				
10	115.37	7.19 (s)	6b, 8, 9		
10a	144.25				
11a	184.07				
12	47.13	(m)			
12a	55.13	3.05	5a, 12, 16, 17	13	13, 16
13	28.84	1.70 (dd, 12.8, 9.9)	12a, 13a, 14	12a	
		1.86 (m)	5a		
13a	68.89				
14	175.86				
15		8.55 (s)			
16	24.48	0.83 (s)	6a, 12, 12a, 17		12a, 13
17	21.01	1.12 (s)	6a, 12a, 16		3, 5, 7, 13

Table S17. Reaction data for PhqK variants, including percent conversion (conversion %), FAD cofactor incorporation (%FAD), and amount of protein obtained relative to the wild-type enzyme ([PhqK]) which is representative of the solubility/stability. Reaction conditions are mentioned above.

Variant	Conversion %		%FAD	[PhqK]
	Paraherquamide K (**14**)	Paraherquamide L (**15**)		
WT	97	83	100	100
D47A	3	0	22	58
D47N	0	0	48	84
Q52E	20	23	13	33
R192A	11	0	22	81
R192K	5	0	24	98
F219A	35	42	11	49
Q232A	3	0	10	48
Q232E	2	0	10	40
S241A	14	20	22	40
S322A	9	14	20	62
Q327A	22	35	11	31
Q327E	12	29	14	59
S397A	76	89	56	100

Table S18. Primers for *phqk* gene disruption.

phqK-Cas9	Forward 1	5'-TCATAGCTGTTTCCGCTGA-3'
	Reverse 1	5-CAGACAGGTCTTGCTGGAGAGACGAGCTTACTCGTTTCGTCCTCACGGACTCATCAGTCTCCACGGTGATGTC TGCTCAAG-3'
	Forward 2	5'-TCGTCTCTCCAGCAAGACCTGTCTGGTTTTAGAGCTAGAAATAGCAAGTTAAA-3'
	Reverse 2	5'-ATTCTGCTGTCTCGGCTG-3'

Table S19. Calculated and isolated yields from PhqK-catalyzed spirocyclization reactions

Substrate	**Product**	**Isolated Yield**
Paraherquamide K (**14**)	Paraherquamide M (**16**)	91%*
Paraherquamide L (**15**)	Paraherquamide N (**17**)	90%*
Malbrancheamide (**19**) 2.75 mg	Spiromalbramide (**20**) 0.94 mg	33%
Malbrancheamide B (**21**) 3.95 mg	Spiromalbrancheamide B (**25**) 1.68 mg	41%
Malbrancheamide C (**22**) 1.46 mg	Spiromalbrancheamide C (**26**) 0.27 mg	18%
Malbrancheamide E (**23**) 1.03 mg	Spiromalbrancheamide E (**27**) 0.89 mg	84%
Isomalbrancheamide D (**24**) 2.2 mg	Spiroisomalbrancheamide D (**28**) 0.95 mg	42%

*Conversions determined by UV using standard curves.

Table S20. Calculated pK_a values for Arg192 in the cocrystal structures with paraherquamide K (**14**), paraherquamide L (**15**), malbrancheamide B (**21**), and malbrancheamide C (**22**) using PROPKA on the PDB2PQR server.

Substrate	**pK_a**
Paraherquamide L (**15**)	10.99
Paraherquamide K (**14**)	12.26
Malbrancheamide B (**21**)	12.86
Malbrancheamide C (**22**)	12.93
No ligand	13.04