Supporting Information for

Phosphorescent Zinc Probe for Reversible Turn-On Detection with Bathochromically Shifted Emission

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Table S1. The Cartesian Coordinates of the Optimized Geometry of the Model Structure of ZIrdap S31
Table S2. The Cartesian Coordinates of the optimized geometry of the model structure of the zinc-bound form of ZIrdap S32

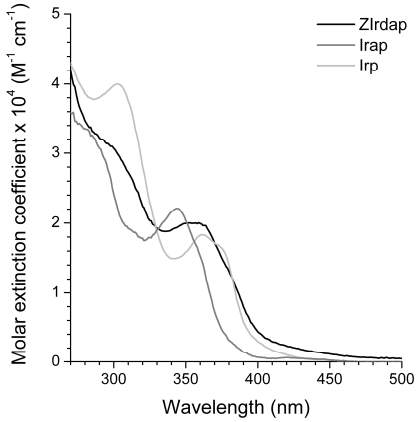


Figure S1. UV–vis absorption spectra of ZIrdap, Irap, and Irp (CH_3CN).

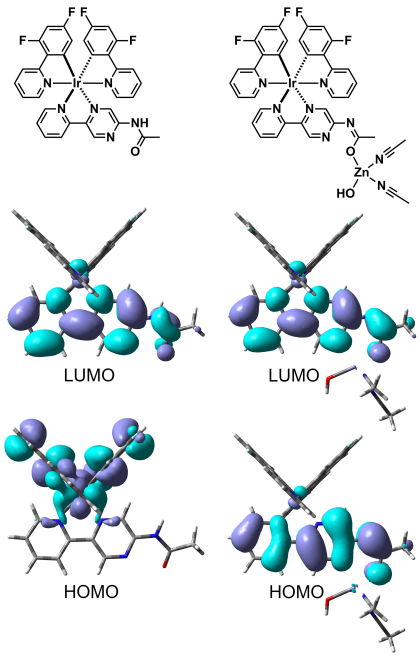
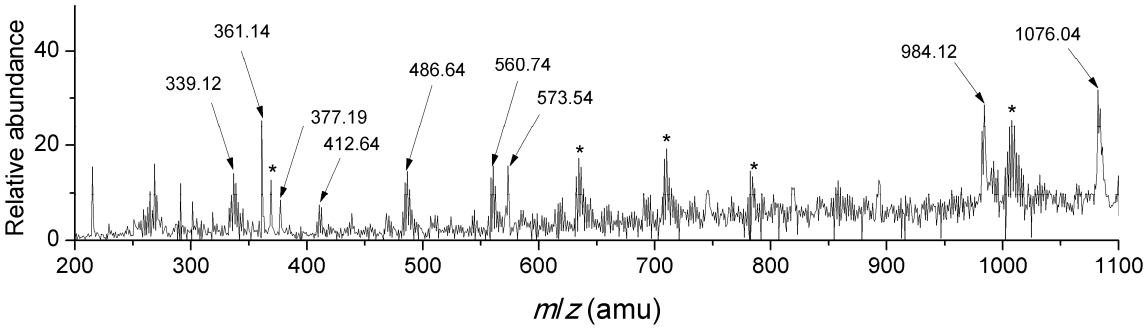


Figure S2. Isosurface of the molecular orbitals participating in the lowest triplet states of ZIrdap in the zinc-free (left) and zinc-bound (right) states. TD–CAM–B3LYP/LANL2DZ:6-311+G(d,p) calculations were performed for the optimized (B3LYP/LANL2DZ:6-311+G(d,p)) geometries. The DPA unit was removed from the model structures in order to reduce calculation costs. T_1 (3.60 eV) of the zinc-free form is constructed from the HOMO → LUMO (0.61) transition. Similarly, HOMO → LUMO (0.59) transition leads to T_1 (3.52 eV) of the zinc-bound form. The starting geometry for the optimization of the zinc-bound form of ZIrdap was constructed on the basis of the ESI MS result (SI, Figure S7). The Cartesian coordinates for the optimized structures are listed in Tables S1 & S2.



List of the Major Peaks and Their Assignments

Assignments	Found (*m*/*z*, amu)	Calcd. (amu)
$[(dap–pic)(H_2O)H^+]^+$	339.12	339.16
$[Na(dap–pic)(H_2O)]^+$	361.14	361.14
$[K(dap–pic)(H_2O)]^+$	377.19	377.11
$[(dap)H^+]^+$	412.64	412.19
$[K(dap)(H_2O)_2]^+$	486.64	486.17
$[K_2(dap)Cl_2H^+]^+$	560.74	560.05
$[Ir(C\wedge N)_2]^+$	573.54	573.06
$[ZIrdap]^+$	984.12	984.24
$[K(ZIrdap)Cl(H_2O)]^+$	1076.04	1076.18

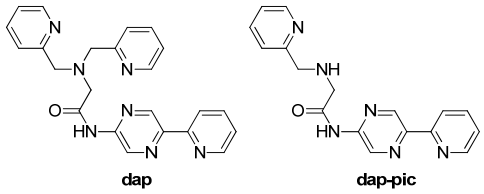


Figure S3. ESI MS spectrum for 50 μM ZIrdap. The solution was incubated in a pH 4.0 aqueous solution for 1 h prior to the measurement. The table lists the observed peaks and their assignments. The peaks marked with asterisks could not be assigned.

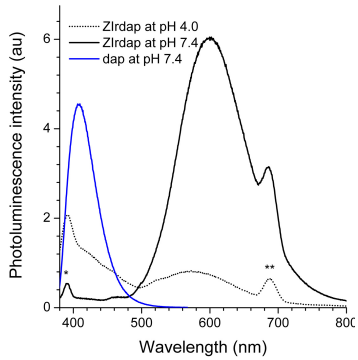


Figure S4. Photoluminescence spectra of 10 μM ZIrdap (dotted black line, after 1 h incubation at pH 4.0; solid black line, pH 7.4) and 10 μM dap ligand (blue). The asterisks, * and **, are due to the Raman scattering of water and the second harmonic of the excitation beam, respectively.

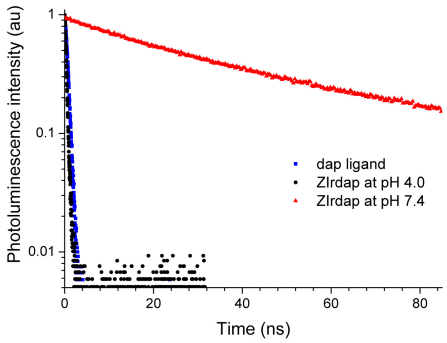


Figure S5. Photoluminescence decay traces of 50 μM dap ligand (blue; λ_{obs} = 400 nm, 20 mM PIPES buffer, pH 7.4) and 50 μM ZIrdap (black; λ_{obs} = 400 nm, incubated at pH 4.0 for 1 h) after picosecond photoexcitation at 345 nm, and decay traces of 50 μM ZIrdap (red; λ_{obs} = 600 nm, 20 mM PIPES buffer, pH 7.4) after nanosecond photoexcitation.

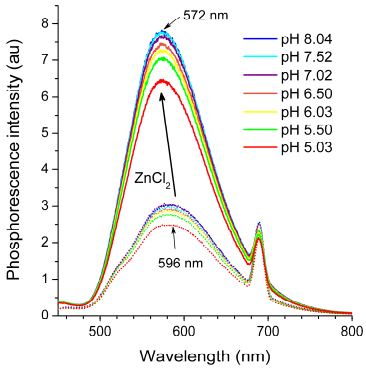


Figure S6. Phosphorescence spectra of 10 μM ZIrdap (air-equilibrated water) before (dotted lines) and after (solid lines) addition of 5 equiv $ZnCl_2$ at different pH values. λ_{ex} = 345 nm.

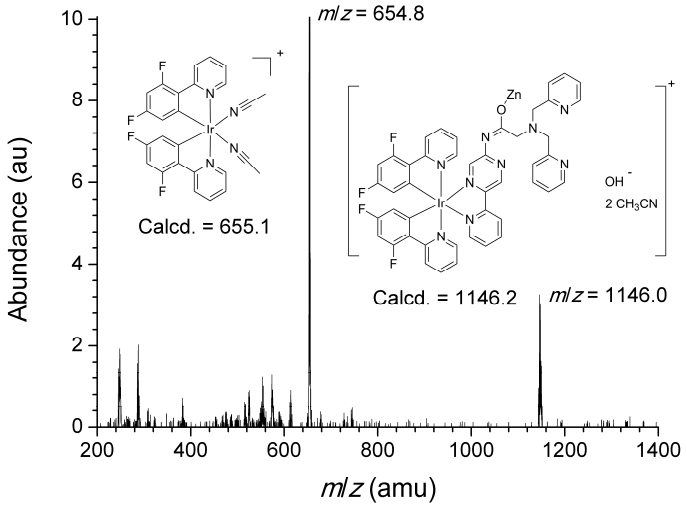


Figure S7. Positive mode ESI MS spectrum of an acetonitrile solution containing 100 μM ZIrdap and 1 mM $Zn(ClO_4)_2$. Inset figures show the proposed structures that correspond to the observed peaks at *m*/*z* = 654.8 and 1146.0.

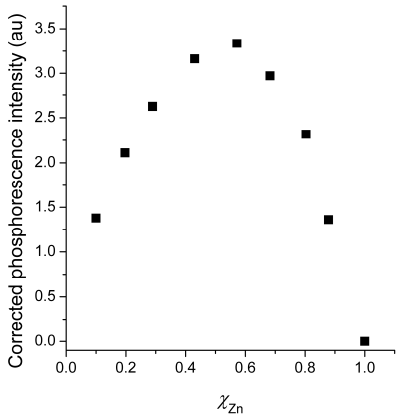


Figure S8. Phosphorescent Job's plot for the zinc binding of ZIrdap. Total salt concentration = 10 μM. Air-equilibrated aqueous buffer solutions were used for the measurement.

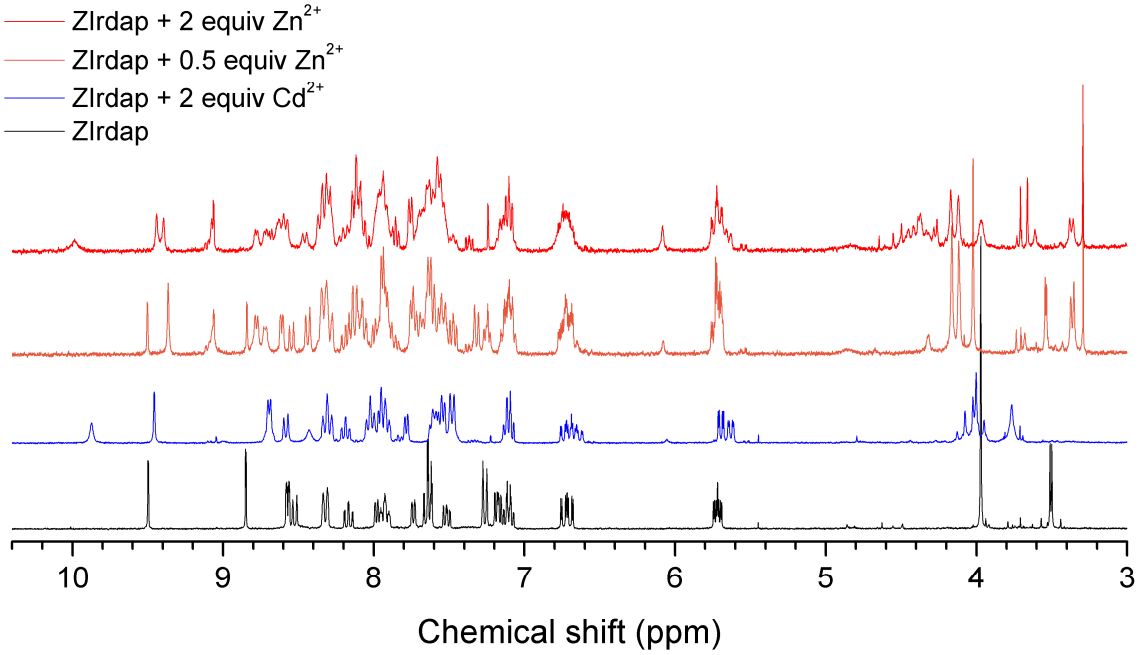


Figure S9. ^{1}H NMR spectra (300 MHz) of CD_3CN solutions of 5.0 mM ZIrdap in the absence (black) and presence of 10 mM Cd ions (blue) and 2.5 mM (orange) and 10 mM (red) zinc ions.

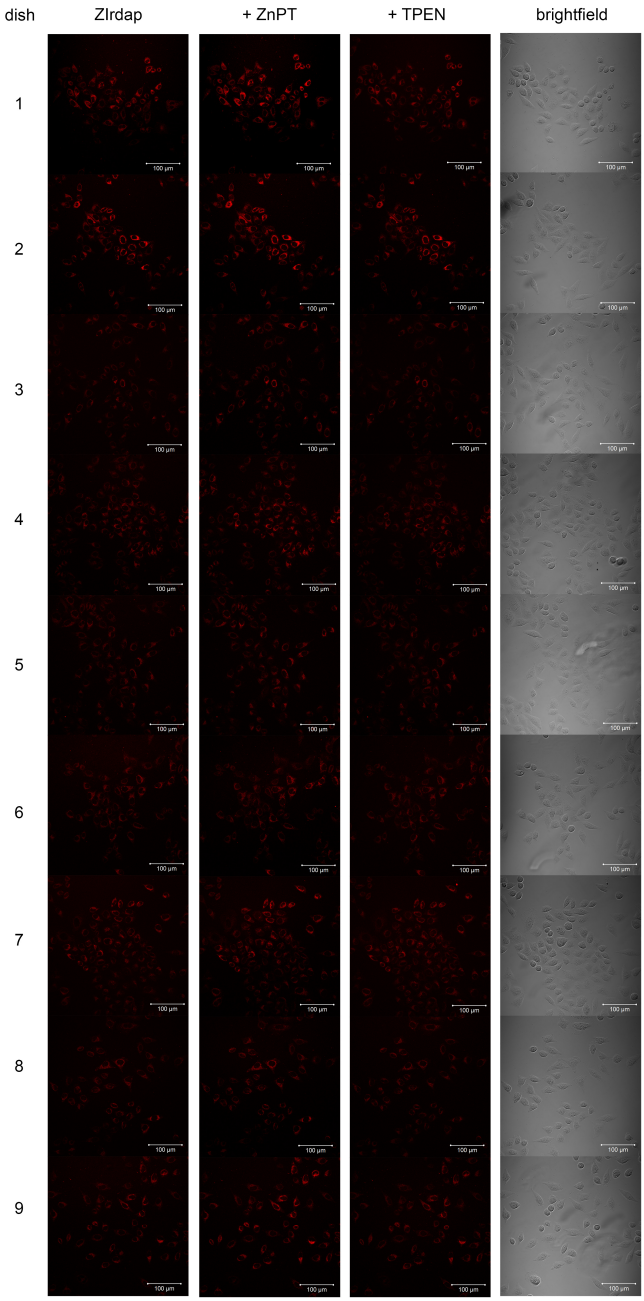
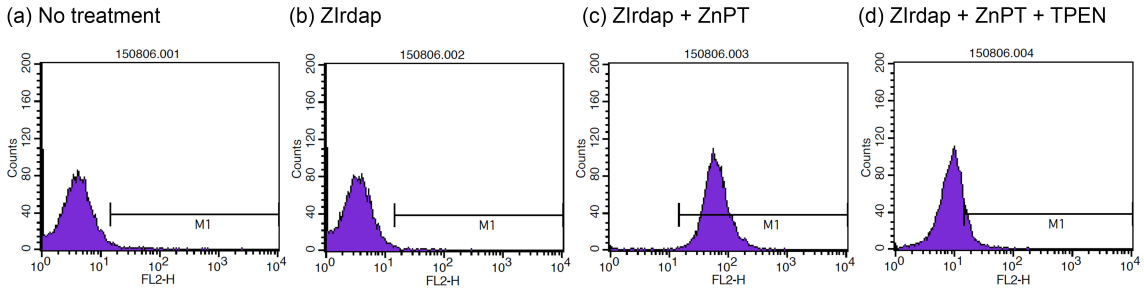


Figure S10. Phosphorescence visualization of exogenously supplied zinc ions in the form of ZnPT ($ZnCl_2$: sodium pyrithione = 1 : 2, mol/mol): From the left: HeLa cells incubated with 10 μM ZIrdap for 10 min, cells subsequently treated with 50 μM ZnPT, after the successive treatment with 100 μM TPEN, bright field image. λ_{em} = 441–691 nm (λ_{ex} = 405 nm). Scale bar = 100 μm.



	Events within the M1 marker (%)	Geo mean
(a) No treatment	1.20	27.86
(b) ZIrdap	0.49	26.34
(c) ZIrdap + ZnPT	99.60	60.06
(d) ZIrdap + ZnPT + TPEN	10.92	18.33

Figure S11. FACS analysis data for HeLa cells: (a) untreated cells, (b) cells incubated with 10 μM ZIrdap, (c) cells incubated with 10 μM ZIrdap and 50 μM ZnPT, and (d) cells incubated with 10 μM ZIrdap, 50 μM ZnPT, and 100 μM TPEN. After the treatments, the cells were washed with fresh DPBS. The cells were detached from the bottom by the addition of trypsin, and the suspensions were transferred into 1.4 mL Eppendorf tubes. The cell suspensions were centrifuged, after which the supernatant liquids were decanted. The cells were re-suspended in fresh DMEM, and transferred into FACS tubes for the measurements. FACS analyses were performed using a BD, FACSCalibur instrument using a FL2 channel (i.e., excitation at 488 nm and a 585/42 nm emission filter).

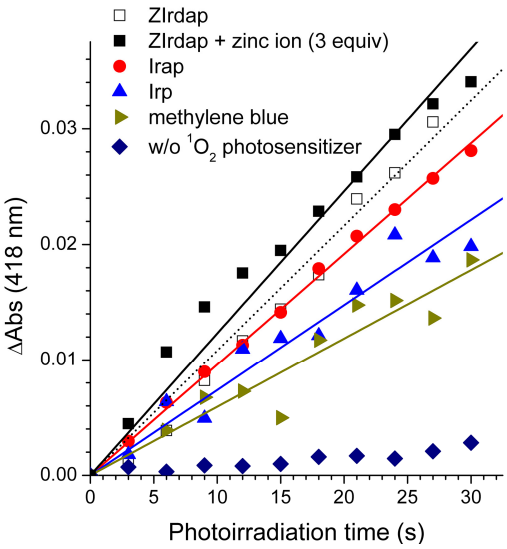


Figure S12. Determination of the quantum yield for singlet oxygen generation by zinc-free ZIrdap (black empty squares), zinc-bound ZIrdap (black filled squares), Irap (red), and Irp (blue), using the methylene blue standard (dark yellow; reference compounds, Φ_Δ = 0.52). DMSO solutions containing either the Ir(III) complex or MB and 1,3-diphenylisobenzofuran (DPBF) as an oxidation substrate were irradiated under UV light (365 nm). A DMSO solution of DPBF without a singlet oxygen generating sensitizer was used as a control (navy). Changes were monitored in the 418 nm absorption band (ΔAbs(418 nm)) of DPBF as a function of irradiation time. Singlet oxygen quantum yields (Φ_Δ) were calculated by the equation S1, in which *m* is the slope of a linear fit to the ΔAbs(418 nm) data and $F = 1-10^{-\text{O.D.}}$, where O.D. refers to the optical density of the Ir(III) complex or MB at 365 nm.

$$\Phi_\Delta = \Phi_\Delta(\text{ref}) \times \frac{m \cdot F_{\text{ref}}}{m_{\text{ref}} \cdot F} \tag{S1}$$

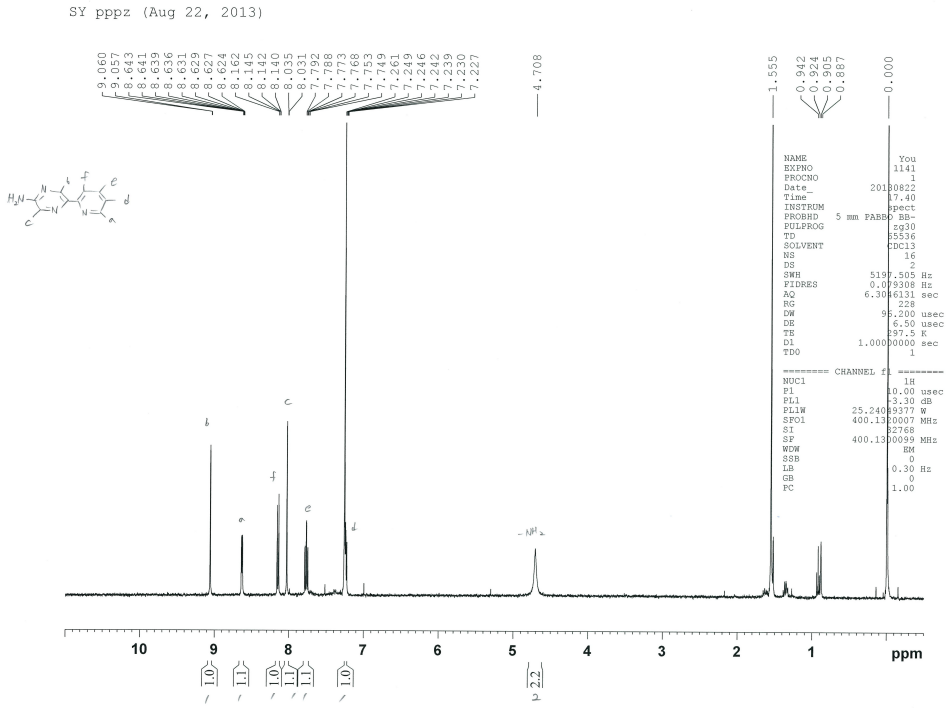


Figure S13. ^{1}H NMR (400 MHz, $CDCl_3$) spectrum of 2-amino-5-(2-pyridyl)pyrazine.

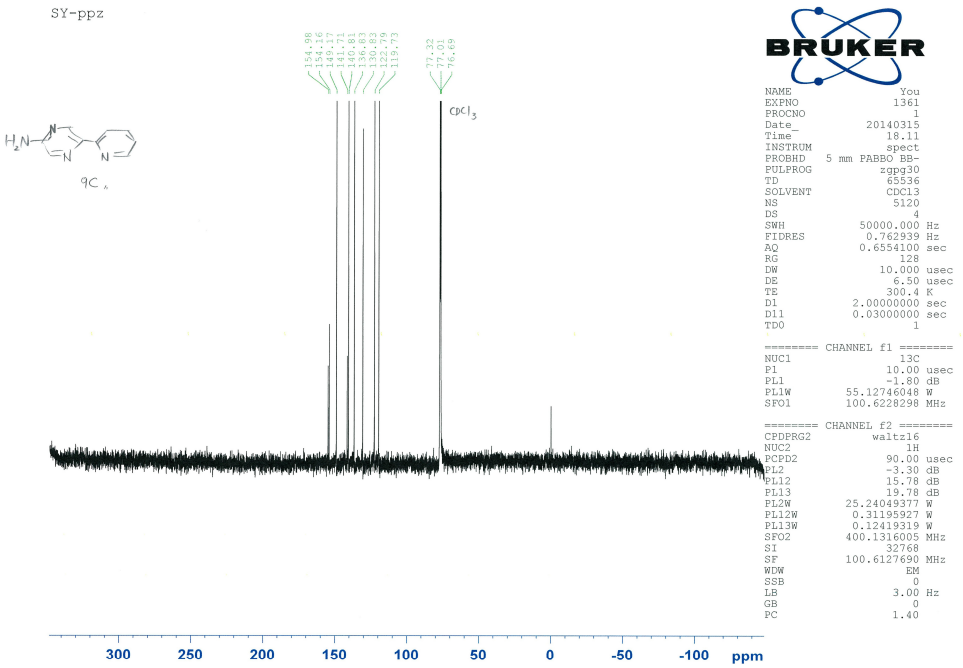


Figure S14. ^{13}C NMR (100 MHz, $CDCl_3$) spectrum of 2-amino-5-(2-pyridyl)pyrazine.

File: 10587
Sample: ppz

Date Run: 05-22-2014

Time Run: 16:51:08

Run By: 공동기기원

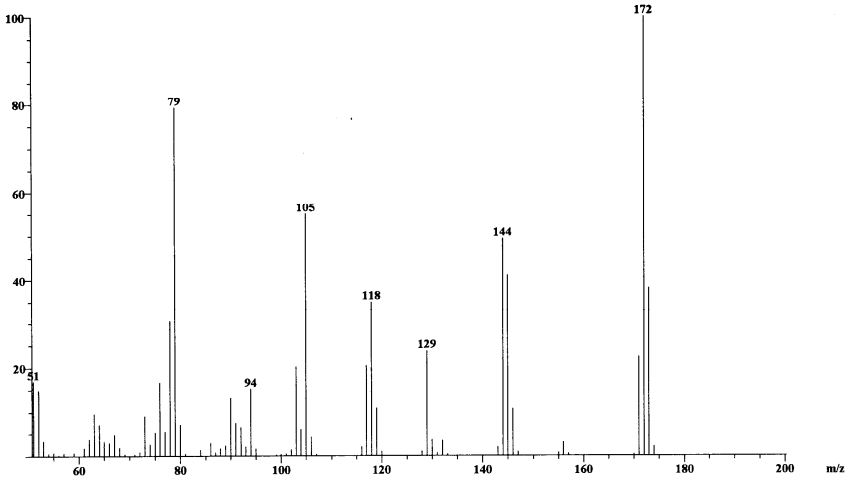
Instrument: JEOL JMS600
Inlet: Direct Probe

Ionization mode: EI+

R.T.: 0:01.95

Scan: 1-2
Base: m/z 172; 100%FS

TIC: 8014496



Page 1

File: 10599
Sample: ppz

Date Run: 05-26-2014

Time Run: 14:08:38

Run By: 공동기기원

Instrument: JEOL JMS600
Inlet: Direct Probe

Ionization mode: EI+

R.T.: 1:51.2

Scan: 93
Base: m/z 217; 8.9%FS

TIC: 1094907

Selected Isotopes : $C_{0\text{-}50}H_{0\text{-}100}N_{0\text{-}5}$

Error Limit : 20 ppm

Measured Mass	*% Base*	*Formula*	*Calculated Mass*	*Error*
172.0746	73.2%	$C_9H_8N_4$	172.0749	1.6

Figure S15. HR MS (EI) spectrum of 2-amino-5-(2-pyridyl)pyrazine.

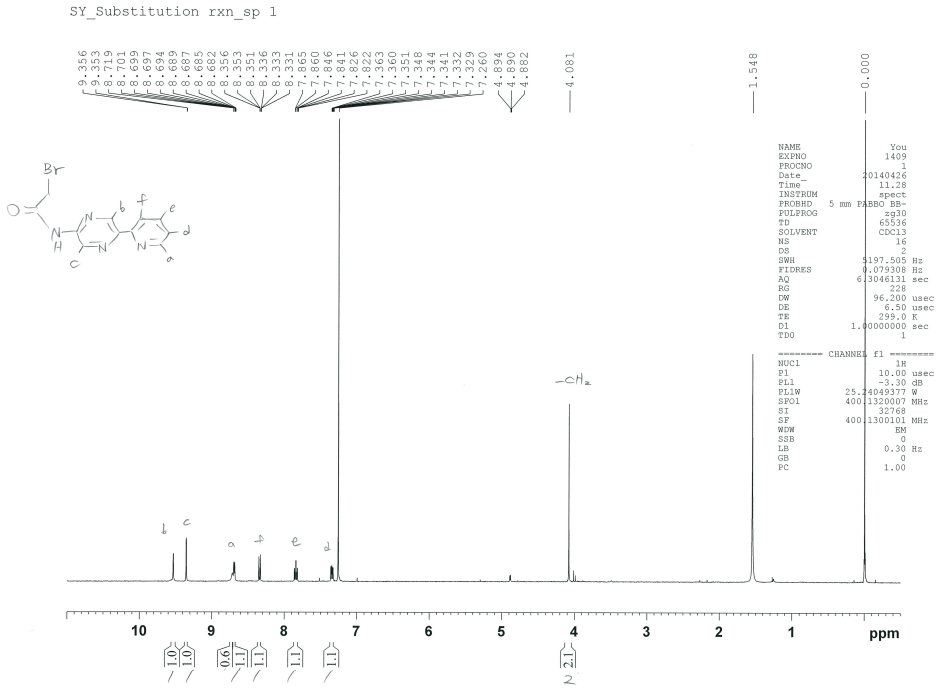


Figure S16. ^{1}H NMR (400 MHz, $CDCl_3$) spectrum of 2-(bromoacetylamino)-5-(2-pyridyl)pyrazine.

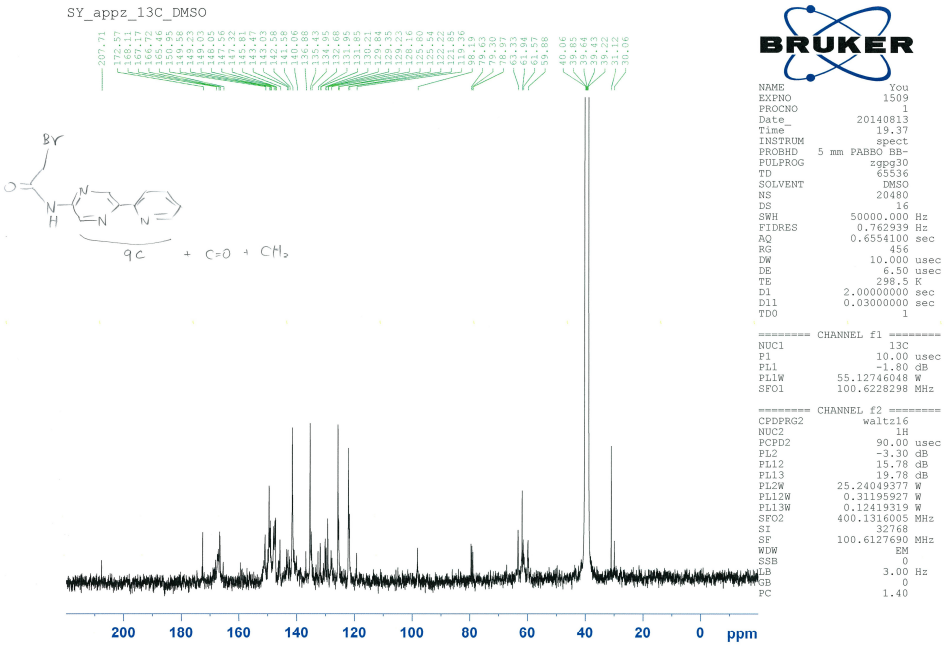


Figure S17. ^{13}C NMR (100 MHz, DMSO-d_6) spectrum of 2-(bromoacetylamino)-5-(2-pyridyl)pyrazine.

File: 10586
Sample: appz

Instrument: JEOL JMS600
Inlet: Direct Probe

Date Run: 05-22-2014

Ionization mode: EI+

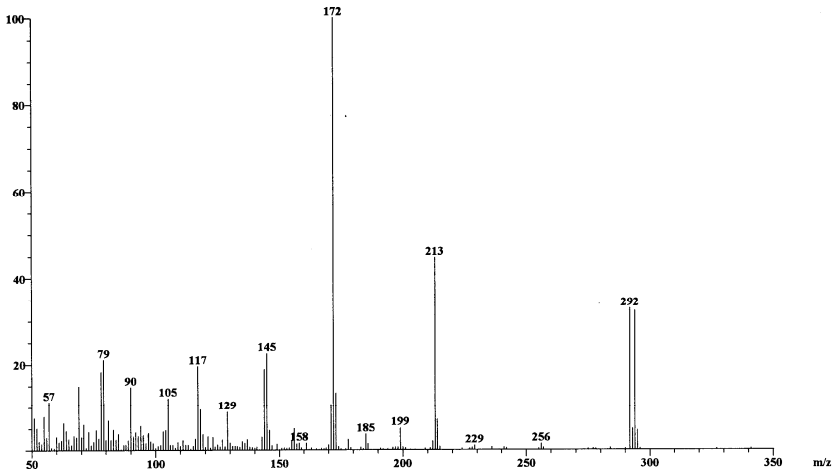
Time Run: 15:27:24

Run By: 공동기기원

R.T.: 0:45.7

Scan: 18-20
Base: m/z 172; 91.2%FS

TIC: 6511264



Page 1

File: 10598
Sample: appz

Instrument: JEOL JMS600
Inlet: Direct Probe

Date Run: 05-23-2014

Ionization mode: EI+

Time Run: 17:42:13

Run By: 공동기기원

R.T.: 1:14.3

Scan: 60-65
Base: m/z 267; 9.4%FS

TIC: 1071910

Selected Isotopes : $H_{0-50}C_{0-50}N_{0-10}O_{0-3}Br_{0-2}{}^{81}Br_{0-2}$

Error Limit : 20 ppm

Measured Mass	*% Base*	*Formula*	*Calculated Mass*	*Error*
291.9957	10.0%	$C_{23}O$	291.9949	-2.7
		$C_{18}N_2O_3$	291.9909	-17.0
		$C_9H_7N_7Br$	291.9946	-3.9
		$C_{11}H_9N_4O\,Br$	291.9959	0.7
		$C_{13}H_{11}N\,O_2Br$	291.9973	5.4
		$C_4H_7N_9O_2Br$	291.9905	-18.0
		$C_6H_9N_6O_3Br$	291.9919	-13.0
		$C_{12}H_9N_3O\,{}^{81}Br$	291.9908	-17.0
		$C_4H_7N_{10}O\,{}^{81}Br$	291.9967	3.4
		$C_{14}H_{11}O_2{}^{81}Br$	291.9922	-12.0
		$C_6H_9N_7O_2{}^{81}Br$	291.9980	8.0
		$C_8H_{11}N_4O_3{}^{81}Br$	291.9994	13.0

Figure S18. HR MS (EI) spectrum of 2-(bromoacetylamino)-5-(2-pyridyl)pyrazine.

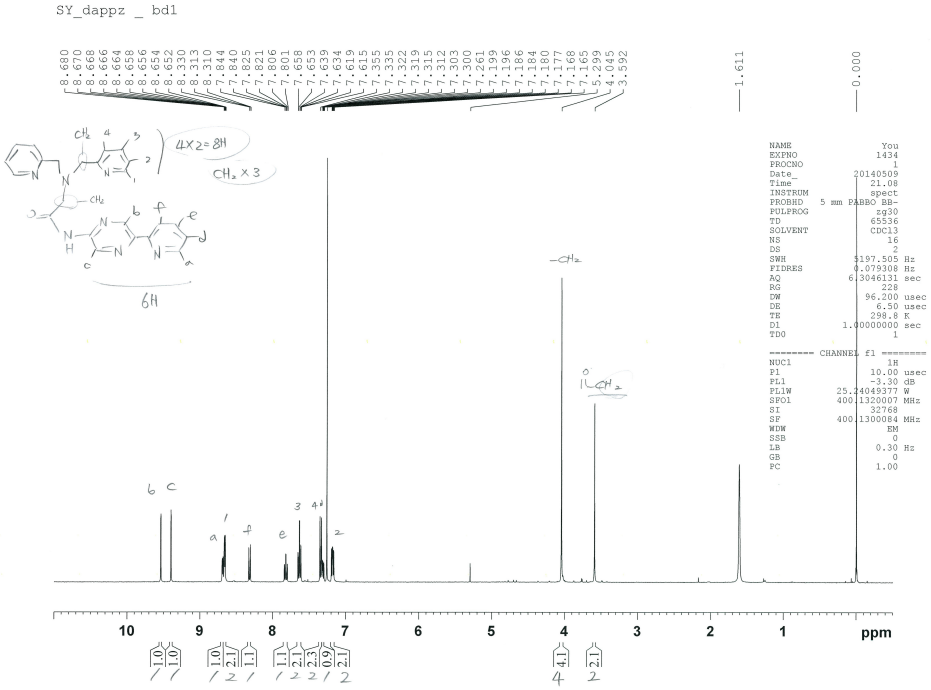


Figure S19. ^{1}H NMR (400 MHz, $CDCl_3$) spectrum of 2-((di(2-picolyl)amino)acetylamino)-5-(2-pyridyl)pyrazine.

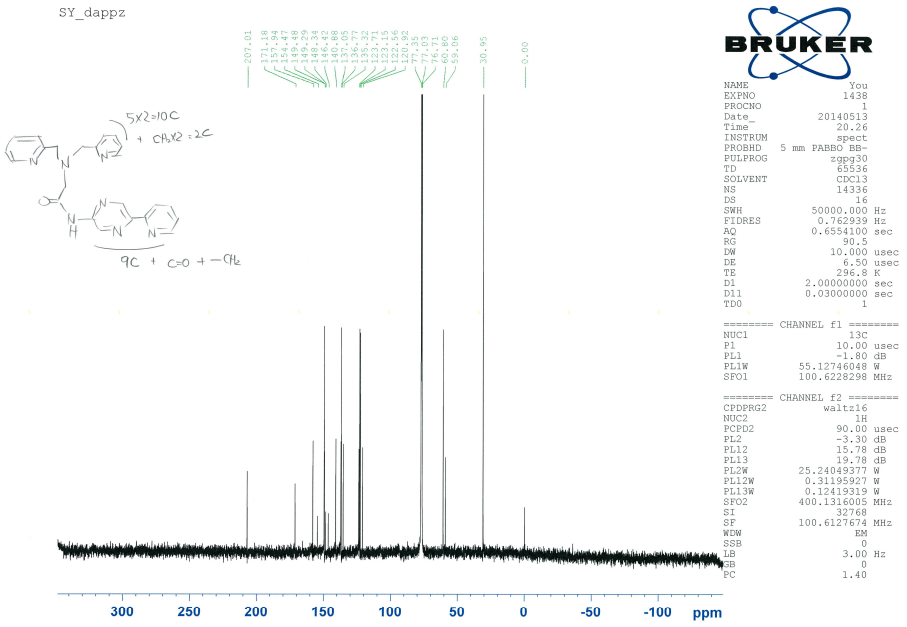


Figure S20. ^{13}C NMR (100 MHz, CDCl3) spectrum of 2-((di(2-picolyl)amino)acetylamino)-5-(2-pyridyl)pyrazine.

Page 1

File: 10588
Sample: dappz

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Time Run: 17:11:18

Run By: 공동기기원

Instrument: JEOL JMS600
Inlet: Direct Probe

Ionization mode: EI+

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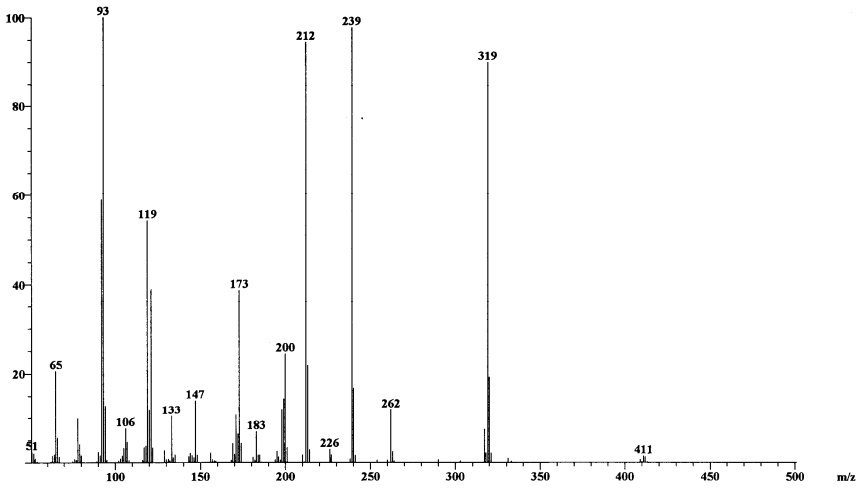
TIC: 6700238



Page 1

File: 10600
Sample: dappz

Date Run: 05-26-2014

Time Run: 14:22:03

Run By: 공동기기원

Instrument: JEOL JMS600
Inlet: Direct Probe

Ionization mode: EI+

R.T.: 3:17.1

Scan: 152
Base: m/z 419; 49.5%FS

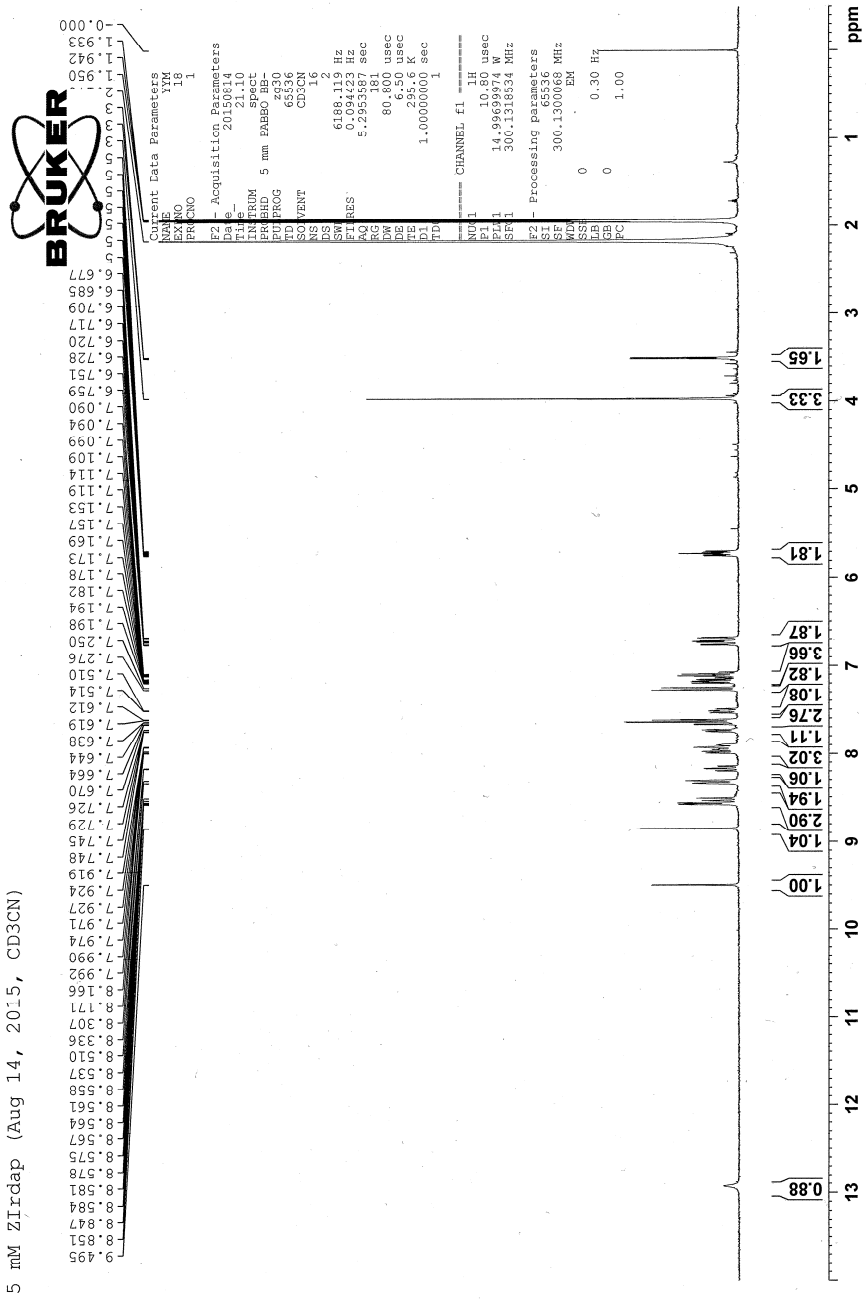
TIC: 6443826

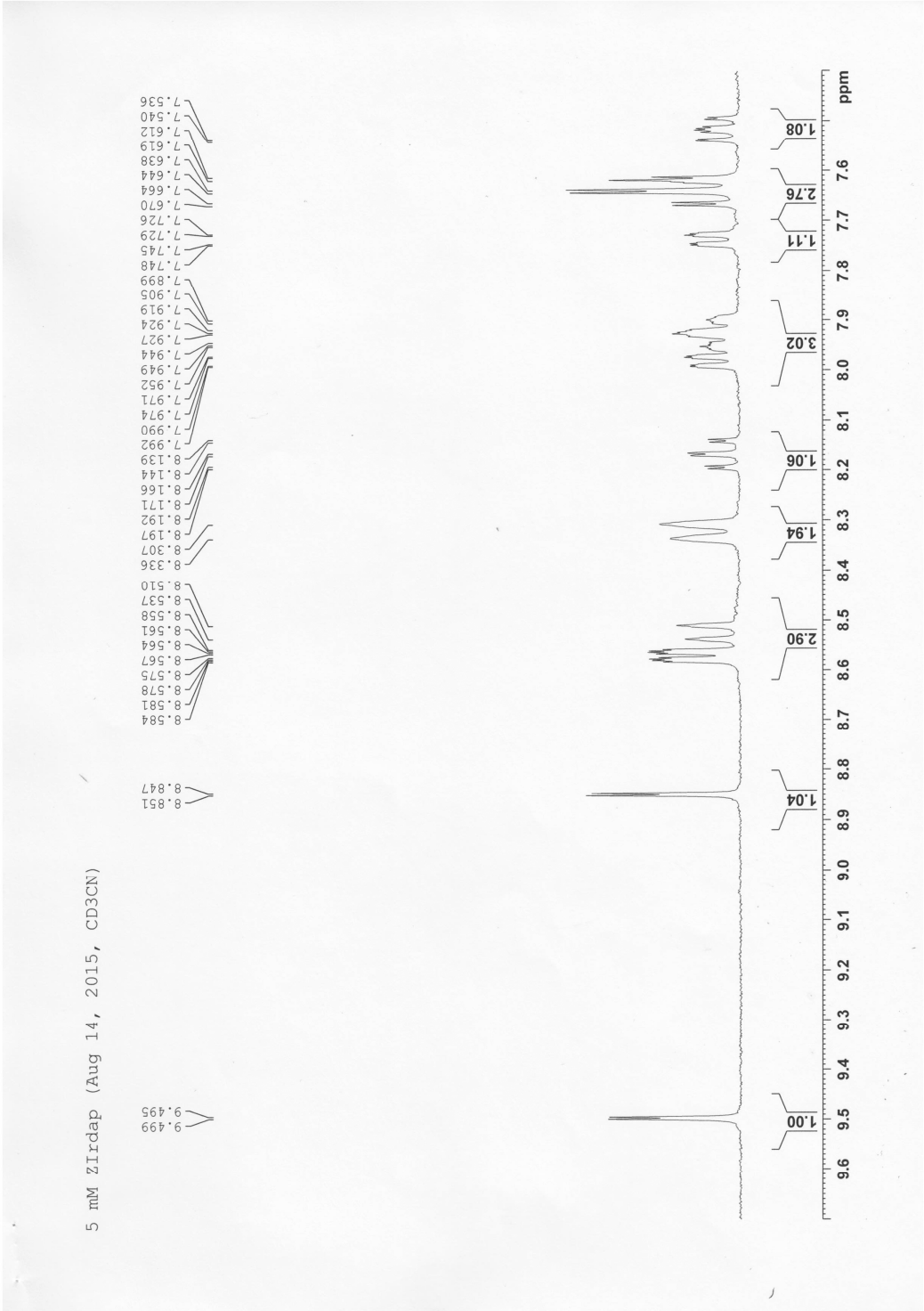
Selected Isotopes : $C_{0-50}H_{0-100}O_{0-10}N_{5-10}$

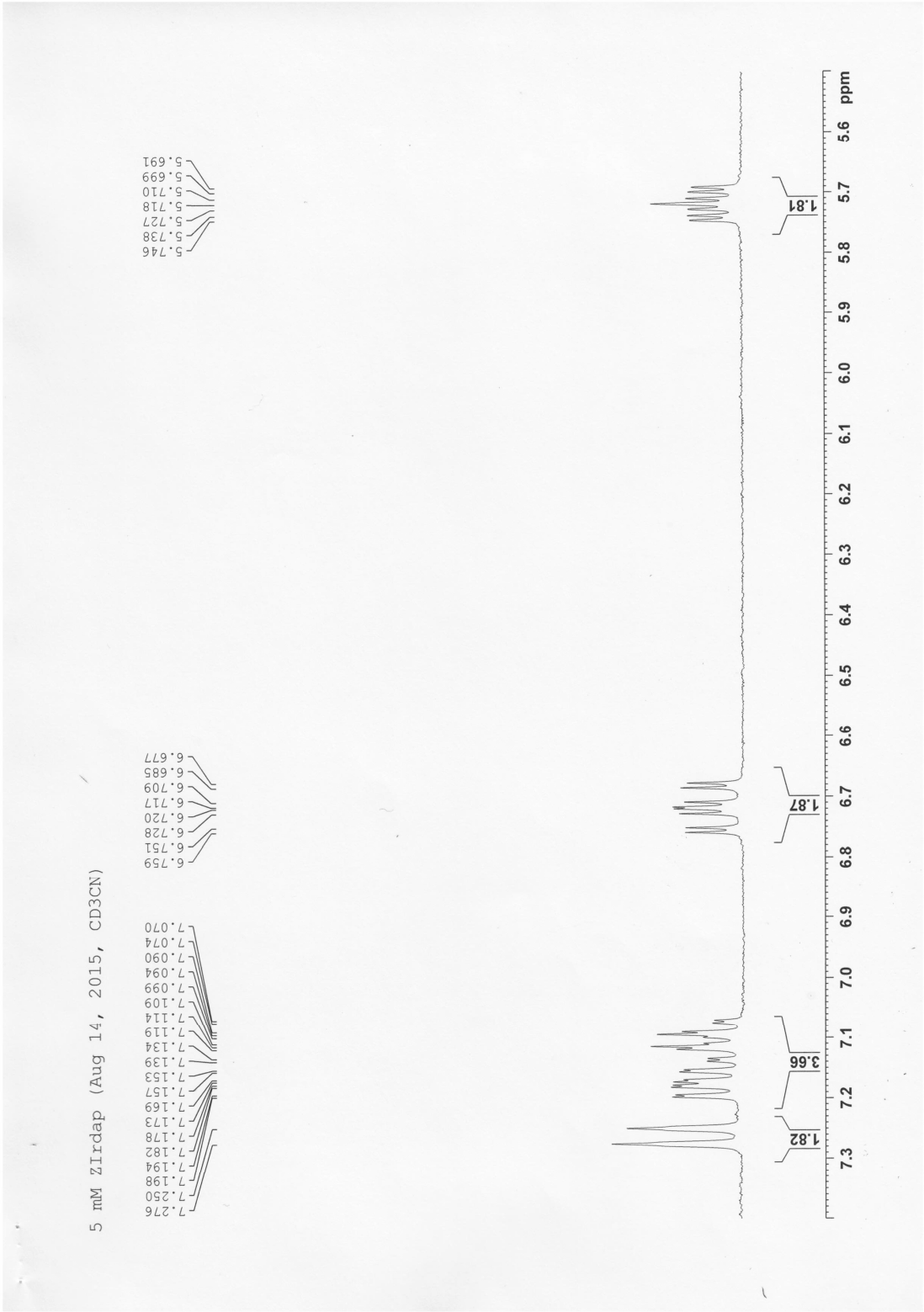
Error Limit : 20 ppm

Measured Mass	*% Base*	*Formula*	*Calculated Mass*	*Error*
411.1818	2.0%	$C_{17}H_{25}O_7N_5$	411.1754	-16.0
		$C_{20}H_{23}O_4N_6$	411.1780	-9.2
		$C_{13}H_{27}O_9N_6$	411.1839	5.1
		$C_{23}H_{21}O\,N_7$	411.1807	-2.6
		$C_{16}H_{25}O_6N_7$	411.1866	12.0
		$C_{19}H_{23}O_3N_8$	411.1893	18.0
		$C_{15}H_{23}O_6N_8$	411.1740	-19.0
		$C_{18}H_{21}O_3N_9$	411.1767	-13.0
		$C_{11}H_{25}O_8N_9$	411.1825	1.8
		$C_{21}H_{19}N_{10}$	411.1794	-5.9
		$C_{14}H_{23}O_5N_{10}$	411.1852	8.3

Figure S21. HR MS (EI) spectrum of 2-((di(2-picolyl)amino)acetylamino)-5-(2-pyridyl)pyrazine.


5 mM ZIrdap (Aug 14, 2015, CD3CN)
BRUKER
Current Data Parameters
NAME YYM
EXPNO 18
PROCNO 1
F2 - Acquisition Parameters
Date_ 20150814
Time 21.10
INSTRUM spect
PROBHD 5 mm PABBO BB-
PULPROG zg30
TD 65536
SOLVENT CD3CN
NS 16
DS 2
SWH 6188.119 Hz
FIDRES 0.094423 Hz
AQ 5.29533887 sec
RG 101
DW 80.800 usec
DE 6.50 usec
TE 295.6 K
D1 1.00000000 sec
TD0 1
======== CHANNEL f1 ========
NUC1 1H
P1 10.60 usec
PLW1 14.99699974 W
SFO1 300.1318534 MHz
F2 - Processing parameters
SI 65536
SF 300.1300069 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
ppm
1.65
3.33
1.81
1.87
3.66
1.82
1.08
2.76
1.11
3.02
1.06
1.94
2.90
1.04
1.00
0.88


5 mM ZIrdap (Aug 14, 2015, CD3CN)
9.499
9.495
8.851
8.847
8.584
8.581
8.578
8.575
8.567
8.564
8.561
8.558
8.537
8.510
8.336
8.307
8.197
8.192
8.171
8.166
8.144
8.139
7.992
7.990
7.974
7.971
7.952
7.949
7.944
7.927
7.924
7.919
7.905
7.899
7.748
7.745
7.729
7.726
7.670
7.664
7.644
7.638
7.619
7.612
7.540
7.536
1.00
1.04
2.90
1.94
1.06
3.02
1.11
2.76
1.08
ppm
9.6
9.5
9.4
9.3
9.2
9.1
9.0
8.9
8.8
8.7
8.6
8.5
8.4
8.3
8.2
8.1
8.0
7.9
7.8
7.7
7.6


5 mM ZIrdap (Aug 14, 2015, CD3CN)
7.276
7.250
7.198
7.194
7.182
7.178
7.173
7.169
7.157
7.153
7.139
7.134
7.119
7.114
7.109
7.099
7.094
7.090
7.074
7.070
6.759
6.751
6.728
6.720
6.717
6.709
6.685
6.677
5.746
5.738
5.727
5.718
5.710
5.699
5.691
1.82
3.66
1.87
1.87
1.81
7.3
7.2
7.1
7.0
6.9
6.8
6.7
6.6
6.5
6.4
6.3
6.2
6.1
6.0
5.9
5.8
5.7
5.6
ppm

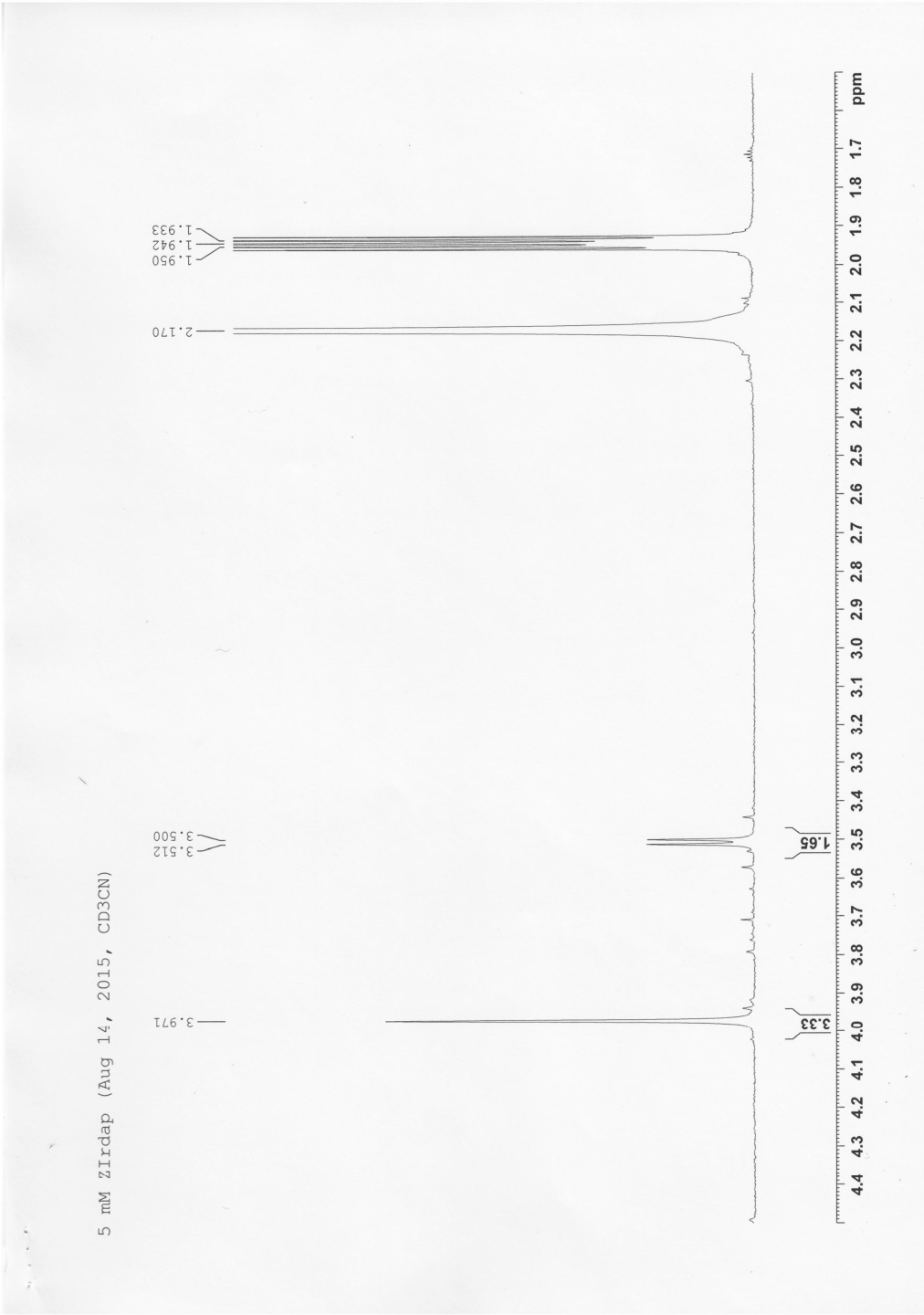


Figure S22. ^{1}H NMR (300 MHz, CD_3CN) spectrum of ZIrdap.

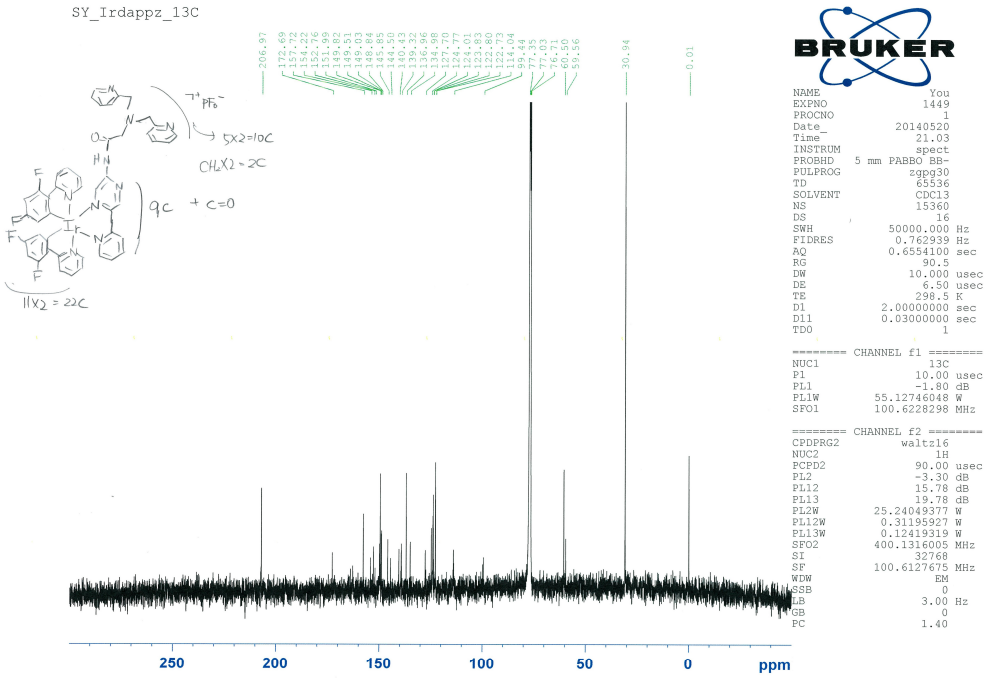


Figure S23. ^{13}C NMR (100 MHz, $CDCl_3$) spectrum of ZIrdap.

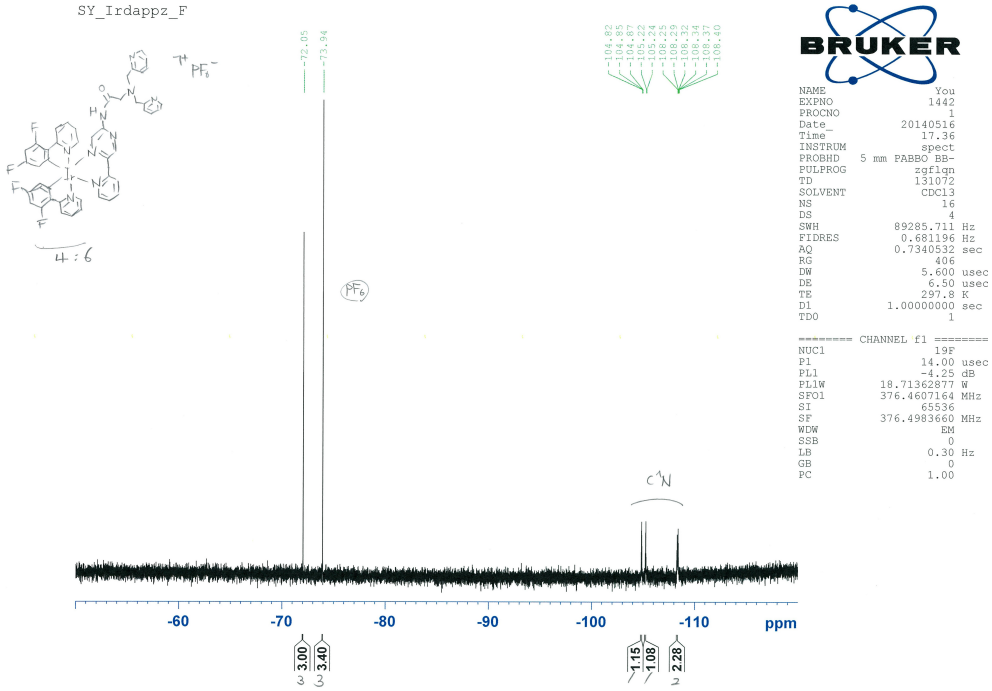
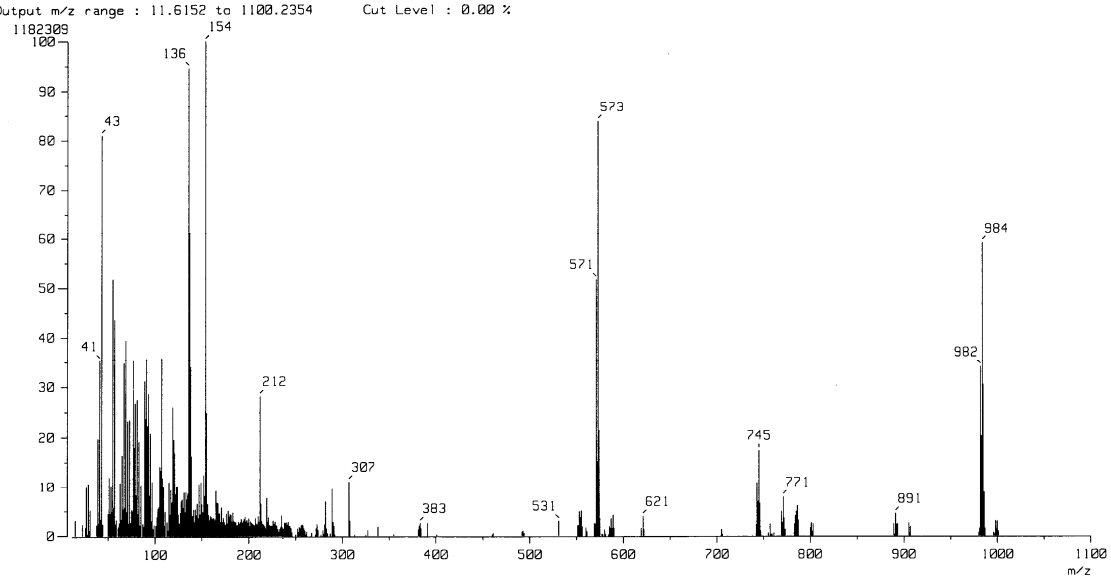


Figure S24. ^{19}F NMR (376 MHz, $CDCl_3$) spectrum of ZIrdap.

[Mass Spectrum]
Data : FAB-H858 Date : 21-May-2014 15:56
Sample: Irdappz
Note : m-NBA
Inlet : Direct Ion Mode : FAB+
Spectrum Type : Normal Ion [MF-Linear]
RT : 0.84 min Scan# : (3,8)
BP : m/z 154.0000 Int. : 112.75
Output m/z range : 11.6152 to 1100.2354 Cut Level : 0.00 %



```
[ Elemental Composition ]                                                    Page: 1
Data  : FAB-H915                        Date : 26-May-2014 17:29
Sample: Irdappz
Note : m-NBA
Inlet : Direct                          Ion Mode : FAB+
RT : 0.62 min                           Scan#: (22,33)
Elements : C 150/0, H 150/0, F 5/3, N 10/0, O 3/0, Ir 2/0(191Ir 2/0, 193Ir 2/0)
Mass Tolerance       : 1000ppm, 3mmu if m/z < 3, 5mmu if m/z > 5
Unsaturation (U.S.)  : 0.0 - 100.0

Observed m/z Int%   Err[ppm / mmu]     U.S.  Composition
  984.2370   100.0     -0.5 /  -0.5    56.5  C 69 H 29 F 3 N 5
                       -0.3 /  -0.3    53.0  C 64 H 28 F 4 N 8
                       +4.5 /  +4.4    52.5  C 69 H 31 F 5 N O
                       -1.9 /  -1.8    56.0  C 71 H 31 F 3 N 2 O
                       -1.7 /  -1.6    52.5  C 66 H 30 F 4 N 5 O
                       -1.5 /  -1.4    49.0  C 61 H 29 F 5 N 8 O
                       +4.9 /  +4.9    53.0  C 62 H 27 F 3 N 10 O
                       -3.0 /  -3.0    52.0  C 68 H 32 F 4 N 2 O 2
                       -2.8 /  -2.8    48.5  C 63 H 31 F 5 N 5 O 2
                       +3.6 /  +3.5    52.5  C 64 H 29 F 3 N 7 O 2
                       +3.8 /  +3.7    49.0  C 59 H 28 F 4 N 10 O 2
                       -4.2 /  -4.1    48.0  C 65 H 33 F 5 N 2 O 3
                       +2.2 /  +2.2    52.0  C 66 H 31 F 3 N 4 O 3
                       +2.4 /  +2.4    48.5  C 61 H 30 F 4 N 7 O 3
                       +2.6 /  +2.6    45.0  C 56 H 29 F 5 N 10 O 3
                       -0.4 /  -0.4    35.5  C 54 H 36 F 5 N 191Ir
                       +0.0 /  +0.0    36.0  C 47 H 32 F 3 N 10 191Ir
                       +4.6 /  +4.6    39.0  C 57 H 36 F 3 O 191Ir
                       +4.8 /  +4.8    35.5  C 52 H 35 F 4 N 3 O 191Ir
                       +5.0 /  +5.0    32.0  C 47 H 34 F 5 N 6 O 191Ir
                       -1.3 /  -1.3    35.5  C 49 H 34 F 3 N 7 O 191Ir
                       -1.1 /  -1.1    32.0  C 44 H 33 F 4 N 10 O 191Ir
                       +3.5 /  +3.4    35.0  C 54 H 37 F 4 O 2 191Ir
                       +3.7 /  +3.6    31.5  C 49 H 36 F 5 N 3 O 2 191Ir
                       -2.7 /  -2.6    35.0  C 51 H 36 F 3 N 4 O 2 191Ir
                       -2.5 /  -2.4    31.5  C 46 H 35 F 4 N 7 O 2 191Ir
                       -2.3 /  -2.2    28.0  C 41 H 34 F 5 N 10 O 2 191Ir
                       +2.3 /  +2.3    31.0  C 51 H 38 F 5 O 3 191Ir
                       -4.0 /  -4.0    34.5  C 53 H 38 F 3 N O 3 191Ir
                       -3.8 /  -3.8    31.0  C 48 H 37 F 4 N 4 O 3 191Ir
                       -3.6 /  -3.6    27.5  C 43 H 36 F 5 N 7 O 3 191Ir
                       +2.8 /  +2.7    31.5  C 44 H 34 F 3 N 9 O 3 191Ir
                       -0.2 /  -0.2    22.0  C 42 H 41 F 3 191Ir 2
                       +0.0 /  +0.0    18.5  C 37 H 40 F 4 N 3 191Ir 2
                       +0.2 /  +0.2    15.0  C 32 H 39 F 5 N 6 191Ir 2
                       -1.4 /  -1.4    18.0  C 39 H 42 F 4 O 191Ir 2
                       -1.2 /  -1.2    14.5  C 34 H 41 F 5 N 3 O 191Ir 2
                       -2.6 /  -2.5    14.0  C 36 H 43 F 5 O 2 191Ir 2
                       +3.8 /  +3.8    18.0  C 37 H 41 F 3 N 2 O 2 191Ir 2
                       +4.0 /  +4.0    14.5  C 32 H 40 F 4 N 5 O 2 191Ir 2
                       +4.2 /  +4.2    11.0  C 27 H 39 F 5 N 8 O 2 191Ir 2
                       -2.1 /  -2.1    14.5  C 29 H 39 F 3 N 9 O 2 191Ir 2
                       +2.7 /  +2.6    14.0  C 34 H 42 F 4 N 2 O 3 191Ir 2
                       +2.9 /  +2.8    10.5  C 29 H 41 F 5 N 5 O 3 191Ir 2
                       -3.5 /  -3.4    14.0  C 31 H 41 F 3 N 6 O 3 191Ir 2
                       -3.3 /  -3.2    10.5  C 26 H 40 F 4 N 9 O 3 191Ir 2
                       +0.3 /  +0.3    36.0  C 55 H 36 F 5 193Ir
                       +0.8 /  +0.8    36.5  C 48 H 32 F 3 N 9 193Ir
                       -0.6 /  -0.6    36.0  C 50 H 34 F 3 N 6 O 193Ir
                       -0.4 /  -0.4    32.5  C 45 H 33 F 4 N 9 O 193Ir
                       +4.4 /  +4.4    32.0  C 50 H 36 F 5 N 2 O 2 193Ir
```

```
[ Theoretical Ion Distribution ]                                             Page: 1
Molecular Formula : C45 H33 F4 Ir N9 O
                    (m/z   984.2374, MW   984.0303, U.S. 32.5)
Base Peak :   984.2376, Averaged MW :   984.0244(a),   984.0259(w)

    m/z        INT.
  982.2350  54.8644 ********************************
  983.2380  29.2947 *****************
  984.2376 100.0000 ************************************************************
```

Figure S25. HR MS (FAB, *m*-NBA) spectrum of ZIrdap.

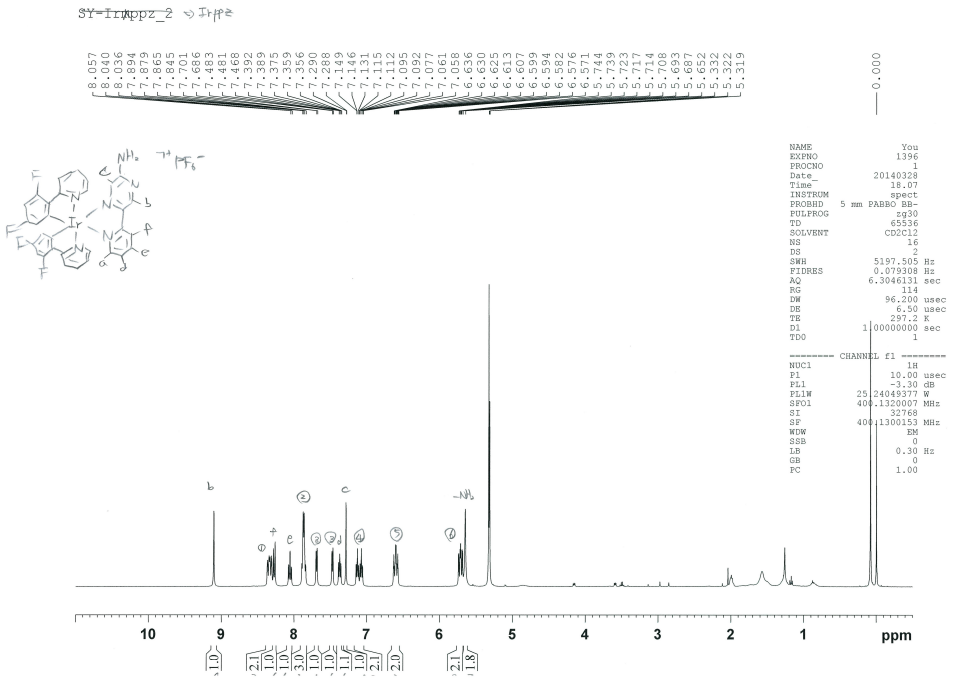


Figure S26. ^{1}H NMR (400 MHz, CD_2Cl_2) spectrum of Irp.

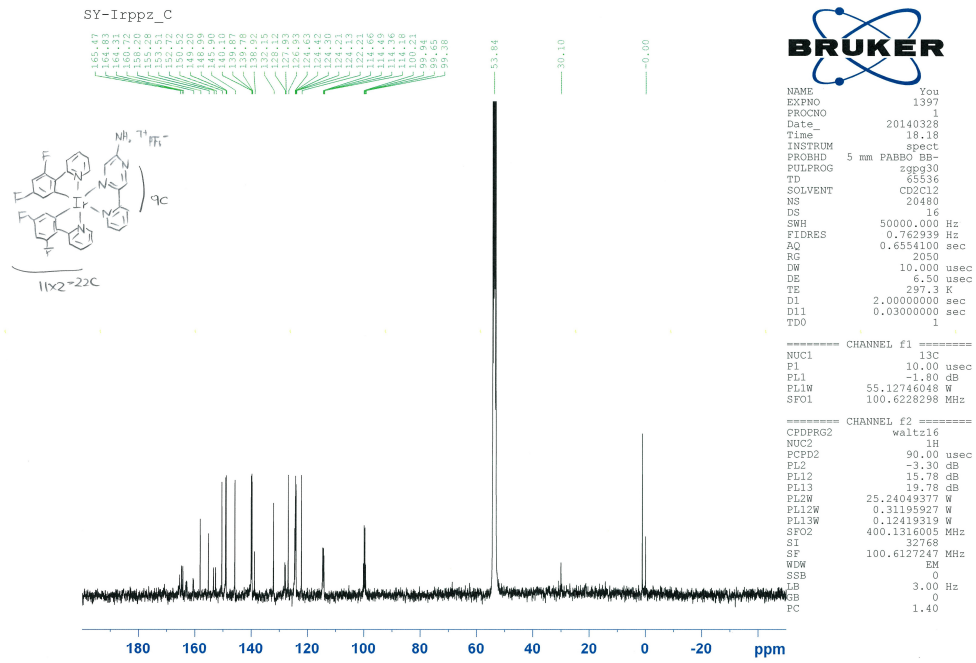


Figure S27. ^{13}C NMR (100 MHz, CD_2Cl_2) spectrum of Irp.

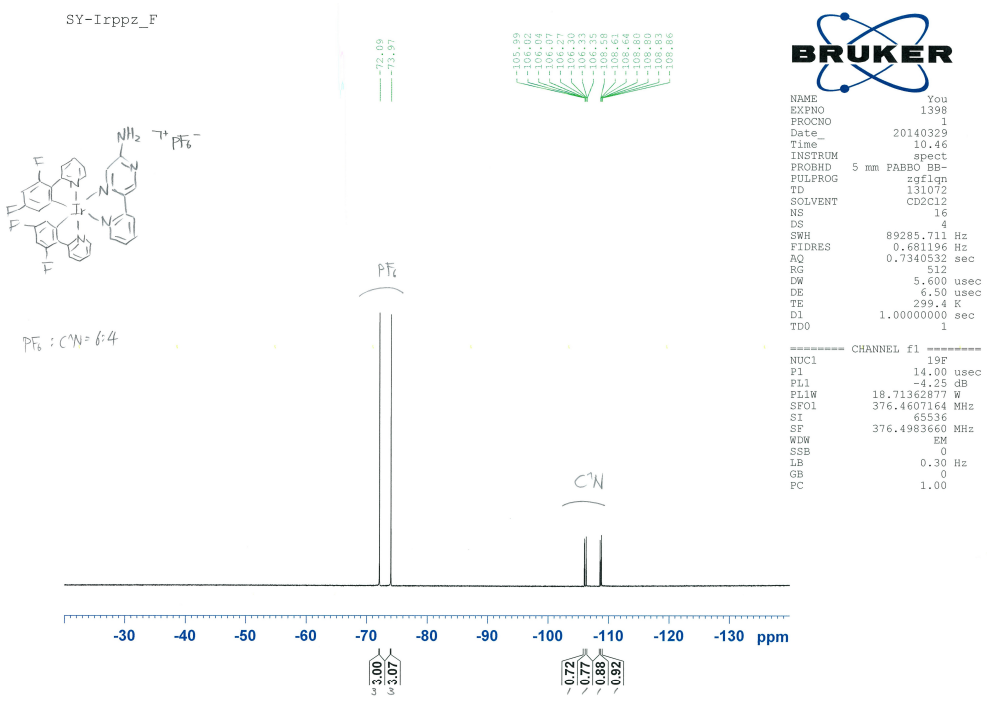
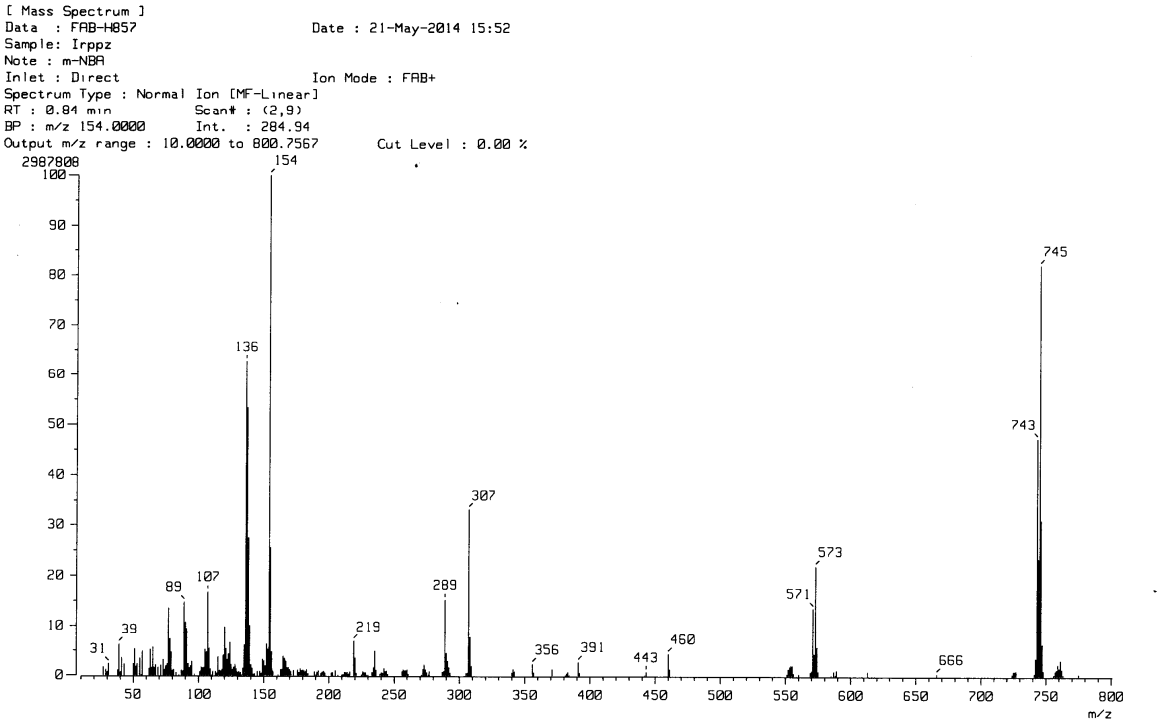


Figure S28. ^{19}F NMR (376 MHz, CD_2Cl_2) spectrum of Irp.



```
[ Elemental Composition ]                                                    Page: 1
Data  : FAB-H913                          Date : 26-May-2014 17:12
Sample: Irppz
Note : m-NBA
Inlet : Direct                            Ion Mode : FAB+
RT : 0.08 min                             Scan#: (3,6)
Elements : C 150/0, H 150/0, F 5/3, N 10/0, Ir 2/0(191Ir 2/0, 193Ir 2/0)
Mass Tolerance      : 1000ppm, 3mmu if m/z < 3, 5mmu if m/z > 5
Unsaturation (U.S.) : 0.0 - 100.0

Observed m/z Int%   Err[ppm / mmu]    U.S.  Composition
  745.1312   100.0     -2.2 /  -1.6   43.5  C 52 H 17 F 4 N 2
                       -1.9 /  -1.4   40.0  C 47 H 16 F 5 N 5
                       +6.6 /  +4.9   44.0  C 48 H 14 F 3 N 7
                       +6.4 /  +4.8   26.5  C 38 H 22 F 4 191Ir
                       +6.7 /  +5.0   23.0  C 33 H 21 F 5 N 3 191Ir
                       -1.7 /  -1.3   26.5  C 35 H 21 F 3 N 4 191Ir
                       -1.4 /  -1.1   23.0  C 30 H 20 F 4 N 7 191Ir
                       -1.2 /  -0.9   19.5  C 25 H 19 F 5 N 10 191Ir
                       -1.6 /  -1.2    5.5  C 20 H 28 F 5 191Ir 2
                       -1.0 /  -0.7    6.0  C 13 H 24 F 3 N 9 191Ir 2
                       -0.7 /  -0.5   27.0  C 36 H 21 F 3 N 3 193Ir
                       -0.4 /  -0.3   23.5  C 31 H 20 F 4 N 6 193Ir
                       -0.2 /  -0.1   20.0  C 26 H 19 F 5 N 9 193Ir
                       +0.0 /  +0.0    6.5  C 14 H 24 F 3 N 8 191Ir 193Ir
                       +1.0 /  +0.8    7.0  C 15 H 24 F 3 N 7 193Ir 2
                       +1.3 /  +1.0    3.5  C 10 H 23 F 4 N 10 193Ir 2


[ Theoretical Ion Distribution ]                                             Page: 1
Molecular Formula : C31 H20 F4 Ir N6
                      (m/z   745.1315, MW   744.7536, U.S. 23.5)
Base Peak :   745.1316, Averaged MW :   744.7487(a),   744.7504(w)

    m/z       INT.
  743.1292  57.2642 *********************************
  744.1321  21.0064 ************
  745.1316 100.0000 ************************************************************
  746.1345  35.7419 *********************
  747.1375   6.3242 ****
  748.1404   0.7265
  749.1433   0.0607
  750.1462   0.0039
  751.1491   0.0002
```

Figure S29. HR MS (FAB, *m*-NBA) spectrum of Irp.

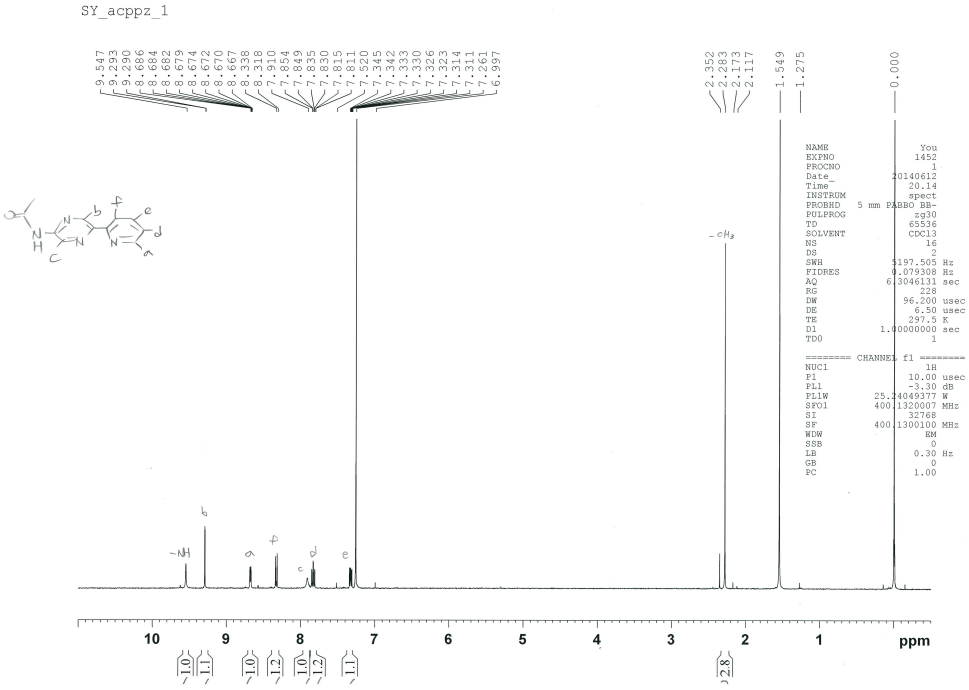


Figure S30. ^{1}H NMR (400 MHz, $CDCl_3$) spectrum of 2-(acetylamino)-5-(2-pyridyl)pyrazine.

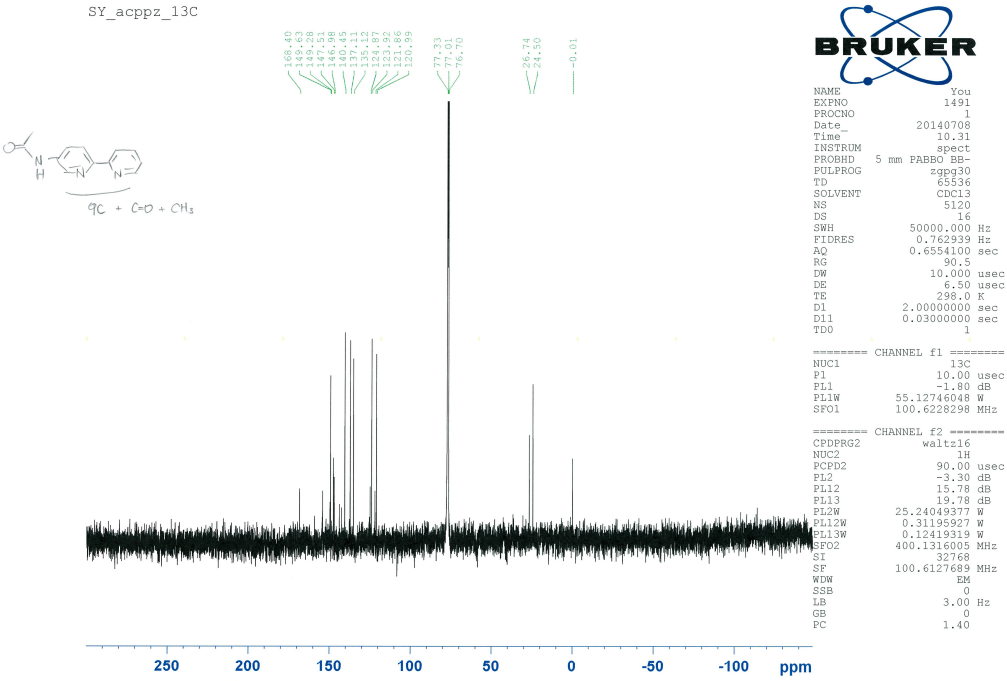


Figure S31. ^{13}C NMR (100 MHz, $CDCl_3$) spectrum of 2-(acetylamino)-5-(2-pyridyl)pyrazine.

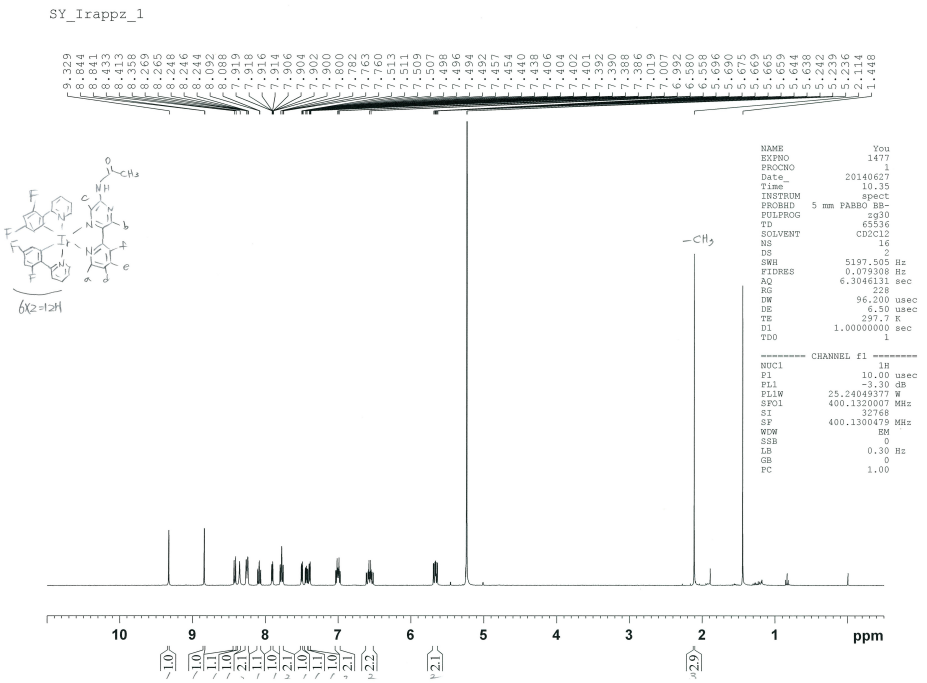


Figure S32. ^{1}H NMR (400 MHz, CD_2Cl_2) spectrum of Irap.

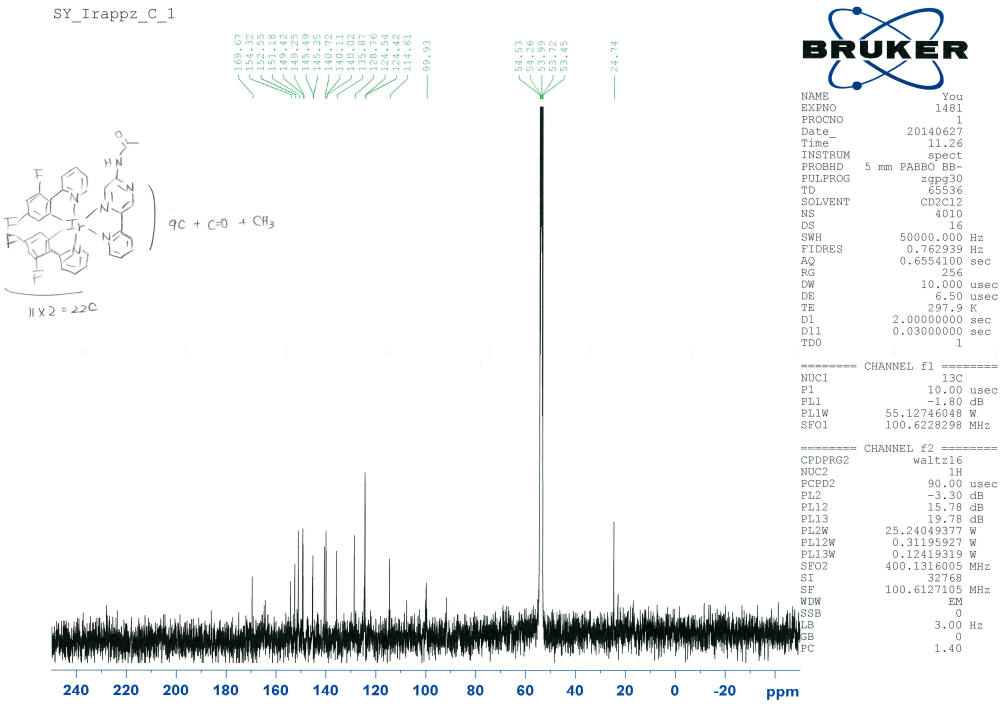


Figure S33. ^{13}C NMR (100 MHz, CD_2Cl_2) spectrum of Irap.

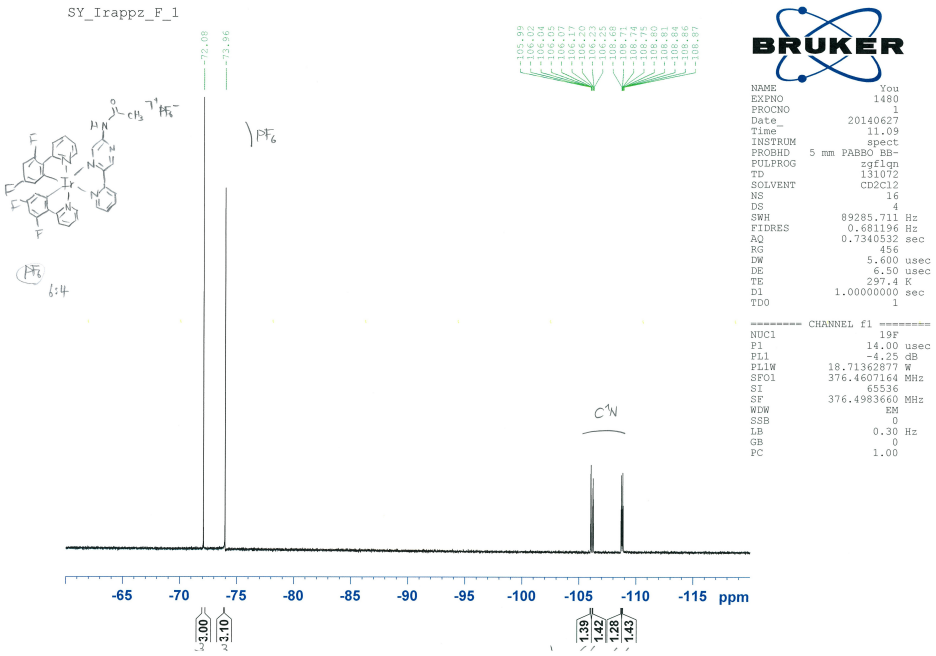


Figure S34. ^{19}F NMR (376 MHz, CD_2Cl_2) spectrum of Irap.

Table S1. The Cartesian Coordinates of the Optimized Geometry of the Model Structure of ZIrdap

Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	77	0	-0.395657	-0.216866	-0.115455
2	6	0	-2.429036	-0.886173	1.916303
3	6	0	-0.289930	-1.548233	2.630376
4	6	0	-2.968858	-1.368367	3.119007
5	6	0	-0.775553	-2.037012	3.828967
6	1	0	0.764545	-1.592522	2.396878
7	6	0	-2.144609	-1.941789	4.073580
8	1	0	-4.029230	-1.285312	3.294481
9	1	0	-0.095891	-2.475564	4.547612
10	1	0	-2.566226	-2.309014	5.001639
11	6	0	1.924048	-2.155030	-0.395098
12	6	0	2.691995	-0.162579	0.500972
13	6	0	3.237687	-2.631809	-0.347819
14	6	0	4.004586	-0.678091	0.525766
15	1	0	2.460188	0.840938	0.837860
16	1	0	3.479747	-3.633759	-0.684103
17	6	0	0.195311	0.145812	-3.087078
18	6	0	0.402034	2.087379	-1.777868
19	6	0	0.530191	0.821067	-4.246444
20	1	0	-0.040305	-0.908960	-3.103100
21	6	0	0.748542	2.814333	-2.926771
22	6	0	0.812295	2.183037	-4.158858
23	1	0	0.563742	0.290213	-5.188628
24	1	0	0.959296	3.868299	-2.843746
25	1	0	1.076082	2.749863	-5.043729
26	6	0	-0.121434	1.668477	0.575906
27	6	0	0.278277	2.607950	-0.417445
28	6	0	-0.257643	2.099632	1.897259
29	6	0	0.517646	3.932103	-0.024168
30	6	0	-0.004608	3.425674	2.218174
31	1	0	-0.569441	1.432914	2.690003
32	6	0	0.385228	4.369569	1.279662
33	1	0	0.572767	5.400271	1.547178
34	6	0	-2.376864	0.177116	-0.286178
35	6	0	-3.014303	0.789375	-1.366859
36	6	0	-3.160792	-0.253554	0.822019
37	6	0	-4.390658	0.964266	-1.344756
38	1	0	-2.469437	1.147137	-2.230036
39	6	0	-4.546735	-0.045045	0.778985
40	6	0	-5.187979	0.557645	-0.285395
41	1	0	-6.259105	0.704780	-0.284831
42	6	0	-1.513863	-2.903400	-1.359638
43	6	0	0.771737	-2.922268	-0.899292
44	6	0	-1.484449	-4.214519	-1.813261
45	1	0	-2.431302	-2.328922	-1.345950
46	6	0	0.866080	-4.244718	-1.343141
47	6	0	-0.270526	-4.896076	-1.803677
48	1	0	-2.394978	-4.683734	-2.162715
49	1	0	1.814685	-4.763256	-1.327304
50	1	0	-0.208085	-5.921092	-2.148497
51	7	0	-1.081114	-0.986971	1.696412
52	7	0	0.131119	0.749249	-1.884915
53	7	0	-0.417626	-2.266844	-0.914637
54	7	0	1.676928	-0.896584	0.056326
55	7	0	4.257801	-1.920935	0.113205
56	9	0	-5.326247	-0.444584	1.811653
57	9	0	-4.985103	1.556143	-2.397783
58	9	0	0.902962	4.853936	-0.938398
59	9	0	-0.141419	3.820445	3.498620
60	7	0	5.030435	0.108647	1.014533
61	1	0	4.754460	0.922939	1.544535
62	6	0	6.413998	0.007680	0.695791
63	8	0	6.828548	-0.752577	-0.136334
64	6	0	7.277840	0.939174	1.516023
65	1	0	7.436784	0.504415	2.507605
66	1	0	8.242361	1.045645	1.024003
67	1	0	6.824097	1.924873	1.648002

Table S2. The Cartesian Coordinates of the Optimized Geometry of the Model Structure of the Zinc-Bound Form of ZIrdap

Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	77	0	-1.714437	-0.136617	-0.151107
2	6	0	-3.397084	-1.959547	1.492764
3	6	0	-1.159054	-2.002019	2.207757
4	6	0	-3.726075	-2.888237	2.492421
5	6	0	-1.435291	-2.915563	3.208976
6	1	0	-0.160606	-1.614578	2.061413
7	6	0	-2.746892	-3.365035	3.349137
8	1	0	-4.745646	-3.223187	2.591621
9	1	0	-0.641260	-3.257281	3.859715
10	1	0	-3.006462	-4.078179	4.122553
11	6	0	1.120011	-1.027972	-0.755410
12	6	0	1.180894	0.804916	0.660617
13	6	0	2.507210	-1.046596	-0.747618
14	6	0	2.615493	0.812706	0.650221
15	1	0	0.660340	1.557028	1.237994
16	1	0	3.106441	-1.754814	-1.314305
17	6	0	-1.315945	1.042798	-2.904873
18	6	0	-1.779429	2.583117	-1.189125
19	6	0	-1.243292	2.038850	-3.861529
20	1	0	-1.170344	0.004896	-3.168504
21	6	0	-1.712755	3.627576	-2.124758
22	6	0	-1.446441	3.356920	-3.456862
23	1	0	-1.035410	1.781850	-4.891786
24	1	0	-1.874071	4.642049	-1.797456
25	1	0	-1.400479	4.167857	-4.174035
26	6	0	-2.094603	1.497416	0.973062
27	6	0	-2.059063	2.714854	0.239462
28	6	0	-2.351251	1.539078	2.345444
29	6	0	-2.283548	3.913598	0.928498
30	6	0	-2.564462	2.761078	2.965075
31	1	0	-2.397677	0.642633	2.948831
32	6	0	-2.538058	3.969679	2.284541
33	1	0	-2.707849	4.911876	2.786677
34	6	0	-3.714106	-0.410822	-0.376994
35	6	0	-4.529640	0.205429	-1.330111
36	6	0	-4.302444	-1.348799	0.523134
37	6	0	-5.880412	-0.101541	-1.386030
38	1	0	-4.140832	0.930806	-2.031936
39	6	0	-5.675024	-1.610718	0.411144
40	6	0	-6.487261	-1.006494	-0.528139
41	1	0	-7.543355	-1.230281	-0.585316
42	6	0	-1.867001	-2.618984	-2.113405
43	6	0	0.294451	-1.969856	-1.514959
44	6	0	-1.394516	-3.661692	-2.893463
45	1	0	-2.926514	-2.421780	-2.012327
46	6	0	0.833763	-3.005563	-2.291438
47	6	0	-0.015789	-3.854491	-2.982448
48	1	0	-2.092026	-4.302795	-3.416590
49	1	0	1.905693	-3.138161	-2.349477
50	1	0	0.390976	-4.658050	-3.584769
51	7	0	-2.102569	-1.533438	1.371433
52	7	0	-1.576311	1.294565	-1.608087
53	7	0	-1.050048	-1.788135	-1.440956
54	7	0	0.466129	-0.073138	-0.013339
55	7	0	3.243907	-0.165155	-0.058976
56	9	0	-6.273766	-2.497023	1.245147
57	9	0	-6.647201	0.504755	-2.314835
58	9	0	-2.254433	5.097234	0.264700
59	9	0	-2.810349	2.784491	4.291139
60	7	0	3.198999	1.793726	1.349108
61	6	0	4.522976	2.035098	1.318957
62	8	0	5.408139	1.414148	0.669423
63	6	0	4.970816	3.226437	2.129526
64	1	0	5.962330	3.044370	2.545704
65	1	0	5.044843	4.093787	1.465292
66	1	0	4.254873	3.460628	2.915715
67	30	0	5.437633	-0.395344	-0.293336

68	8	0	5.105838	-1.740143	-1.632753
69	1	0	5.839441	-2.073026	-2.153381
70	6	0	6.650020	-2.132946	2.338718
71	6	0	8.388843	0.799444	-1.599132
72	6	0	9.561250	1.381945	-2.227993
73	1	0	9.437867	1.386912	-3.312585
74	1	0	9.697130	2.407784	-1.879603
75	1	0	10.448482	0.798466	-1.974661
76	6	0	7.199363	-2.945092	3.409479
77	1	0	7.104552	-4.002812	3.157692
78	1	0	8.254729	-2.704511	3.552532
79	1	0	6.662252	-2.747679	4.339568
80	7	0	7.457809	0.348205	-1.098253
81	7	0	6.213687	-1.487336	1.493379