Oxyonium phosphobetaines – unusually stable nucleophilic catalyst-phosphate complexes formed from *H*-phosphonates and *N*-oxides

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1. General experimental information

NMR spectra were recorded on Bruker Avance II 400 MHz machine. All reagents were of analytical grade, obtained from commercial suppliers and used without further purification. Anhydrous solvents used for reactions were stored over molecular sieves 4 Å and the content of water was controlled by Karl Fischer coulometric titration, (Metrohm 684 KF coulometer). TLC analyses were carried out on Merck silica gel 60 F 254 precoated plates using DCM–MeOH 7:3 v/v solvent system.
Powdered molecular sieves were activated by heating for 24 h at 150 ºC under vacuum (<0.1 Torr). Pivaloyl chloride was distilled and used within one month. Aliphatic *N*-oxides were rendered anhydrous using the method by Soderquist and Anderson.[1]
Immediately prior to reactions, all solid reactants were dissolved in pyridine and evaporated to dryness to evacuate any residual moisture (2x). The procedure was repeated with toluene (1x) to remove remains of pyridine. Insufficient removal of water leads to formation of increased amounts of phenyl *H*-phosphonate **2a** (the main or sole by-product) in the reaction mixtures.

The rates and yields of formation of phosphobetaines were higher in polar solvents (DMF, acetonitrile) than in DCM; however, precipitation was more effective in DCM and this solvent was used typically for preparative purposes. For *in situ* generation of phosphobetaines, DMF or acetonitrile are recommended.

Caution! The reactions of *N*-oxides with aryl *H*-phosphonate diesters are highly exothermic and dichloromethane (DCM) used as a solvent may boil violently upon mixing the reactants.

2. Experimental procedures

2.1. Aliphatic phosphobetaines

General procedure
N-Oxide **3** (1.5 mmol) was dissolved in anhydrous DCM (5 mL). Diphenyl *H*-phosphonate **1a** (96 µl, 0.5 mmol) was added while stirring and the stirring was continued for 20 min at room temperature. White microcrystalline precipitate was collected by filtration, washed with DCM, and dried under vacuum.

N-methylmorpholino-4-ium phenyl phosphate (*5b*)
Yield: 80 mg (60%). M.p. 103–105 ºC; R_f 0.25;
^{1}H NMR (400 MHz, D_2O): δ_H 3.94 (s, 3H), 3.98 (d, *J*= 11.9 Hz, 2H), 4.16 (quint, *J*= 12.2 Hz, 4H), 4.35 (d, *J*= 13.6 Hz, 2H), 7.31 (d, ^3J= 8.1 Hz, 2H), 7.35 (t, ^3J= 7.4 Hz, 1H), 7.52 (t, ^3J= 7.9 Hz, 2H);
^{13}C NMR (100 MHz, D_2O): δ_C 55.7, 61.0, 65.2, 120.0 (d, $^3J_{CP}$= 3.8 Hz), 125.4, 130.0, 150.8 (d, $^2J_{CP}$= 8.0 Hz);
^{31}P NMR (162 MHz, H_2O): δ_P −9.4 (s);
HRMS ESI (*m/z* calcd. for $C_{11}H_{16}NO_5P$: 273.0766), negative ion mode, found 272.0667 $[M–H]^-$; positive ion mode, found 296.0662 $[M+Na]^+$.

In order to obtain crystals suitable for X-ray analysis, the reaction was carried out in 15 ml of DCM without stirring and left for a few hours at room temperature in an open flask protected with a soft paper towel to allow slow evaporation of the solvent. The crystals were collected, washed briefly with DCM, dried under vacuum, and kept under nitrogen in a refrigerator.

Trimethylammonium phenyl phosphate (*5e*)
Yield: 58 mg (50%). M.p. 112–114 ºC; R_f 0.22;
^{1}H NMR (400 MHz, D_2O): δ_H 3.87 (s, 9H), 7.30 (d, ^3J= 8.2 Hz, 2H), 7.34 (t, ^3J= 7.5 Hz, 1H), 7.51 (t, ^3J= 7.9 Hz, 2H);
^{13}C NMR (100 MHz, D_2O): δ_C 58.1, 119.9 (d, $^3J_{CP}$= 3.8 Hz), 125.2, 130.0, 150.9 (d, $^2J_{CP}$= 7.9 Hz);
^{31}P NMR (162 MHz, H_2O): δ_P −9.4 (s);
HRMS ESI (*m/z* calcd. for $C_9H_{14}NO_4P$: 231.0660), negative ion mode, found 230.0565 $[M–H]^-$; positive ion mode, found 254.0544 $[M+Na]^+$.

In order to obtain crystals suitable for X-ray analysis, the reaction was carried out in 5 ml of DCM without stirring and left overnight at 4 ºC in an open flask protected with a soft paper towel to allow slow evaporation of the solvent. The crystals were collected, washed briefly with DCM, dried under vacuum, and kept under nitrogen in a refrigerator.

2.2. Aromatic phosphobetaines

4-methoxypyridin-1-ium-1-yl phenyl phosphate (*5a*)
4-Methoxypyridine *N*-oxide **3a** (188 mg, 1.5 mmol) was dissolved in anhydrous acetonitrile (4 mL). Diphenyl *H*-phosphonate **1a** (96 µl, 0.5 mmol) was added while stirring and the stirring was continued for 10 min at 40 ºC. Then, the reaction mixture was diluted with diethyl ether (8 mL) and left at 4 ºC overnight. Yellowish precipitate was collected by filtration, washed with ether, and dried under vacuum.
Yield: 93 mg (66%). M.p. 150–153 ºC; R_f 0.74;
^{1}H NMR (400 MHz, DMF-d_7) δ_H 4.22 (s, 3H), 7.09 (m, 1H), 7.30 (m, 4H), 7.70 (d, ^3J= 7.8 Hz, 2H), 9.02 (d, ^3J= 7.3 Hz, 2H);
^{13}C NMR (100 MHz, DMF-d_7) δ_C 58.8, 113.9, 120.6 (d, $^3J_{CP}$= 4.4 Hz), 123.8, 129.9, 144.3, 153.9 (d, $^2J_{CP}$= 7.3 Hz), 170.6;
^{31}P NMR (162 MHz, DMF) δ_P -5.5 (s);
HRMS ESI (*m/z* calcd. for $C_{13}H_{15}N_2O_4P$: 281.0453) positive ion mode, found 304.0355 $[M+Na]^+$. No expected molecular ion was found in the negative ion mode.

4-methylpyridin-1-ium-1-yl phenyl phosphate (*5c*)
4-Methylpyridine *N*-oxide **3c** (164 mg, 1.5 mmol) was dissolved in anhydrous acetonitrile (4 mL). Diphenyl *H*-phosphonate **1a** (96 µl, 0.5 mmol) was added while stirring. Compound **5c** (δ_P -5.6 ppm) was formed in ca. 40% yield (^{31}P NMR; Figs. S7a/b). Attempts to isolate it were unsuccessful.

4-(Dimethylamino)pyridin-1-ium-1-yl phenyl phosphate (*5d*)
4-(*N,N*-Dimethylamino)pyridine *N*-oxide **3d** (218 mg, 1.5 mmol) was dissolved in anhydrous DCM/diethyl ether (1:1 v/v, 20 mL) solvent system. Diphenyl *H*-phosphonate **1a** (96 µl, 0.5 mmol) was added while stirring and the stirring was continued for 30 min at room temperature (last 20 min in an open flask). White microcrystalline precipitate was collected by filtration, washed with ether and dried under vacuum.
Yield: 120 mg (81%). M.p. 185–188 ºC; R_f 0.38;
^{1}H NMR (400 MHz, D_2O): δ_H 3.22 (s, 6H), 6.86 (d, ^3J= 7.3 Hz, 2H), 7.20 (d, ^3J= 7.7 Hz, 2H), 7.27 (t, ^3J= 7.4 Hz, 1H), 7.43 (t, ^3J= 7.9 Hz, 2H), 8.19 (d, ^3J= 8.0 Hz, 2H);
^{13}C NMR (100 MHz, D_2O): δ_C 39.3, 106.5, 119.3 (d, $^3J_{CP}$= 4.1 Hz), 124.7, 129.6, 138.1, 150.5 (d, $^2J_{CP}$= 7.7 Hz), 155.3;
^{31}P NMR (162 MHz, H_2O): δ_P −4.8 (s);
HRMS ESI (*m/z* calcd. for $C_{13}H_{15}N_2O_4P$: 294.0769), positive ion mode, found 317.0723 $[M+Na]^+$. No expected molecular ion was found in the negative ion mode.

2.3. Nucleoside phosphobetaine

N-methylmorpholino-4-ium 5'-O-dimethoxytritylthymidin-3'-yl phosphate (*5f*)

5'-O-(4,4'-Dimethoxytrityl)thymidine (272 mg, 0.5 mmol) was dissolved in anhydrous dimethylformamide/pyridine (7:3, v/v) solvent system (5 mL) in the presence of activated powdered molecular sieves 4 Å. Diphenyl *H*-phosphonate **1a** (239 µl, 1.25 mmol) was added while stirring and the stirring was continued for 20 min at room temperature. *N*-Methylmorpholine *N*-oxide **3b** (352 mg, 3.0 mmol) was dissolved in the same solvent system over activated powdered molecular sieves 4 Å (5 mL), added to the reaction mixture and left stirred for 10 min at room temperature. Molecular sieves were removed by filtration and washed with toluene (ca. 25 ml). Combined solutions were concentrated to *ca* 10–15 ml in a rotary evaporator. Cold diethyl ether (50 ml) was added dropwise while stirring and the mixture was left at 0 ºC for 1 h. The solution was decanted, the precipitate was dissolved in DCM and washed twice with freshly prepared saturated aqueous solution of sodium bicarbonate. The organic layer was dried with anhydrous Na_2SO_4 and evaporated to dryness. The residue was dissolved in DCM and precipitated with cold diethyl ether. The white solid obtained was dried under vacuum.
Yield: 159 mg (44%). R_f 0.85;
^{1}H NMR (400 MHz, DMSO-d_6) δ_H 1.40 (s, 3H), 2.40 (m, 2H), 3.25 (dd, *J*= 9.0 and 9.6 Hz, 2H), 3.39 (br, 4H), 3.69 (s, 3H), 3.74 (s, 6H), 3.91 (t, *J*= 7.7 Hz, 2H), 4.09 (t, *J*= 9.9 Hz, 2H), 4.17 (br, 1H), 4.88 (br, 1H), 6.21 (t, *J*= 7.1 Hz, 1H), 6.89 (d, *J*= 8.9 Hz, 4H), 7.24 (t, 1H), 7.25 (d, *J*= 8.8 Hz, 4H), 7.31 (t, *J*= 7.5 Hz, 2H), 7.38 (d, *J*= 7.6 Hz, 2H), 7.51 (s, 1H), 11.38 (br, 1H);
^{13}C NMR (100 MHz, DMSO-d_6) δ_C 11.5, 38.1, 55.0, 56.5, 60.5, 62.8, 63.8, 74.4, 83.7, 84.4, 85.9, 109.6, 113.2, 126.8, 127.6, 127.9, 129.7, 135.1, 135.4, 144.6, 150.3, 158.1, 163.6;
^{31}P NMR (162 MHz, DMSO) δ_P −3.7 (d, ^3J= 7.0 Hz);
HRMS ESI (*m/z* calcd. for $C_9H_{14}NO_4P$: 723.2556), negative ion mode, found 722.2450 $[M-H]^-$; positive ion mode, found 746.2413 $[M+Na]^+$.

2.4. X-ray diffraction measurements

The X-ray diffraction measurements on monocrystals were performed at 100 K on beamline 14.2 at the BESSY synchrotron in Berlin using a Mar Research MX-225 detector. The resolution range of the reflections for both compounds was 20-0.81 Å and the X-ray wavelength was 0.88561 Å. The data were integrated and scaled using the HKL200 software.[2] The structures were solved by direct methods using SHELXT[3] and refined using SHELXL.[4] The R-factor for the final model of **5b** and for all data was 0.0475, and for **5e** it was 0.0256. The crystallographic data have been deposited at the Cambridge Crystallographic Data Centre and allocated with the deposition numbers: CCDC 1425902 for **5b** and 1425903 for **5e**.

2.5. References

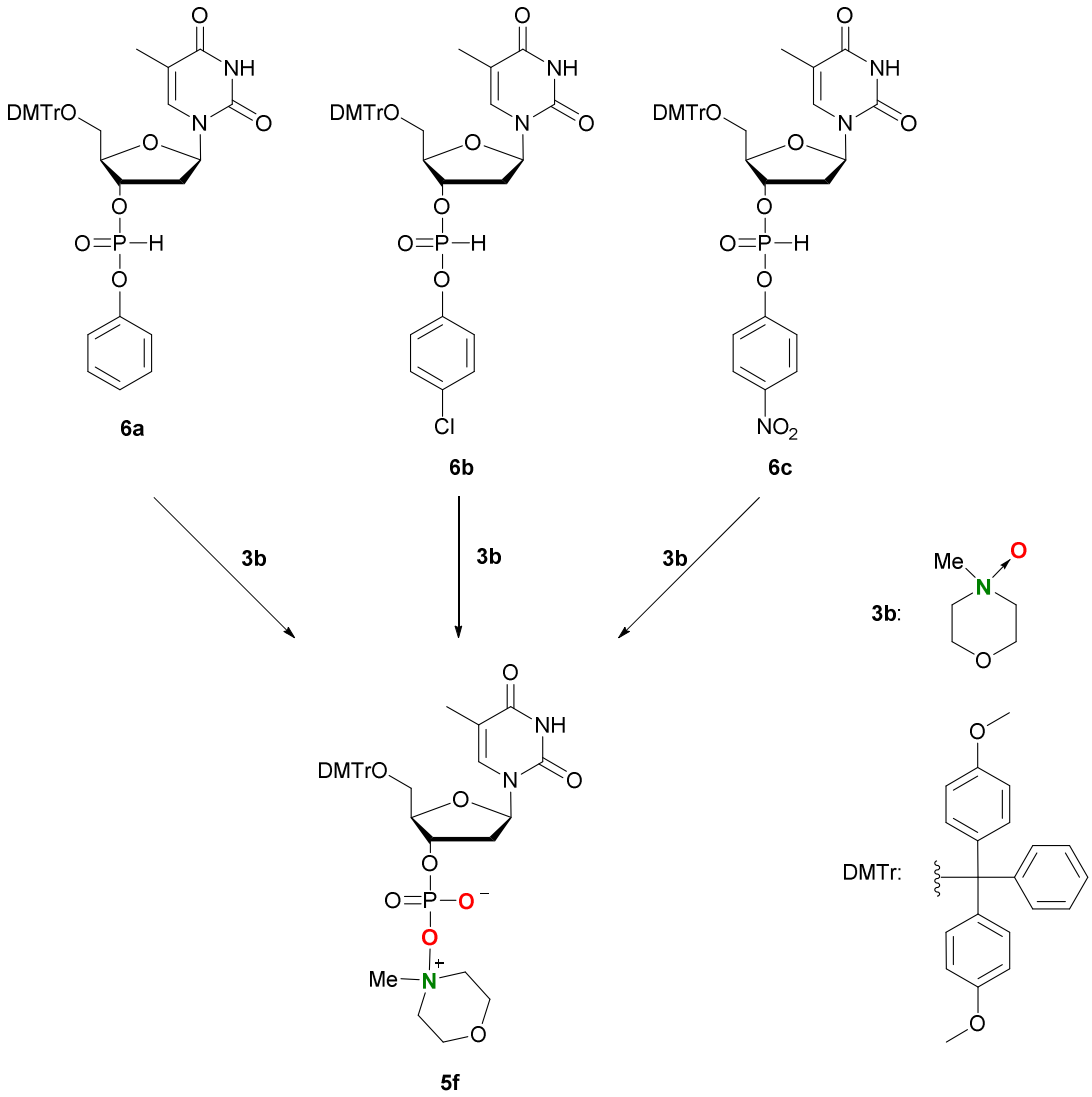
1 J. A. Soderquist, C. L. Anderson, *Tetrahedron Lett.*, 1986, **27**, 3961.

2 Z. Otwinowski, W. Minor, Processing of X-Ray Diffraction Data Collected in Oscillation Mode, in *Methods in Enzymology*, vol. **276**: *Macromolecular Crystallography Part A*; Academic Press: 1997, pp 307-326.

(3) G. M. Sheldrick, *Acta Crystallographica Section A*, 2015, **71***,* 3.

(4) G. M. Sheldrick, *Acta Crystallographica Section A*, 2008, **64***,* 112.

3. Figures and schemes



Scheme S1 Reactions of aryl nucleoside *H*-phosphonate diesters with *N*-methylmorpholine *N*-oxide (**3b**). For ^{31}P NMR spectra of crude **5f**, see Figs. S9a/b.

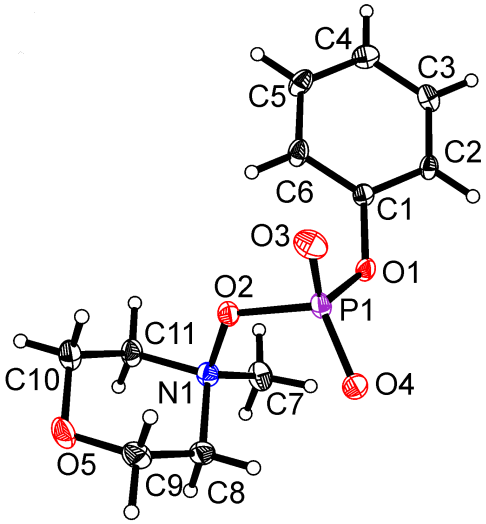


Fig. S1a Crystal structure of betaine **5b**. Displacement ellipsoids are drawn at the 50% probability level and H atoms are shown as small spheres of arbitrary radii.

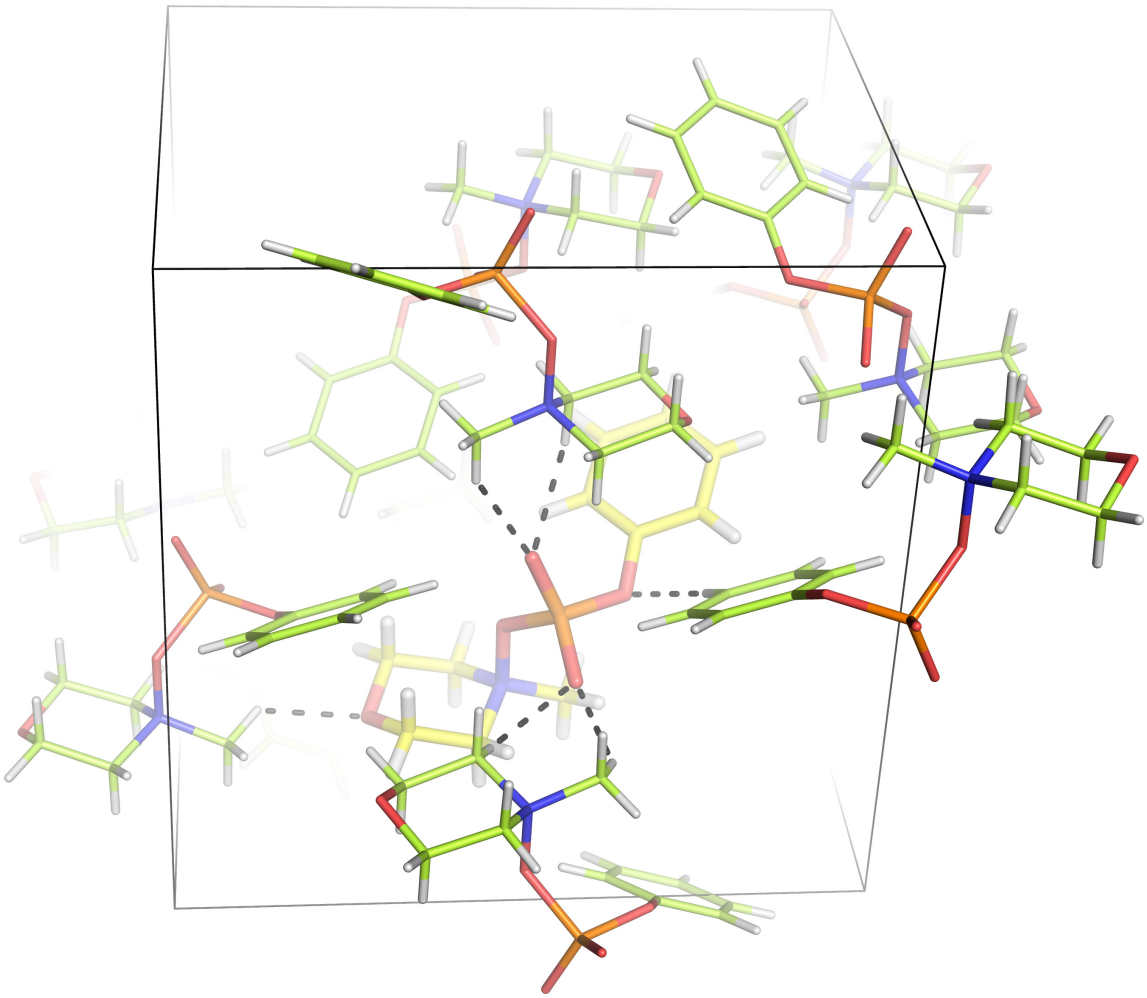


Fig. S1b The unit cell of betaine **5b**. Weak C–H···O interactions found are shown with dotted lines.

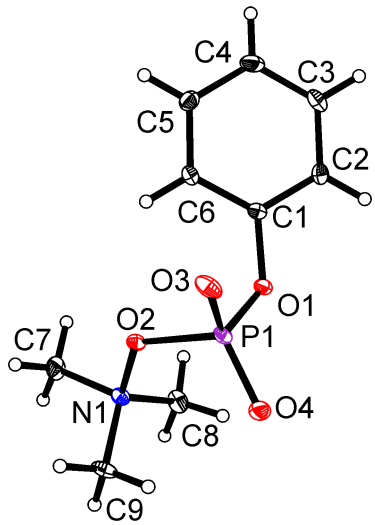


Fig. S2a Crystal structure of betaine **5e**. Displacement ellipsoids are drawn at the 50% probability level and H atoms are shown as small spheres of arbitrary radii.

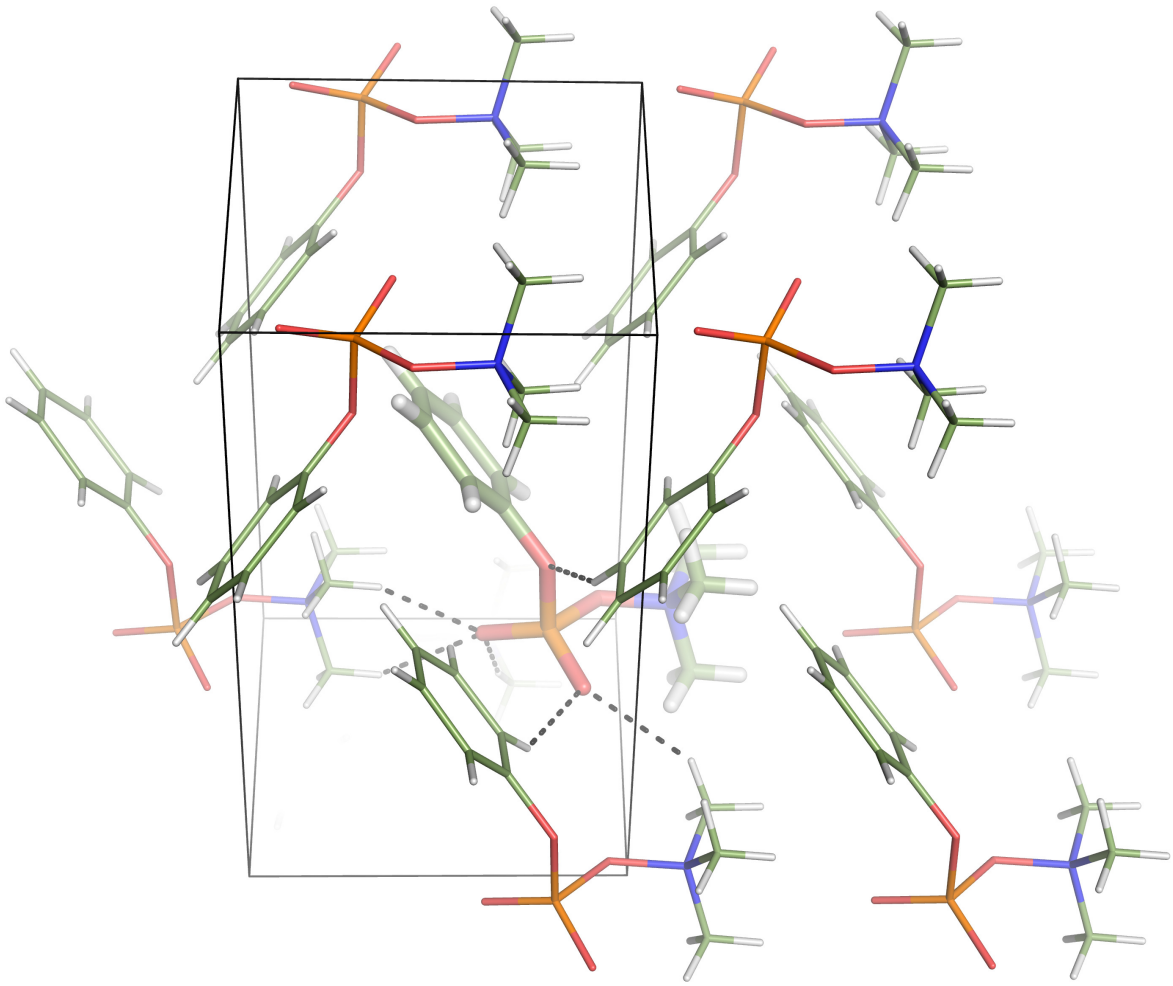


Fig. S2b The unit cell of betaine **5e**. Weak C–H···O interactions found are shown with dotted lines.

4. ^{31}P NMR spectra of the reaction mixtures

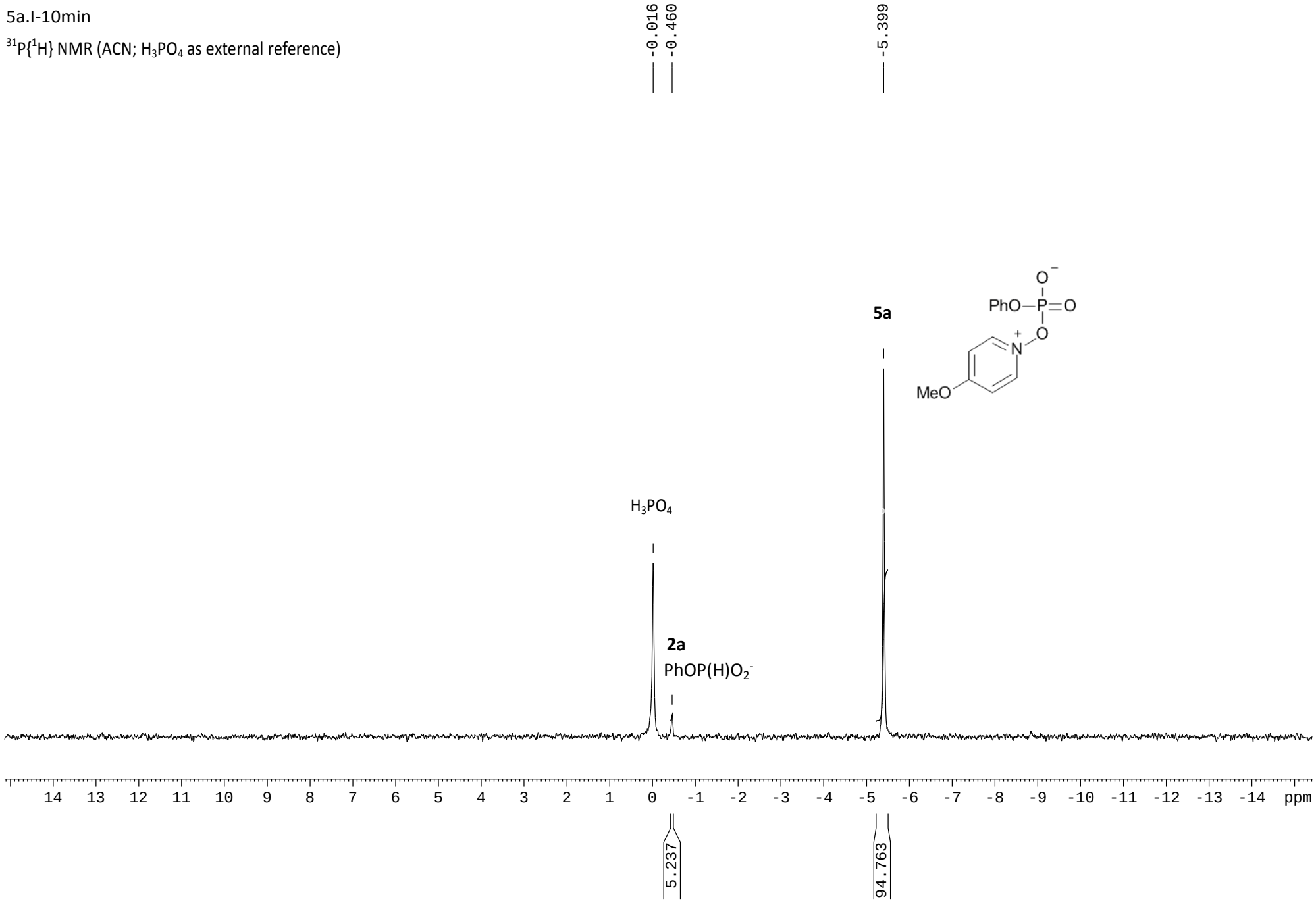


Fig. S3a Reaction of $(PhO)_2P(H)O$ **1a** with *p*MeOPy *N*-oxide **3a** in acetonitrile.
Proton-decoupled ^{31}P NMR spectrum of the reaction mixture after ca. 10 min.

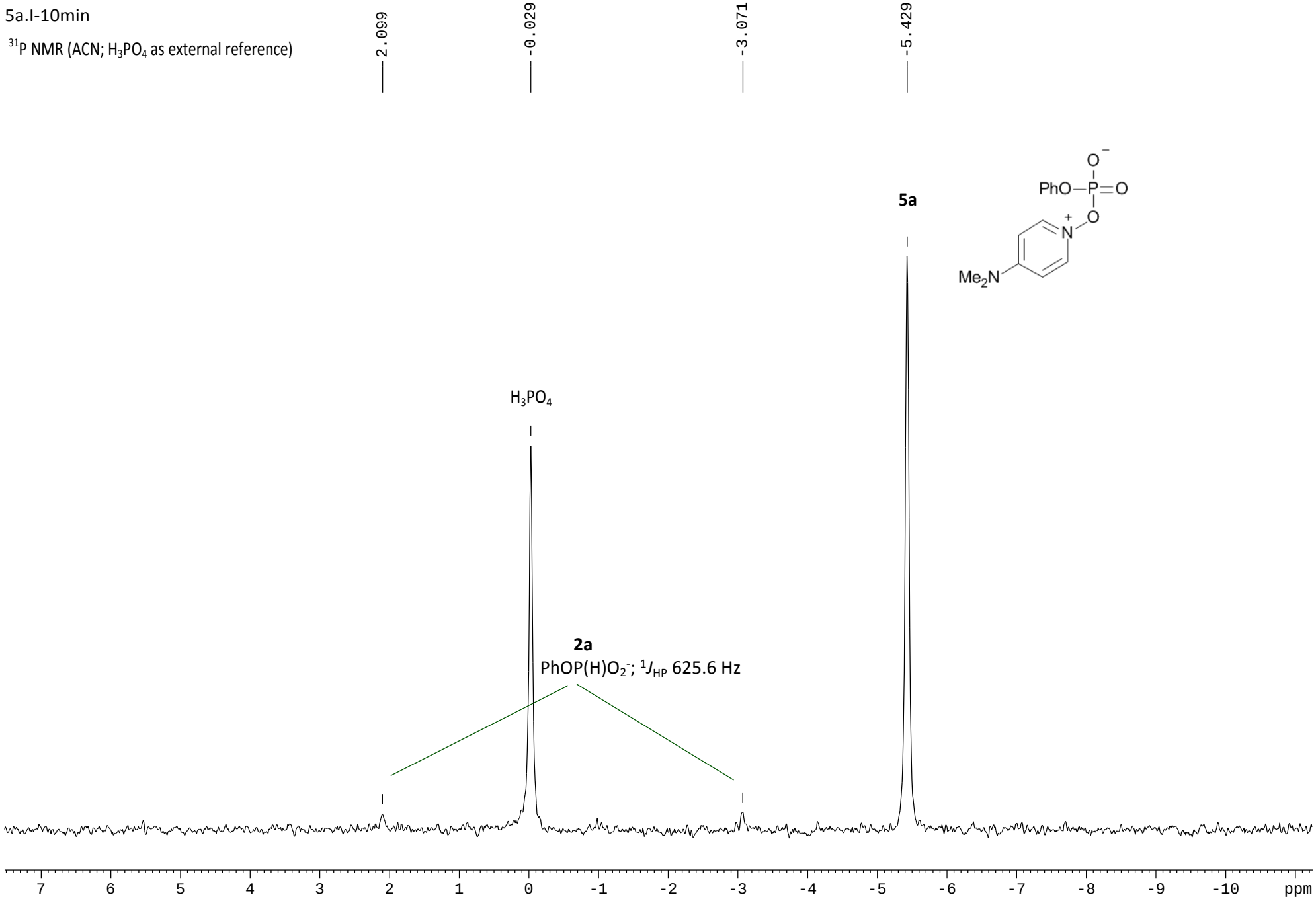


Fig. S3b Reaction of $(PhO)_2P(H)O$ **1a** with *p*MeOPy *N*-oxide **3a** in acetonitrile.
^{31}P NMR spectrum of the reaction mixture after ca. 10 min.

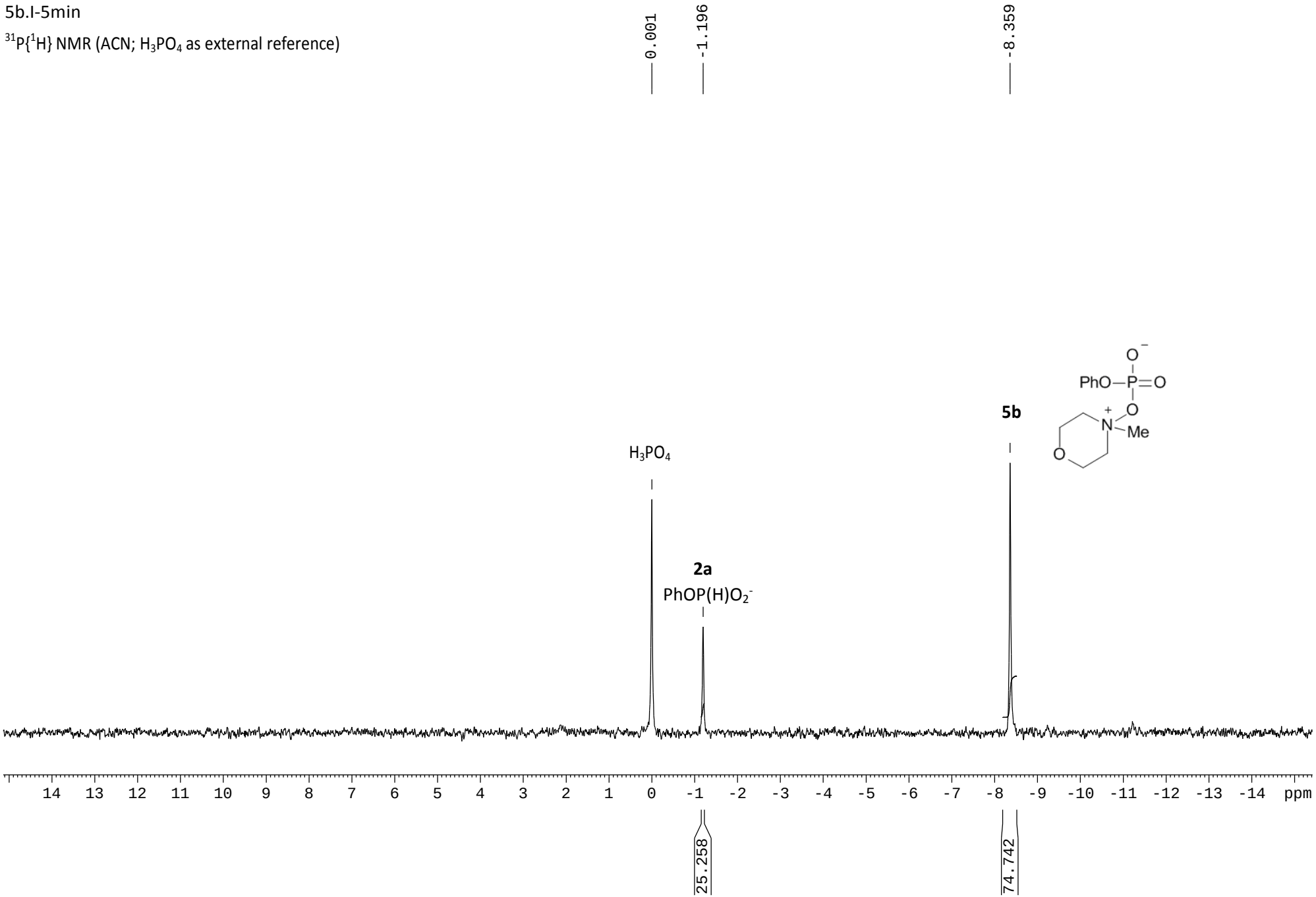


Fig. S4a Reaction of $(PhO)_2P(H)O$ **1a** with NMM *N*-oxide **3b** in acetonitrile. Proton-decoupled ^{31}P NMR spectrum of the reaction mixture after ca. 5 min.

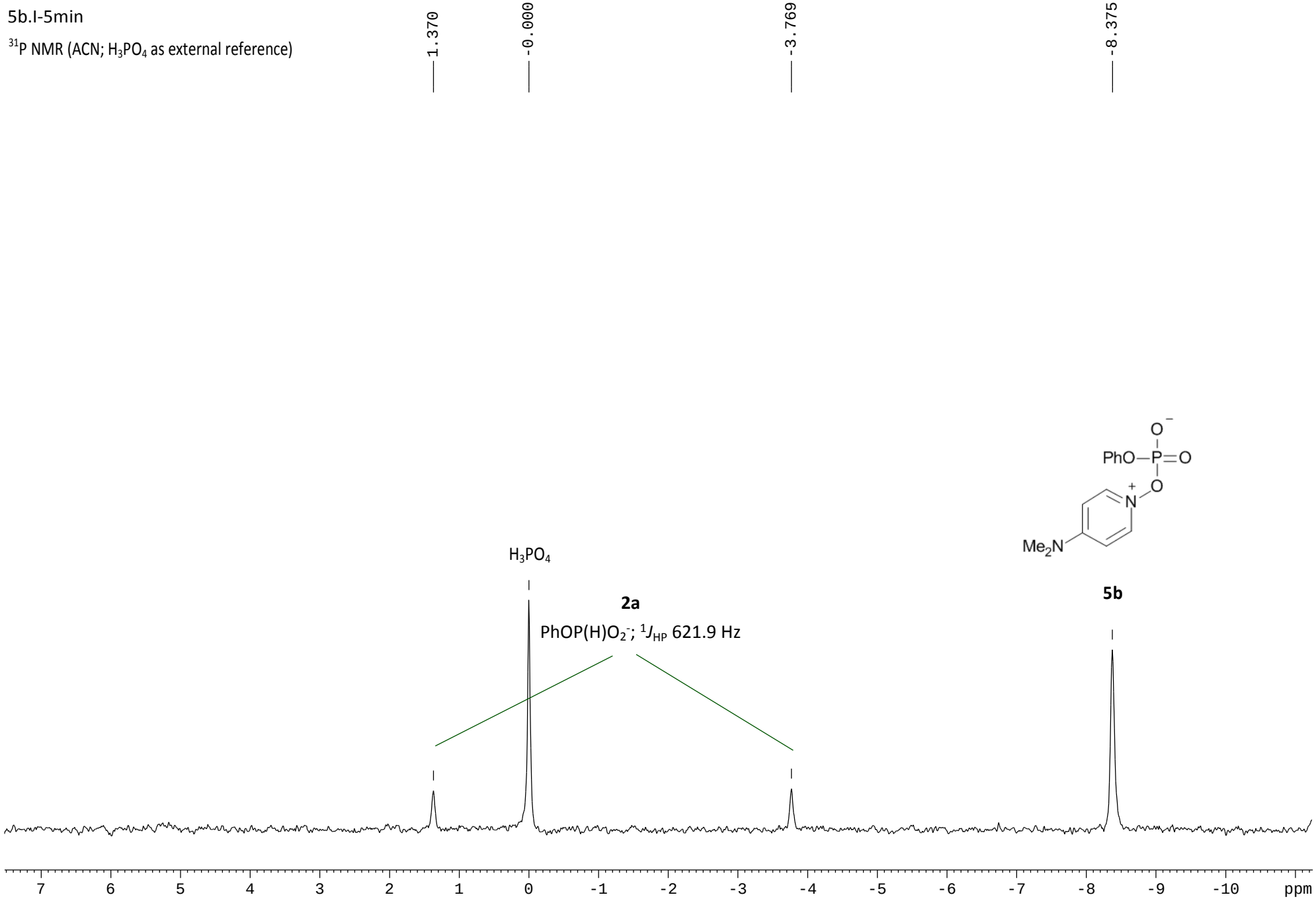


Fig. S4b Reaction of $(PhO)_2P(H)O$ **1a** with NMM *N*-oxide **3b** in acetonitrile.
^{31}P NMR spectrum of the reaction mixture after ca. 5 min.

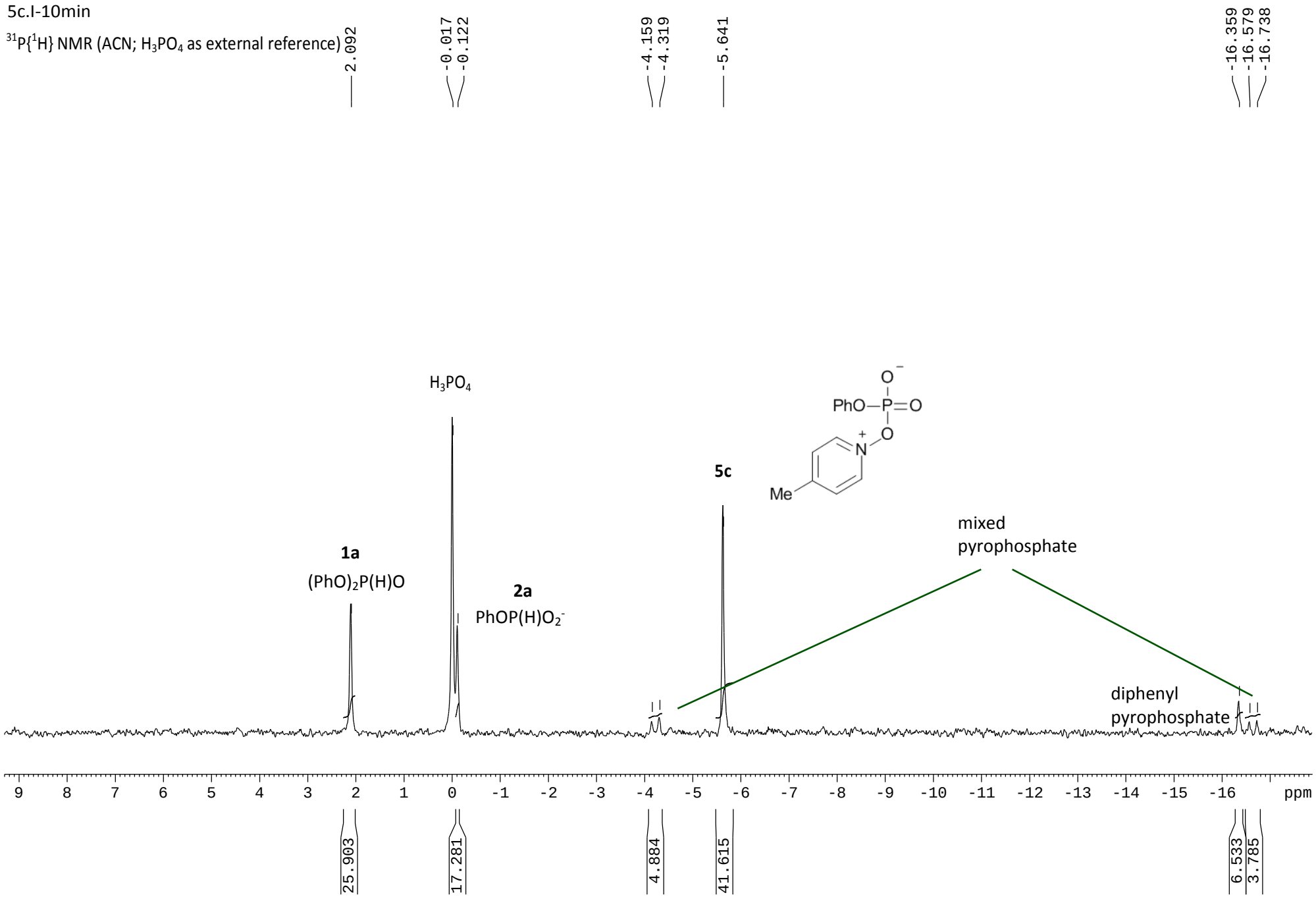


Fig. S5a Reaction of $(PhO)_2P(H)O$ **1a** with *p*MePy *N*-oxide **3c** in acetonitrile.
Proton-decoupled ^{31}P NMR spectrum of the reaction mixture after ca. 10 min.

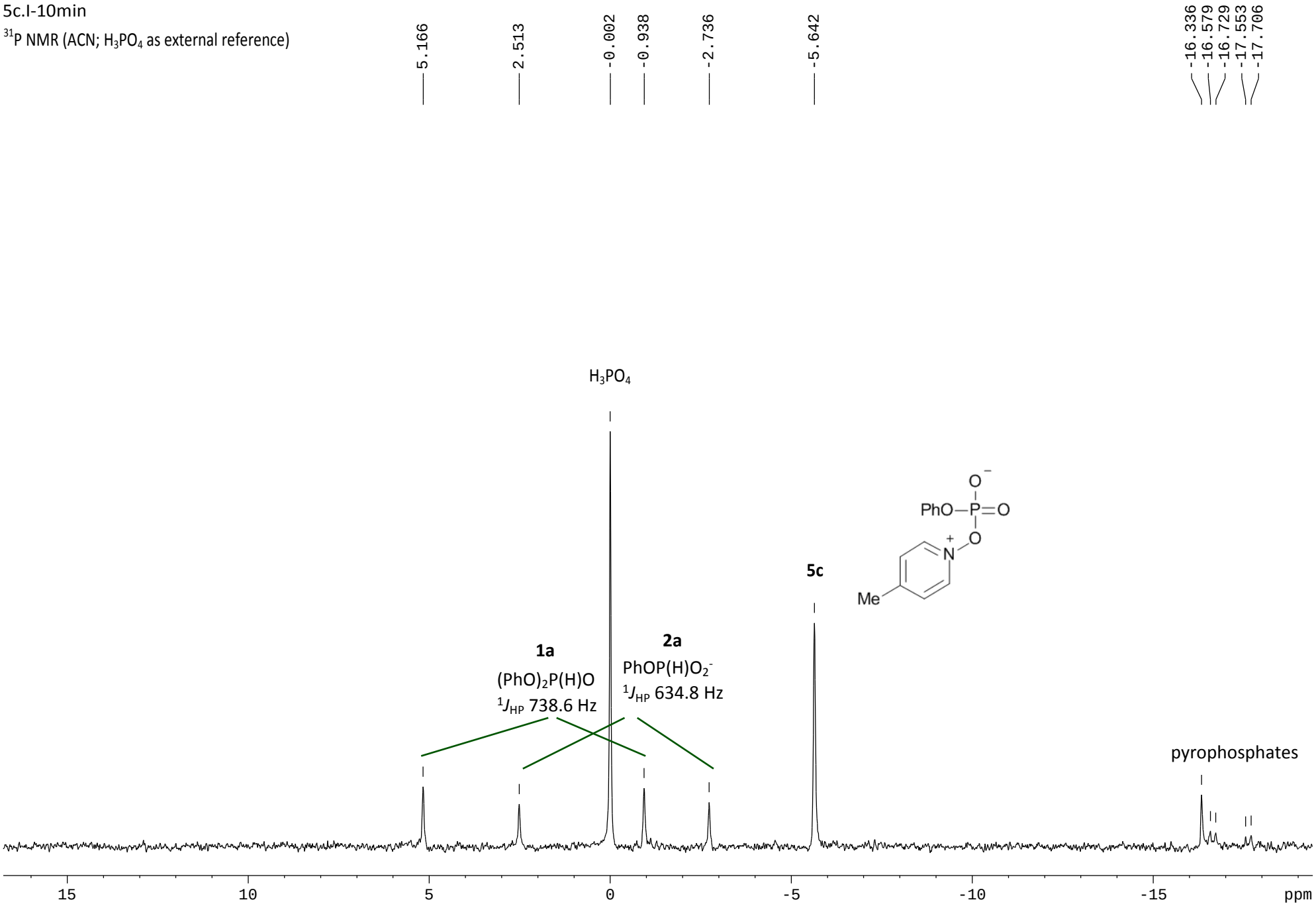


Fig. S5b Reaction of $(PhO)_2P(H)O$ **1a** with *p*MePy *N*-oxide **3c** in acetonitrile.
^{31}P NMR spectrum of the reaction mixture after ca. 10 min.

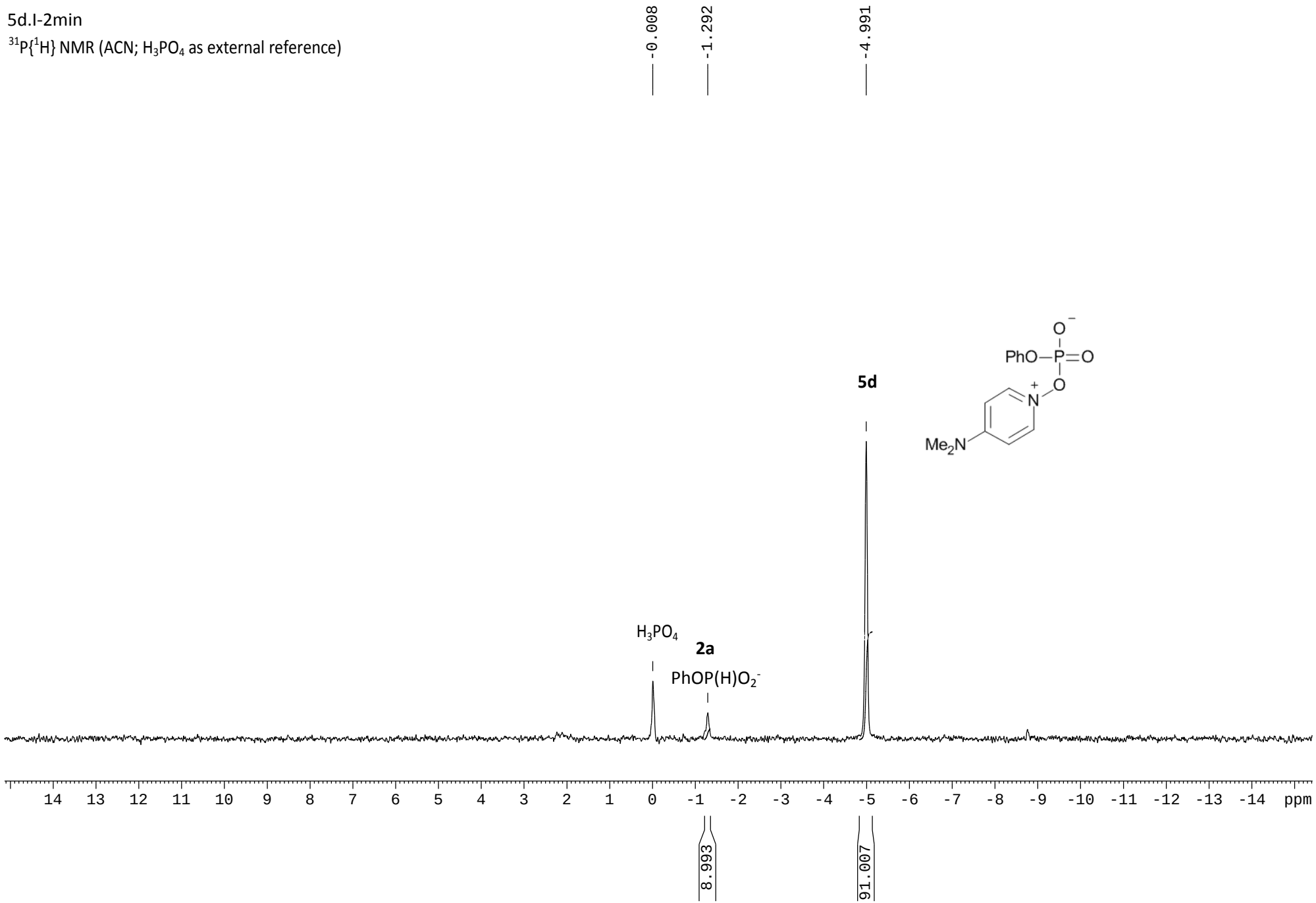


Fig. S6a Reaction of $(PhO)_2P(H)O$ **1a** with DMAP *N*-oxide **3d** in acetonitrile. Proton-decoupled ^{31}P NMR spectrum of the reaction mixture after ca. 2 min.

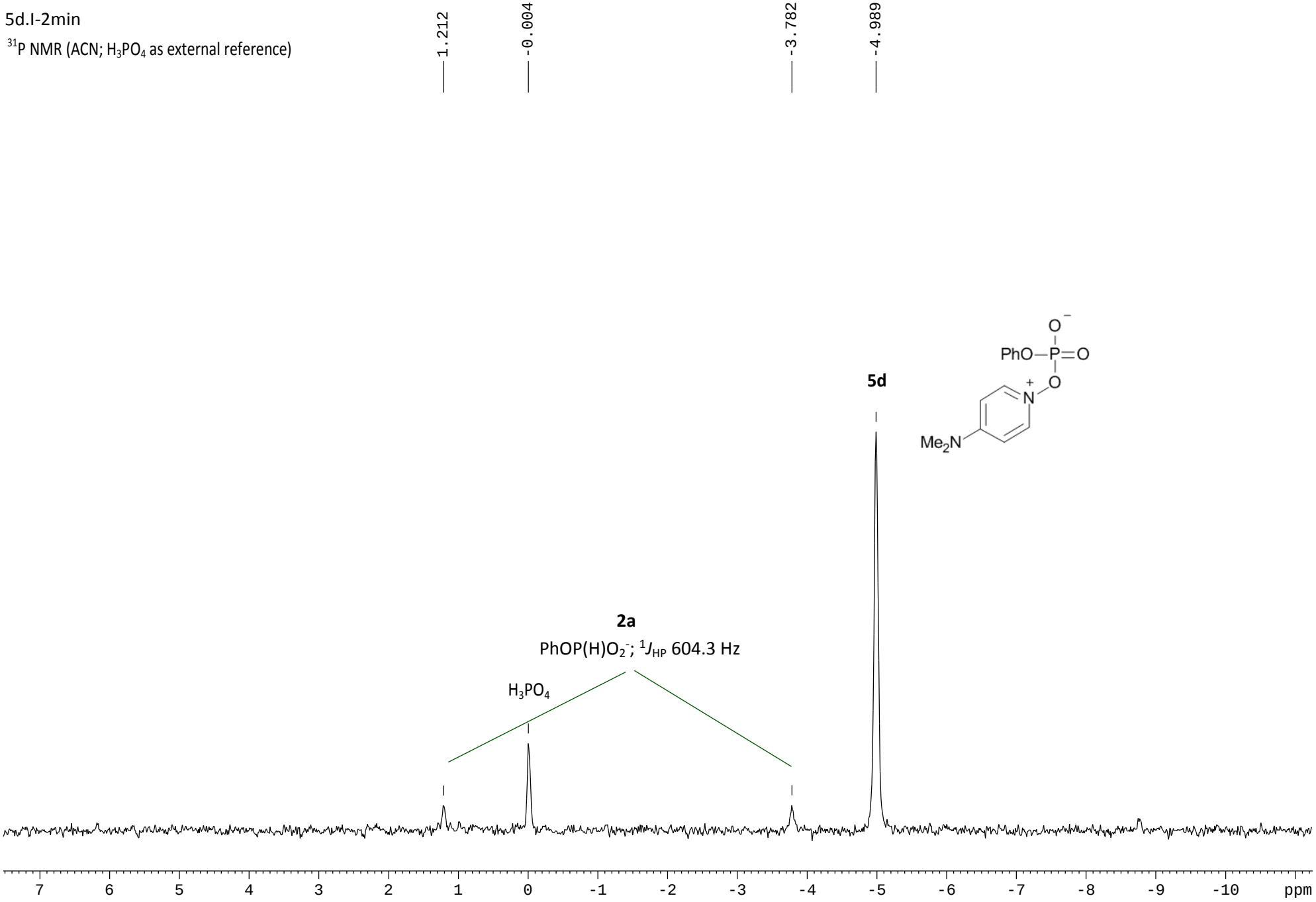


Fig. S6b Reaction of $(PhO)_2P(H)O$ **1a** with DMAP *N*-oxide **3d** in acetonitrile.
^{31}P NMR spectrum of the reaction mixture after ca. 2 min.

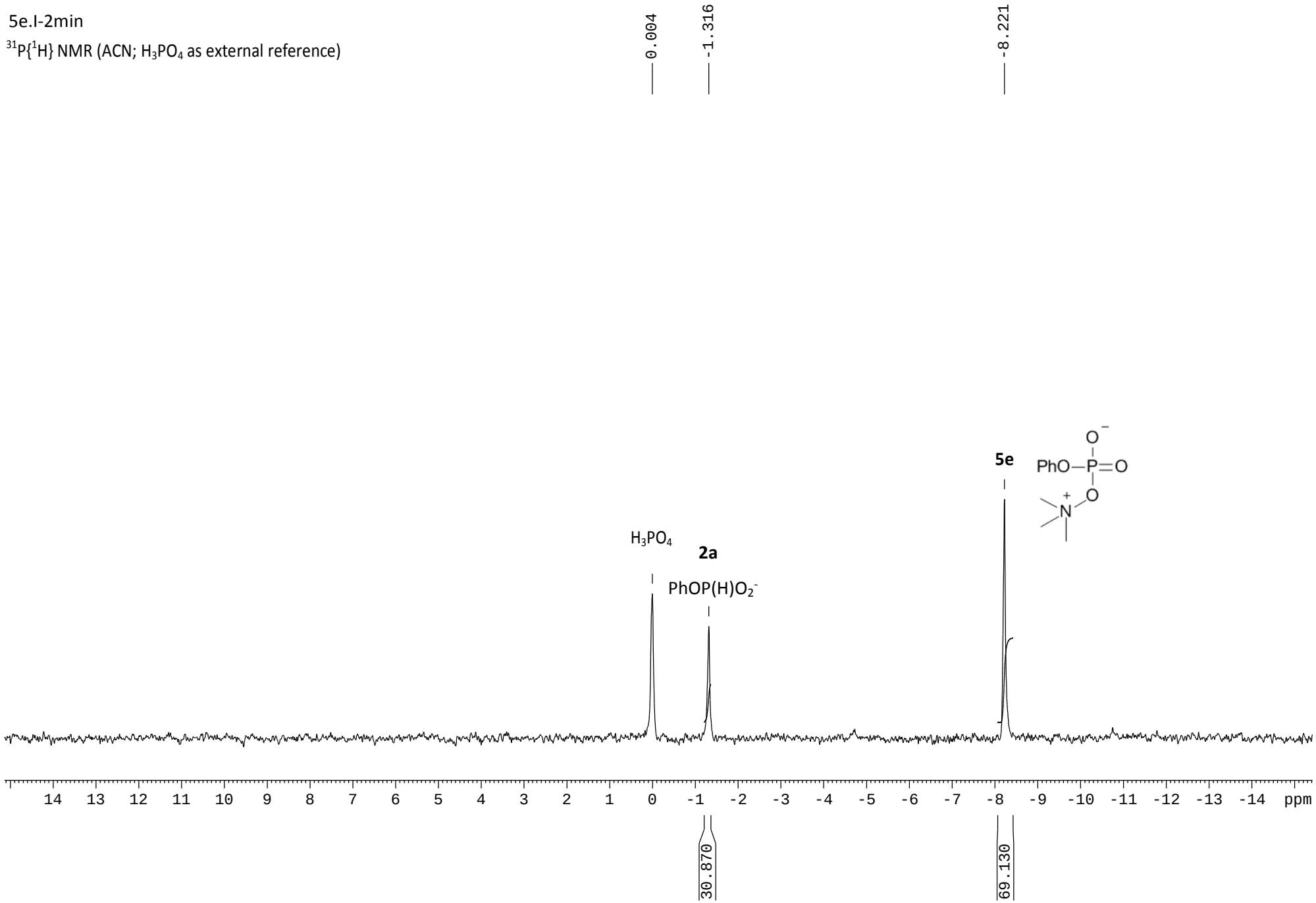


Fig. S7a Reaction of $(PhO)_2P(H)O$ **1a** with Me_3N *N*-oxide **3e** in acetonitrile.
Proton-decoupled ^{31}P NMR spectrum of the reaction mixture after ca. 2 min.

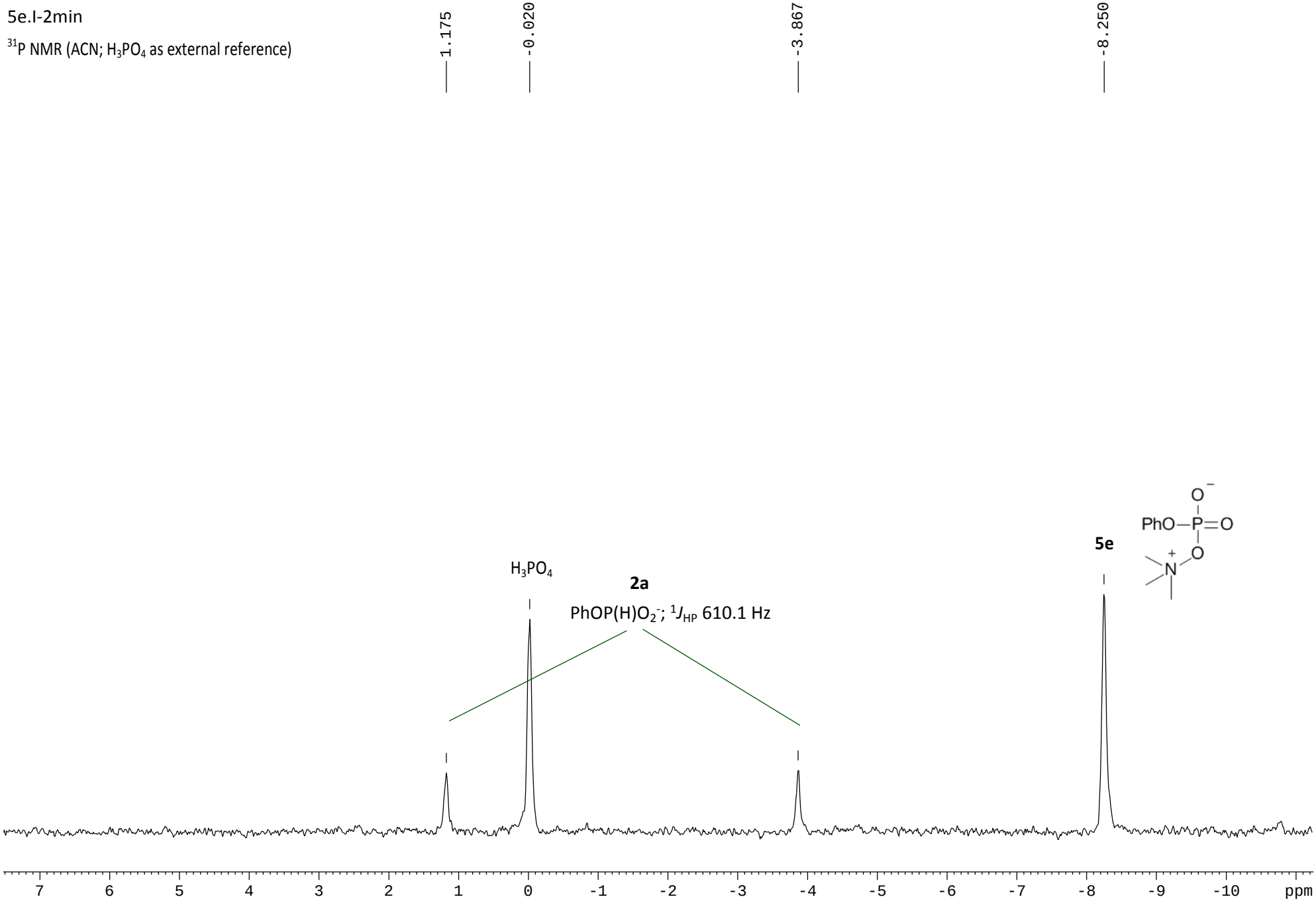


Fig. S7b Reaction of $(PhO)_2P(H)O$ **1a** with Me_3N *N*-oxide **3e** in acetonitrile.
^{31}P NMR spectrum of the reaction mixture after ca. 2 min.

5f.II-10min

$^{31}P\{^{1}H\}$ NMR (DMF/Py (7:3); H_3PO_4 as external reference)

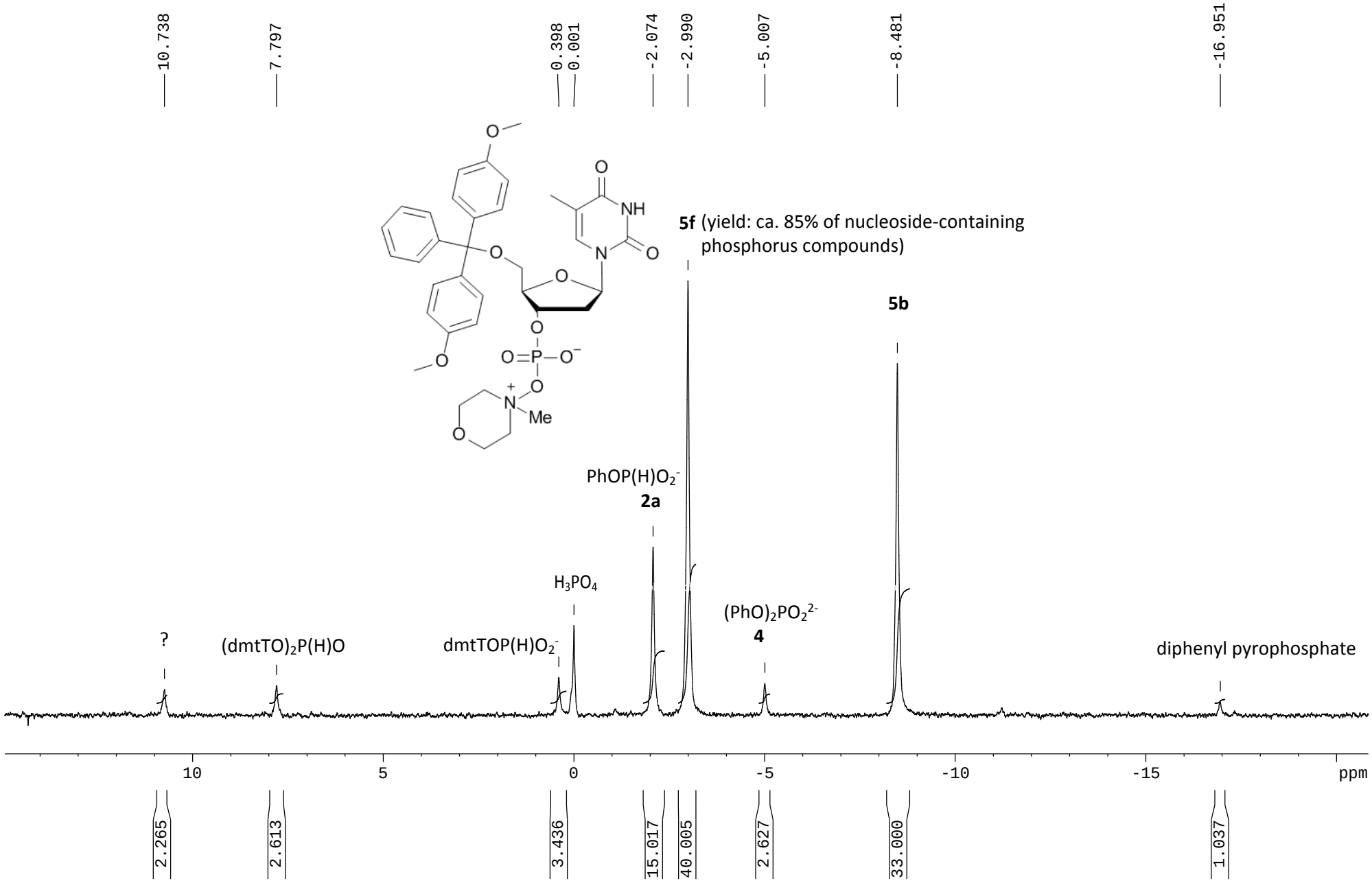


Fig. S8a Reaction of an *in situ* generated (PhO)(dmtTO)P(H)O (ca. 0.5 mmol) and the excess of $(PhO)_2P(H)O$ (ca. 0.75 mmol) with NMM *N*-oxide **3b** (3 mmol) in DMF-pyridine 7:3 (v/v). Proton-decoupled ^{31}P NMR spectrum of the reaction mixture after ca. 10 min.

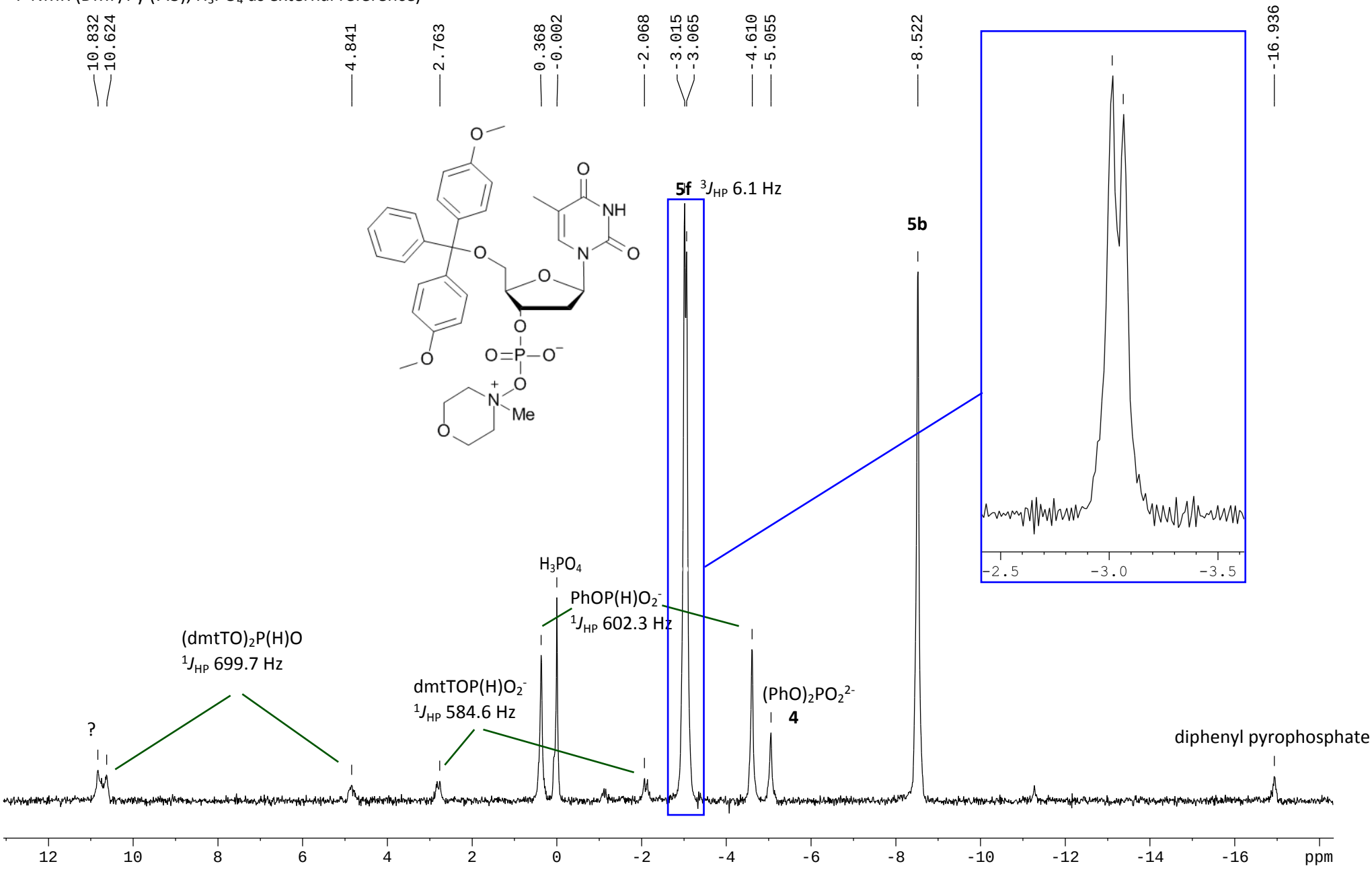
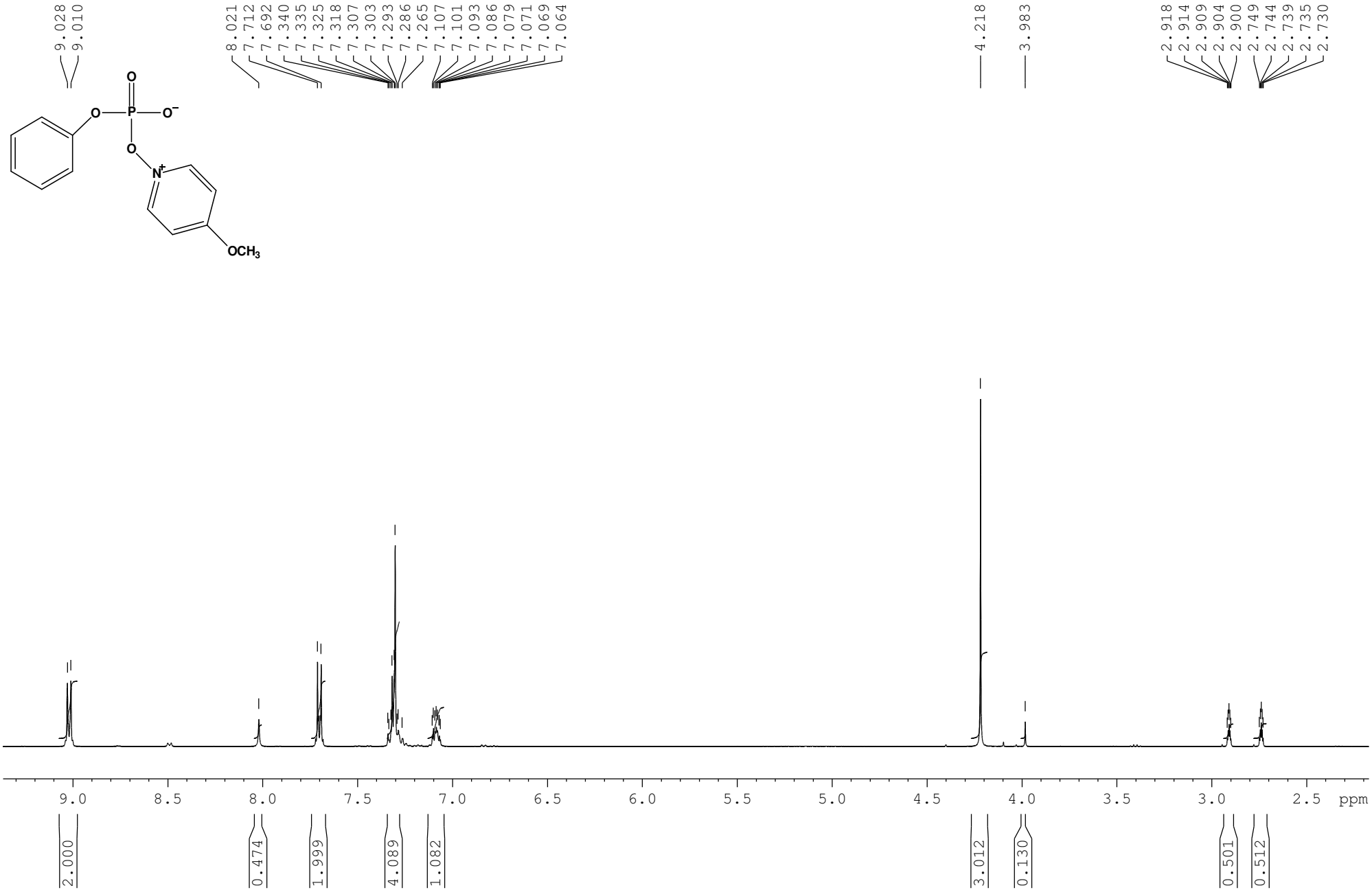


Fig. S8b Reaction of an *in situ* generated (PhO)(dmtTO)P(H)O (ca. 0.5 mmol) and the excess of $(PhO)_2P(H)O$ (ca. 0.75 mmol) with NMM *N*-oxide **3b** (3 mmol) in DMF-pyridine 7:3 (v/v). ^{31}P NMR spectrum of the reaction mixture after ca. 10 min.

5. ^{1}H, ^{13}C, ^{31}P, ^{1}H-^{1}H COSY, ^{1}H-^{13}C HSQC NMR and MS spectra of the isolated products

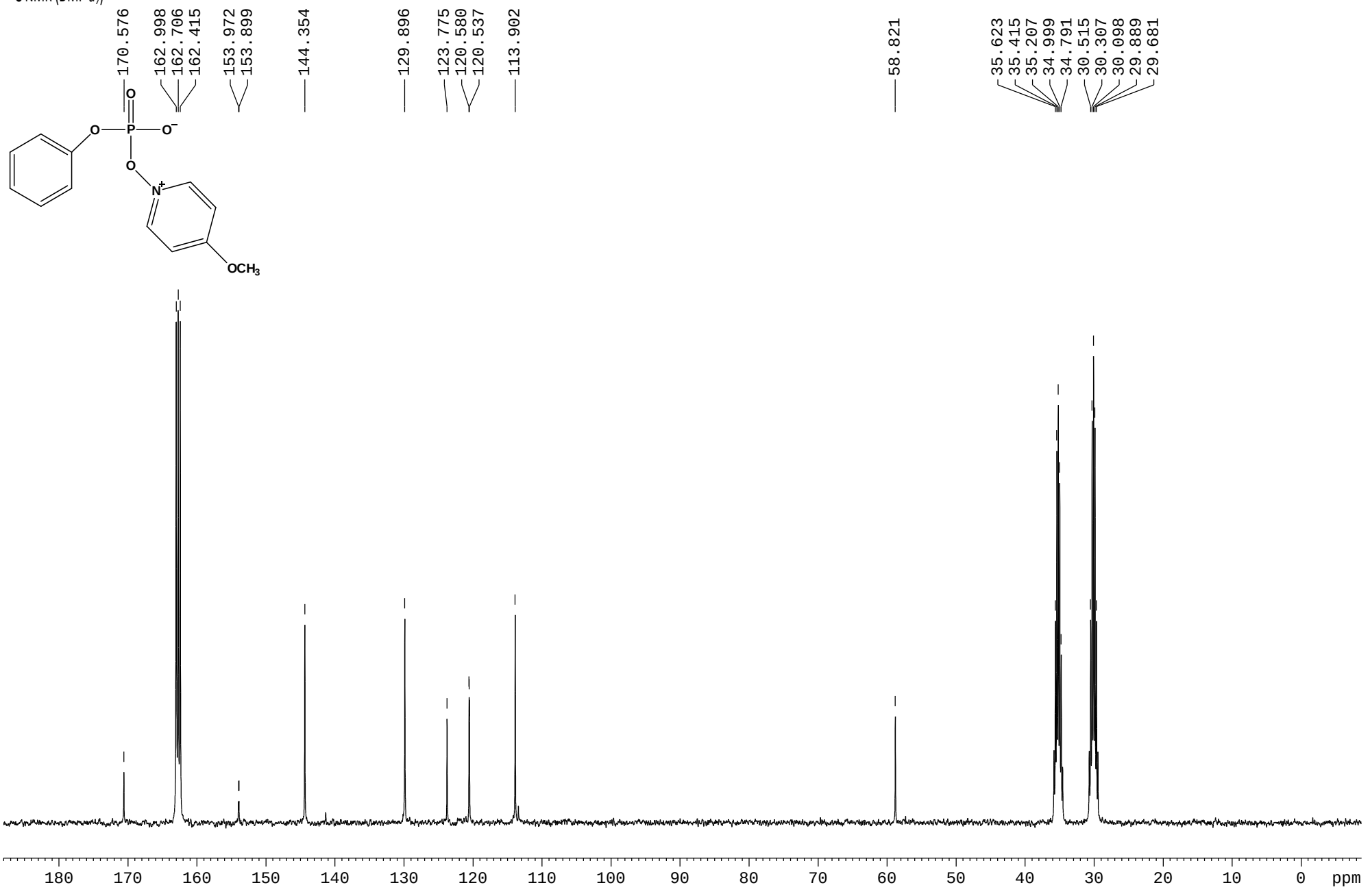
4-methoxypyridin-1-ium-1-yl phenyl phosphate **5a**

^{1}H NMR (DMF-d_7)



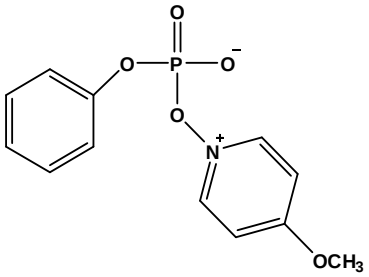
4-methoxypyridin-1-ium-1-yl phenyl phosphate **5a**

^{13}C NMR (DMF-d_7)



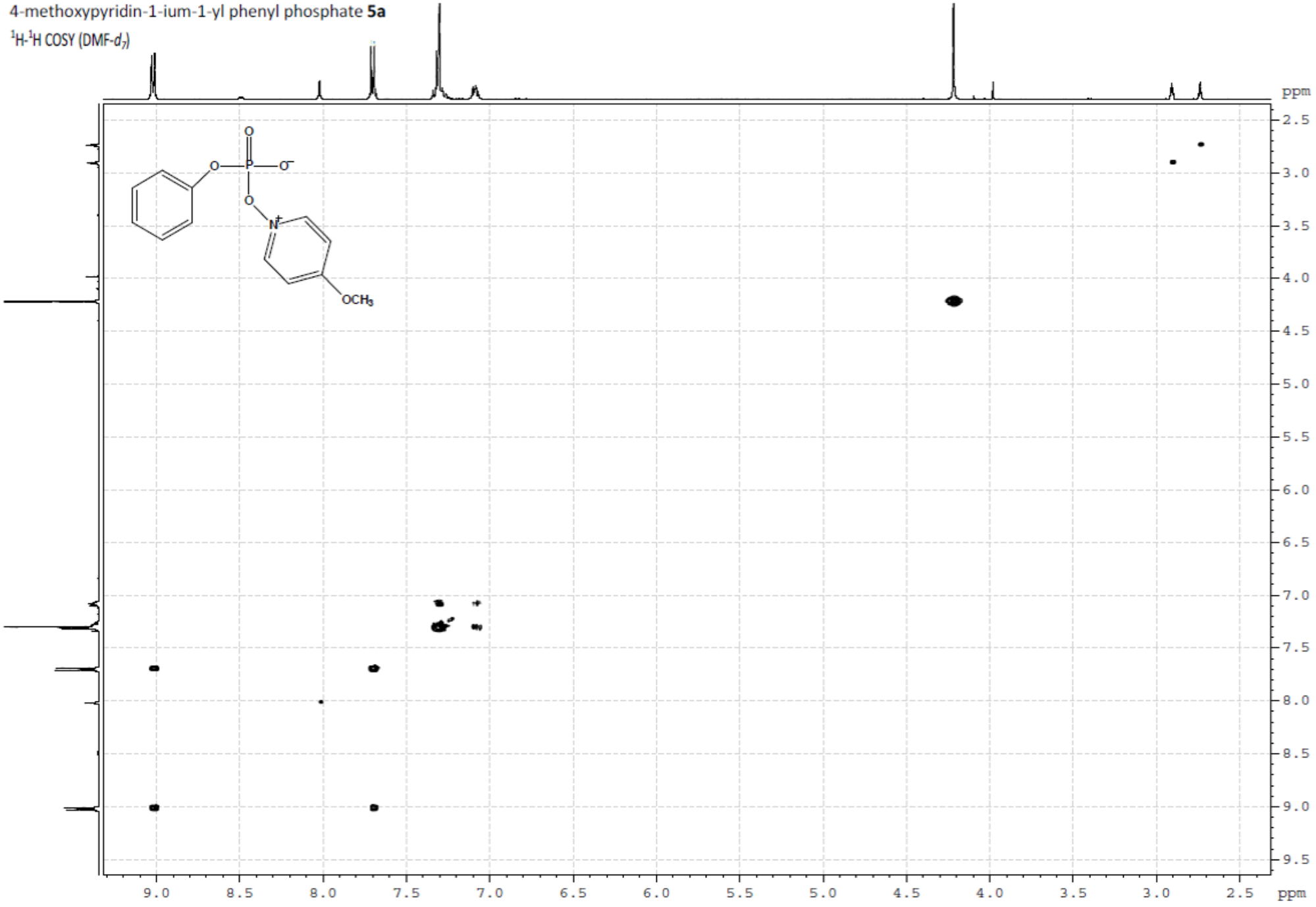
4-methoxypyridin-1-ium-1-yl phenyl phosphate **5a**

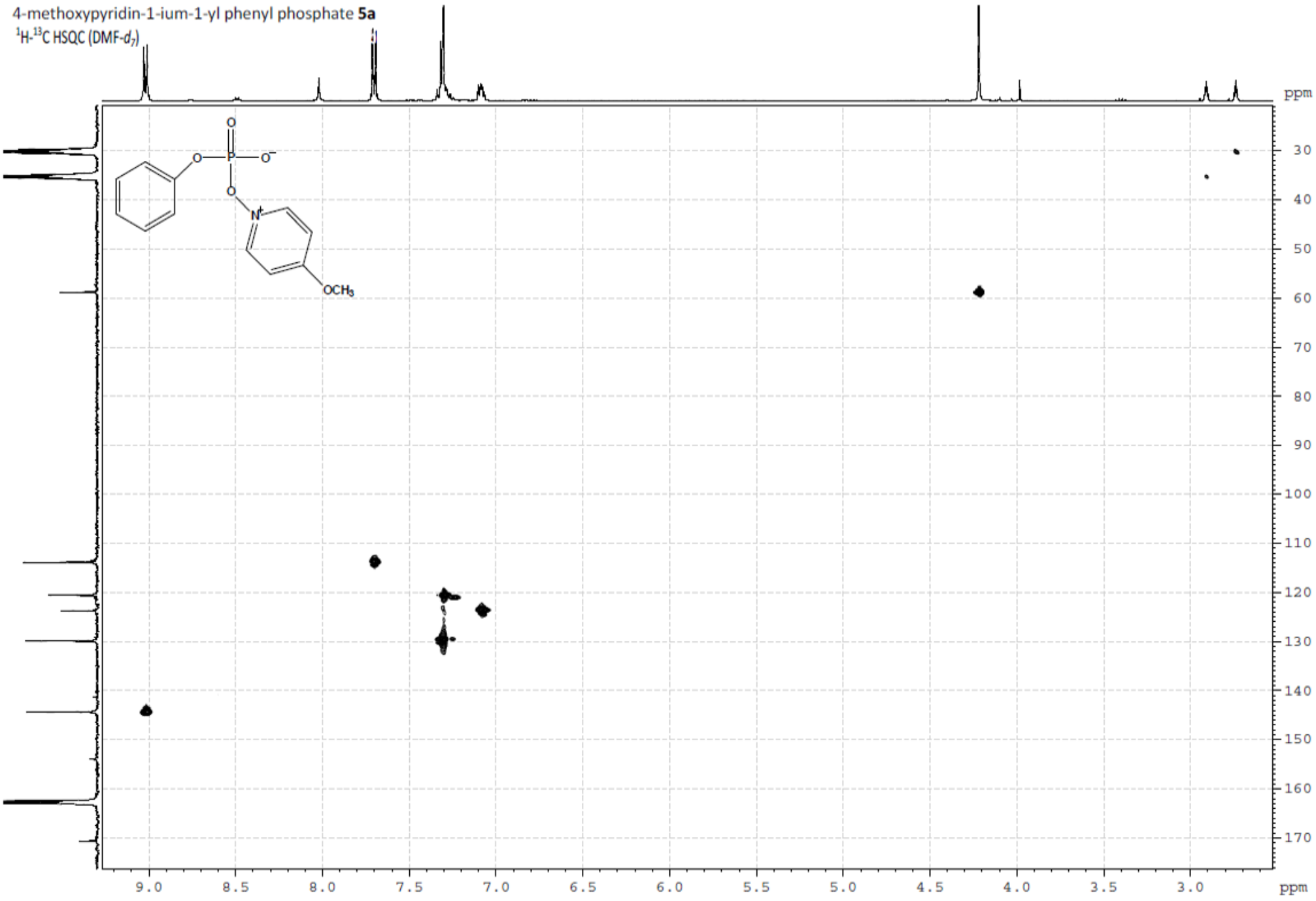
$^{31}P\{^{1}H\}$ NMR (DMF; H_3PO_4 as external reference)



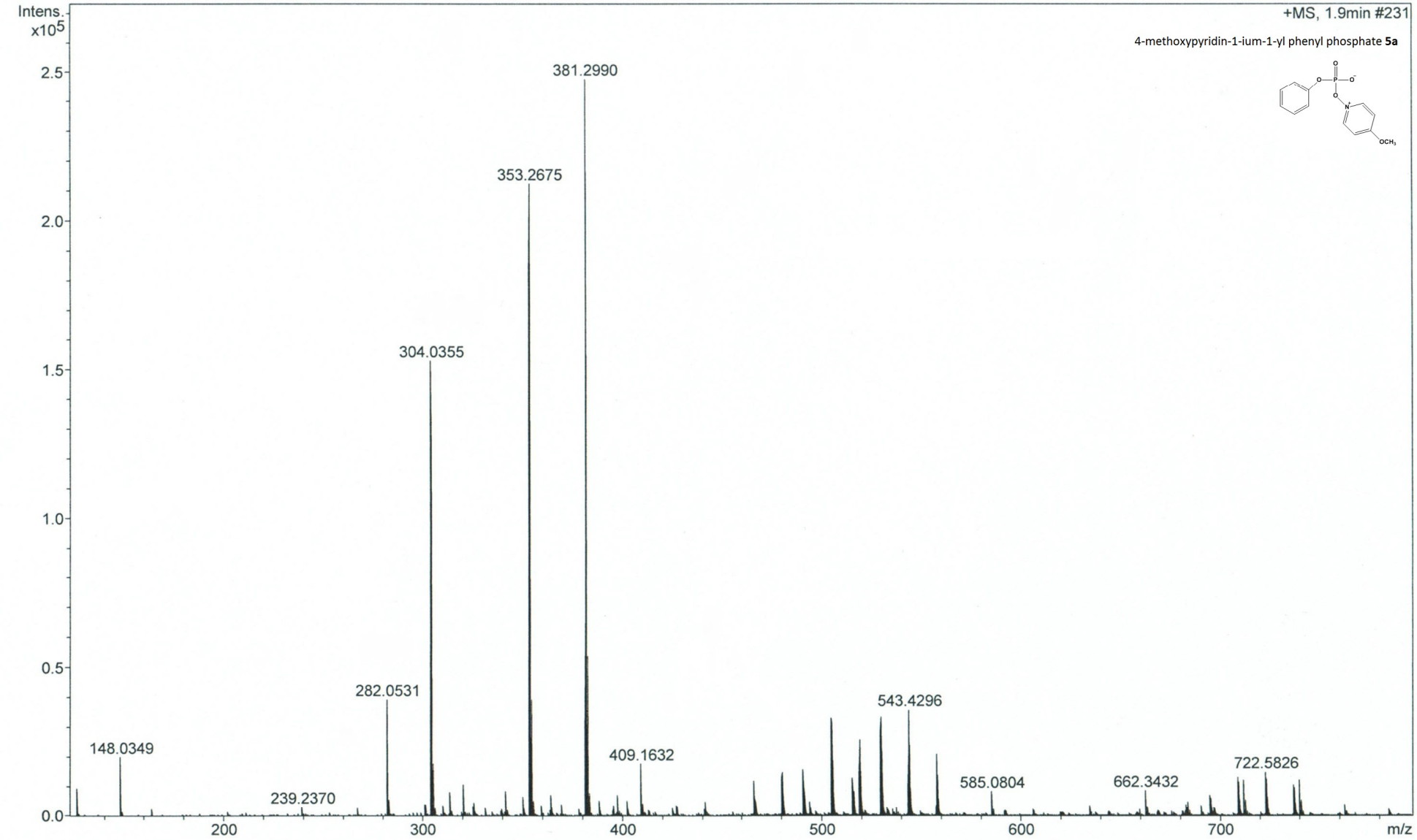
4-methoxypyridin-1-ium-1-yl phenyl phosphate **5a**

^{1}H-^{1}H COSY (DMF-d_7)




4-methoxypyridin-1-ium-1-yl phenyl phosphate **5a**
^{1}H-^{13}C HSQC (DMF-d_7)
ppm
30
40
50
60
70
80
90
100
110
120
130
140
150
160
170
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
ppm

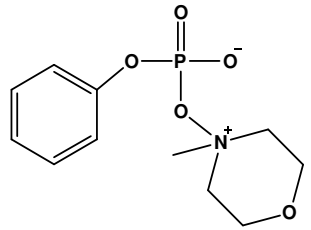
Generic Display Report (all)



Bruker Compass DataAnalysis 4.0 printed: 9/10/2015 2:45:56 PM Page 1 of 1

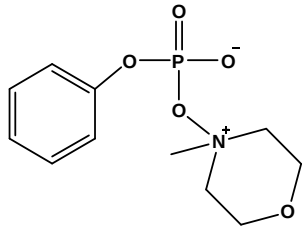
N-methylmorpholino-4-ium phenyl phosphate **5b**

^{1}H NMR (D_2O)



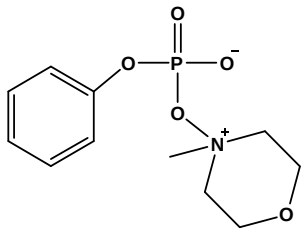
N-methylmorpholino-4-ium phenyl phosphate **5b**

^{13}C NMR (D_2O)



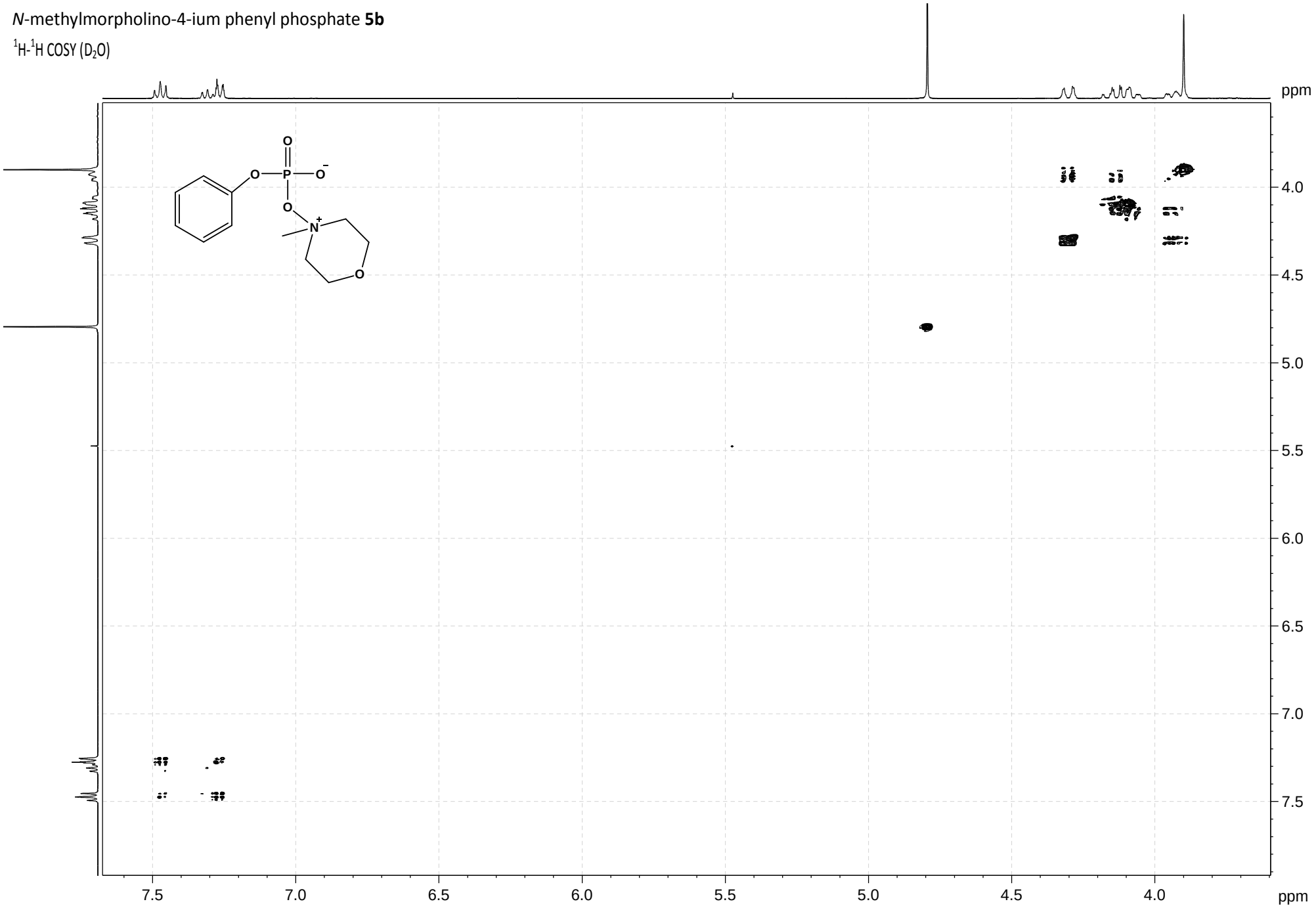
N-methylmorpholino-4-ium phenyl phosphate **5b**

$^{31}P\{^{1}H\}$ NMR (H_2O; H_3PO_4 as external reference)



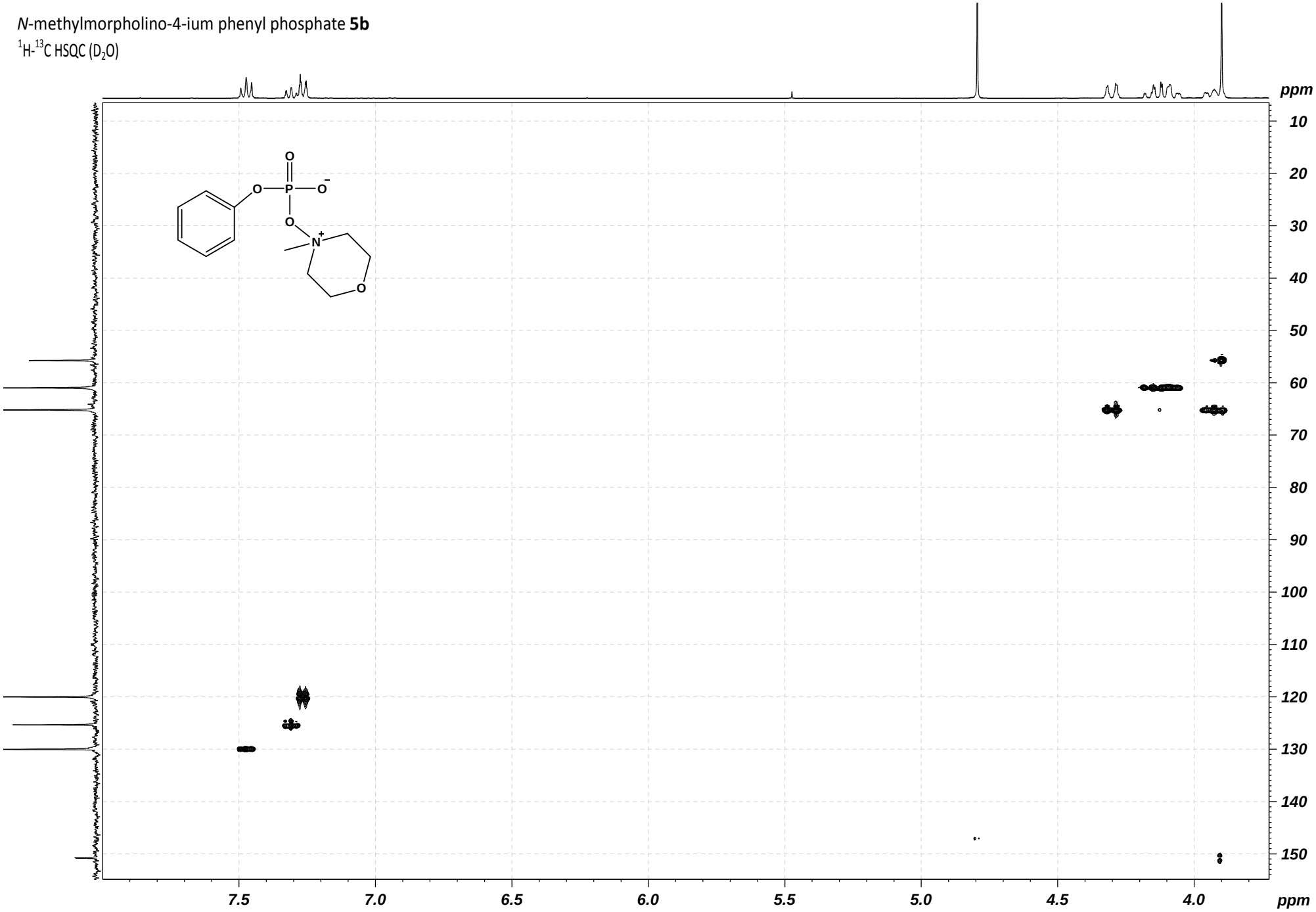
N-methylmorpholino-4-ium phenyl phosphate **5b**

^{1}H-^{1}H COSY (D_2O)



N-methylmorpholino-4-ium phenyl phosphate **5b**

^{1}H-^{13}C HSQC (D_2O)



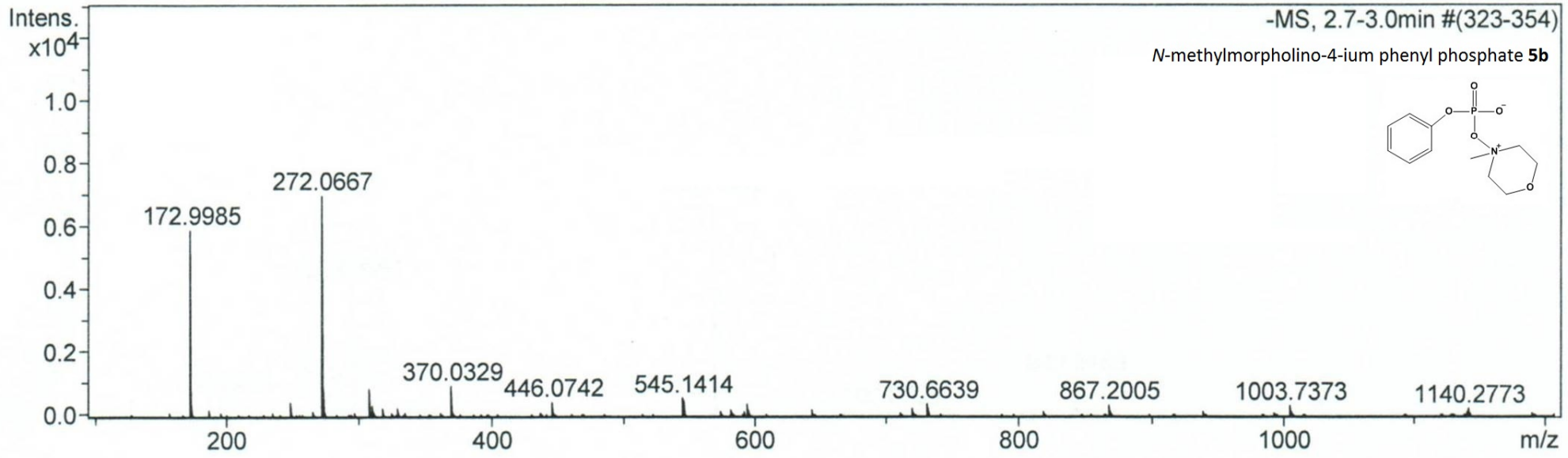
Mass Spectrum SmartFormula Report

Analysis Info

Analysis Name	D:\Data\Basia\zlecone\15_02_23_A1neg.d	Acquisition Date	2/23/2015 11:12:55 AM
Method	ewelina.m	Operator	Bruker Customer
Sample Name		Instrument / Ser#	micrOTOF-Q 128
Comment			

Acquisition Parameter

Source Type	ESI	Ion Polarity	Negative	Set Nebulizer	0.4 Bar
Focus	Not active	Set Capillary	3200 V	Set Dry Heater	220 °C
Scan Begin	100 m/z	Set End Plate Offset	-500 V	Set Dry Gas	4.0 l/min
Scan End	1200 m/z	Set Collision Cell RF	200.0 Vpp	Set Divert Valve	Source



Meas. m/z	#	Formula	m/z	err [ppm]	Mean err [ppm]	rdb	N-Rule	e⁻ Conf	mSigma	Std I	Std Mean m/z	Std I VarNorm	Std m/z Diff	Std Comb Dev

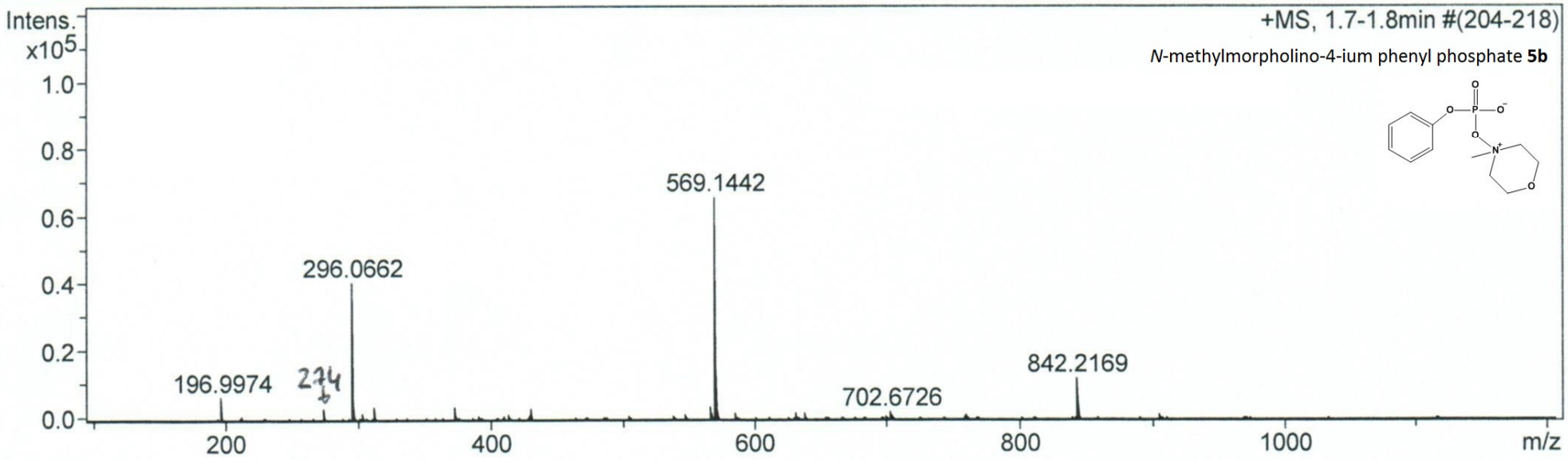
Mass Spectrum SmartFormula Report

Analysis Info

Analysis Name	D:\Data\Basia\zlecone\15_02_23_A1pos.d	Acquisition Date	2/23/2015 10:51:18 AM
Method	ewelina.m	Operator	Bruker Customer
Sample Name		Instrument / Ser#	micrOTOF-Q 128
Comment			

Acquisition Parameter

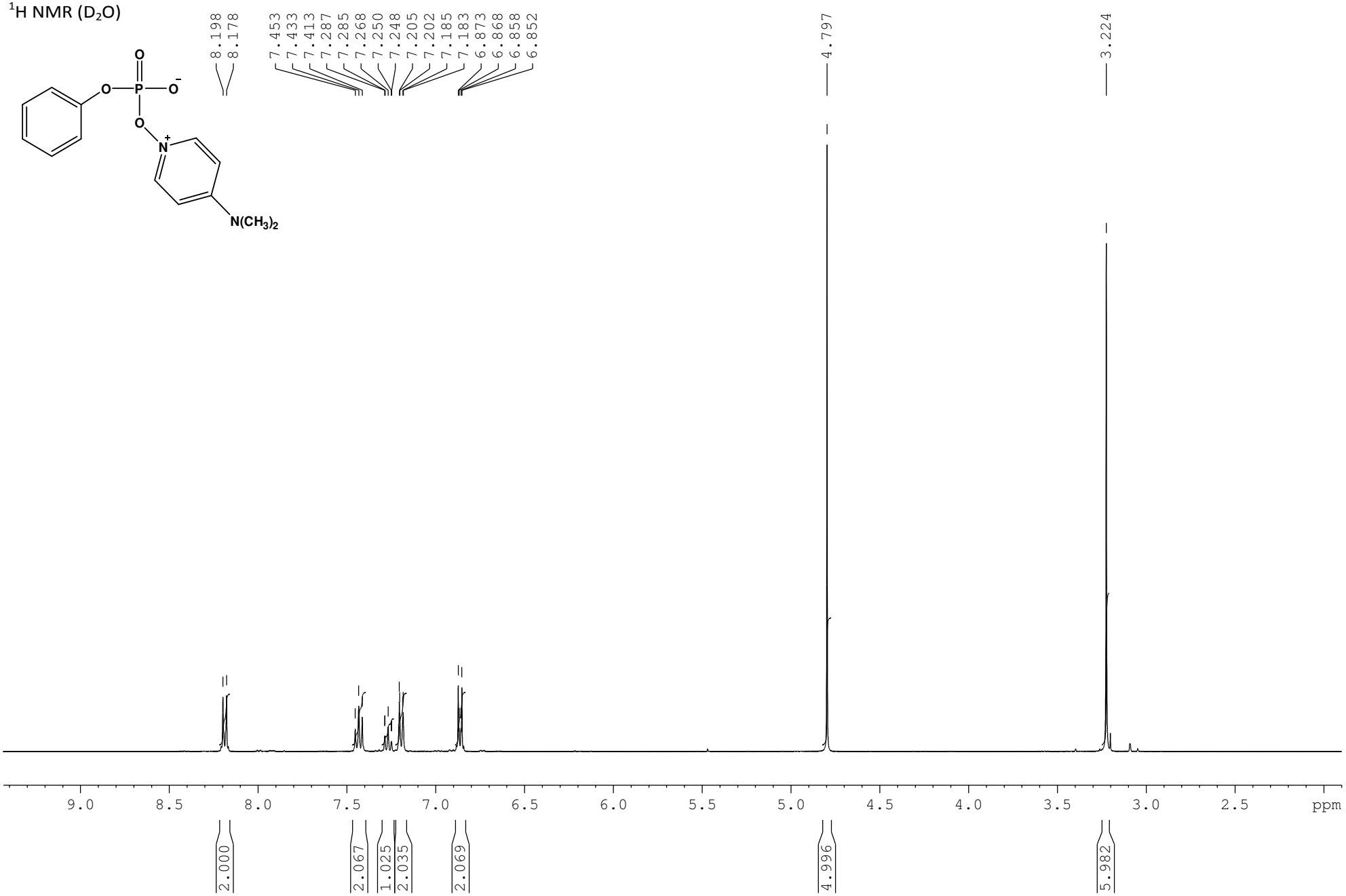
Source Type	ESI	Ion Polarity	Positive	Set Nebulizer	0.4 Bar
Focus	Not active	Set Capillary	4500 V	Set Dry Heater	220 °C
Scan Begin	100 m/z	Set End Plate Offset	-500 V	Set Dry Gas	4.0 l/min
Scan End	1200 m/z	Set Collision Cell RF	200.0 Vpp	Set Divert Valve	Source



Meas. m/z	#	Formula	m/z	err [ppm]	Mean err [ppm]	rdb	N-Rule	e⁻ Conf	mSigma	Std I	Std Mean m/z	Std I VarNorm	Std m/z Diff	Std Comb Dev

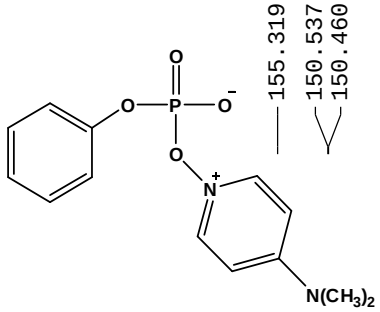
4-(dimethylamino)pyridin-1-ium-1-yl phenyl phosphate **5d**

^{1}H NMR (D_2O)

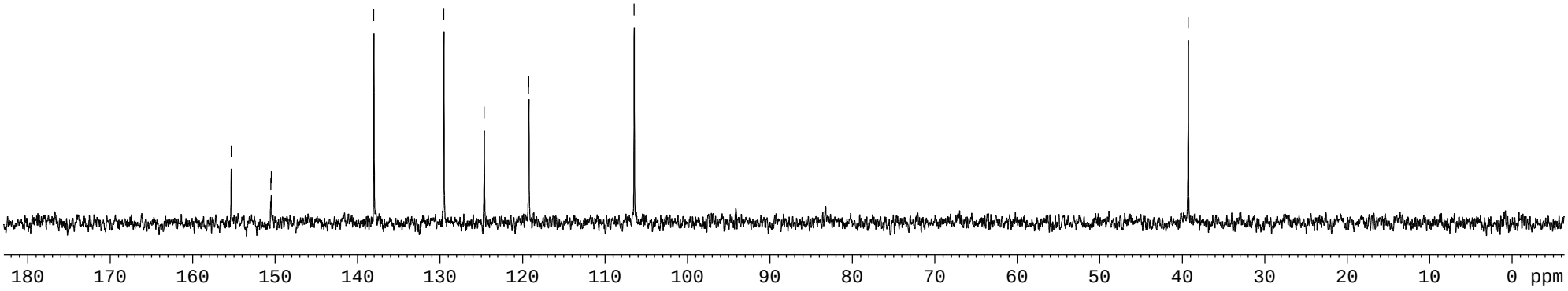


4-(dimethylamino)pyridin-1-ium-1-yl phenyl phosphate **5d**

^{13}C NMR (D_2O)

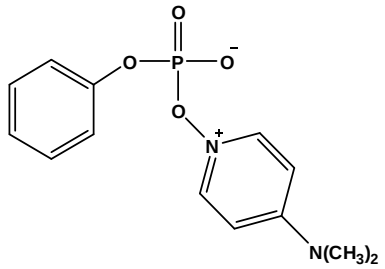


155.319
150.537
150.460
138.046
129.559
124.664
119.295
119.248
106.478
39.288



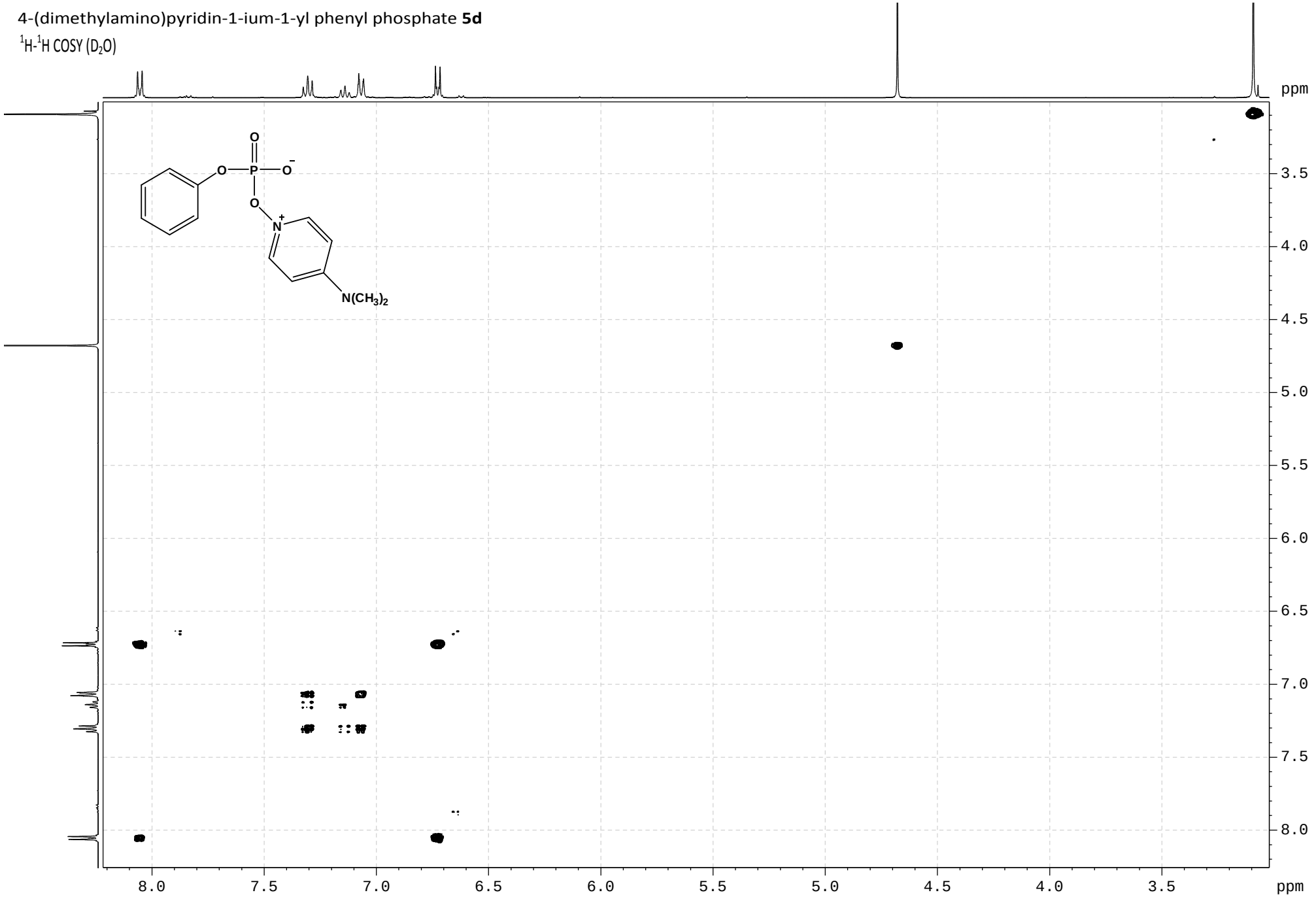
4-(dimethylamino)pyridin-1-ium-1-yl phenyl phosphate **5d**

$^{31}P\{^{1}H\}$ NMR (H_2O; H_3PO_4 as external reference)



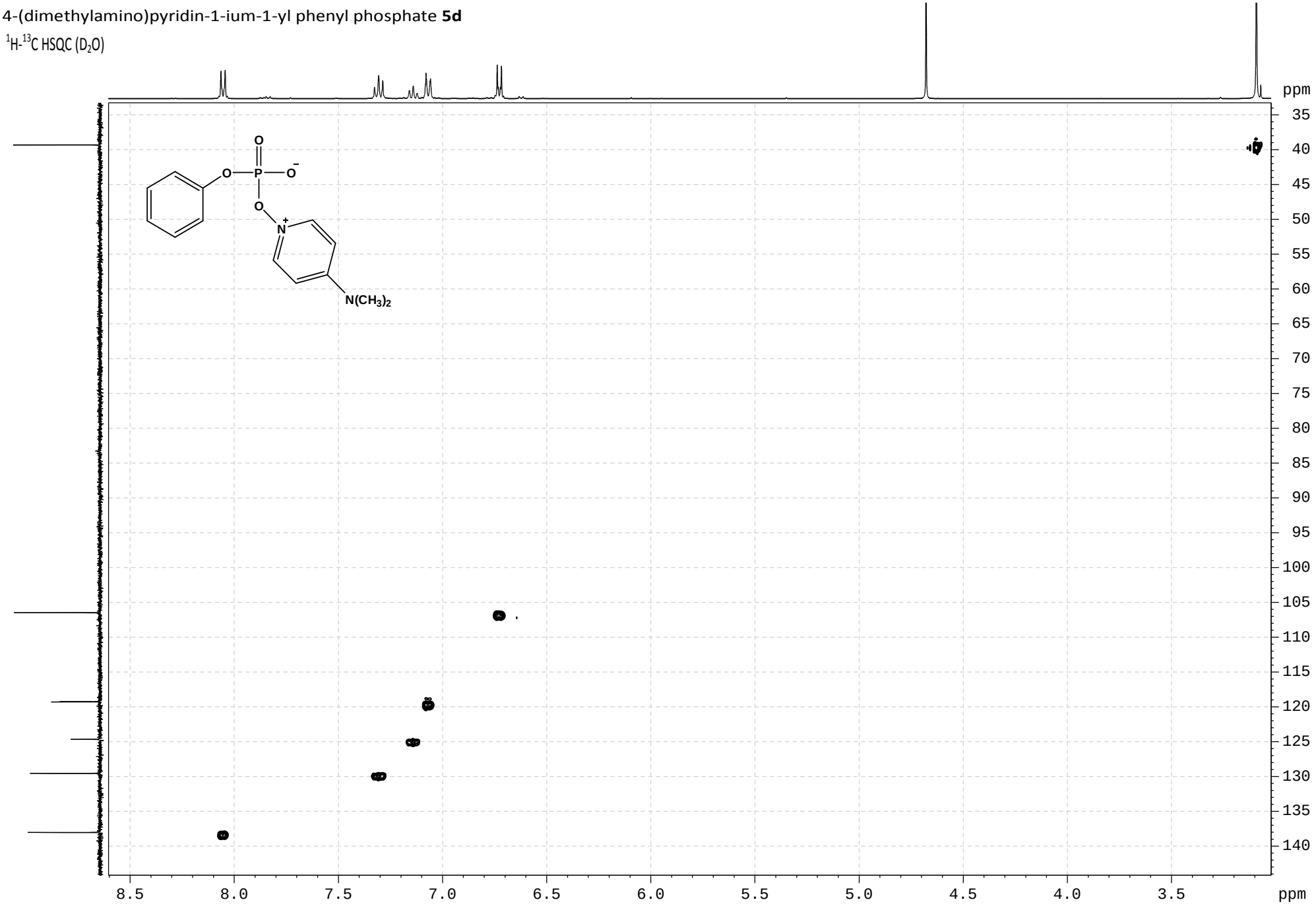
4-(dimethylamino)pyridin-1-ium-1-yl phenyl phosphate **5d**

^{1}H-^{1}H COSY (D_2O)



4-(dimethylamino)pyridin-1-ium-1-yl phenyl phosphate **5d**

^{1}H-^{13}C HSQC (D_2O)



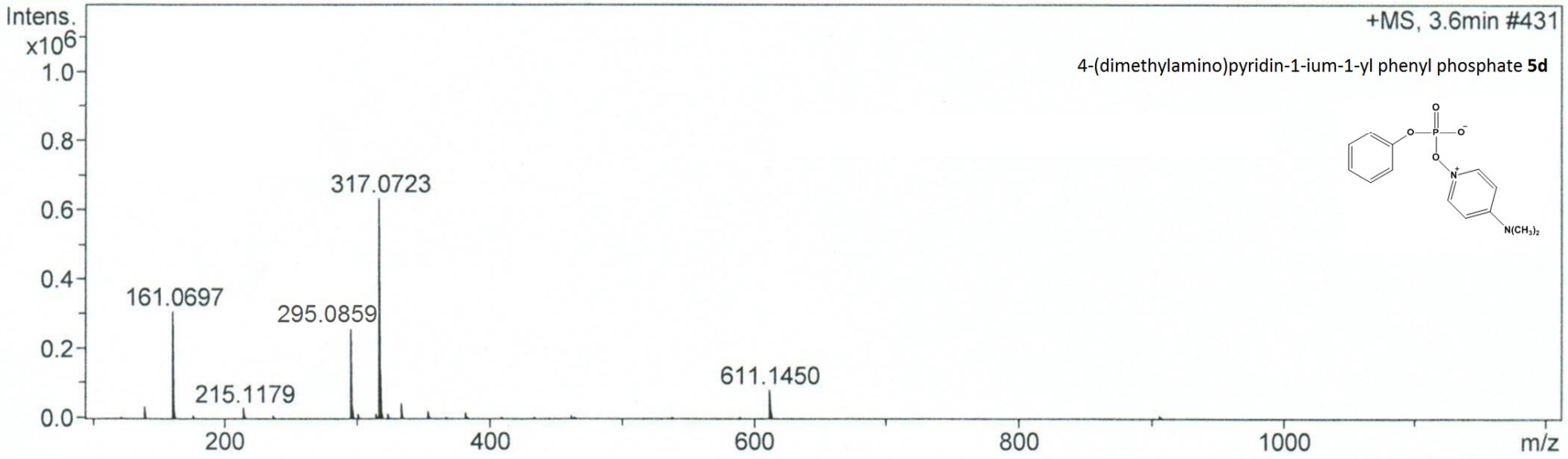
Mass Spectrum SmartFormula Report

Analysis Info

Analysis Name	D:\Data\Basia\zlecone\15_08_31_a3_pos2_kal.d	Acquisition Date	8/31/2015 1:37:30 PM
Method	ewelina.m	Operator	Bruker Customer
Sample Name	NO2	Instrument / Ser#	micrOTOF-Q 128
Comment			

Acquisition Parameter

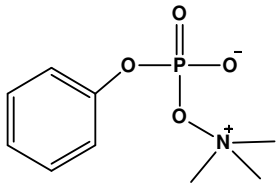
Source Type	ESI	Ion Polarity	Positive	Set Nebulizer	0.4 Bar
Focus	Not active	Set Capillary	4500 V	Set Dry Heater	220 °C
Scan Begin	100 m/z	Set End Plate Offset	-500 V	Set Dry Gas	4.0 l/min
Scan End	1200 m/z	Set Collision Cell RF	200.0 Vpp	Set Divert Valve	Waste



Meas. m/z	#	Formula	m/z	err [ppm]	Mean err [ppm]	rdb	N-Rule	e⁻ Conf	mSigma	Std I	Std Mean m/z	Std I VarNorm	Std m/z Diff	Std Comb Dev

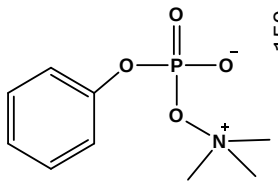
Trimethylammonium phenyl phosphate **5e**

^{1}H NMR (D_2O)



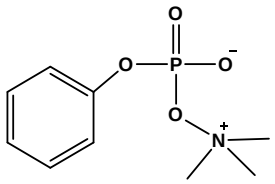
Trimethylammonium phenyl phosphate **5e**

^{13}C NMR (D_2O)



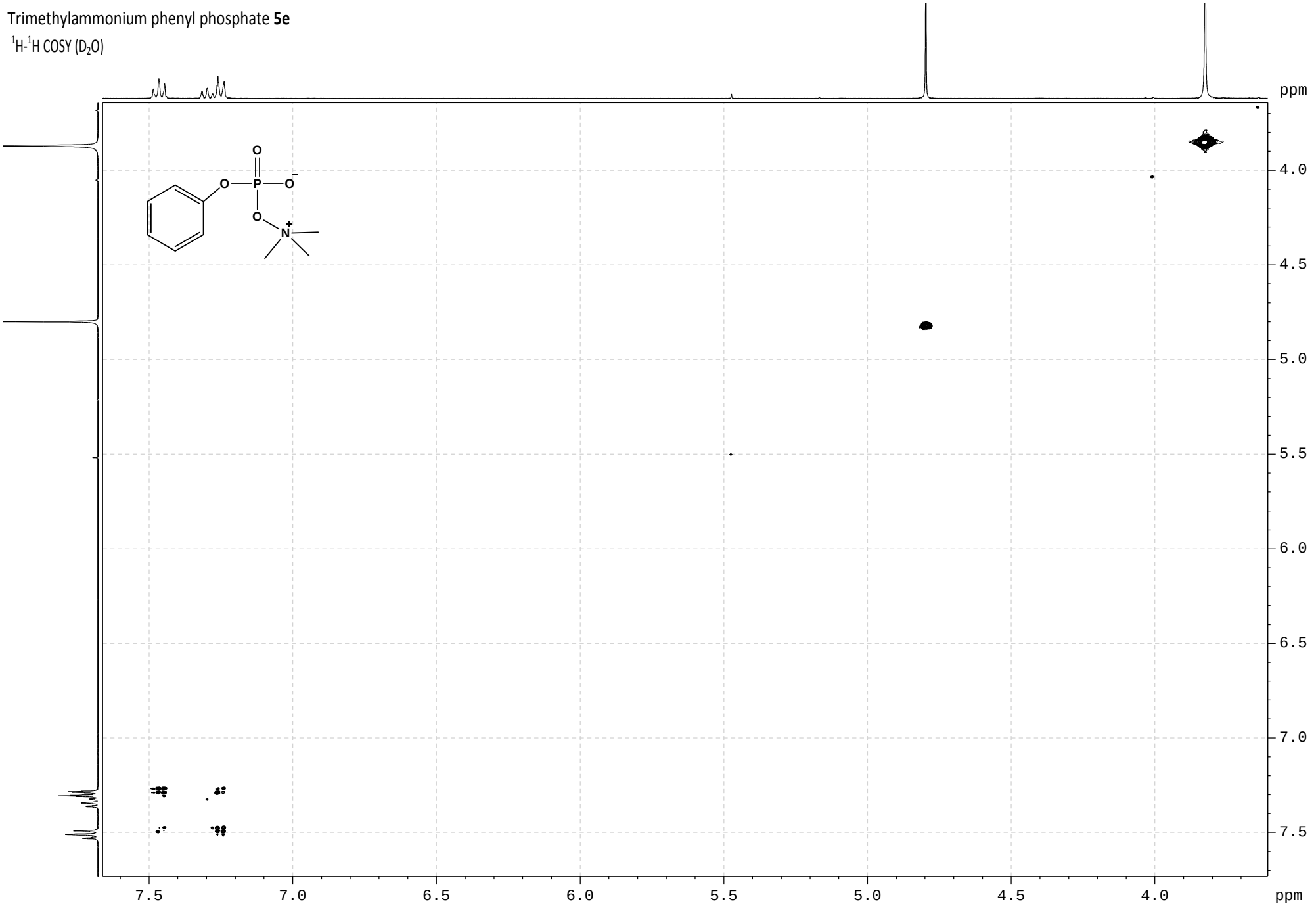
Trimethylammonium phenyl phosphate **5e**

$^{31}P\{^{1}H\}$ NMR (H_2O; H_3PO_4 as external reference)



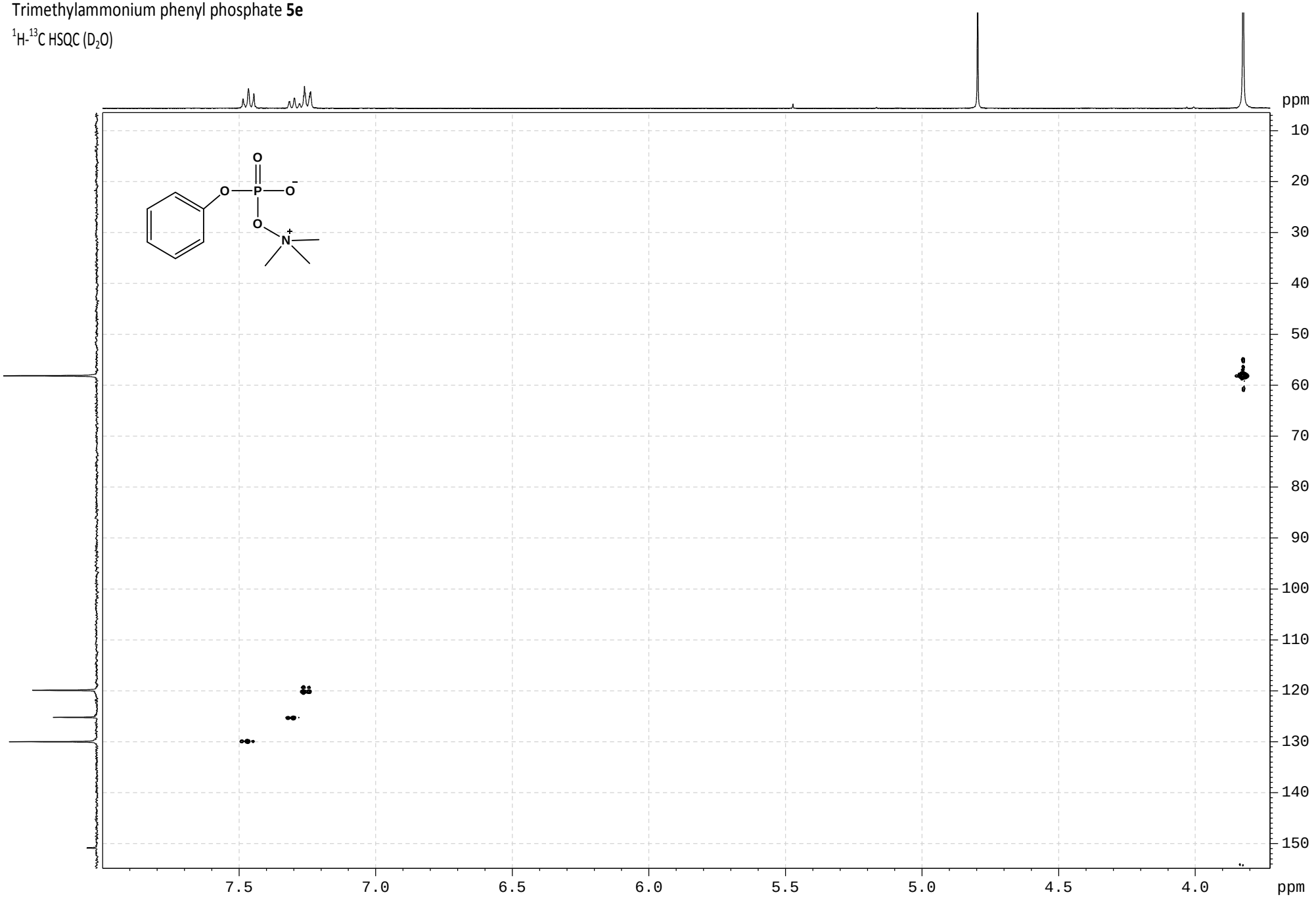
Trimethylammonium phenyl phosphate **5e**

^{1}H-^{1}H COSY (D_2O)



Trimethylammonium phenyl phosphate **5e**

^{1}H-^{13}C HSQC (D_2O)



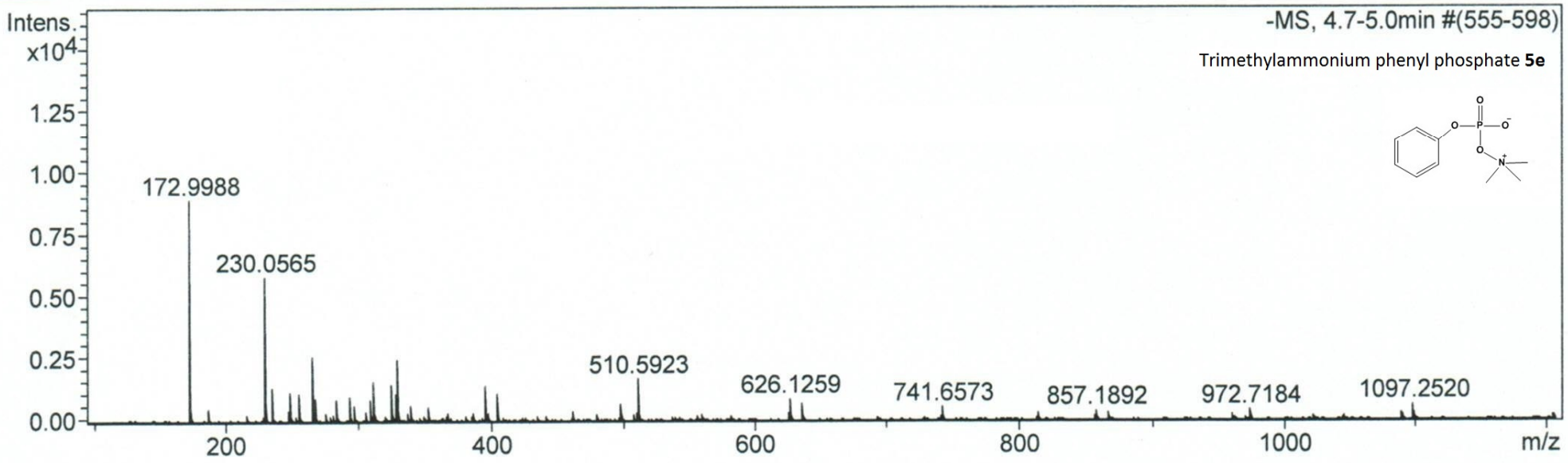
Mass Spectrum SmartFormula Report

Analysis Info

Analysis Name	D:\Data\Basia\zlecone\15_02_24_A2_neg.d	Acquisition Date	2/24/2015 12:57:13 PM
Method	ewelina.m	Operator	Bruker Customer
Sample Name		Instrument / Ser#	micrOTOF-Q 128
Comment			

Acquisition Parameter

Source Type	ESI	Ion Polarity	Negative	Set Nebulizer	0.4 Bar
Focus	Not active	Set Capillary	3200 V	Set Dry Heater	220 °C
Scan Begin	100 m/z	Set End Plate Offset	-500 V	Set Dry Gas	4.0 l/min
Scan End	1200 m/z	Set Collision Cell RF	200.0 Vpp	Set Divert Valve	Source



Meas. m/z	#	Formula	m/z	err [ppm]	Mean err [ppm]	rdb	N-Rule	e⁻ Conf	mSigma	Std I	Std Mean m/z	Std I VarNorm	Std m/z Diff	Std Comb Dev

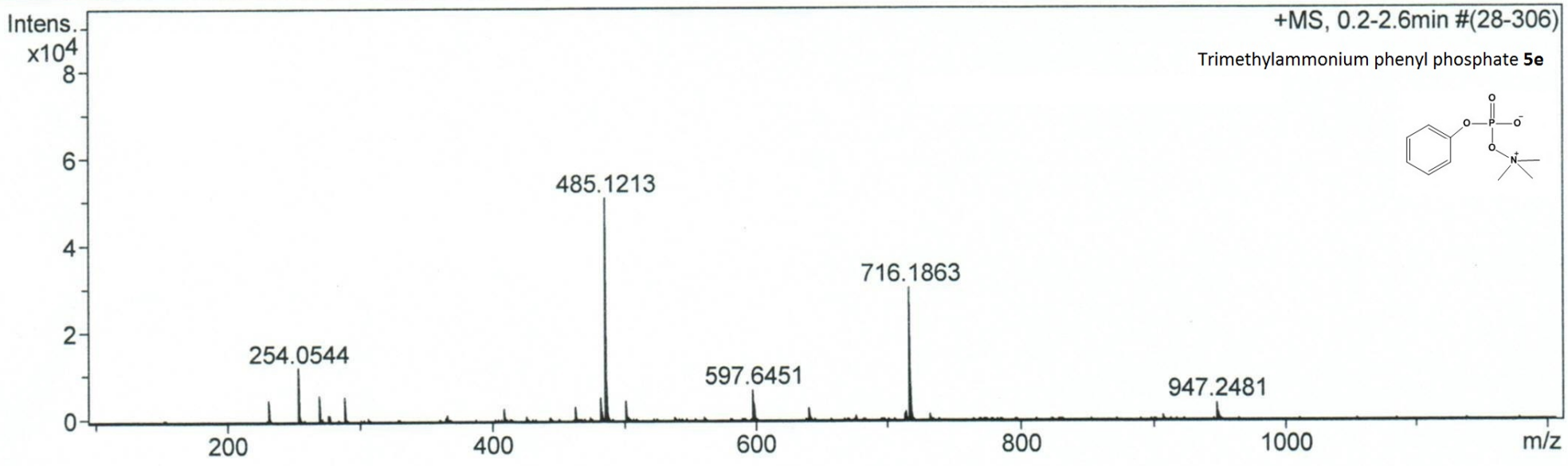
Mass Spectrum SmartFormula Report

Analysis Info

Analysis Name	D:\Data\Basia\zlecone\15_02_24_A2pos.d	Acquisition Date	2/24/2015 12:40:31 PM
Method	ewelina.m	Operator	Bruker Customer
Sample Name		Instrument / Ser#	micrOTOF-Q 128
Comment			

Acquisition Parameter

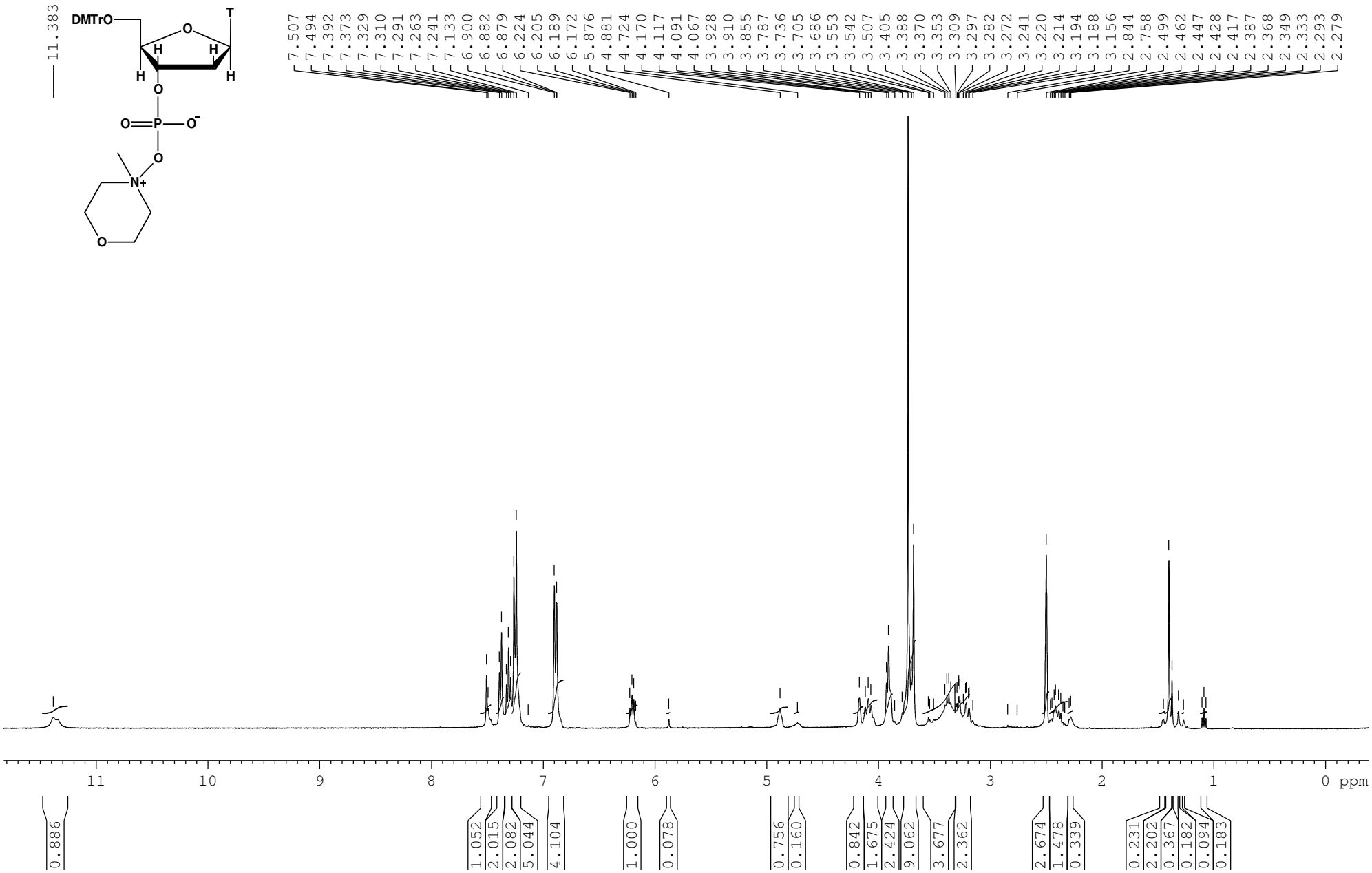
Source Type	ESI	Ion Polarity	Positive	Set Nebulizer	0.4 Bar
Focus	Not active	Set Capillary	4500 V	Set Dry Heater	220 °C
Scan Begin	100 m/z	Set End Plate Offset	-500 V	Set Dry Gas	4.0 l/min
Scan End	1200 m/z	Set Collision Cell RF	200.0 Vpp	Set Divert Valve	Source



Meas. m/z	#	Formula	m/z	err [ppm]	Mean err [ppm]	rdb	N-Rule	e⁻ Conf	mSigma	Std I	Std Mean m/z	Std I VarNorm	Std m/z Diff	Std Comb Dev

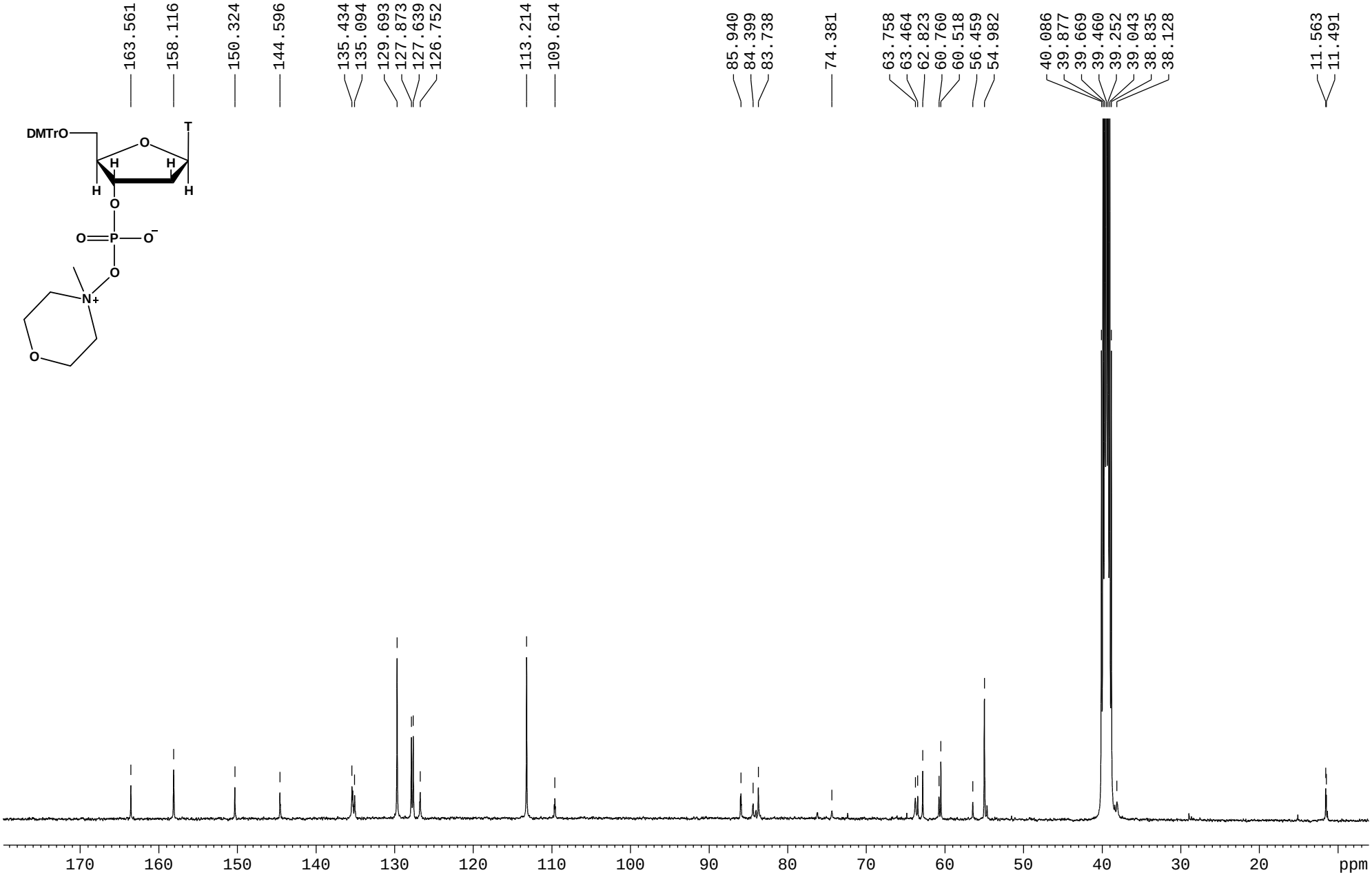
N-methylmorpholino-4-ium 5'-*O*-dimethoxytritylthymidin-3'-yl phosphate **5f**

^{1}H NMR (DMSO-d_6)



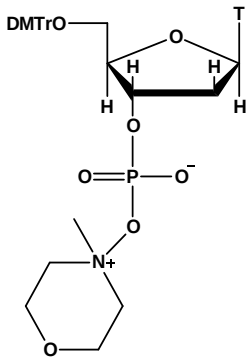
N-methylmorpholino-4-ium 5'-*O*-dimethoxytritylthymidin-3'-yl phosphate **5f**

^{13}C NMR (DMSO-d_6)



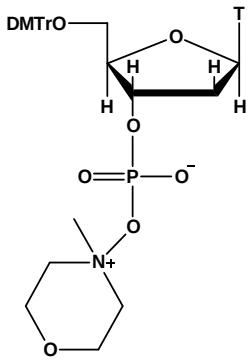
N-methylmorpholino-4-ium 5'-*O*-dimethoxytritylthymidin-3'-yl phosphate **5f**

$^{31}P\{^{1}H\}$ NMR (DMSO; H_3PO_4 as external reference)



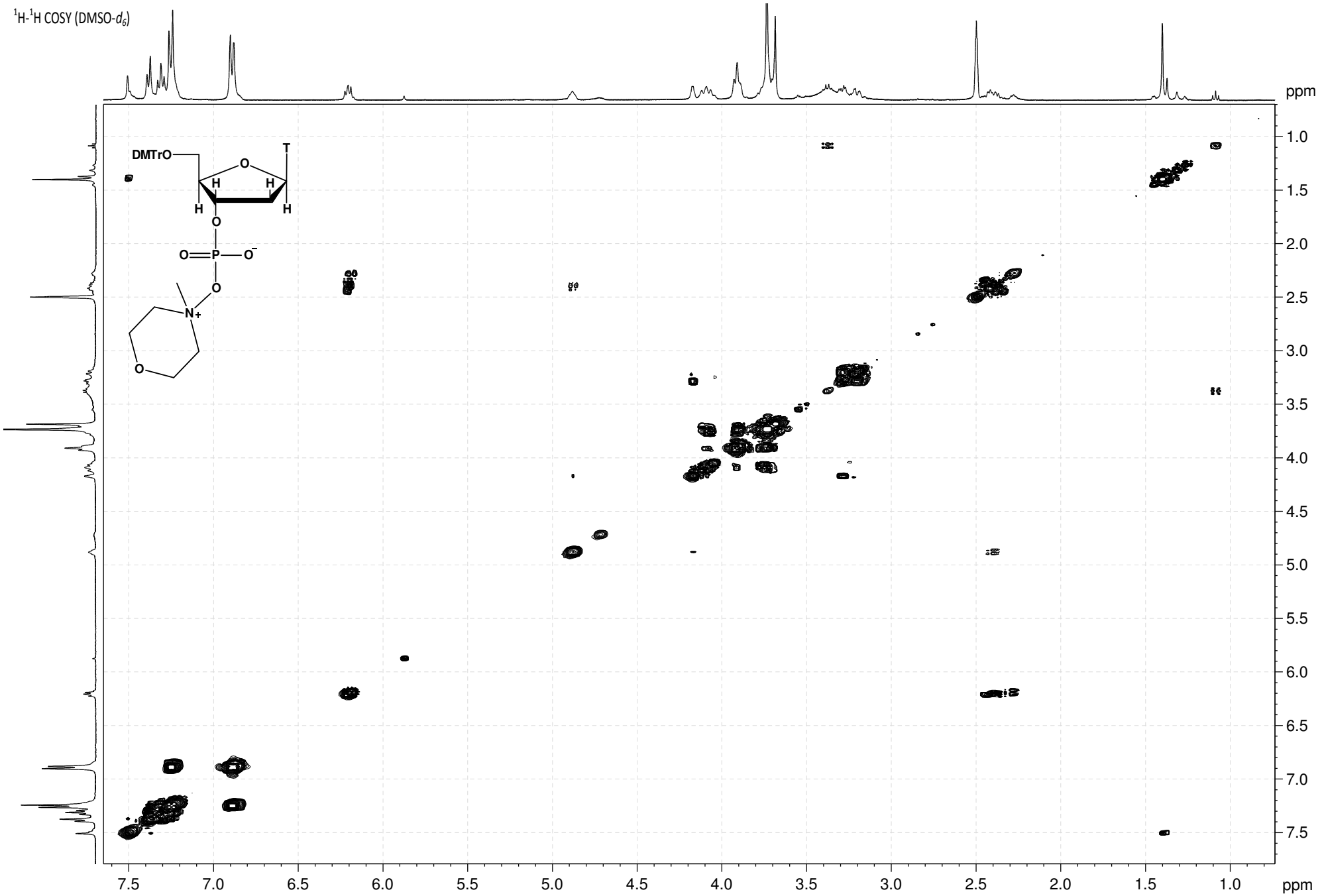
N-methylmorpholino-4-ium 5'-*O*-dimethoxytritylthymidin-3'-yl phosphate **5f**

^{31}P NMR (DMSO; H_3PO_4 as external reference)



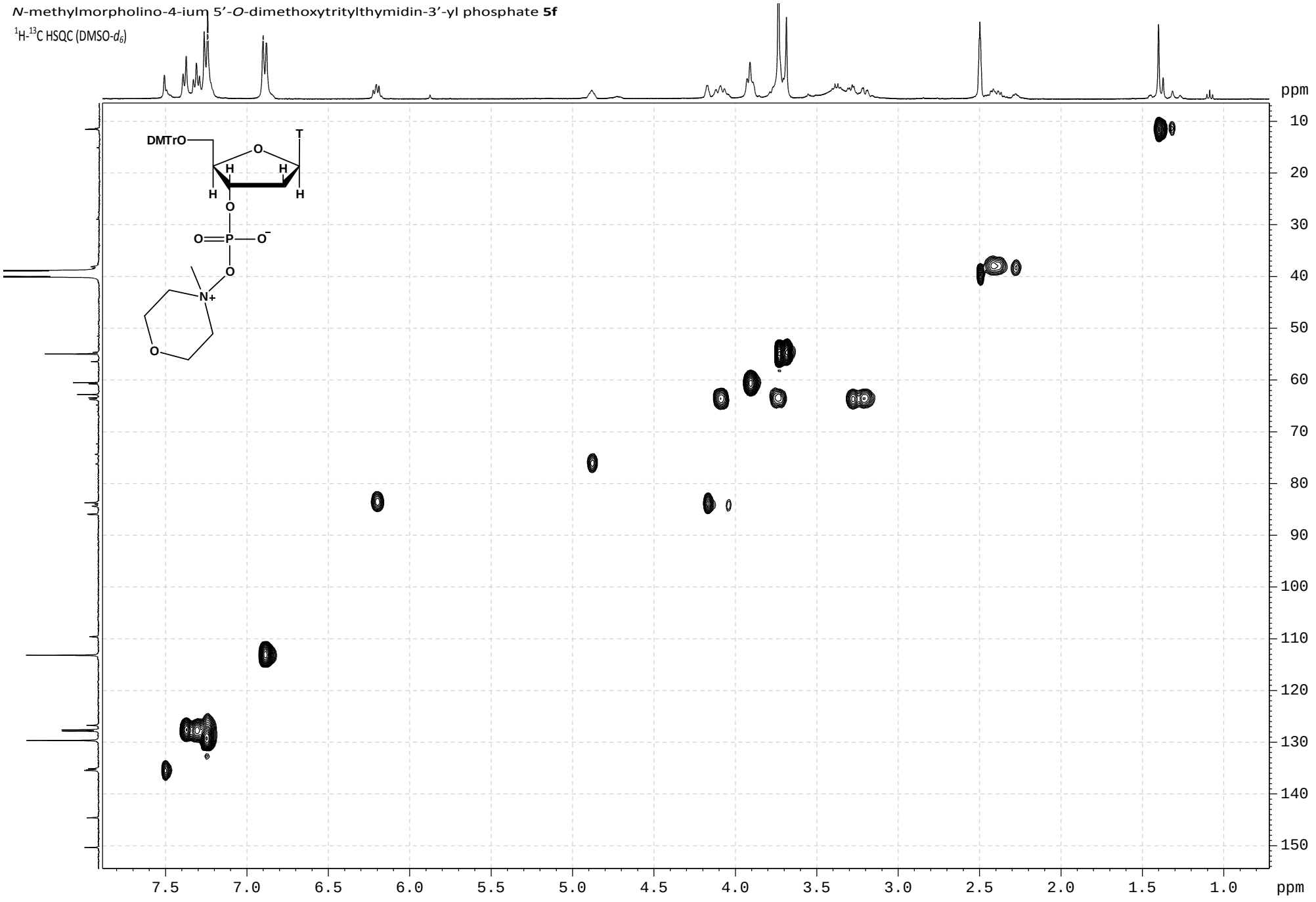
N-methylmorpholino-4-ium 5'-*O*-dimethoxytritylthymidin-3'-yl phosphate **5f**

^{1}H-^{1}H COSY (DMSO-d_6)



N-methylmorpholino-4-ium 5′-*O*-dimethoxytritylthymidin-3′-yl phosphate **5f**

^{1}H-^{13}C HSQC (DMSO-d_6)



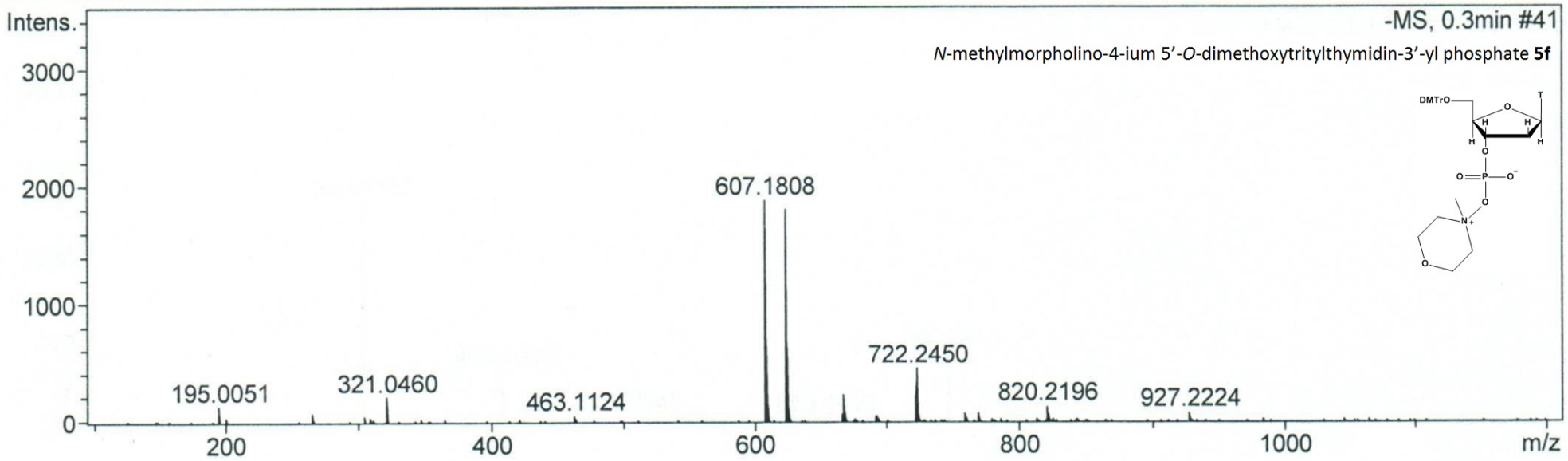
Mass Spectrum SmartFormula Report

Analysis Info

Analysis Name	D:\Data\Basia\zlecone\15_02_23_A6neg.d	Acquisition Date	2/23/2015 11:37:44 AM
Method	ewelina.m	Operator	Bruker Customer
Sample Name		Instrument / Ser#	micrOTOF-Q 128
Comment			

Acquisition Parameter

Source Type	ESI	Ion Polarity	Negative	Set Nebulizer	0.4 Bar
Focus	Not active	Set Capillary	3200 V	Set Dry Heater	220 °C
Scan Begin	100 m/z	Set End Plate Offset	-500 V	Set Dry Gas	4.0 l/min
Scan End	1200 m/z	Set Collision Cell RF	200.0 Vpp	Set Divert Valve	Source



N-methylmorpholino-4-ium 5'-*O*-dimethoxytritylthymidin-3'-yl phosphate **5f**

Meas. m/z	#	Formula	m/z	err [ppm]	Mean err [ppm]	rdb	N-Rule	e⁻ Conf	mSigma	Std I	Std Mean m/z	Std I VarNorm	Std m/z Diff	Std Comb Dev

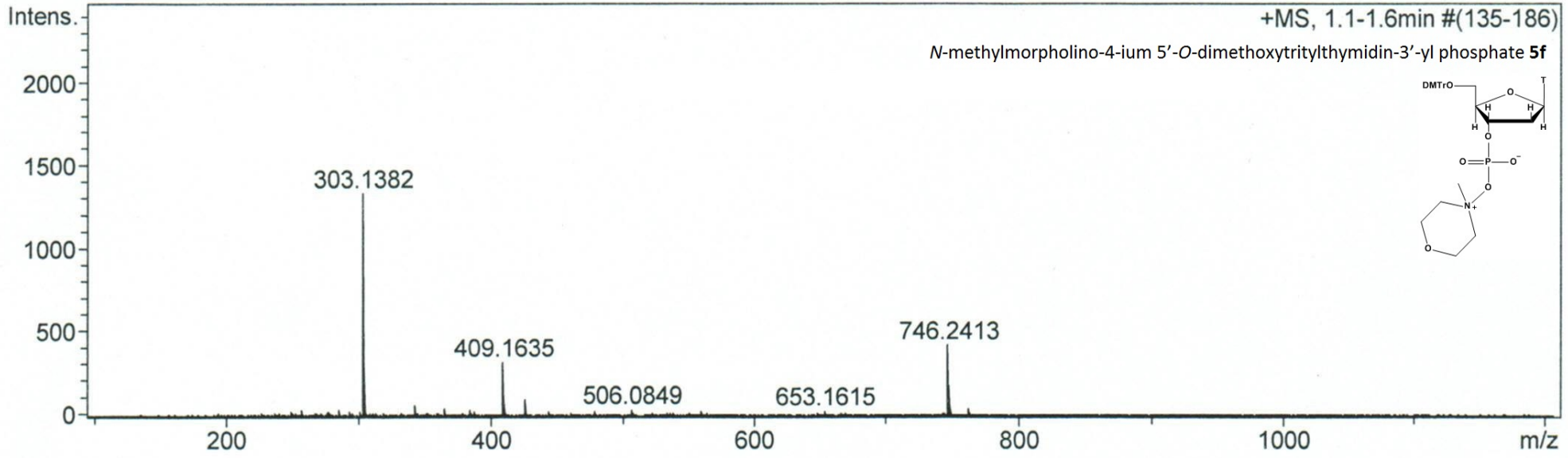
Mass Spectrum SmartFormula Report

Analysis Info

Analysis Name	D:\Data\Basia\zlecone\15_02_23_A6.d	Acquisition Date	2/23/2015 10:31:52 AM
Method	ewelina.m	Operator	Bruker Customer
Sample Name	blank	Instrument / Ser#	micrOTOF-Q 128
Comment			

Acquisition Parameter

Source Type	ESI	Ion Polarity	Positive	Set Nebulizer	0.4 Bar
Focus	Not active	Set Capillary	4500 V	Set Dry Heater	220 °C
Scan Begin	100 m/z	Set End Plate Offset	-500 V	Set Dry Gas	4.0 l/min
Scan End	1200 m/z	Set Collision Cell RF	200.0 Vpp	Set Divert Valve	Source



N-methylmorpholino-4-ium 5'-*O*-dimethoxytritylthymidin-3'-yl phosphate **5f**

Meas. m/z	#	Formula	m/z	err [ppm]	Mean err [ppm]	rdb	N-Rule	e⁻ Conf	mSigma	Std I	Std Mean m/z	Std I VarNorm	Std m/z Diff	Std Comb Dev