antibiotics

Supplementary Information

General

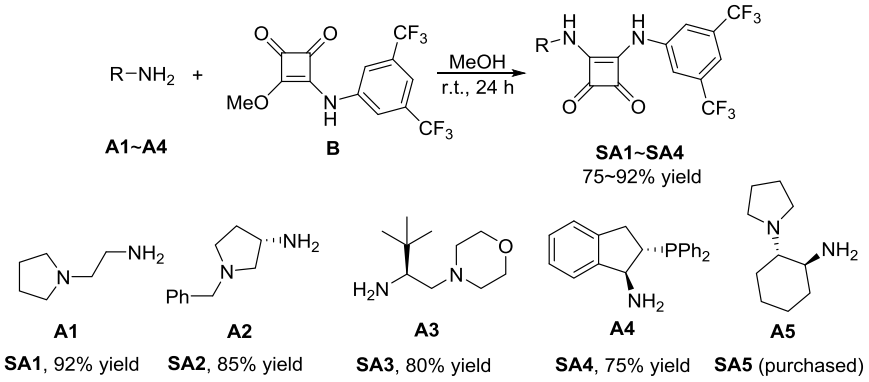
The ^{1}H NMR, ^{13}C NMR, ^{19}F NMR spectra were measured in $CDCl_3$ and MeOD solution on a Bruker AV-400 spectrometer using TMS as an internal reference. Coupling constant (*J*) values are given in Hz. Multiplicities are designated by the following abbreviations: s, singlet; d, doublet; t, triplet; q, quartet; br, broad; m, multiplet. High-resolution mass spectra (HRMS) were performed on a Bruker microTOF-Q II Mass Spectrometer with ES ionization (ESI). All commercially available reagents were used as received. Thin-layer chromatography on silica (with GF_{254}) was used to monitor all reactions. Products were purified by flash column chromatography on silica gel purchased from Qingdao Haiyang Chemical Co., Ltd. All solvents, organic and inorganic reagents were from commercial sources and used without purification unless otherwise noted. The primary aminess **A1~A4** and **SA5** are purchased from Daicel chiral technologies (China) Co. Ltd. The mono-methyl ester **B** was prepared according to literatures reported method.[1] **SA1**, **SA2** and **SA3** are known compounds.[2-4] The characterization data of all new compounds were listed in the Supporting Information.

The preparation of mono-methyl ester B[1]

The mixture of 3,4-dimethoxy-3-cyclobutene-1,2-dione (1.00 g, 7 mmol) and 3,5-bis(trifluoromethyl)aniline (1.20 mL, 7.7 mmol, 1.1 equiv) in MeOH (10 mL) was stirred at r.t. for 2d to give yellow precipitate. The yellow precipitate was filtered with the aid of cold MeOH to provide a yellow powder and dried in vacuo to (2.15 g, 90%).

mono-methyl ester B: ^{1}H NMR (400 MHz, MeOD) δ: 8.05 (s, 2H), 7.68 (s, 1H), 4.51 (s, 3H, OCH_3).

General procedure for preparation of squaramides (SA1-SA4) [2-4]



To a solution of mono-metyl ester (**B**, 1.02 g, 3.0 mmol) in MeOH (15 mL) was added a solution of corresponding amine (3.0 mmol) in MeOH (5 mL) at room temperature (r.t.). The mixture was stirred for 24 h. The reaction mixture was filtered, and the precipitate was washed with cold MeOH (2 × 5.0 mL) to afford pure squaramides **SA1-SA4**, respectively. They are analytical pure and used in the *vitro* and *vivo* experiments without further purification.

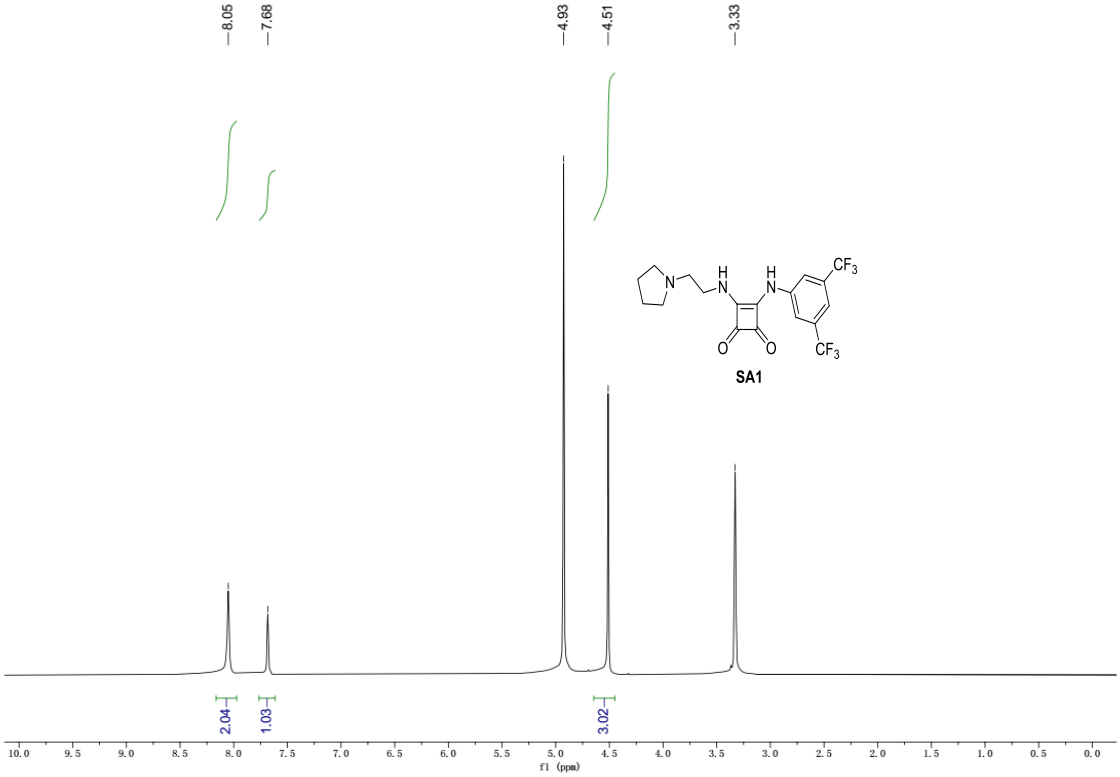
SA1: white powder, 92% yield; m.p. 145.2-146.1 ^{o}C; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.31 (s, 1H), 8.06 (s, 2H), 7.80 (s, 1H), 7.66 (s, 1H), 3.75 (s, 2H), 3.35 (4H, merged), 2.65 (s, 2H), 2.51 (4H, merged); ^{13}C NMR (101 MHz, DMSO-d_6) δ 185.4, 178.5, 162.3, 141.2, 131.5, 124.5, 121.8, 117.9, 114.6, 55.7, 53.5, 42.8, 23.1; ^{19}F NMR (376 MHz, DMSO-d_6) δ: -61.8; HRMS (ESI) *m/z* calcd. for $C_{18}H_{18}F_6N_3O_2^+$ $[M+H]^+$: 422.1298, found 422.1302.

SA2: light yellow powder, 85% yield; m.p. 167.1-168.5 ^{o}C; [α] $^{20}_{D}$ +63.7^{o} (c = 0.25, CH_2Cl_2); ^{1}H NMR (400 MHz, MeOD) δ: 8.05 (bs, 2H), 7.55 (bs, 1H), 7.34-7.26 (m, 5H), 4.78 (s, 1H), 3.70 (s, 1H), 3.33 (s, 1H), 2.94 (br, 1H), 2.77 (bs, 2H), 2.45 (bs, 2H), 1.84 (bs, 1H), 4.73 (q, *J* = 6.9 Hz, 1H), 4.38-4.26 (m, 2H), 3.82 (dd, *J* = 12.3, 6.8 Hz, 1H), 2.86 (t, *J* = 7.0 Hz, 1H), 2.71 (s, 3H), 1.95 (bs, 1H), 1.52 (s, 3H), 1.35 (s, 3H); ^{13}C NMR (100 MHz, MeOD) δ: 169.0, 162.6, 140.9, 132.7, 128.8, 128.1, 127.1, 124.5, 121.8, 117.9, 115.2, 60.5, 59.5, 53.8, 52.2, 32.8; ^{19}F NMR (376 MHz, MeOD) δ: -64.5; HRMS (ESI) *m/z* calcd. for $C_{23}H_{20}F_6N_3O_2^+$ $[M+H]^+$: 484.1454, found 484.1453.

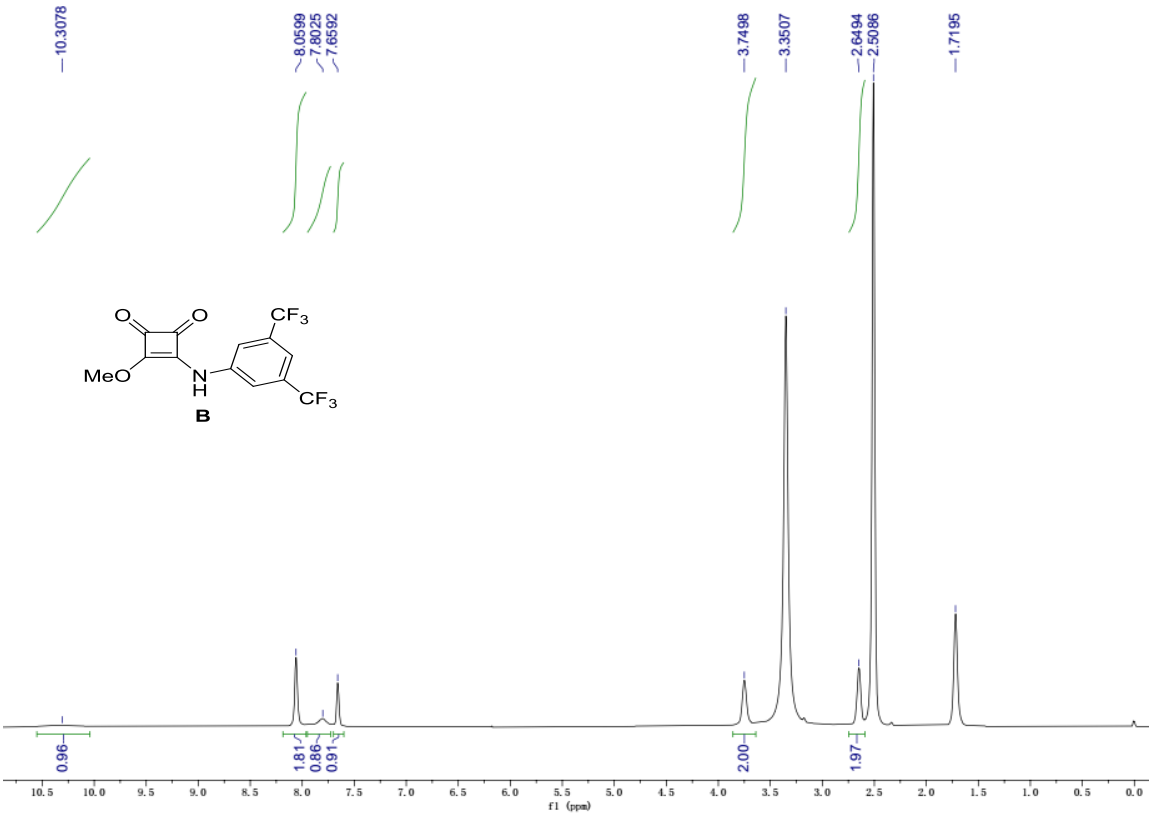
SA3: white powder, 80% yield; m.p. > 230 ^{o}C; [α] $^{20}_{D}$ +5.3^{o} (c = 0.22, CH_2Cl_2); ^{1}H NMR (400 MHz, MeOD) δ: 8.14 (s, 2H), 7.59 (s, 1H), 4.18 (d, J = 10.28 Hz, 1H), 3.62 (br, 4H), 2.70-2.65 (m, 3H), 2.48 (t, *J* = 12.0 Hz, 1H), 2.37 (br, 2H), 1.05 (s, 9H); ^{13}C NMR (100 MHz, MeOD) δ: 184.3, 180.5, 170.7, 161.9, 140.8, 132.8 (d, $J_{C\text{-}F}$ = 33 Hz), 124.5, 121.8, 117.8, 115.5, 66.8, 60.4, 59.5, 53.9. 33.7, 25.7; ^{19}F NMR (376 MHz, MeOD) δ: -64.0; HRMS (ESI) *m/z* calcd. for $C_{22}H_{26}F_6N_3O_3^+$ $[M+H]^+$: 494.1873, found 494.1886.

SA4: white powder，75% yield; m.p. 141.9- 143.1^{o}C; [α] $^{20}_{D}$ +149.4^{o} (c = 0.05, CH_2Cl_2); ^{1}H NMR (400 MHz, Chloroform-*d*) δ: 9.79 (s, 1H), 8.04 (s, 2H), 7.82 (d, *J* = 6.9 Hz, 2H), 7.74 (d, *J* = 9.9 Hz, 1H), 7.58 (s, 1H), 7.53 (t, *J* = 7.7 Hz, 2H), 7.37-7.30 (m, 1H), 7.24 (t, *J* = 7.5 Hz, 2H), 7.17 (s, 2H), 7.03-6.86 (m, 2H), 6.88-6.81 (m, 2H), 5.65-5.40 (m, 1H), 3.62 (s, 1H), 2.81 (s, 1H), 2.72-2.51 (m, 1H); ^{13}C NMR (101 MHz, Chloroform-*d*) δ: 184.7, 178.9, 167.8, 163.7, 141.0 (dd, *J* = 29.3, 8.08 Hz), 139.4, 136.7 (d, *J* = 12.5 Hz), 135.3 (dd, *J* = 32.3, 12.1 Hz), 132.8 (d, *J* = 20.2 Hz), 132.7 (q, *J* = 33.3 Hz), 129.3 (d, *J* = 33.3 Hz), 128.7 (d, *J* = 7.4 Hz), 128.5 (t, *J* = 8.1 Hz), 126.9, 124.4, 123.7, 123.0 (q, *J*C-F = 272 Hz), 122.9, 119.3, 117.1, 64.5 (d, *J* = 26.8 Hz), 44.6 (d, *J* = 10.7 Hz), 35.4 (d, *J* = 21.5 Hz); ^{19}F NMR (376 MHz, Chloroform-*d*) δ: -63.1; ^{31}P NMR (162 MHz, Chloroform-*d*) δ: -8.4; HRMS (ESI) *m/z* Calculated for $C_{33}H_{24}F_6N_2O_2P^+$ $[M+H]^+$: 625.1469, found: 625.1470.

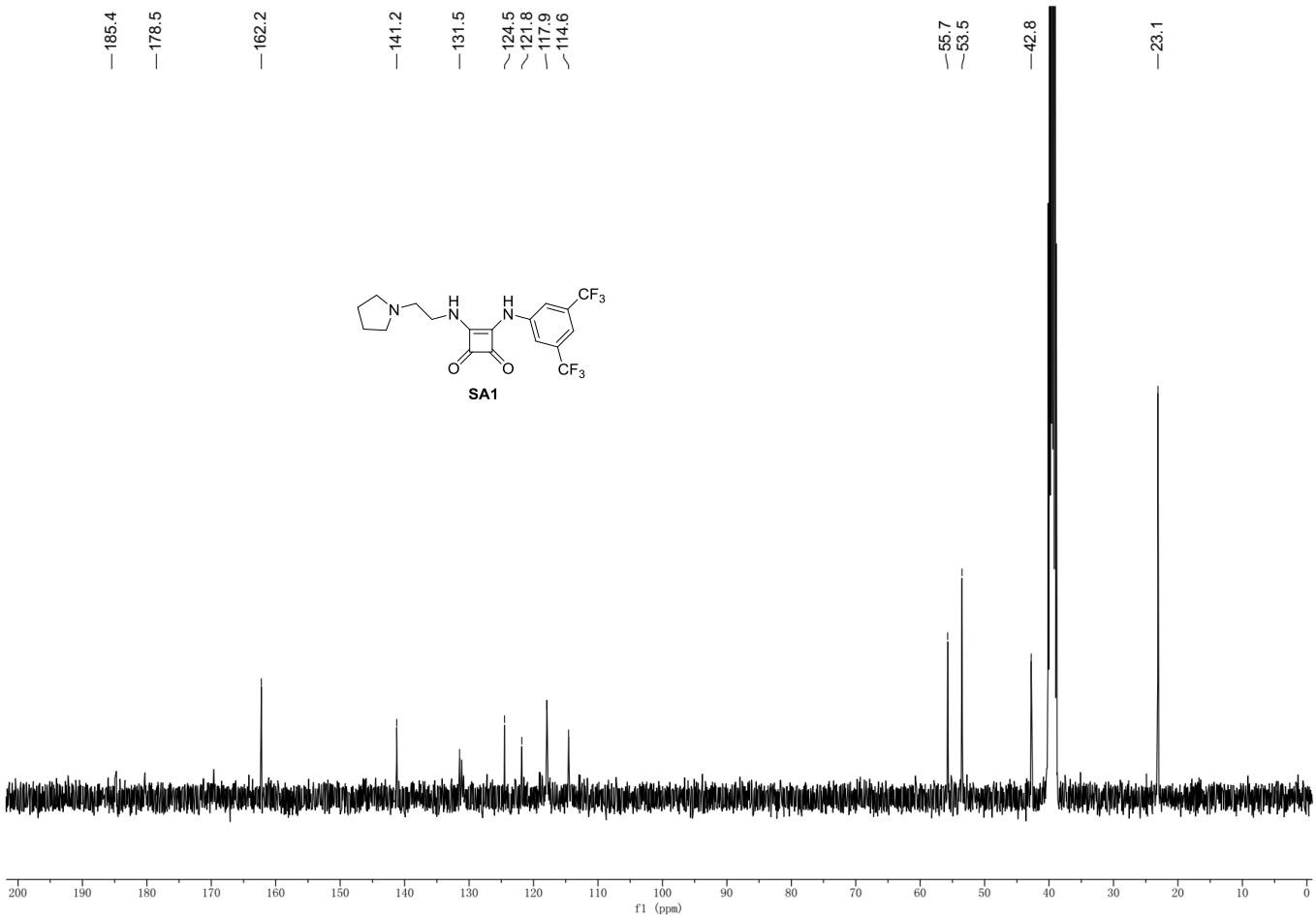
SA5 was purchased from Daicel Chiral Technologies (China) Co., Ltd. for only used in Lab; HRMS (ESI) *m/z* Calculated for $C_{22}H_{24}F_6N_3O_2^+$ $[M+H]^+$: 476.1767, found: 476.1767.



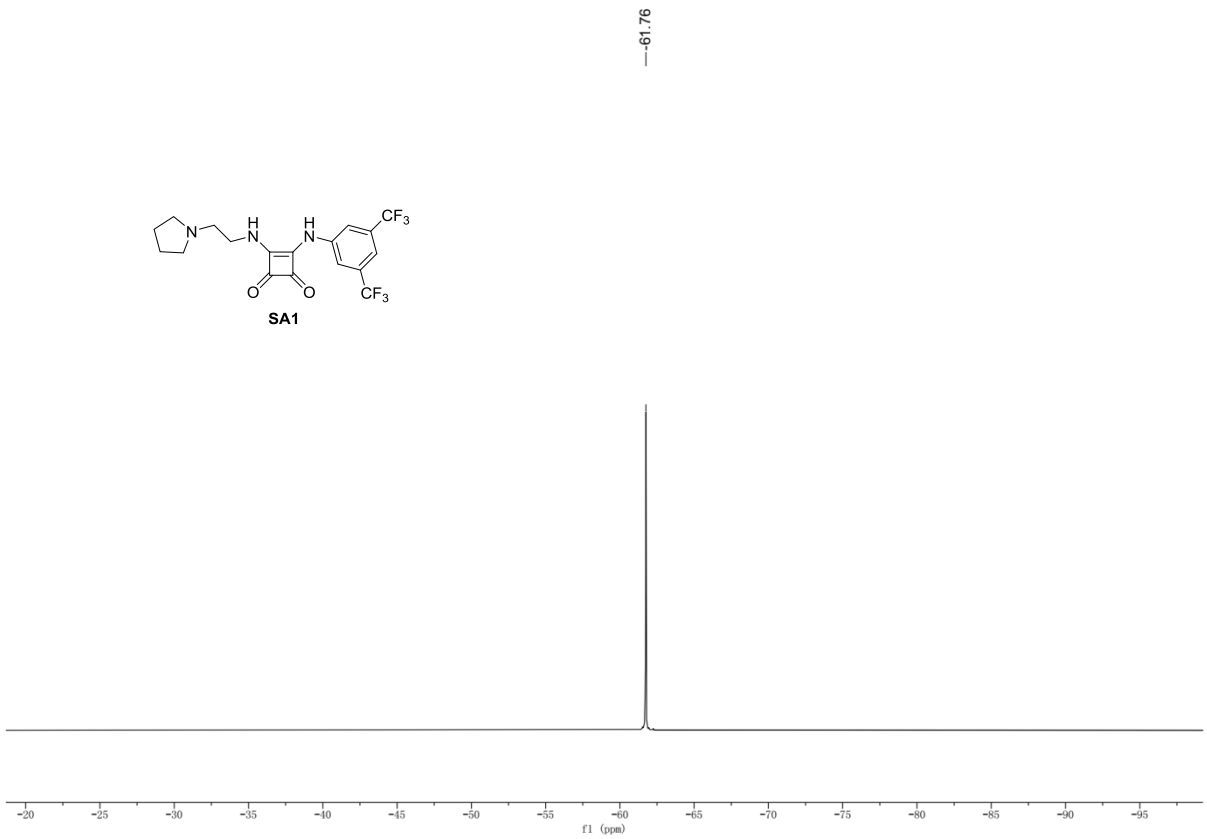
^{1}H NMR for **B**



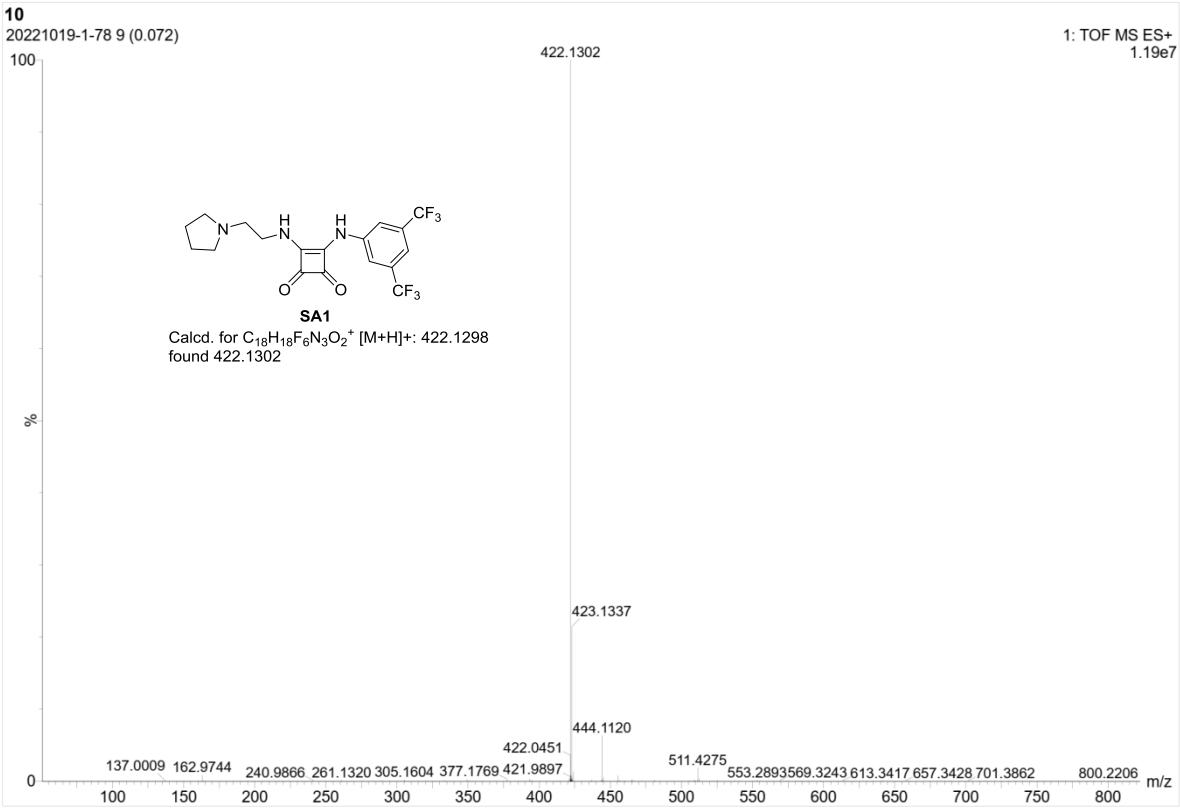
^{1}H NMR for **SA1**



^{13}C NMR for **SA1**



^{19}F NMR for **SA1**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422.0451
421.9897
137.0009
162.9744
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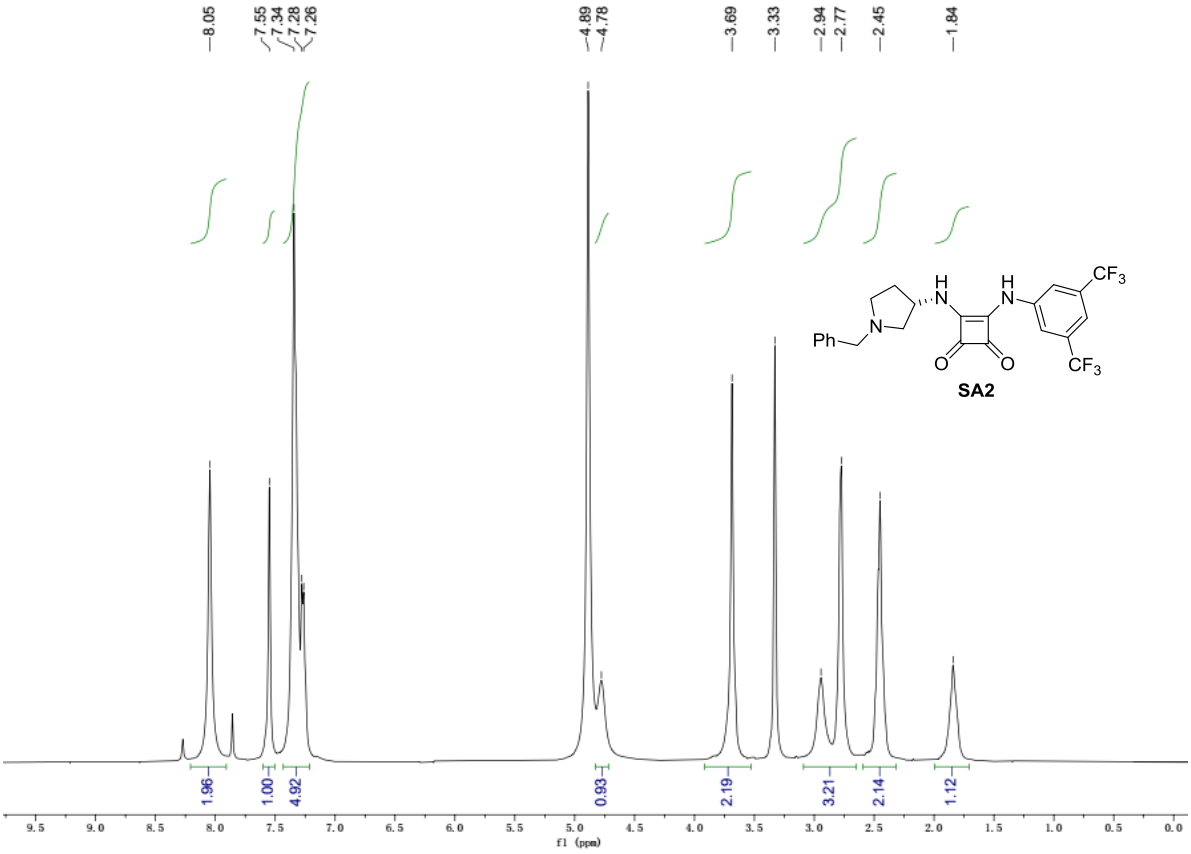
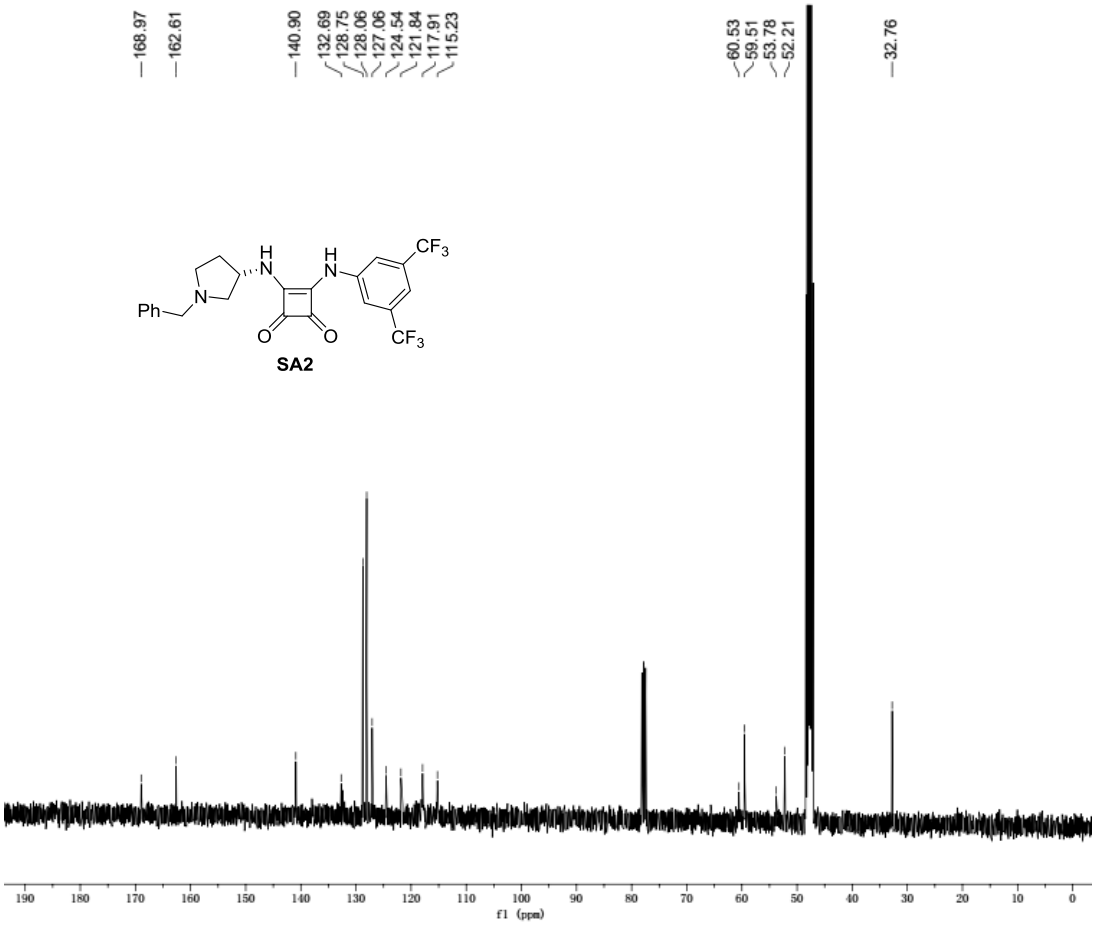
305.1604
377.1769
511.4275
553.2893
569.3243
613.3417
657.3428
701.3862
800.2206
m/z
100 150 200 250 300 350 400 450 500 550 600 650 700 750 800
SA1
Calcd. for $C_{18}H_{18}F_6N_3O_2^+$ [M+H]+: 422.1298
found 422.1302

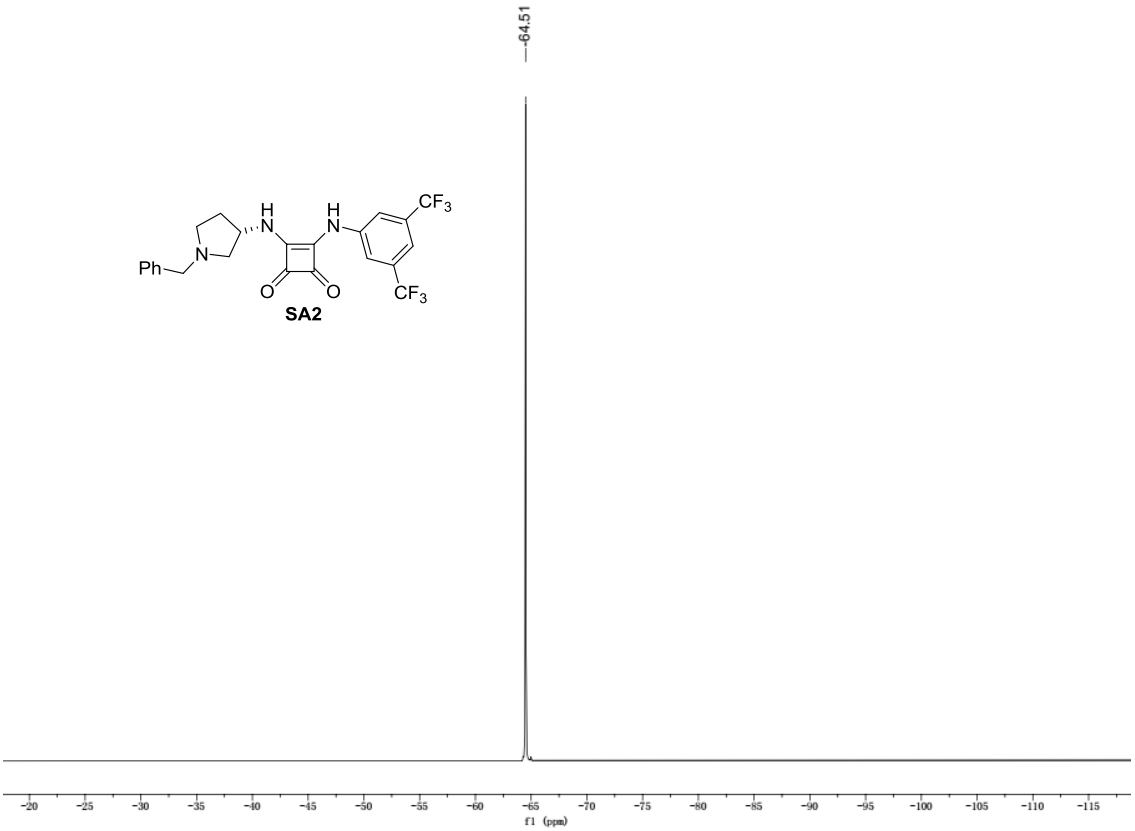
HRMS for **SA1**


8.05
7.55
7.34
7.28
7.26
4.89
4.78
3.69
3.33
2.94
2.77
2.45
1.84
1.96
1.00
4.92
0.93
2.19
3.21
2.14
1.12
f1 (ppm)
Ph
N
H
N
H
N
O
O
CF3
CF3
SA2

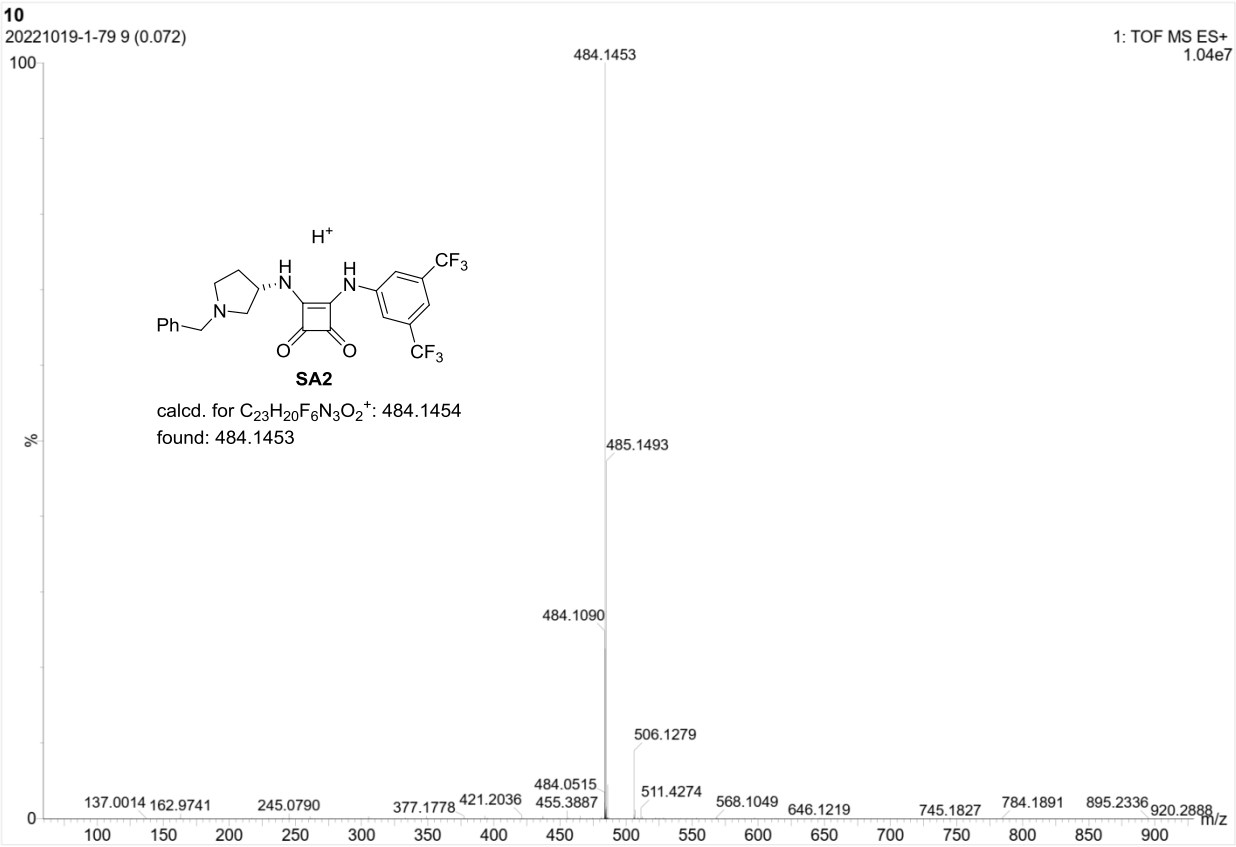
^{1}H NMR of **SA2**



^{13}C NMR of **SA2**



^{19}F NMR of **SA2**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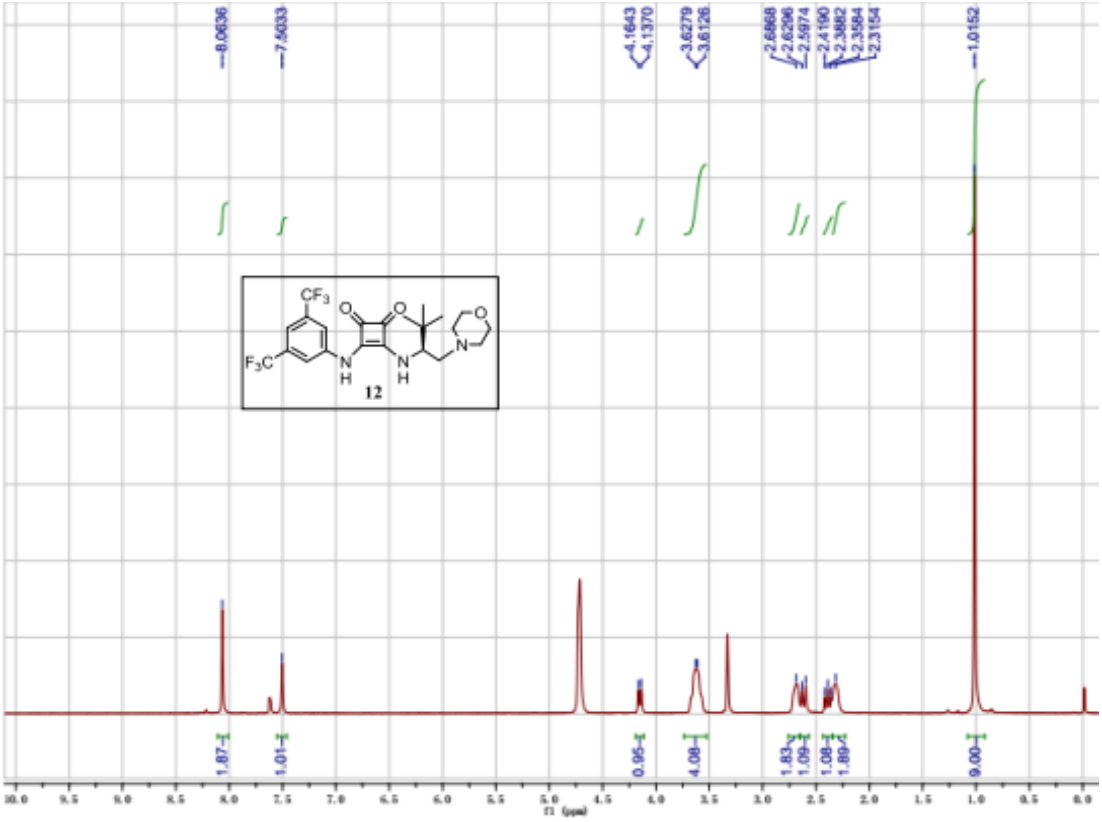
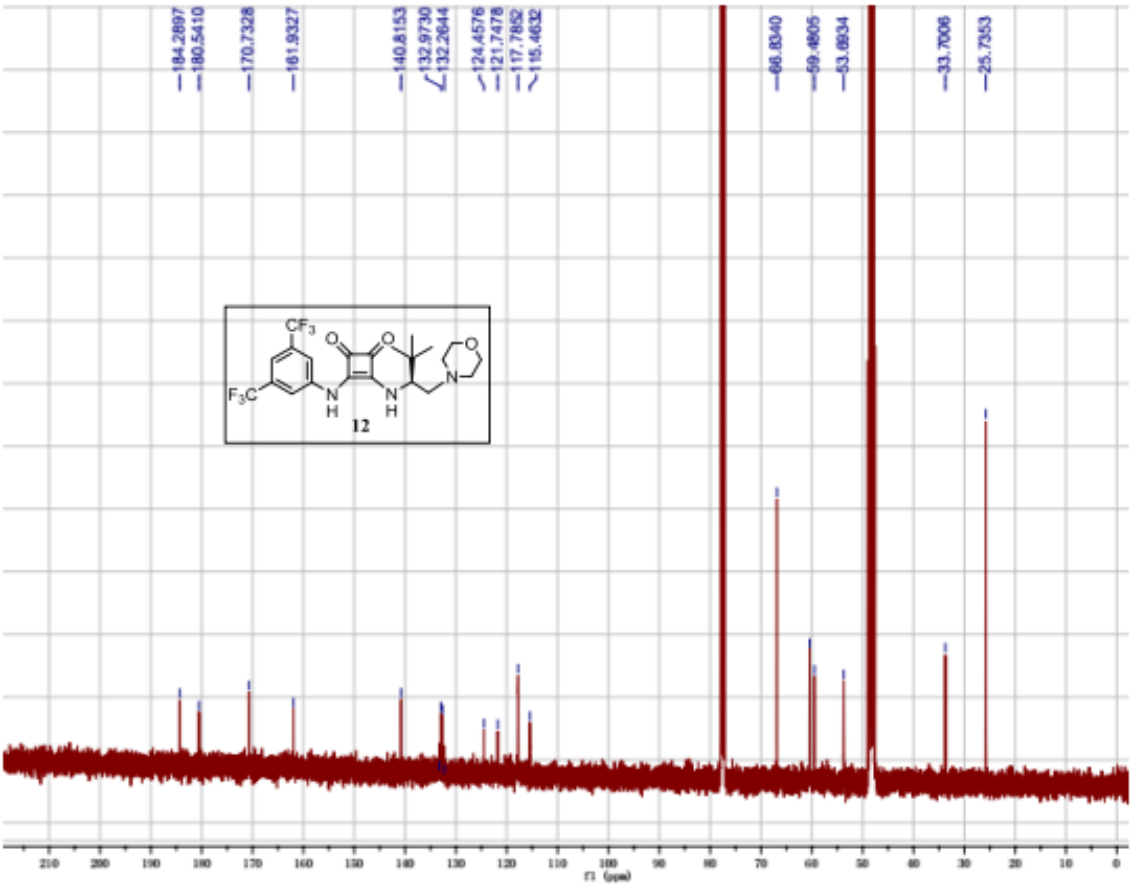
568.1049
646.1219
745.1827
784.1891
895.2336
920.2888
m/z
100 150 200 250 300 350 400 450 500 550 600 650 700 750 800 850 900
H+
H
N
H
N
CF3
CF3
Ph
N
O
O
SA2
calcd. for C23H20F6N3O2+: 484.1454
found: 484.1453

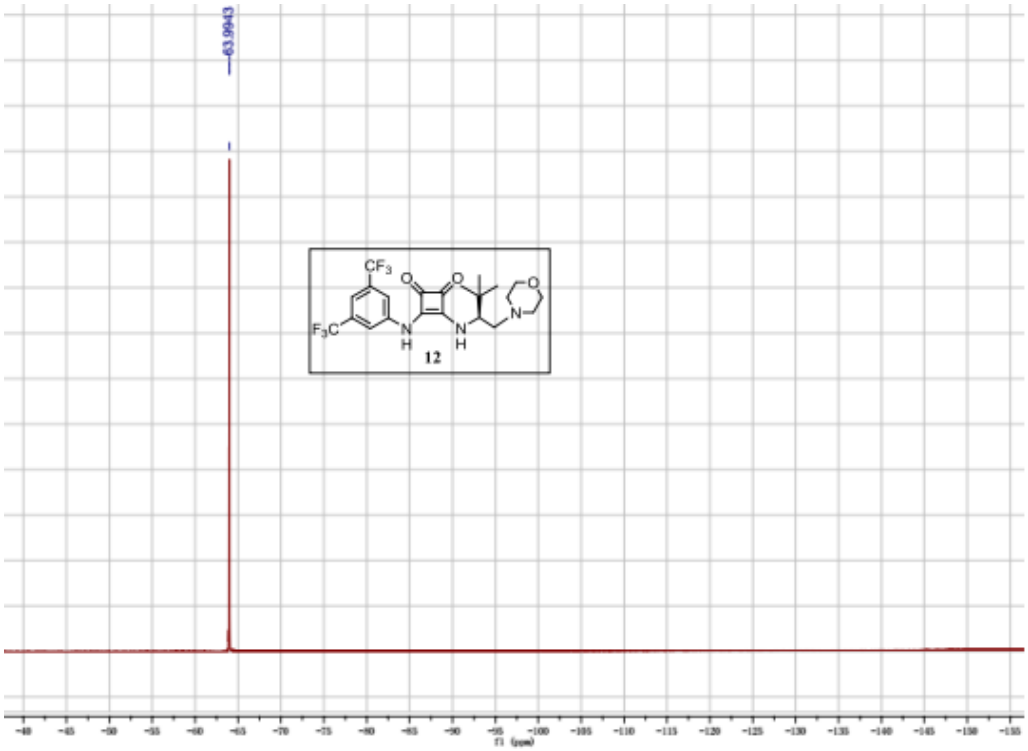
HRMS of **SA2**



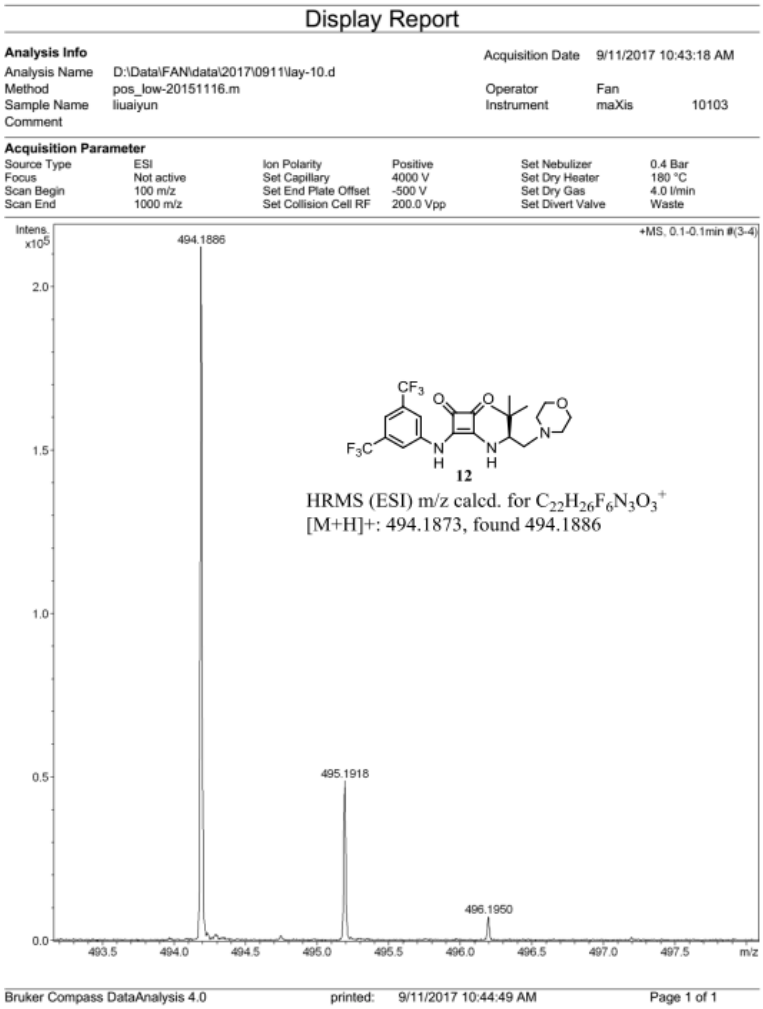
^{1}H NMR of **SA3**



^{13}H NMR of **SA3**



^{19}F NMR of **SA3**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Instrument maXis 10103
Acquisition Parameter
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Focus Not active
Scan Begin 100 m/z
Scan End 1000 m/z
Ion Polarity Positive
Set Capillary 4000 V
Set End Plate Offset -500 V
Set Collision Cell RF 200.0 Vpp
Set Nebulizer 0.4 Bar
Set Dry Heater 180 °C
Set Dry Gas 4.0 l/min
Set Divert Valve Waste
Intens. x10^5
+MS, 0.1-0.1min #(3-4)
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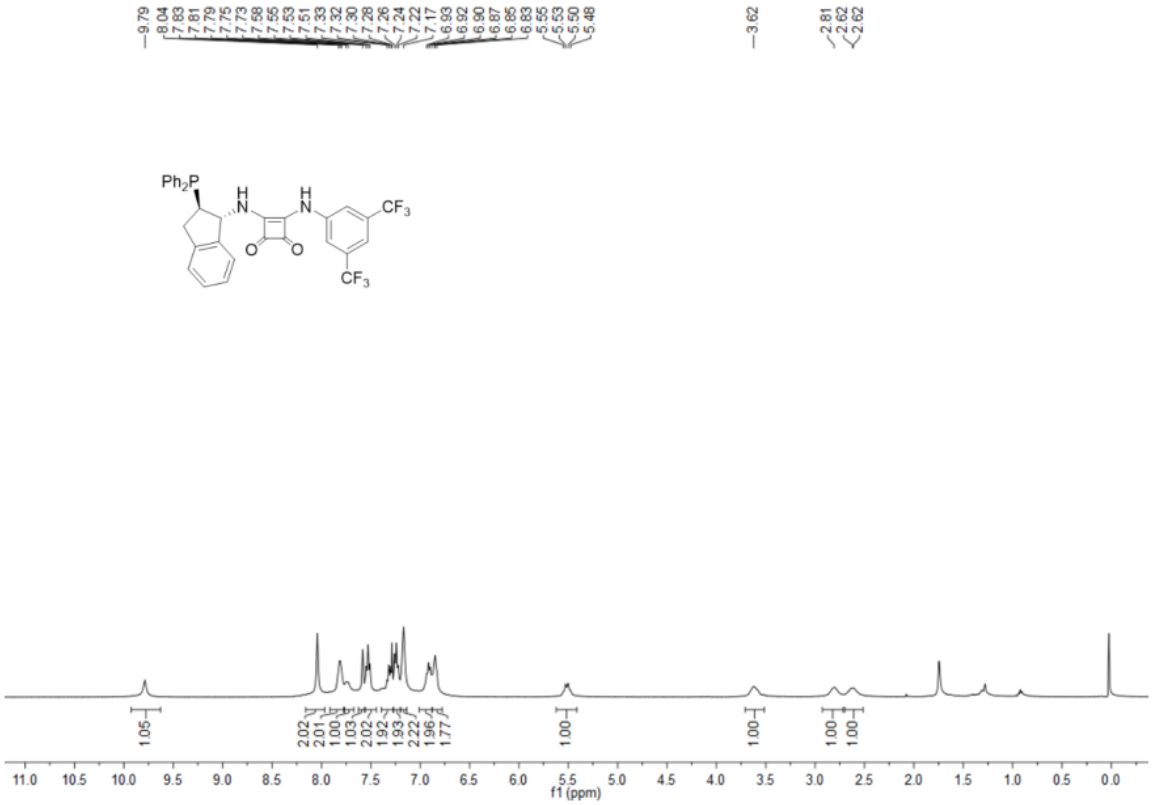
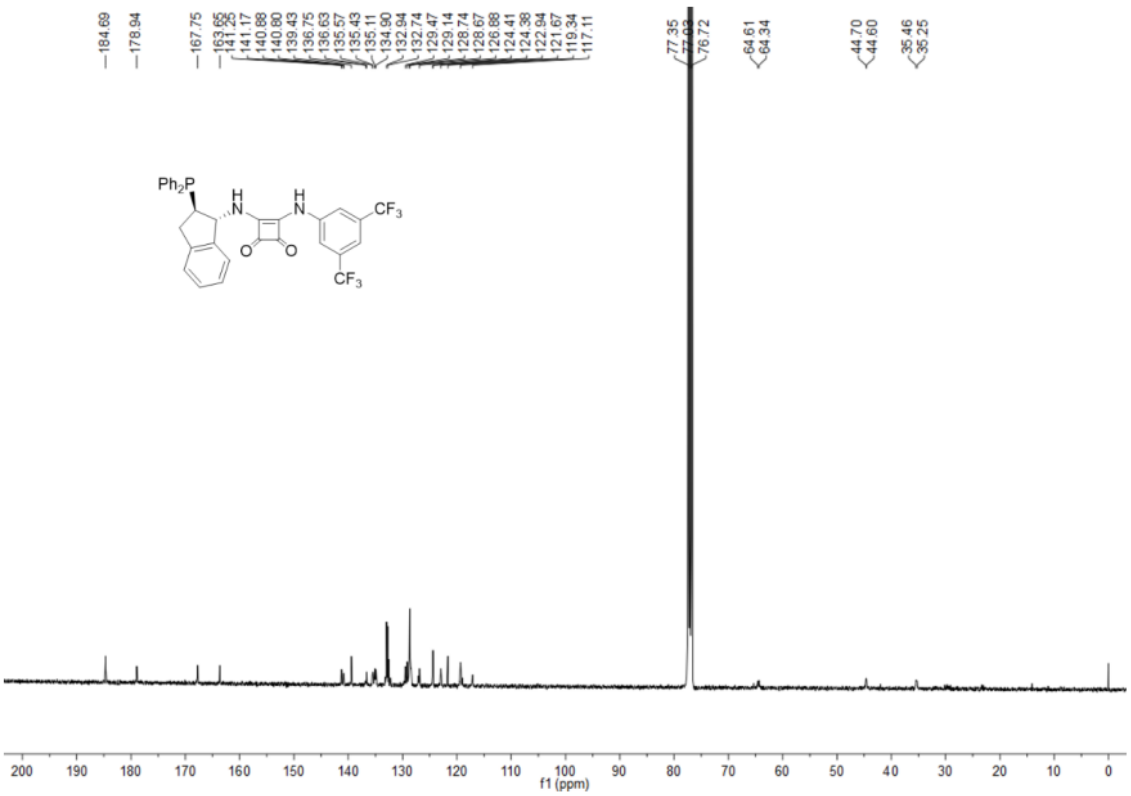
495.1918
496.1950
12
HRMS (ESI) m/z calcd. for C22H26F6N3O3+
[M+H]+: 494.1873, found 494.1886
m/z
Bruker Compass DataAnalysis 4.0
printed: 9/11/2017 10:44:49 AM
Page 1 of 1

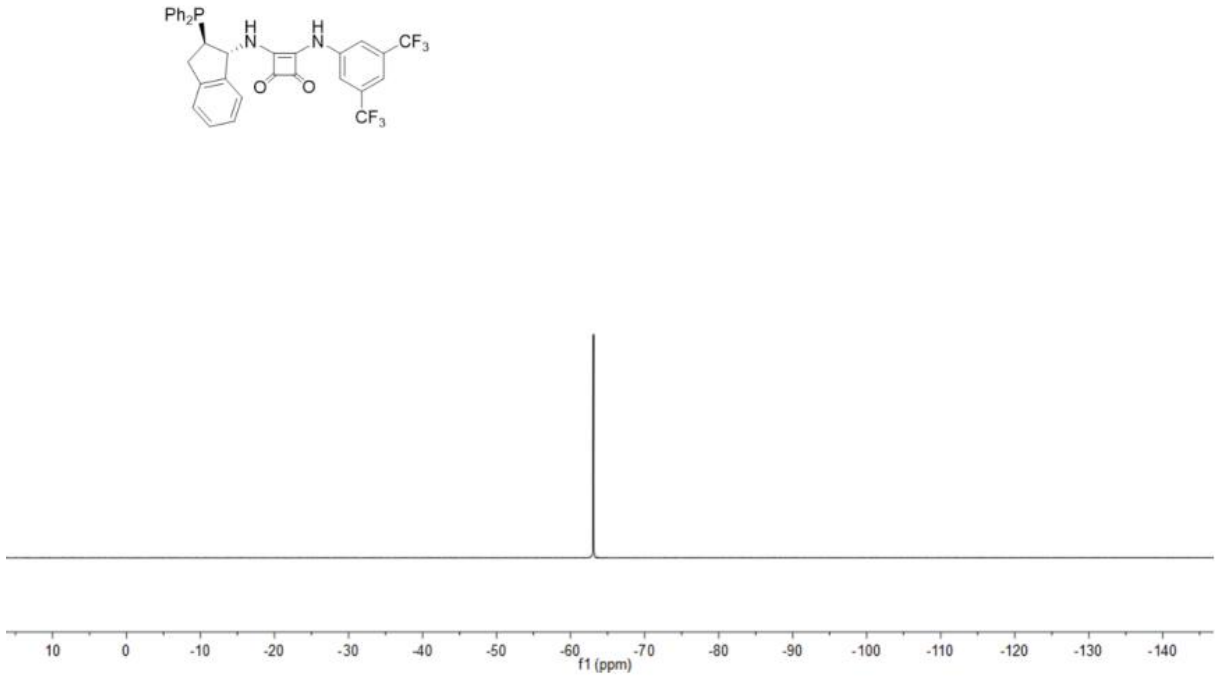
HRMS for **SA3**



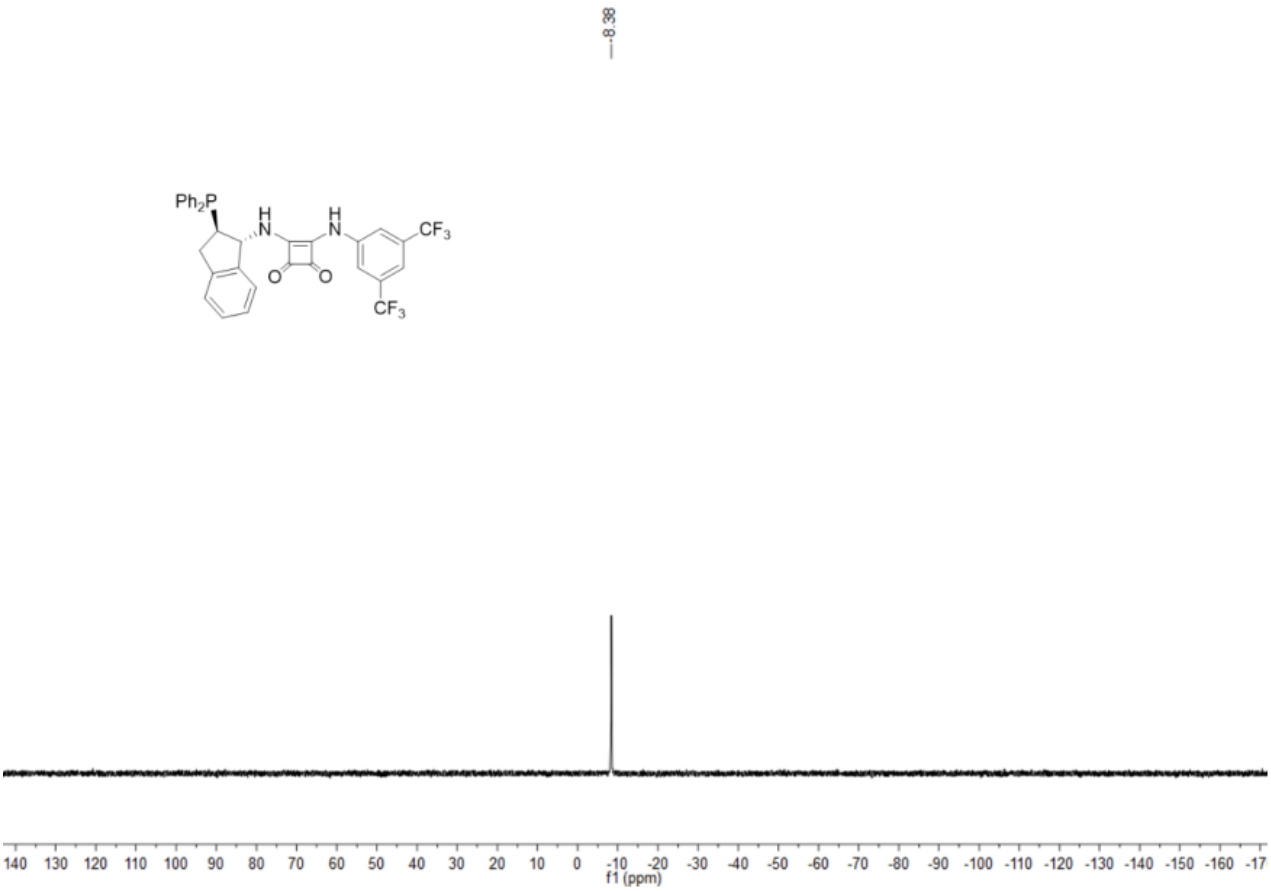
^{1}H NMR for **SA4**



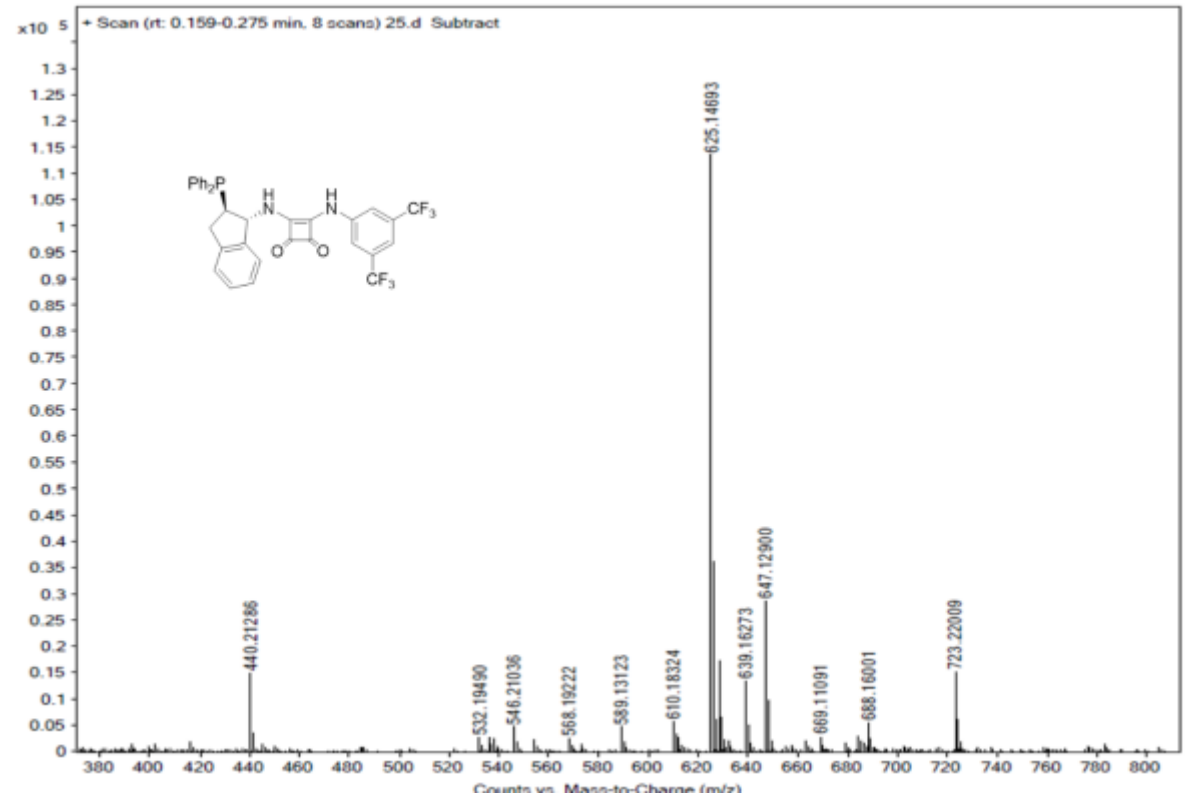
^{13}C NMR for **SA4**



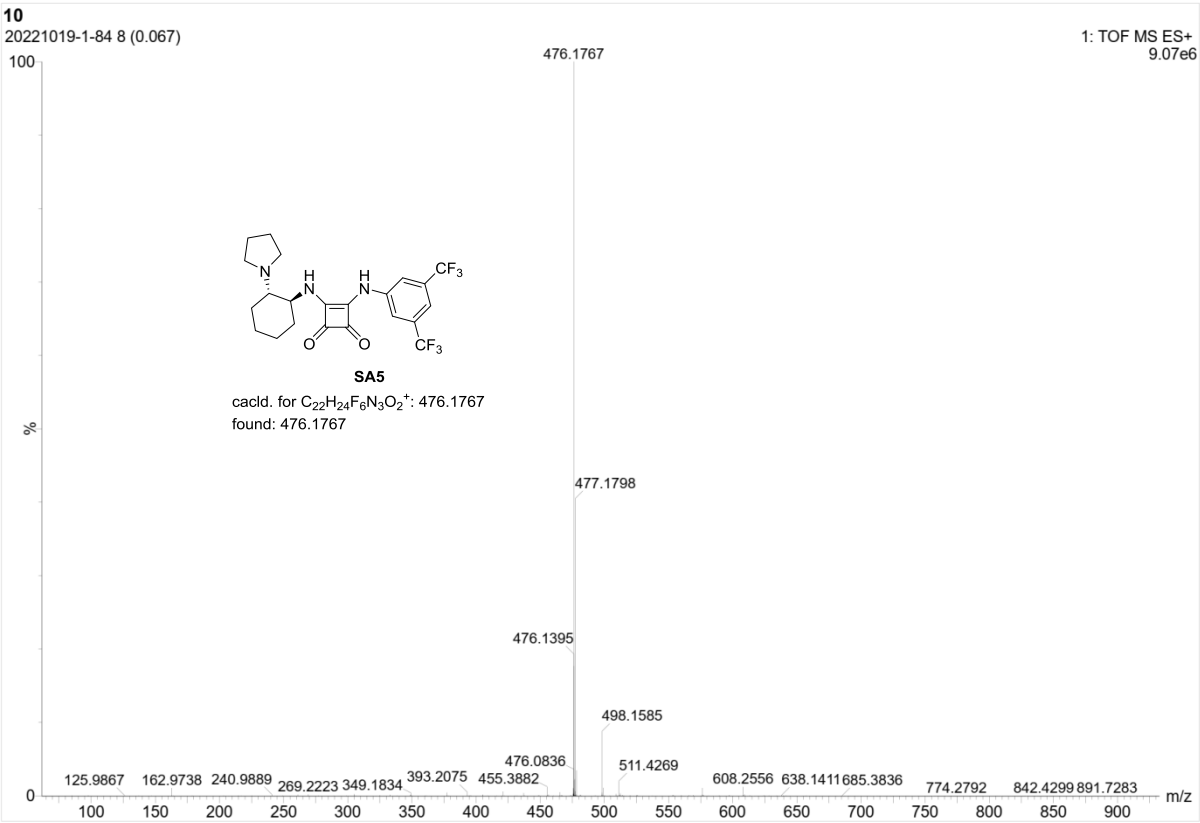
^{19}F NMR for **SA4**



^{31}P NMR for **SA4**



HRMS for **SA4**



HRMS for **SA5**

References

[1] Konishi H.; Lam T.Y.; Malerich J.P.; Rawal V.H. Enantioselective α-amination of 1,3-dicarbonyl compounds using squaramide derivatives as hydrogen bonding catalysts. *Org Lett* **2010**,12,2028-2031.

[2] Kowalczyk R.; Nowak A.E.; Skarzewski J. Organocatalytic asymmetric addition of aliphatic thiols to nitro olefins and nitrodienes. *Tetrahedron: Asymmetry* **2013**,24,505-514.

[3] Zhu J.; Cui D.; Li Y.; He J.; Chen W.; Wang P. Enantioselective amination of nitroolefins under base-free and water-rich conditions using chiral bifunctional phase-transfer catalysts. *Org Biomol Chem* **2018**,16,3012-3017.

[4] Li Y.; Cui D.; Zhu J.; Huang P.; Tian Z.; Jia Y.; Wang P. Bifunctional phase-transfer catalysts for fixation of CO_2 with epoxides under ambient pressure. *Green Chem*.**2019**, 21, 5231-5237.

Supplementary Table S1 Anti-bacterial activity of SA compounds

Compound/drug	MIC (μg/mL)	
	S. aureus (ATCC 29213)	MRSA (USA300)
Compound SA1	32	64
Compound SA2	2-4	4
Compound SA3	64	64
Compound SA4	>256	>256
Compound SA5	64	64
Oxacillin	2	>256
Ceftazidime	4	>256

Supplementary Table S2 Primers of RT-PCR

Gene		Primer sequence (5'-3')
norB	Forward	GCTACACCATCAACAGATACAGC
	Reverse	ACGCCAAATGCTCCACCTAA
ald	Forward	TGTAGCTTGCACACCCGAAA
	Reverse	CAGCTTCCCATGCTTGTTCG
tdcB	Forward	AGGCTATGGGGCAAAGGTTA
	Reverse	CGCCAACTGGTACGATGACT
gyrB	Forward	ACAGGAATCGGTGGCGACTTTG
	Reverse	GCTCCATCCACATCGGCATCAG