Laccase-Catalyzed Green Synthesis and Cytotoxic Activity of Novel Pyrimidobenzothiazoles and Catechol Thioethers

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Electronic Supplementary Information

Contents

1. NMR spectra of compounds 2a-i, 3a-h, 4a-d,g, 5a-k (Figures 1-29)

2,3-Dihydro-6-methyl-2-thioxopyrimidin-4(1*H*)-one (2a)

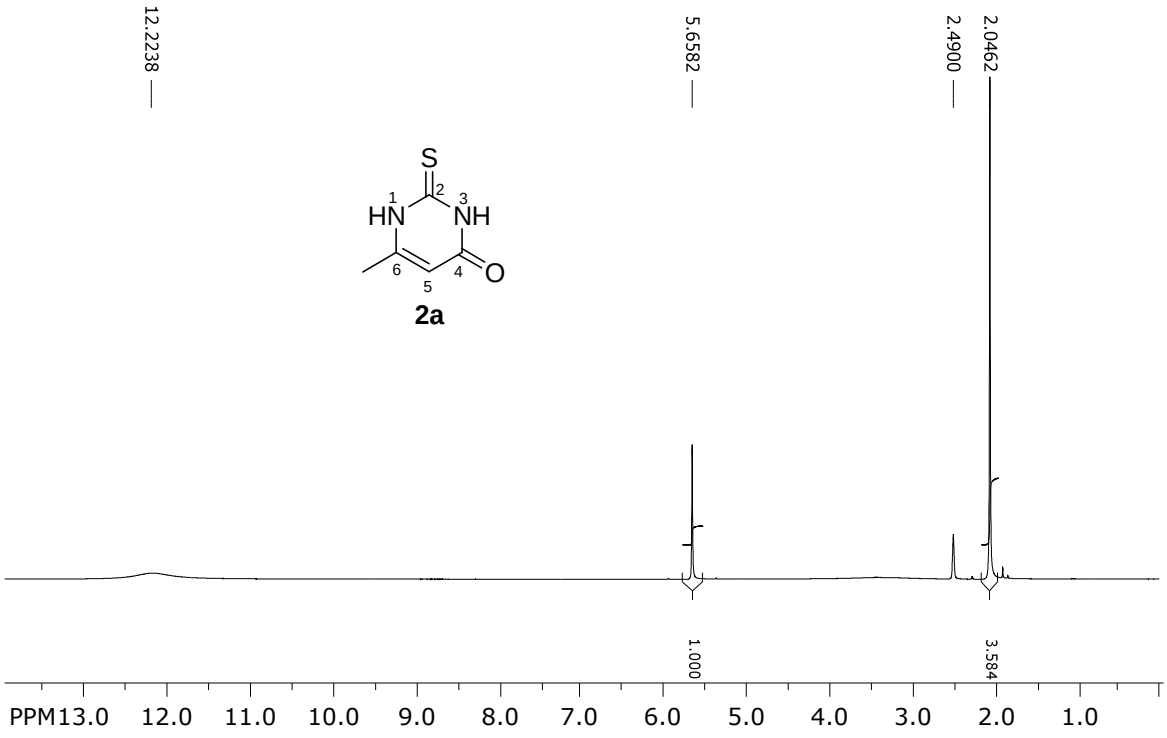


Fig. 1 ^{1}H NMR (300 MHz) spectrum of **2a** in DMSO-d_6

6-Ethyl-2,3-dihydro-2-thioxopyrimidin-4(1*H*)-one (2b)

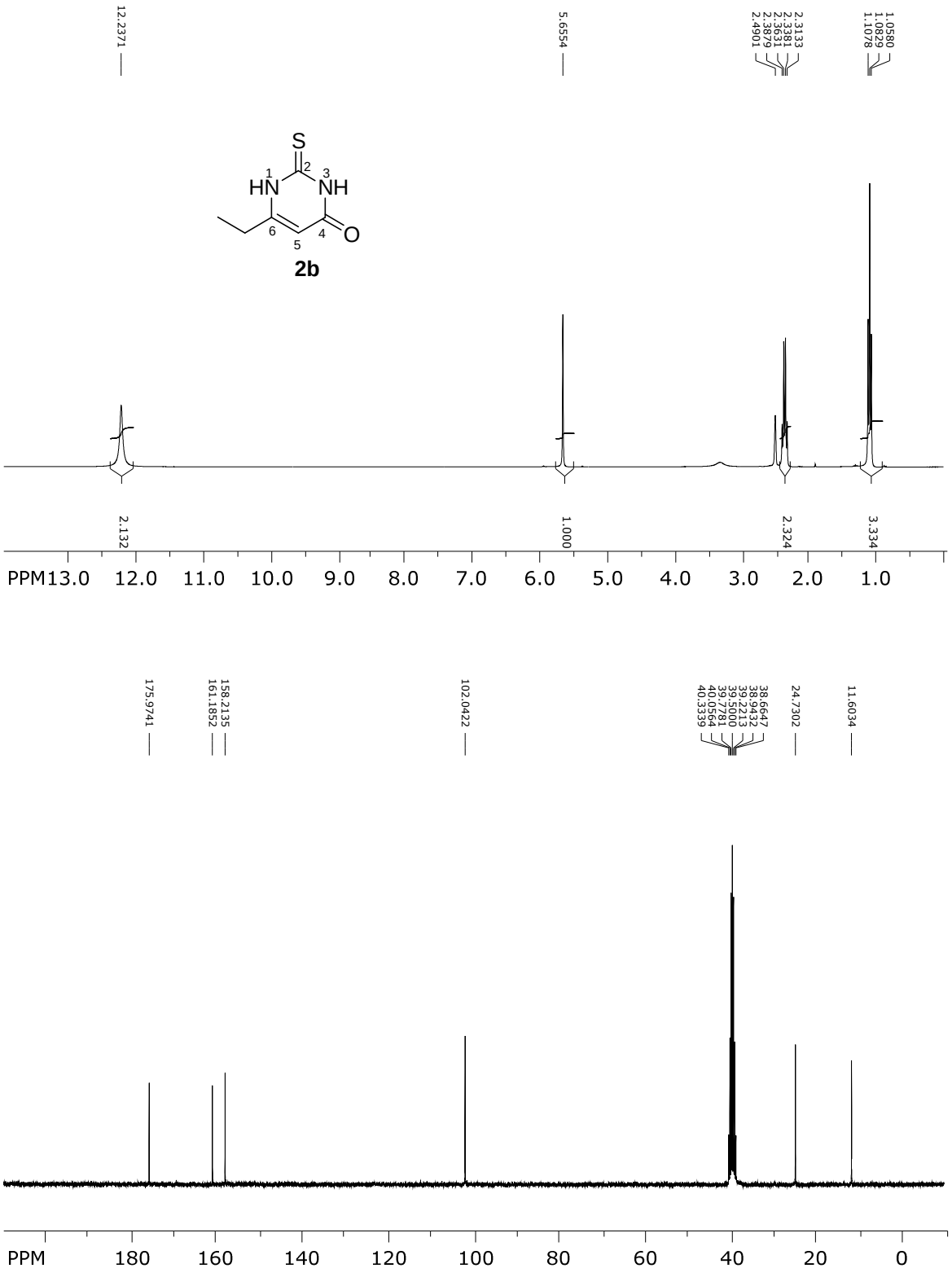


Fig. 2 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **2b** in DMSO-d_6

2,3-Dihydro-6-propyl-2-thioxopyrimidin-4(1*H*)-one (2c)

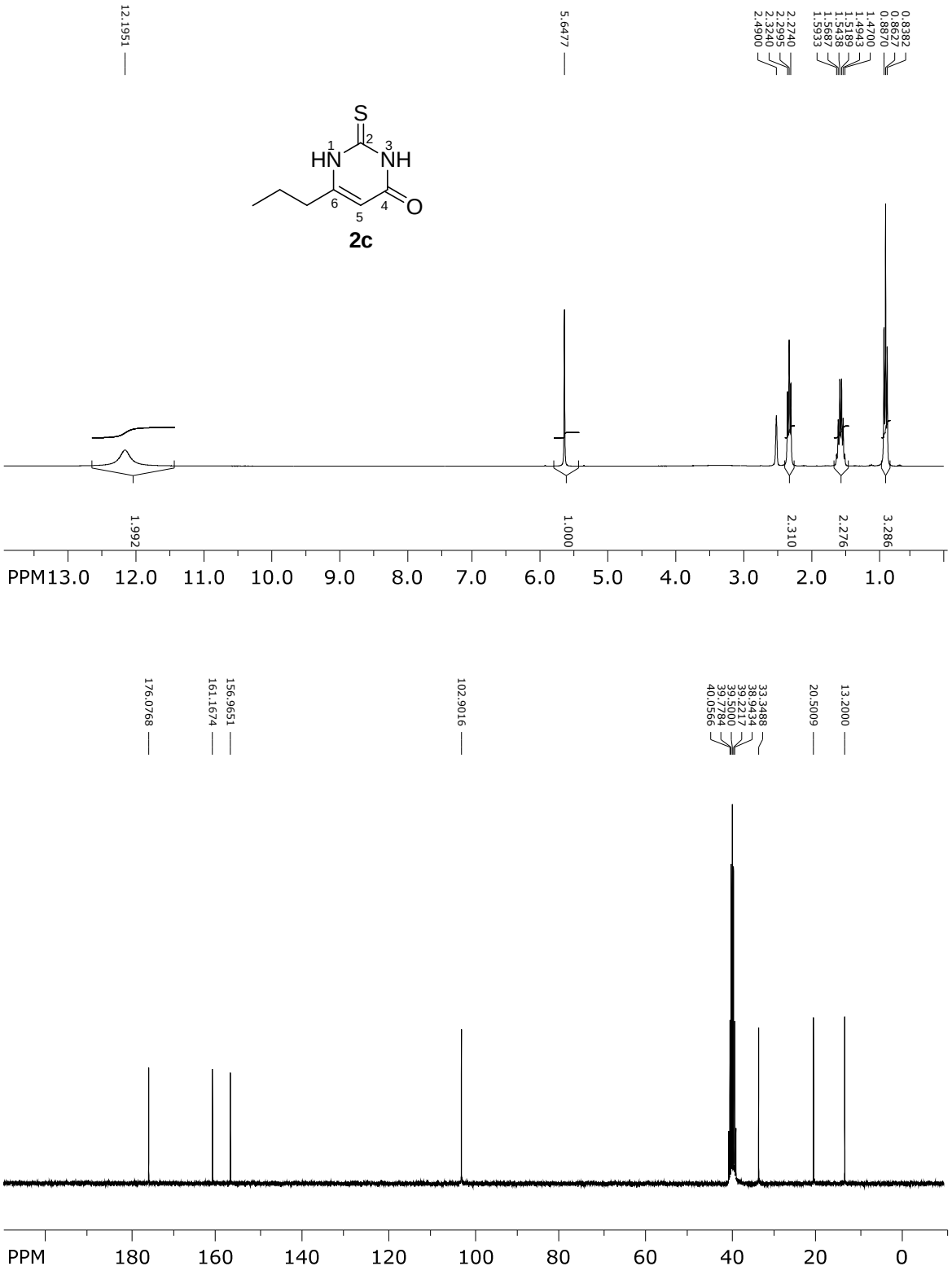


Fig. 3 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **2c** in DMSO-d_6

2,3-Dihydro-6-isopropyl-2-thioxopyrimidin-4(1*H*)-one (2d)

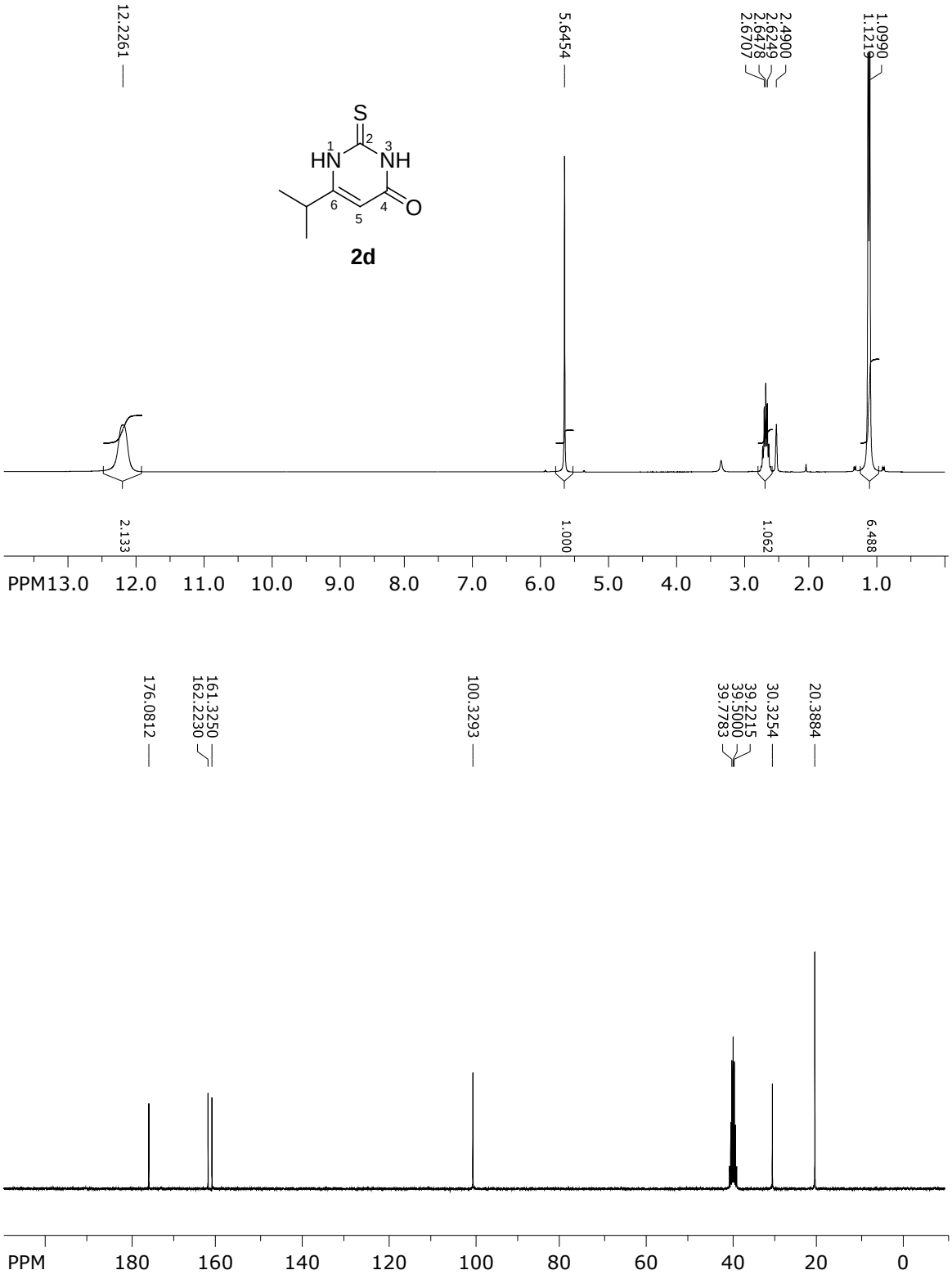


Fig. 4 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **2d** in DMSO-d_6

6-*tert*-Butyl-2,3-dihydro-2-thioxopyrimidin-4(1*H*)-one (2e)

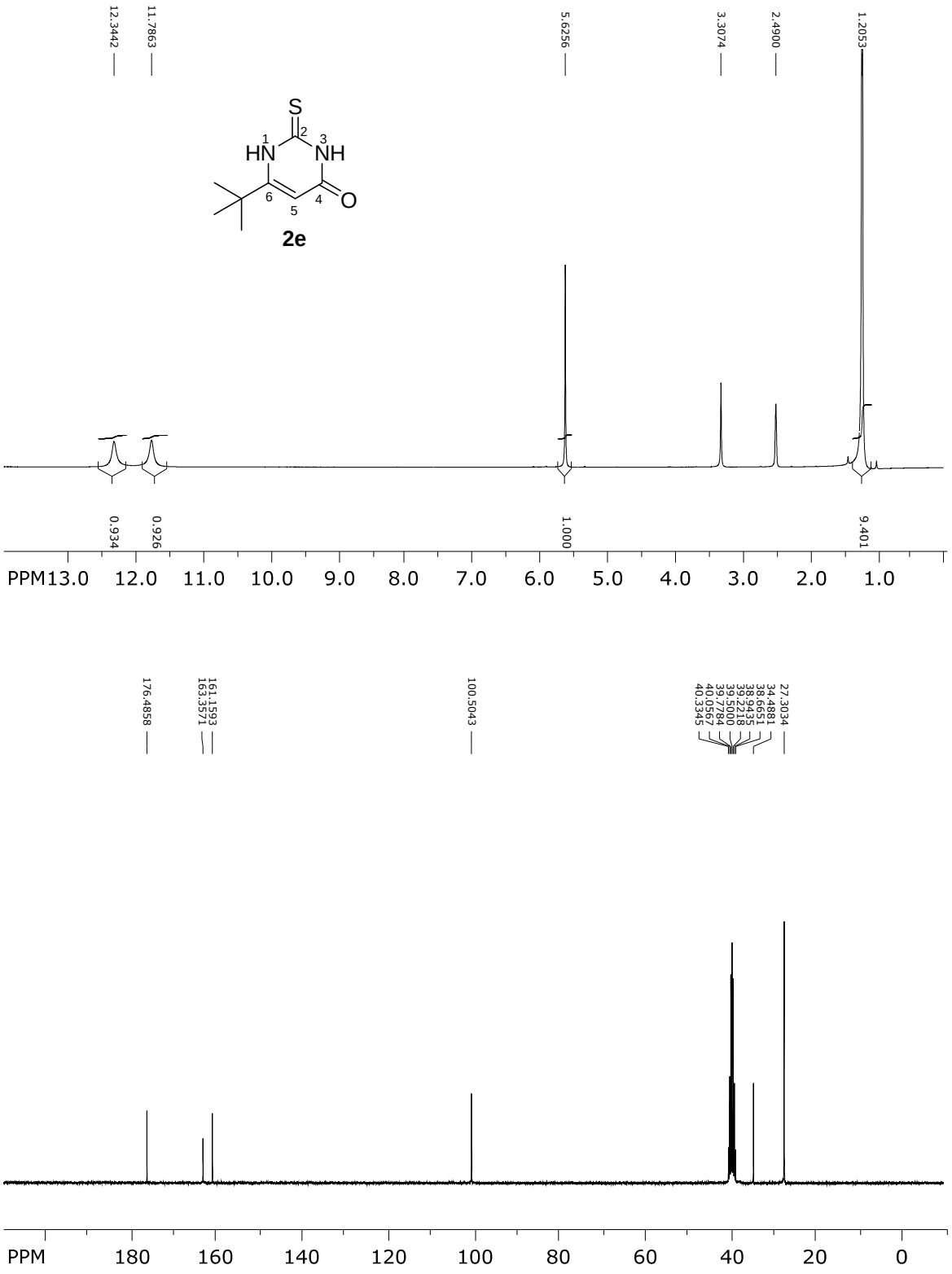


Fig. 5 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **2e** in DMSO-d_6

6-Trifluoromethyl-2,3-dihydro-2-thioxopyrimidin-4(1*H*)-one (2f)

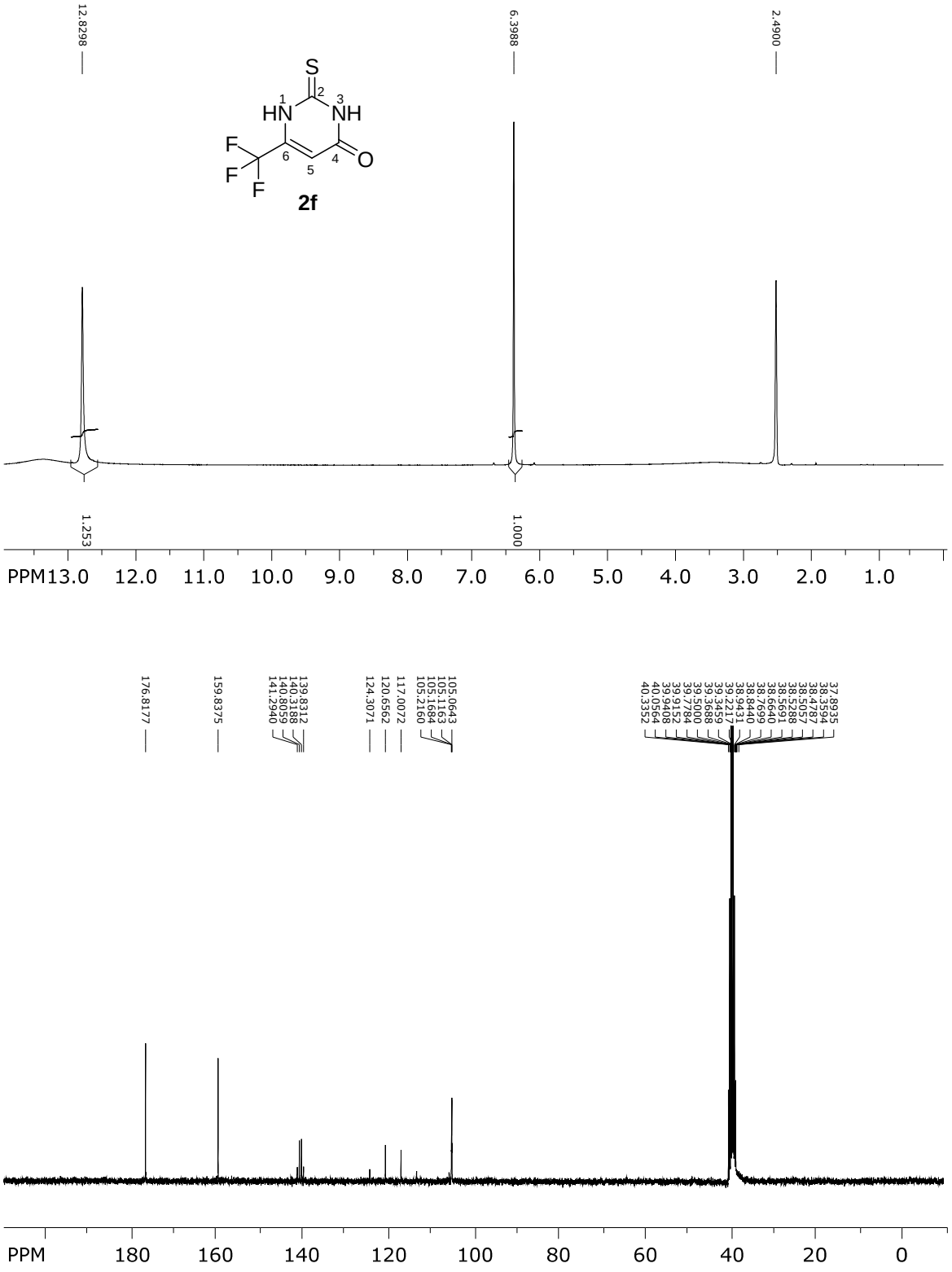


Fig. 6 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **2f** in DMSO-d_6

2,3-Dihydro-5,6-dimethyl-2-thioxopyrimidin-4(1*H*)-one (2g)

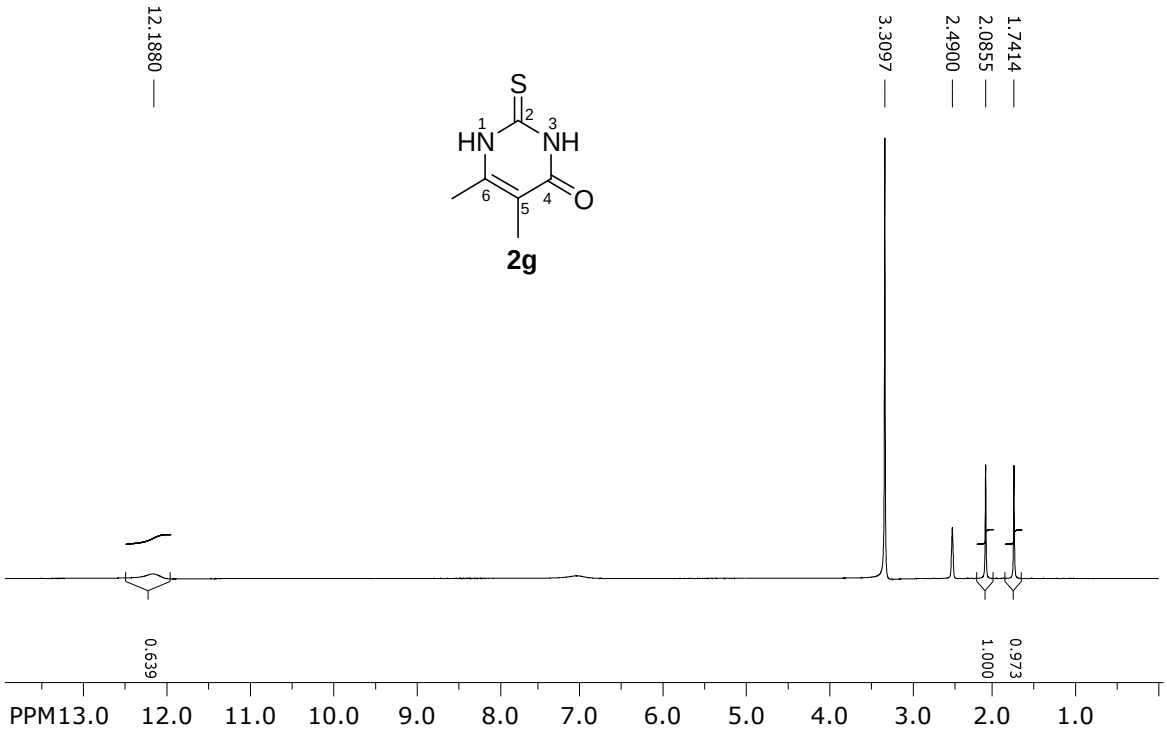


Fig. 7 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **2g** in DMSO-d_6

6-Trifluoromethyl-2,3-dihydro-5-methyl-2-thioxopyrimidin-4(1*H*)-one (2h)

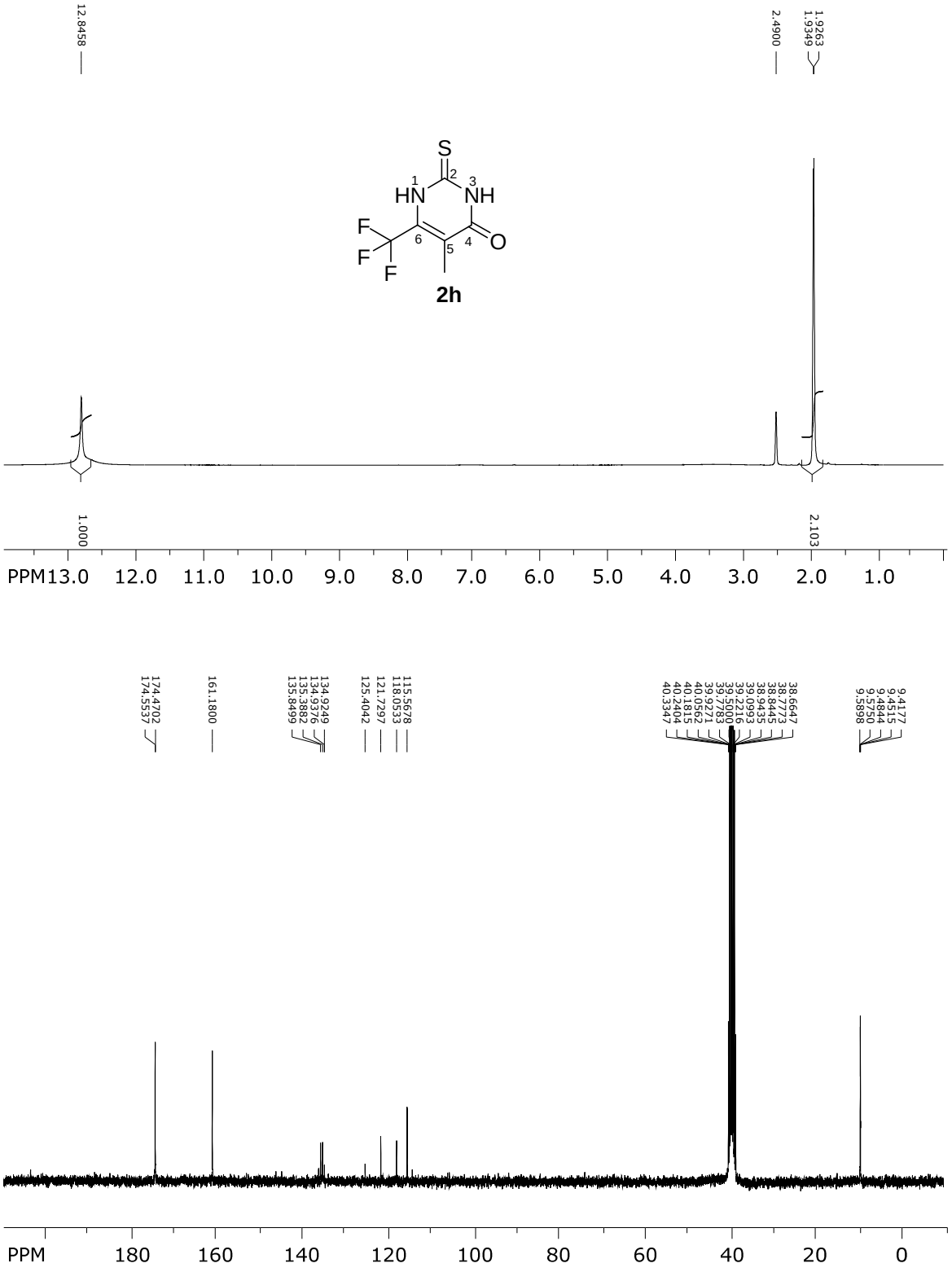


Fig. 8 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **2h** in DMSO-d_6

2,3,6,7-Tetrahydro-2-thioxo-1*H*-cyclopenta[*d*]pyrimidin-4(5*H*)-one (2i)

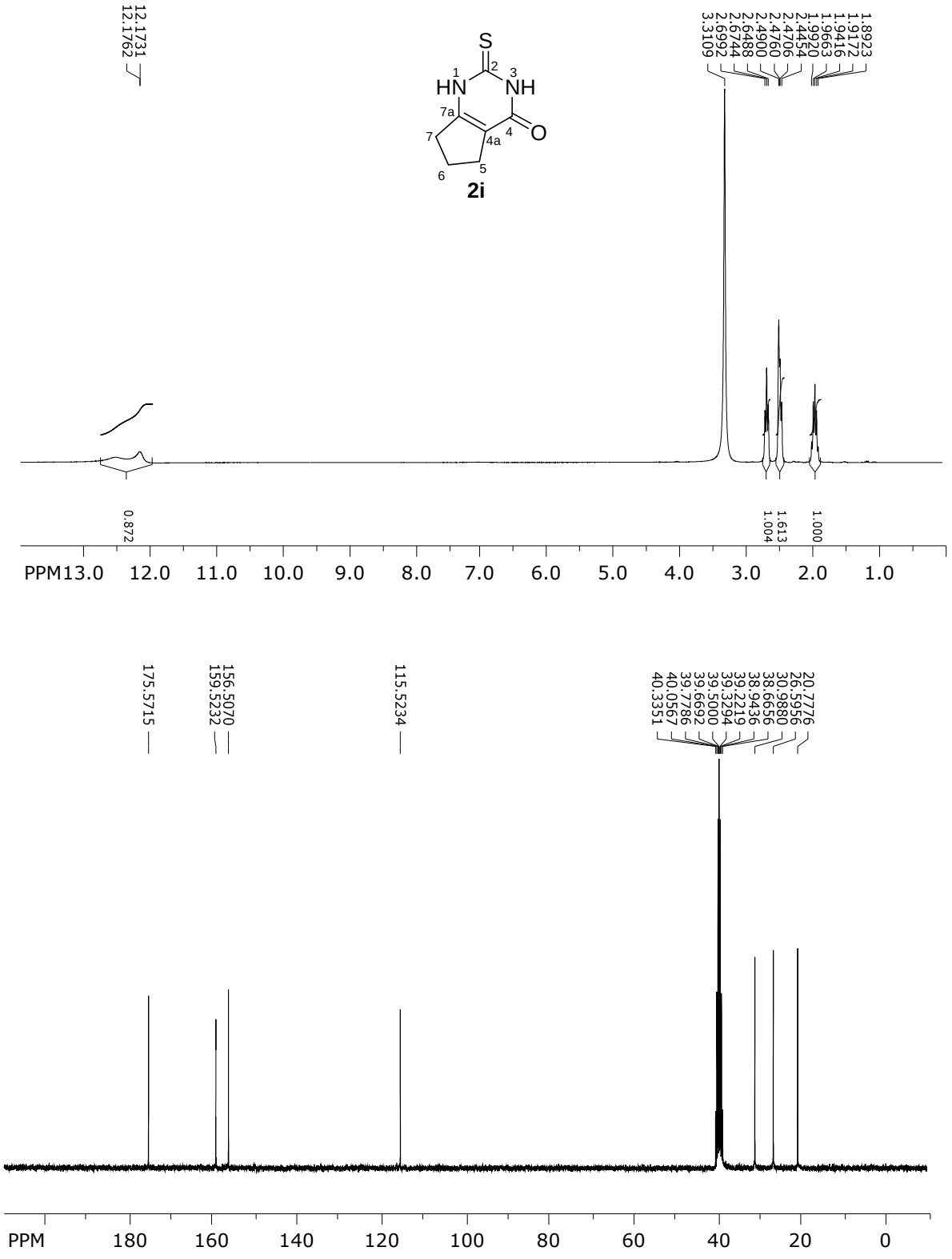


Fig. 9 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **2i** in DMSO-d_6

7,8-Dihydroxy-4*H*-2-methyl-pyrimido[2,1-*b*]benzothiazol-4-one (3a) and 7,8-dihydroxy-2*H*-4-methyl-pyrimido[2,1-*b*]benzothiazol-2-one (4a)

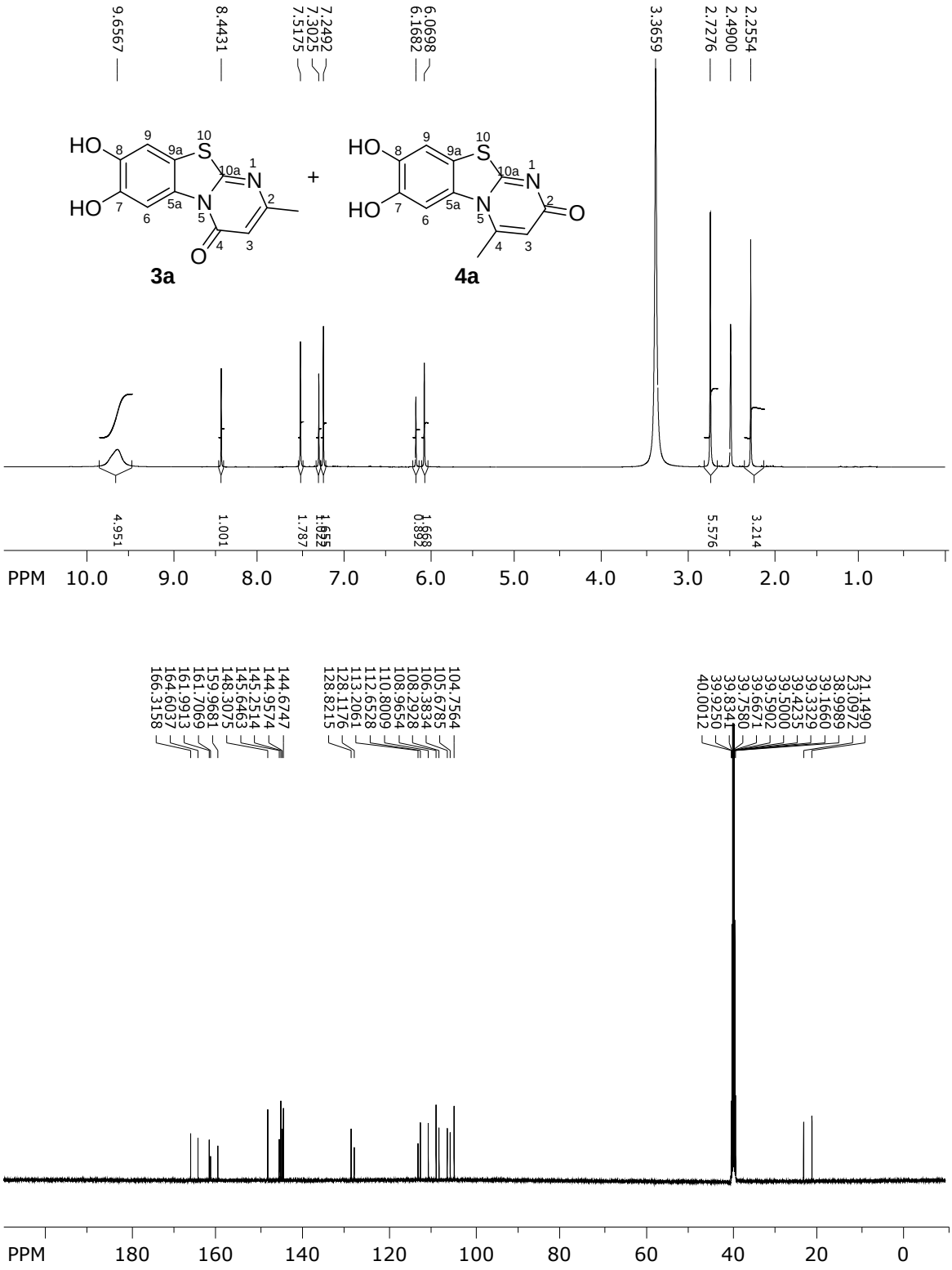


Fig. 10 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **3a** and **4a** in DMSO-d_6

7,8-Dihydroxy-4*H*-2-ethyl-pyrimido[2,1-*b*]benzothiazol-4-one (3b) and 7,8-dihydroxy-2*H*-4-ethyl-pyrimido[2,1-*b*]benzothiazol-2-one (4b).

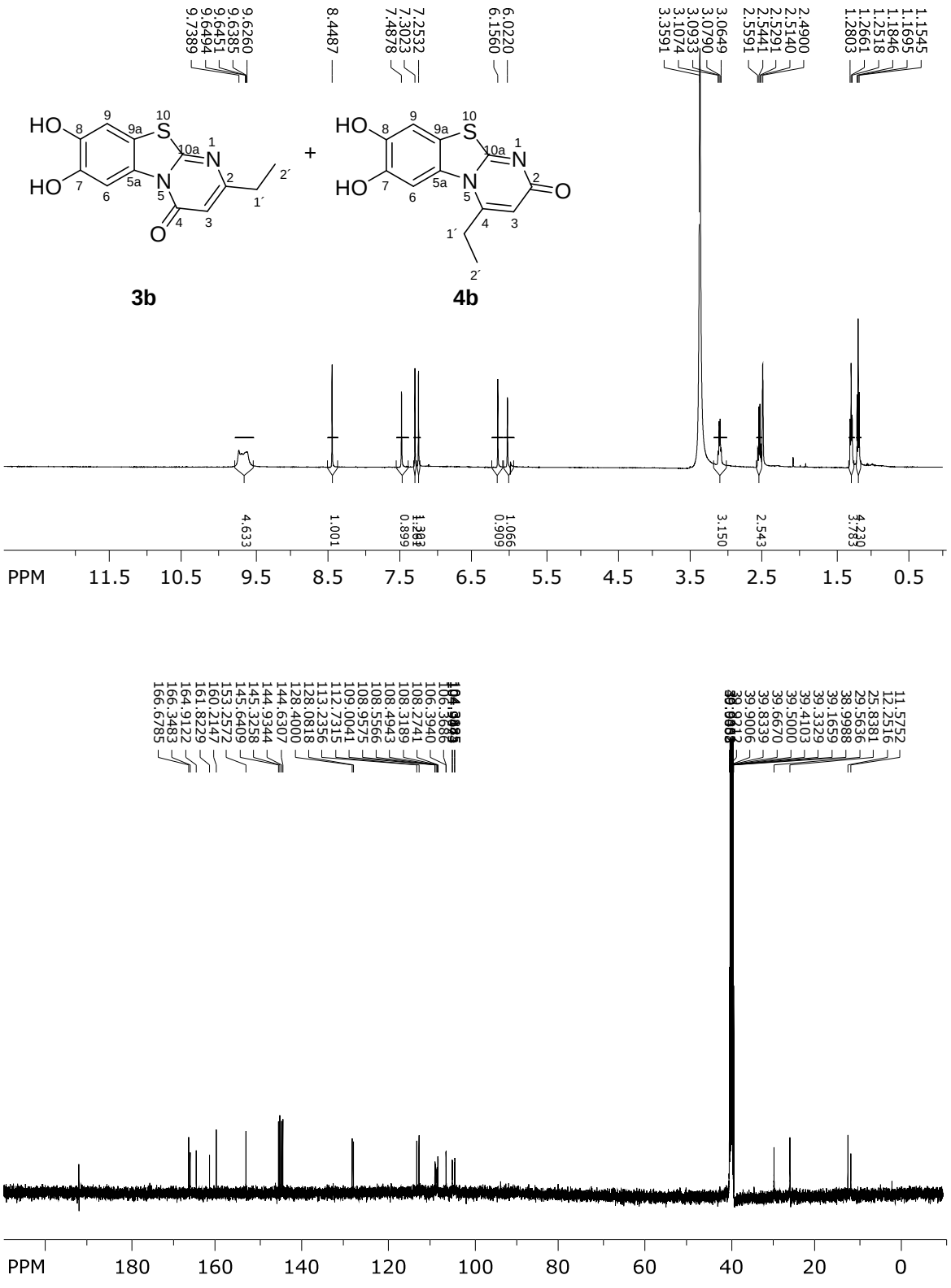


Fig. 11 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **3b** and **4b** in DMSO-d_6

7,8-Dihydroxy-4*H*-2-propyl-pyrimido[2,1-*b*]benzothiazol-4-one (3c) and 7,8-dihydroxy-2*H*-4-propyl-pyrimido[2,1-*b*]benzothiazol-2-one (4c)

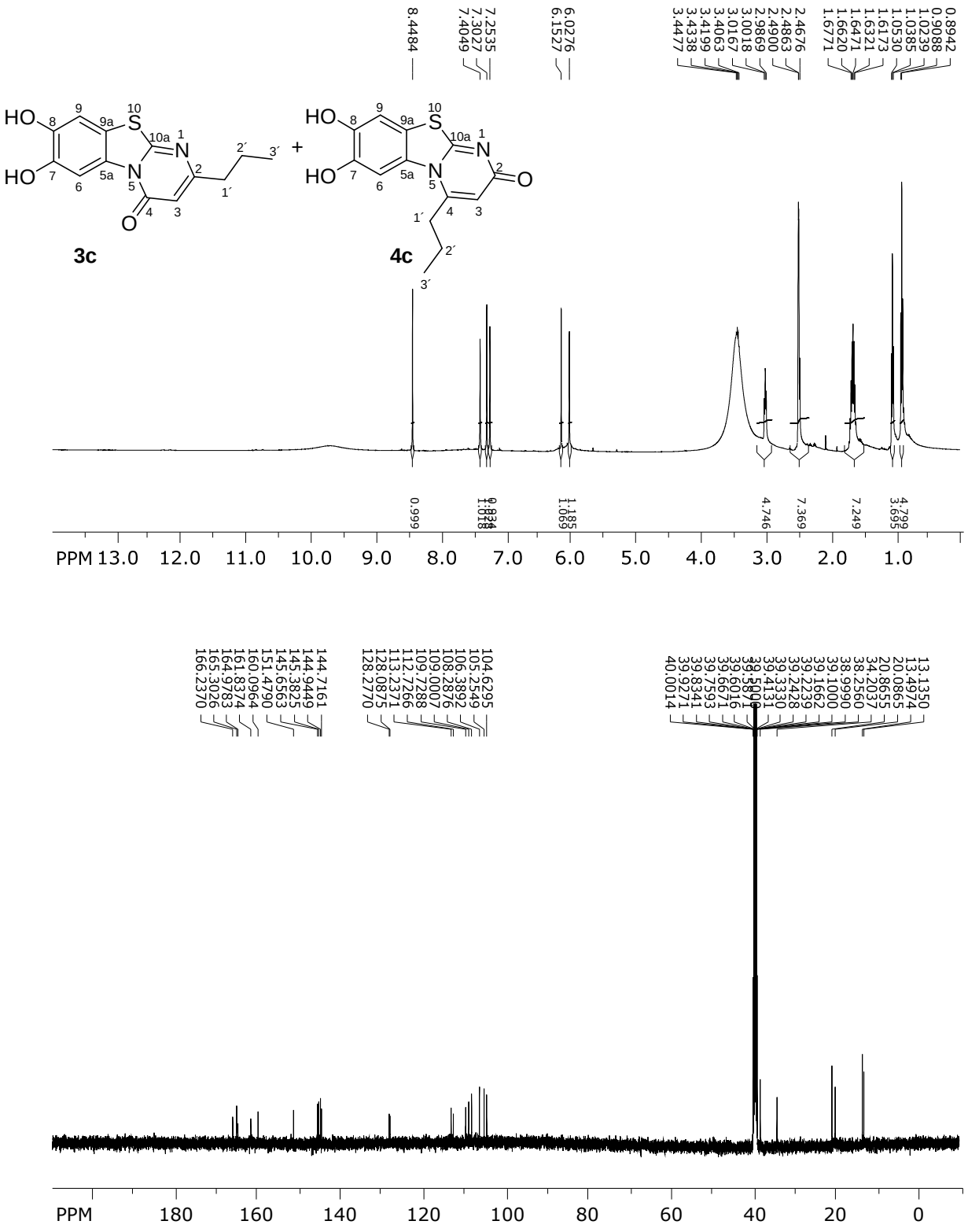


Fig. 12 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **3c** and **4c** in DMSO-d_6

7,8-Dihydroxy-4*H*-2-isopropyl-pyrimido[2,1-*b*]benzothiazol-4-one (3d) and 7,8-dihydroxy-2*H*-4-isopropyl-pyrimido[2,1-*b*]benzothiazol-2-one (4d)

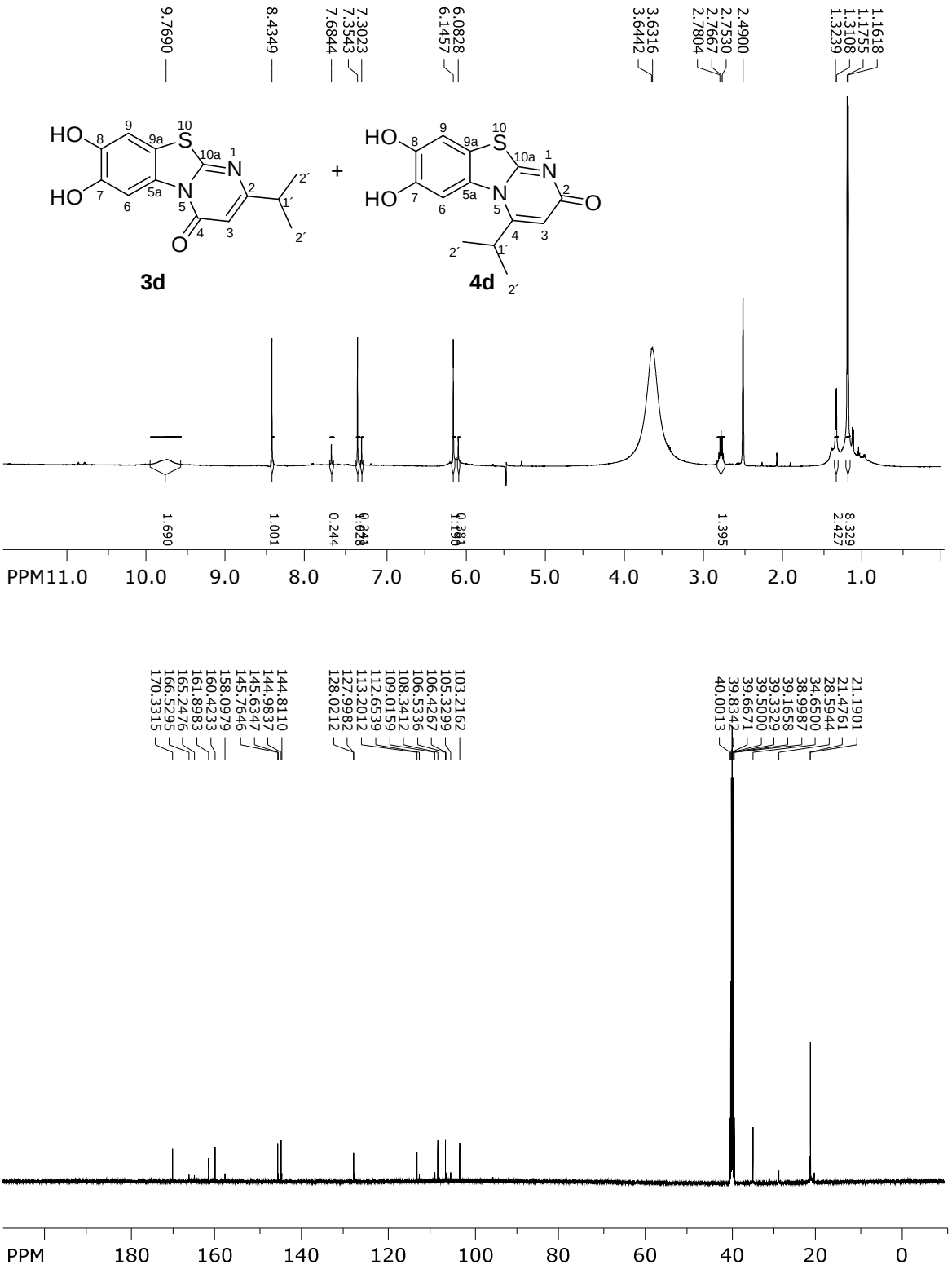


Fig. 13 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **3d** and **4d** in DMSO-d_6

7,8-Dihydroxy-4*H*-2-isopropyl-pyrimido[2,1-*b*]benzothiazol-4-one (3d)

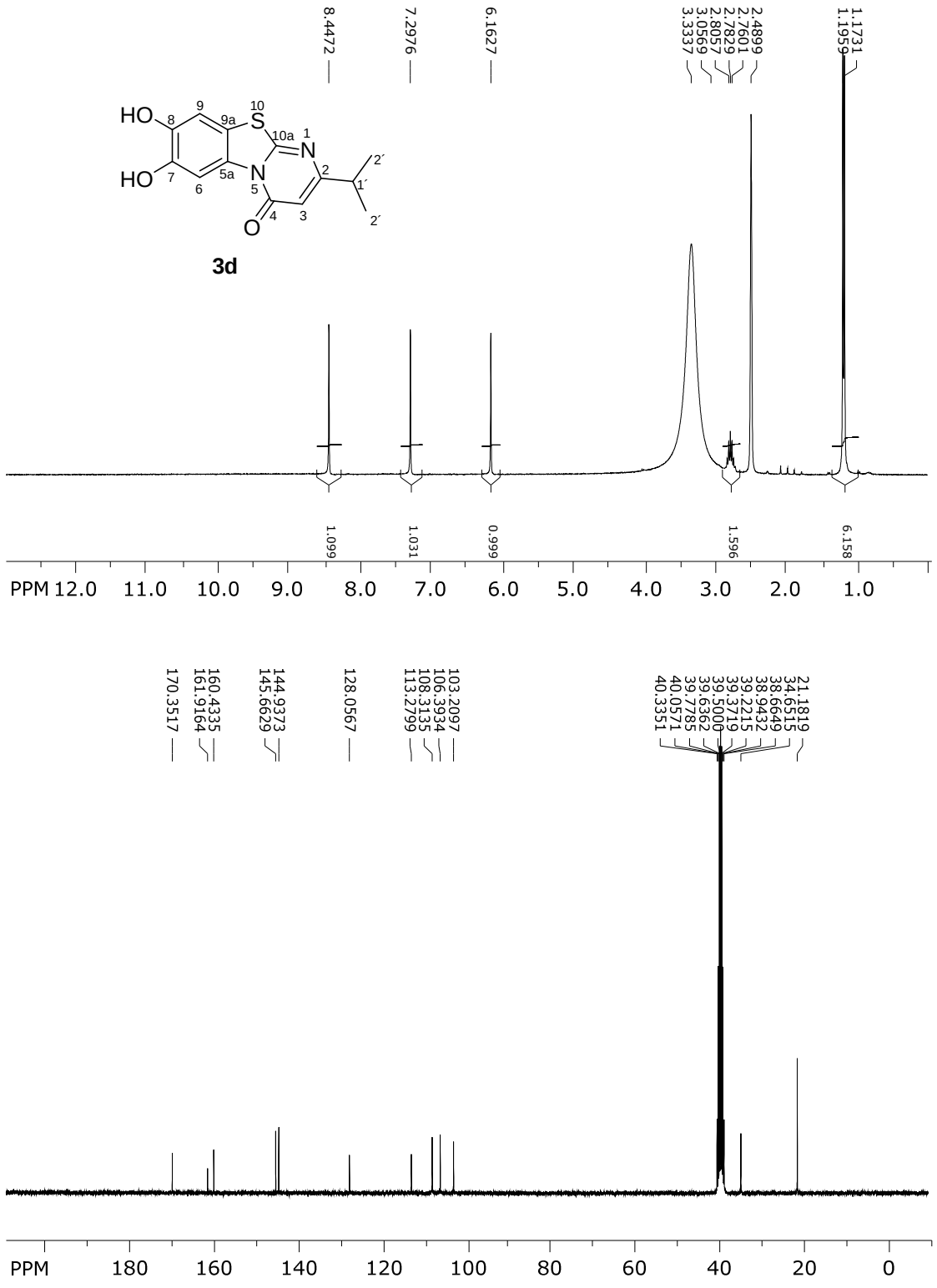


Fig. 14 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **3d** in DMSO-d_6

7,8-Dihydroxy-4*H*-2-*tert.*butyl-pyrimido[2,1-*b*]benzothiazol-4-one (3e)

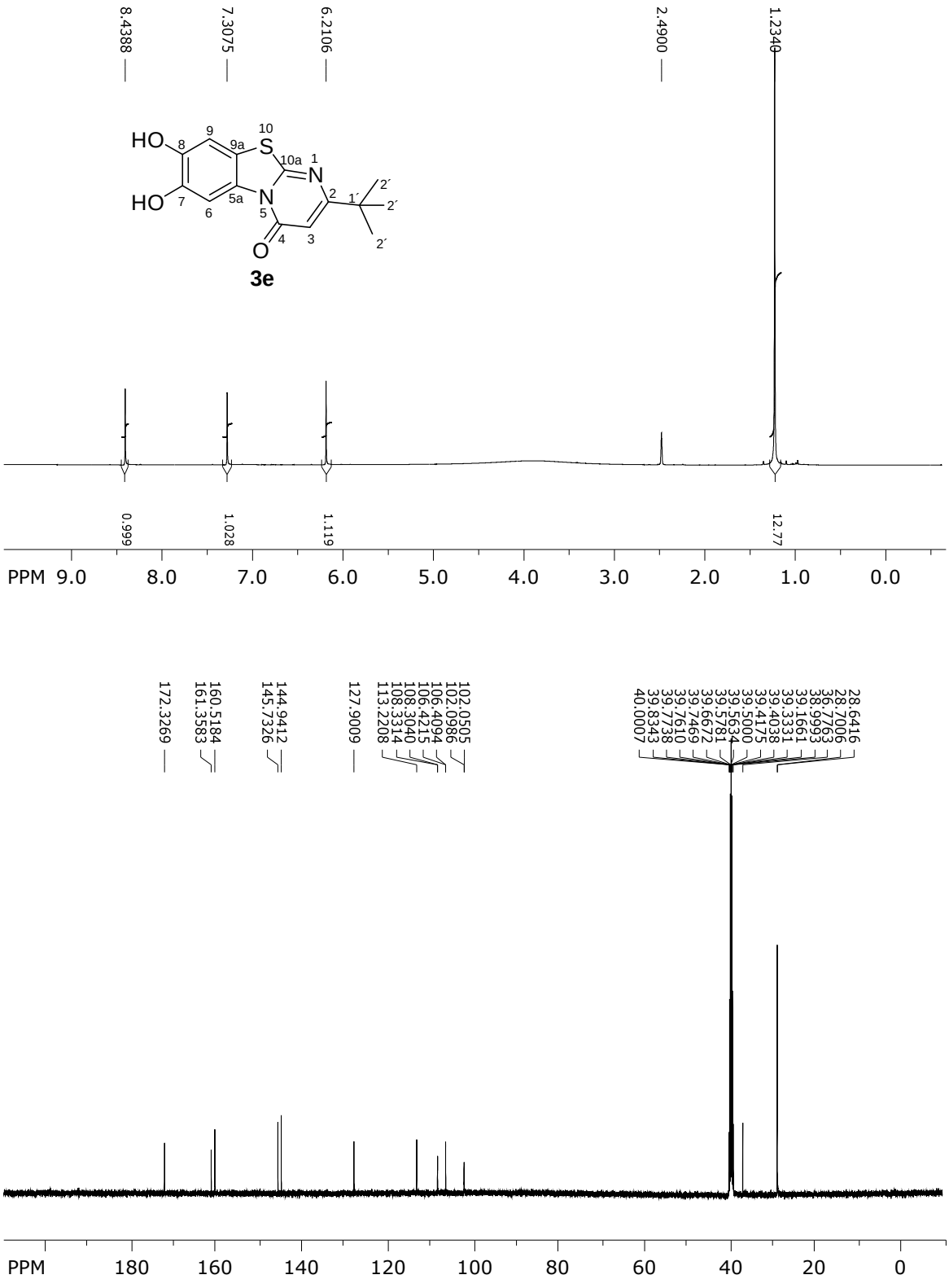


Fig. 15 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **3e** in DMSO-d_6

7,8-Dihydroxy-4*H*-2-trifluoromethyl-pyrimido[2,1-*b*]benzothiazol-4-one (3f)

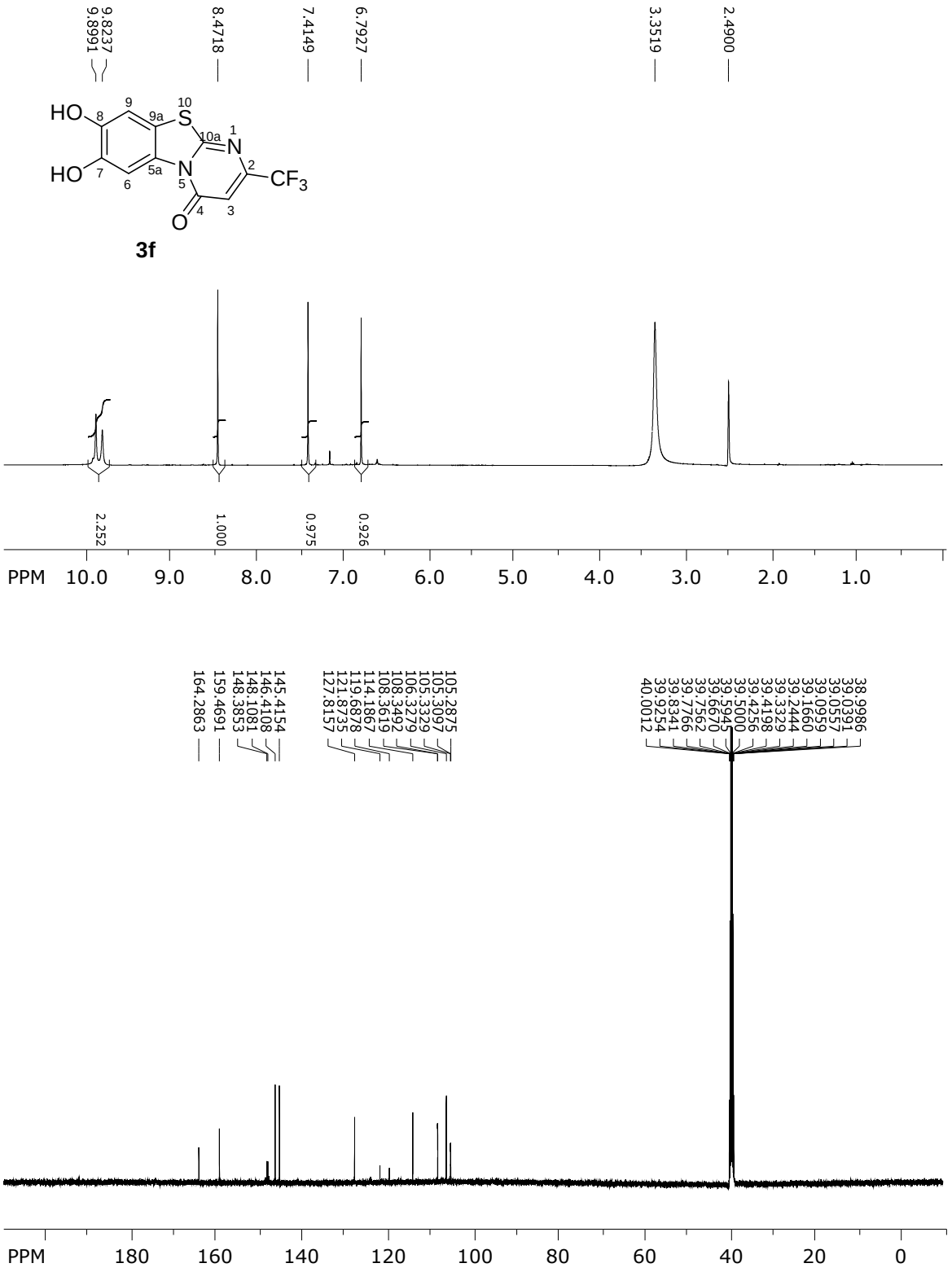


Fig. 16 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **3f** in DMSO-d_6

7,8-Dihydroxy-4*H*-2,3-dimethyl-pyrimido[2,1-*b*]benzothiazol-4-one (3g) and 7,8-dihydroxy-2*H*-3,4-dimethyl-pyrimido[2,1-*b*]benzothiazol-2-one (4g)

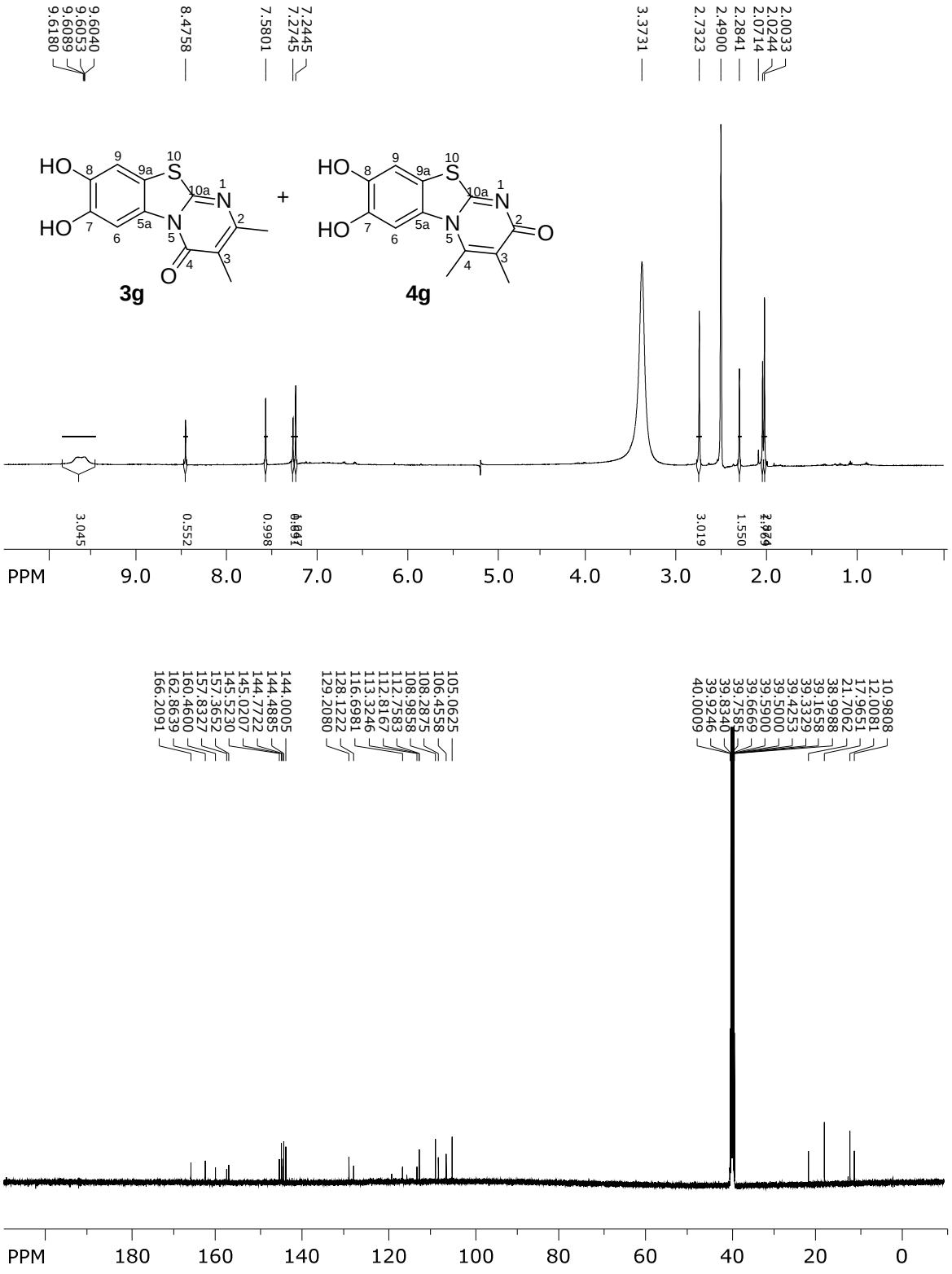


Fig. 17 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **3g** and **4g** in DMSO-d_6

7,8-Dihydroxy-4*H*-3-methyl-2-trifluoromethyl-pyrimido[2,1-*b*]benzothiazol-4-one (3h)

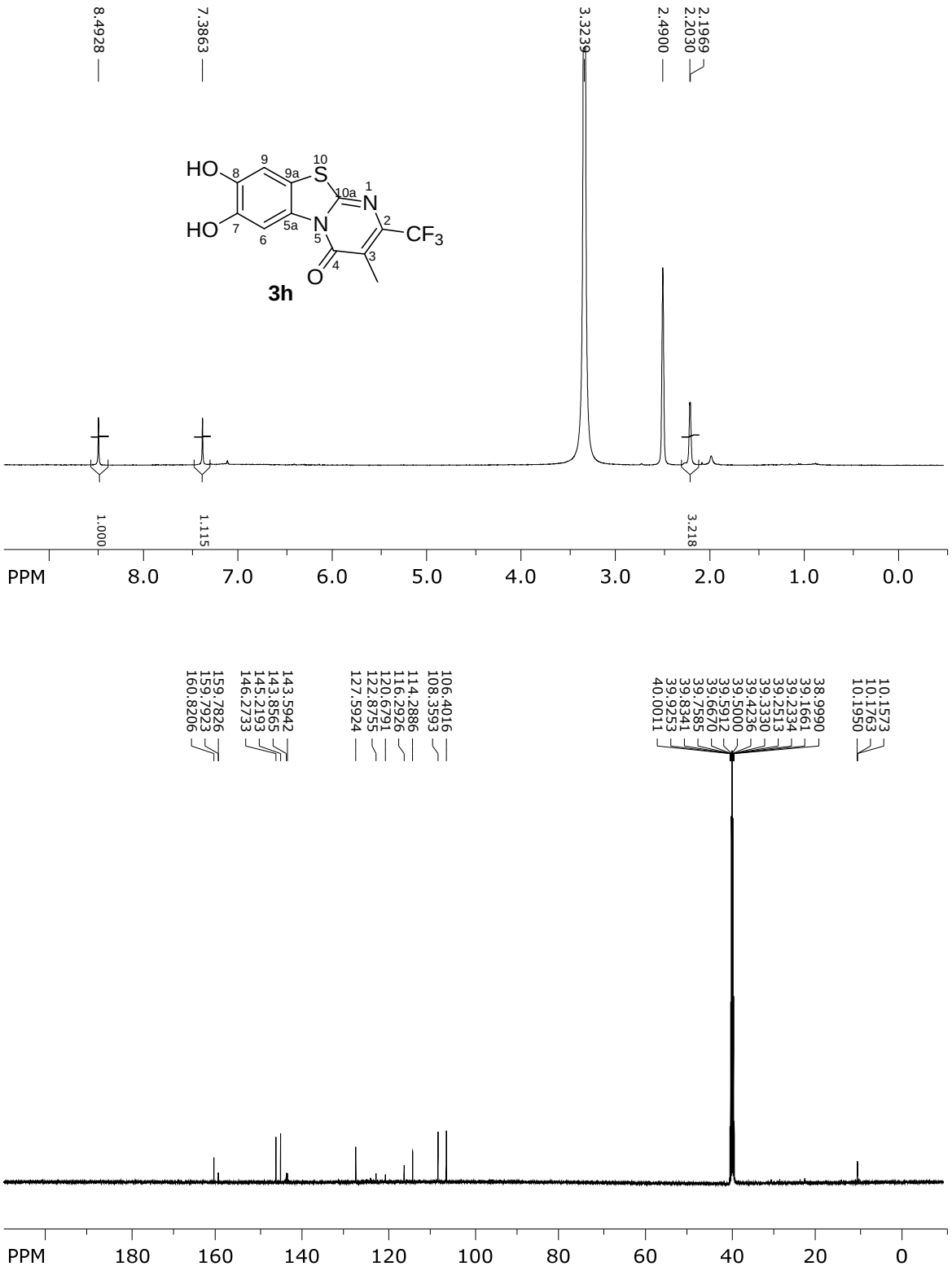


Fig. 18 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **3h** in DMSO-d_6

2-(4,5-Dihydroxy-2-methylphenylthio)-6-methylpyrimidin-4(3*H*)-one (5a)

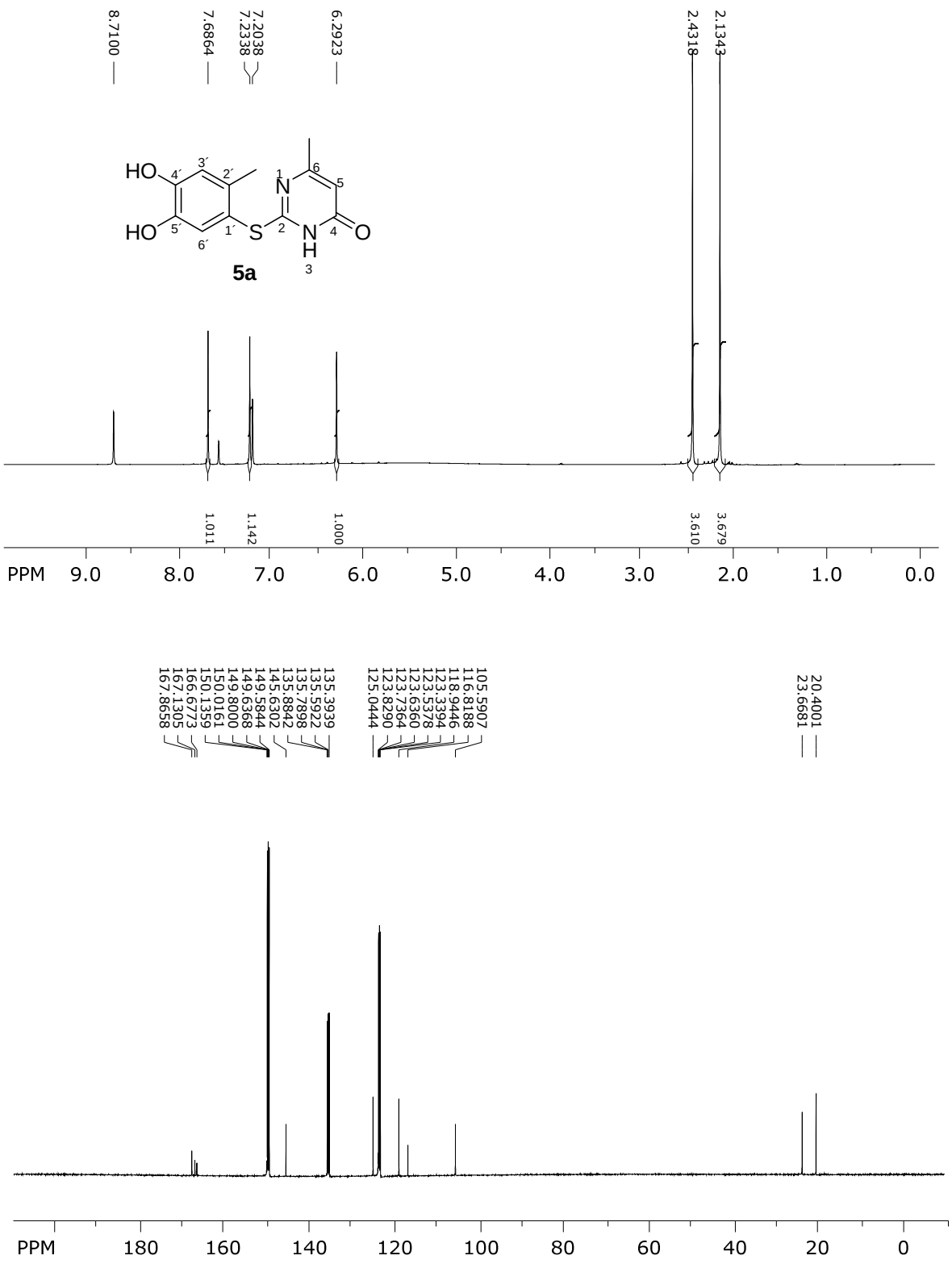


Fig. 19 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **5a** in pyridine-d_5

2-(4,5-Dihydroxy-2-methylphenylthio)-6-propylpyrimidin-4(3*H*)-one (5b)

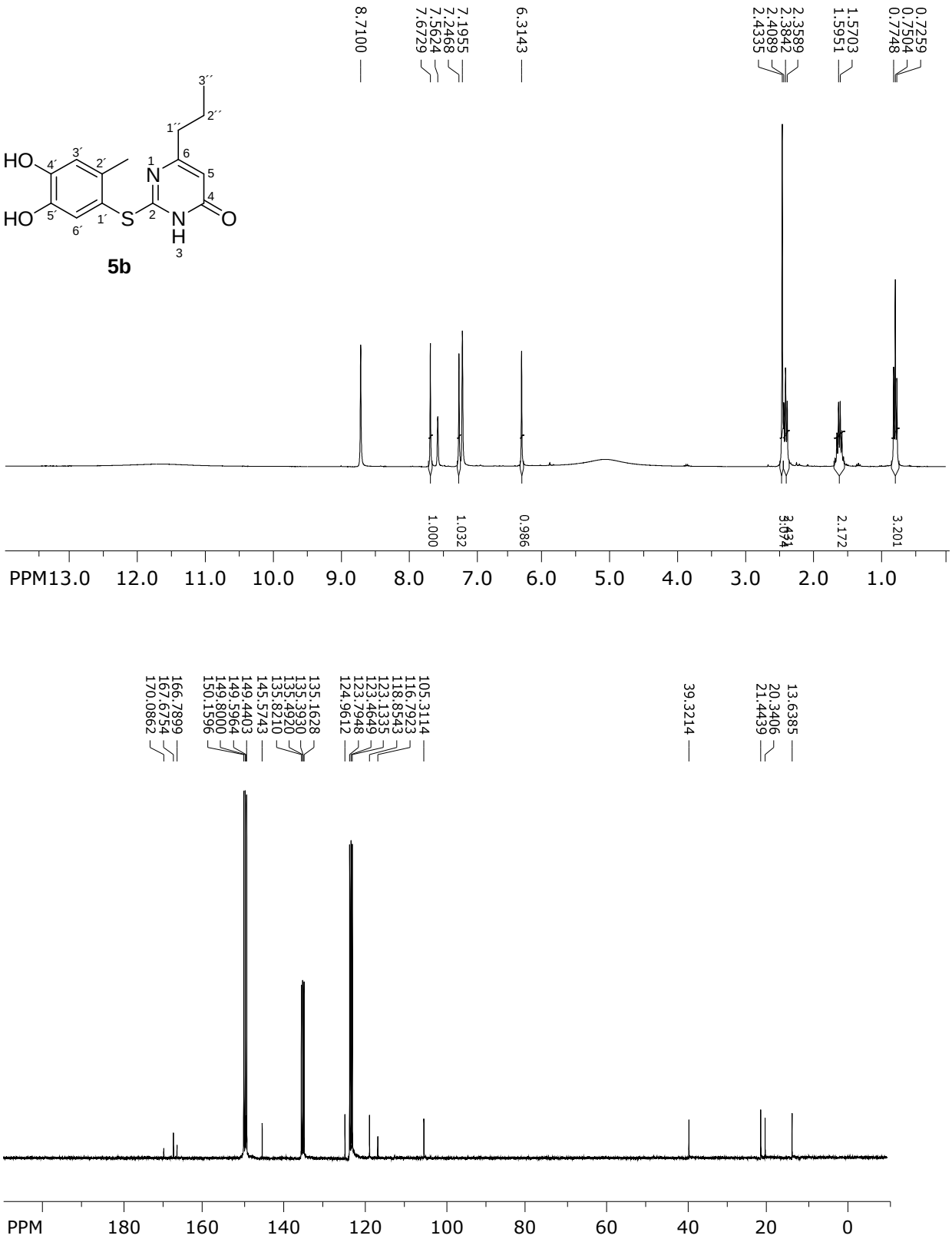


Fig. 20 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **5b** in pyridine-d_5

2-(4,5-Dihydroxy-2-methylphenylthio)-6-isopropylpyrimidin-4(3*H*)-one (5c)

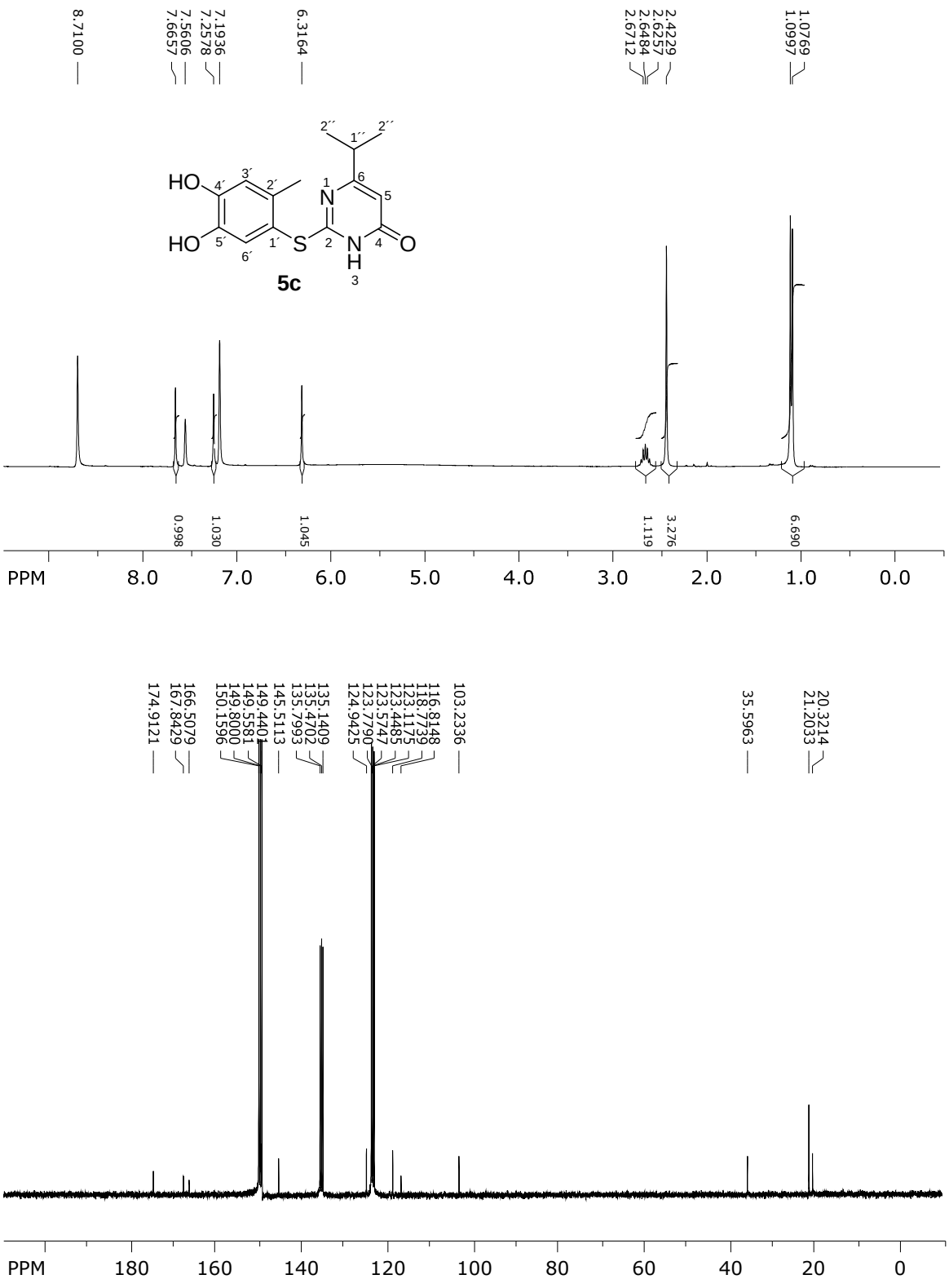


Fig. 21 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **5c** in pyridine-d_5

2-(4,5-Dihydroxy-2-methylphenylthio)-6-*tert*-butylpyrimidin-4(3*H*)-one (5d)

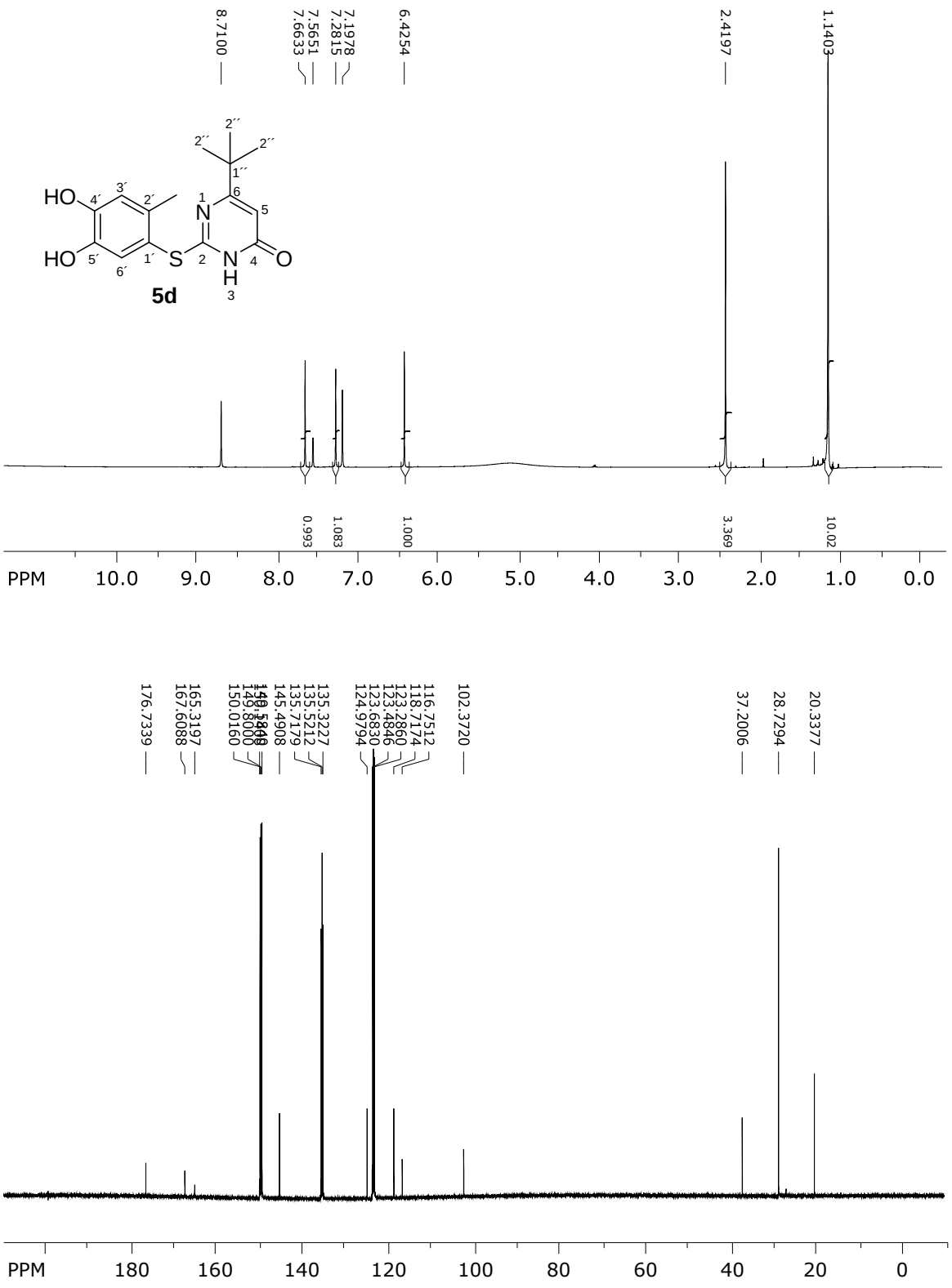


Fig. 22 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **5d** in pyridine-d_5

2-(4,5-Dihydroxy-2-methylphenylthio)-5,6-dimethylpyrimidin-4(3*H*)-one (5e)

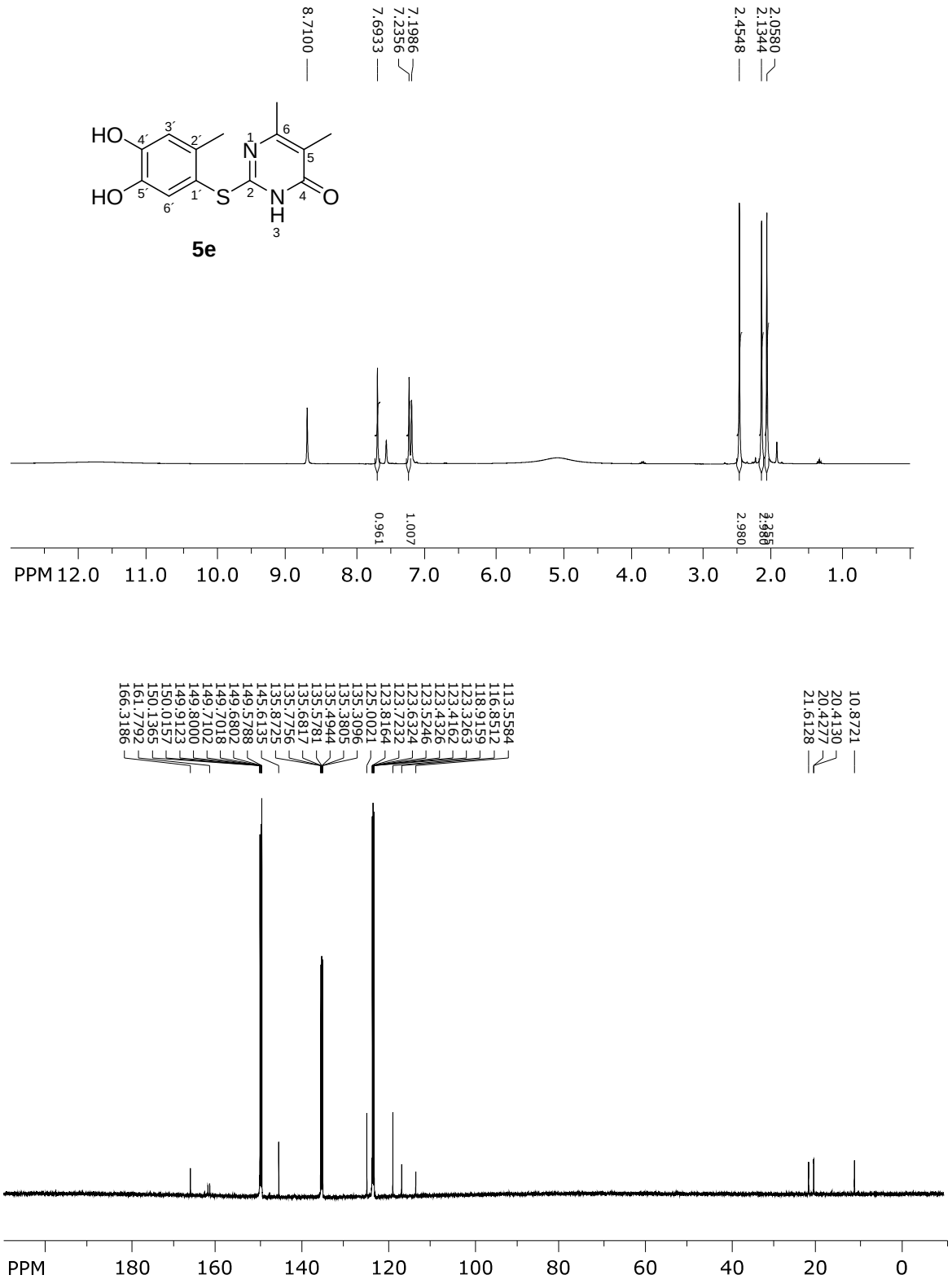


Fig. 23 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **5e** in pyridine-d_5

2-(2-Ethyl-4,5-dihydroxyphenylthio)-6-methylpyrimidin-4(3*H*)-one (5f)

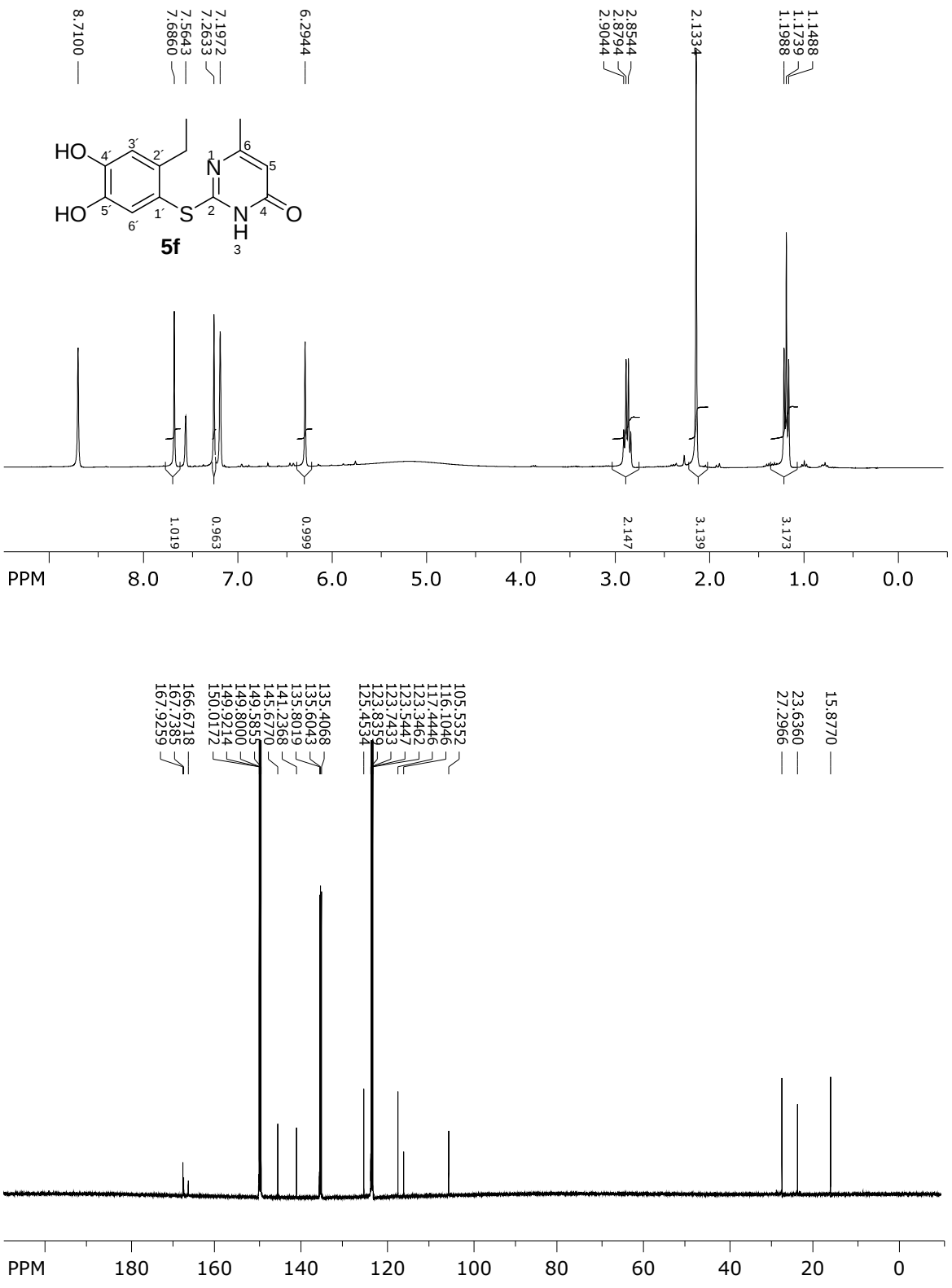


Fig. 24 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **5f** in pyridine-d_5

2-(2-Ethyl-4,5-dihydroxyphenylthio)-6-isopropylpyrimidin-4(3*H*)-one (5g)

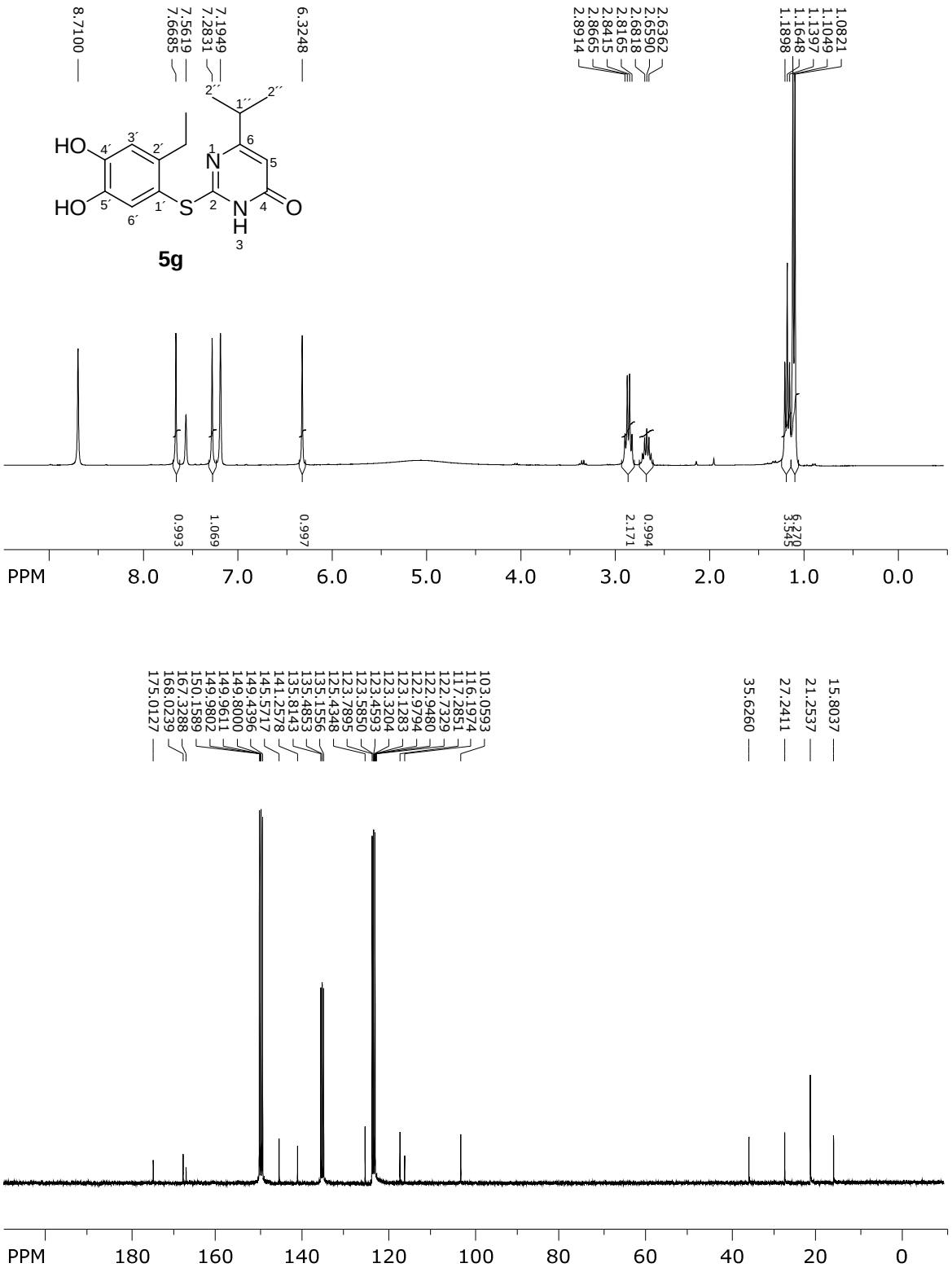


Fig. 25 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **5g** in pyridine-d_5

2-(2-Ethyl-4,5-dihydroxyphenylthio)-6-*tert*-butylpyrimidin-4(3*H*)-one (5h)

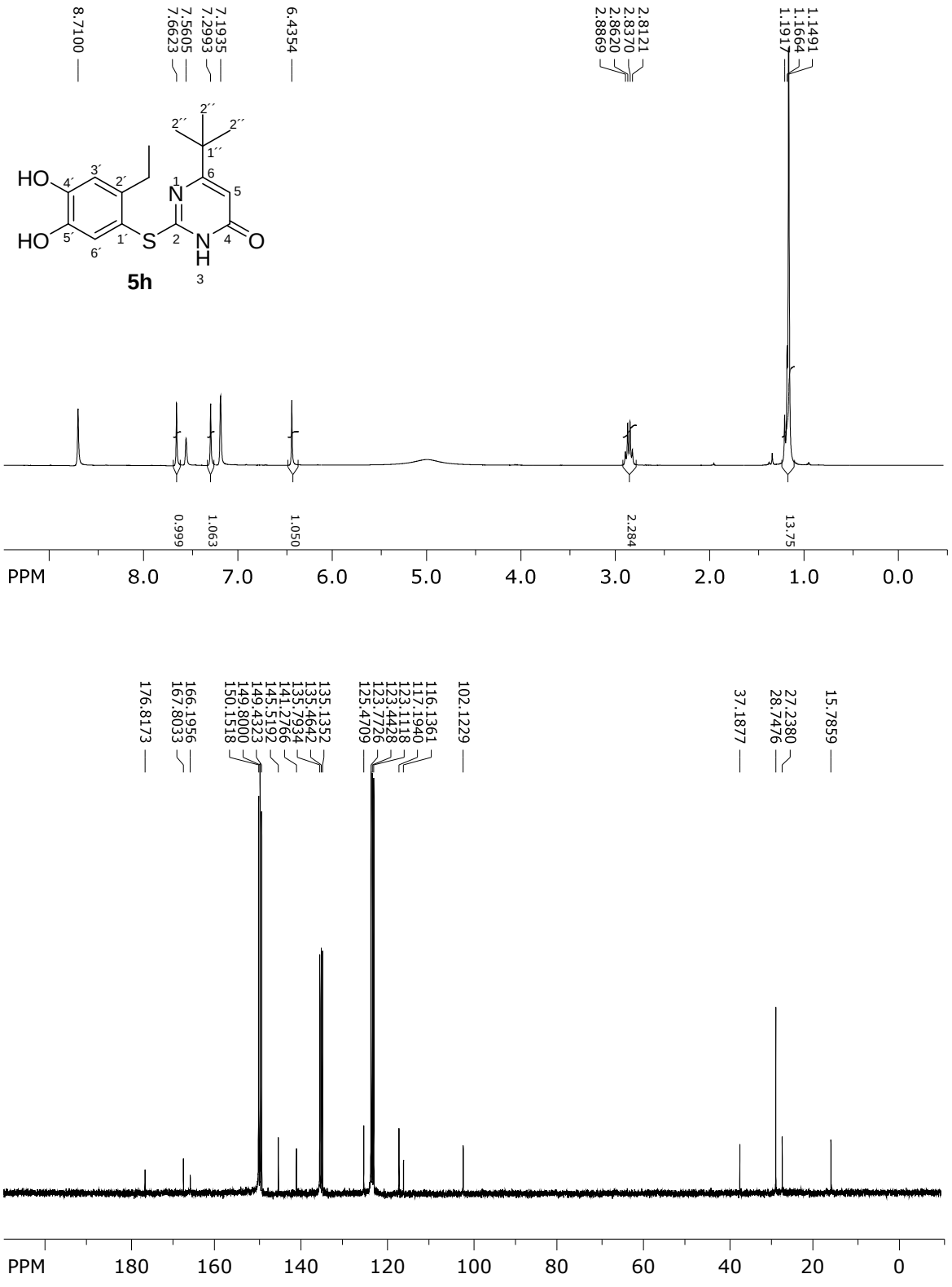


Fig. 26 ^{1}H (300 MHz) and ^{13}C (75 MHz) NMR spectra of **5h** in pyridine-d_5

2-(2-Ethyl-4,5-dihydroxyphenylthio)-5,6-dimethylpyrimidin-4(3*H*)-one (5i)

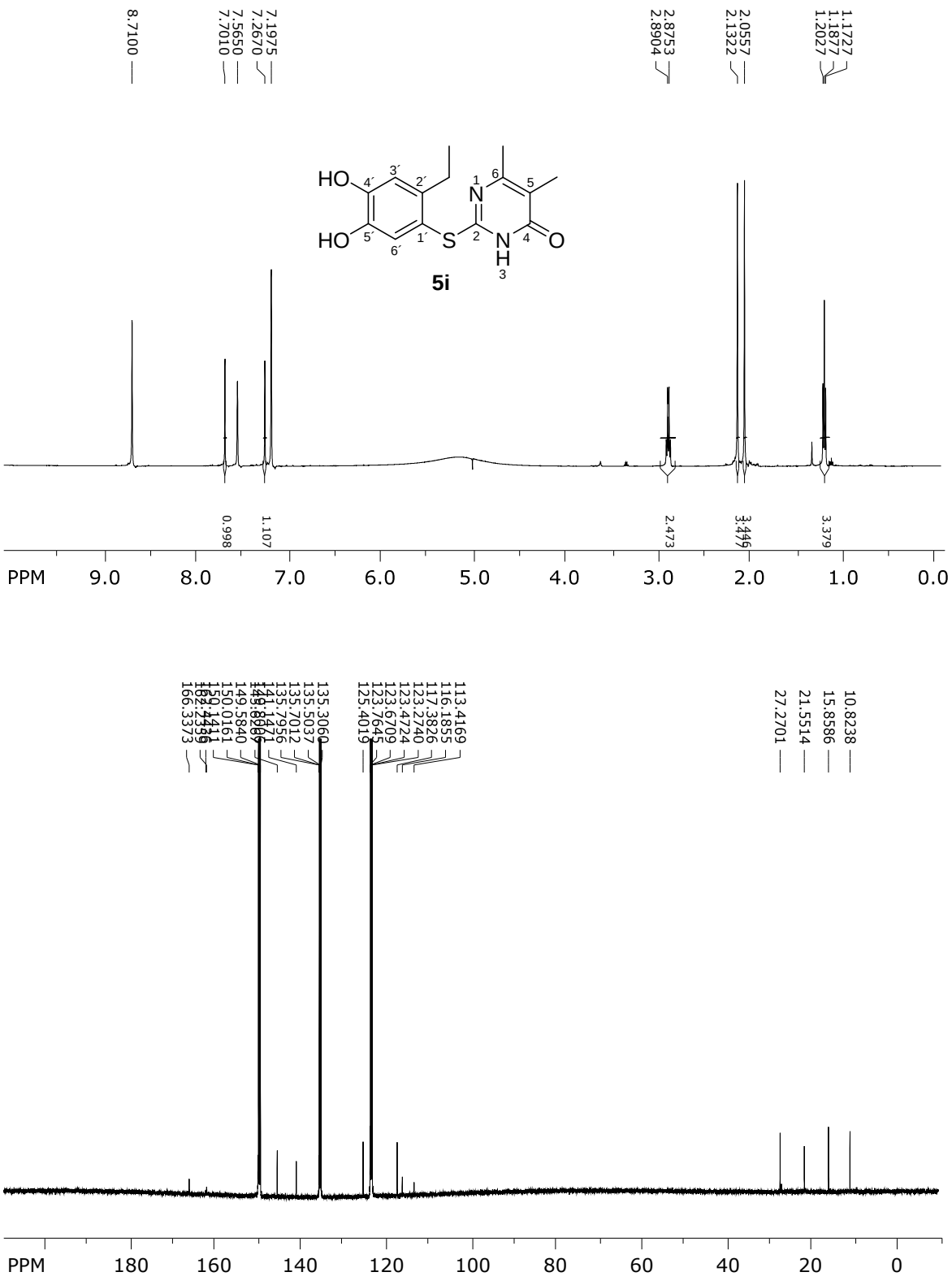


Fig. 27 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **5i** in pyridine-d_5

2-(4,5-Dihydroxy-2-methylphenylthio)-6,7-dihydro-3*H*-cyclopenta[*d*]pyrimidin-4(5*H*)-one (5j)

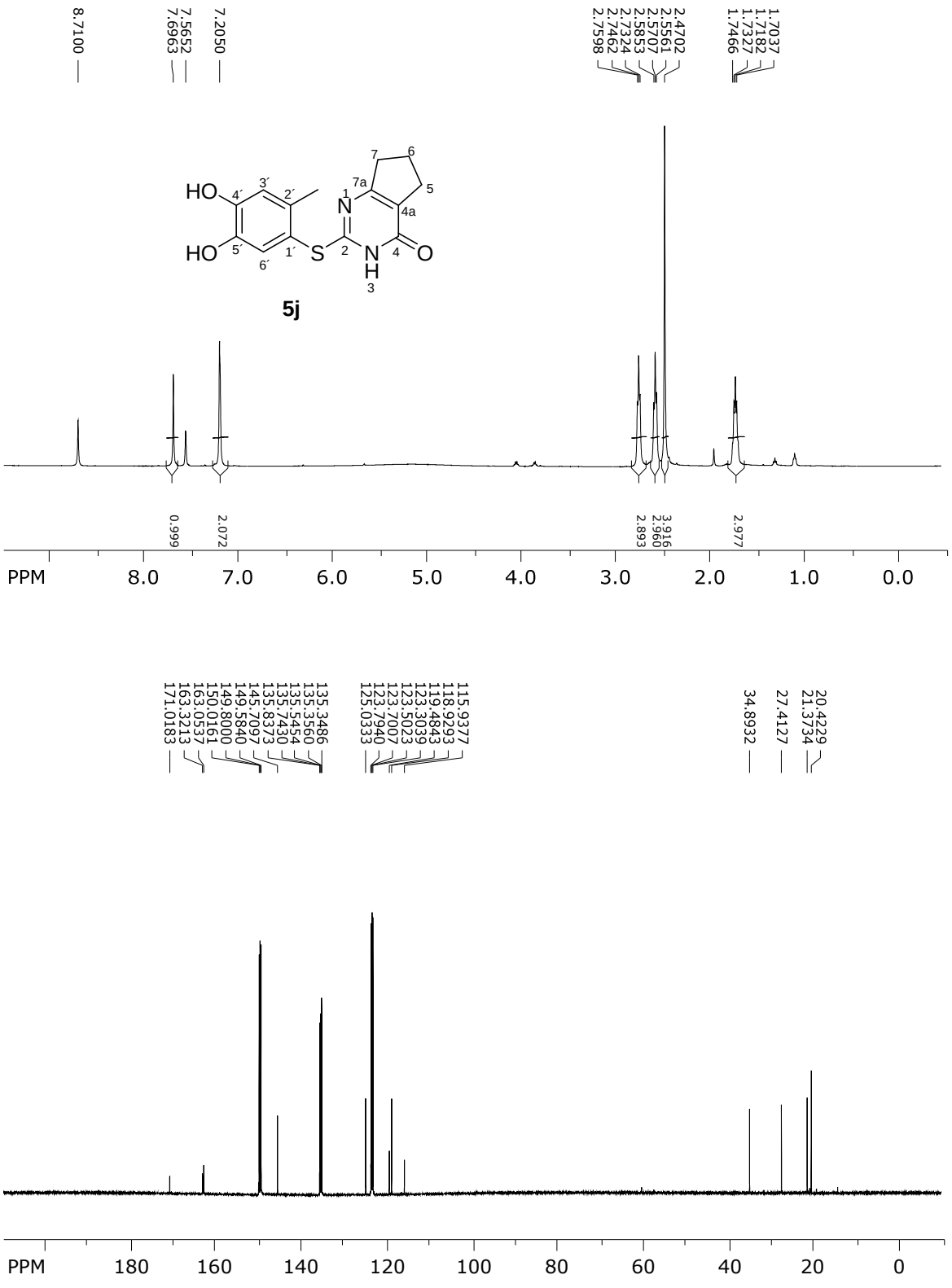


Fig. 28 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **5j** in pyridine-d_5

2-(2-Ethyl-4,5-dihydroxyphenylthio)-6,7-dihydro-3*H*-cyclopenta[*d*]pyrimidin-4(5*H*)-one (5k)

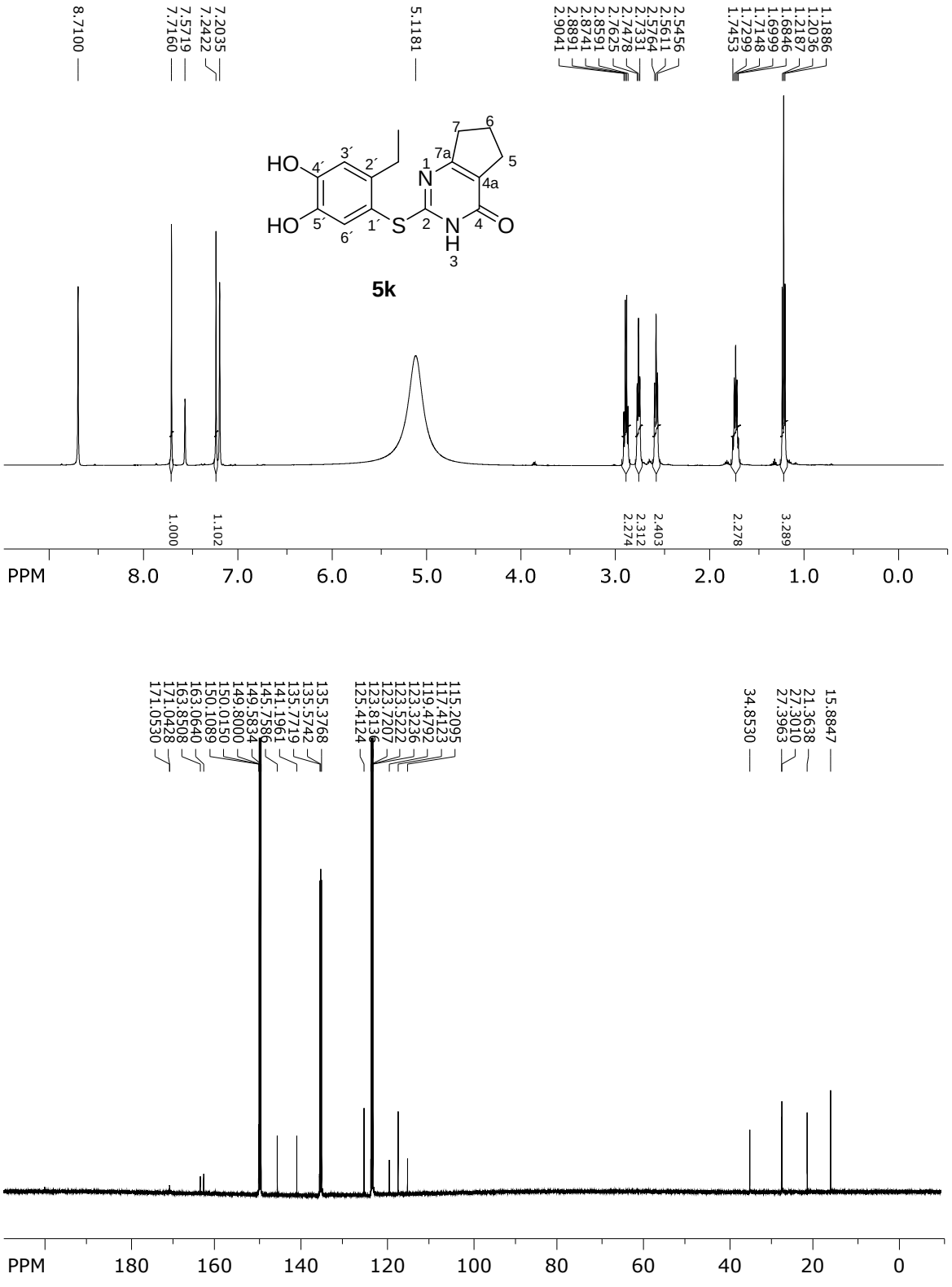


Fig. 29 ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra of **5k** in pyridine-d_5

2. Molecular structures and crystal data of 3f and 5a (Figures 30, 31; Tables 1-12)

2.1. Molecular structure and crystal data of 7,8-dihydroxy-4*H*-2-trifluoromethylpyrimido[2,1-*b*]benzothiazol-4-one (3f)

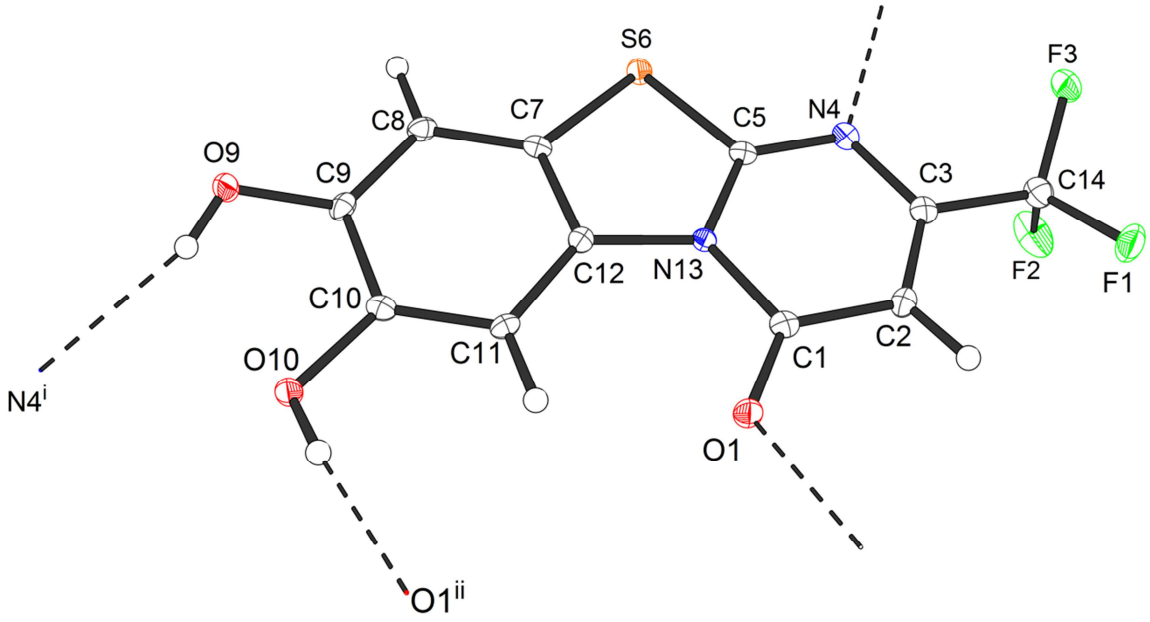


Fig. 30 Molecular structure of 7,8-dihydroxy-4*H*-2-trifluoromethyl-pyrimido[2,1-*b*]benzothiazol-4-one (**3f**), derived from X-ray crystal structure analysis. Thermal ellipsoids are drawn at the 50% level. Broken lines indicate hydrogen bond contacts.

Table 1. Crystal data and structure refinement for **3f**.

Crystal data

Identification code	**3f**	
Habitus, colour	needle, colourless	
Crystal size	0.40 x 0.06 x 0.04 mm^3	
Crystal system	Monoclinic	
Space group	$P2_1/c$	Z = 4
Unit cell dimensions	a = 6.5438(4) Å	α= 90°.
	b = 13.1859(7) Å	β= 93.862(2)°.
	c = 12.5014(7) Å	γ = 90°.
Volume	1076.24(11) $Å^3$	
Cell determination	9932 peaks with Theta 2.2 to 25.2°.	
Empirical formula	C_{11} H_5 F_3 N_2 O_3 S	
Moiety formula	C_{11} H_5 F_3 N_2 O_3 S	
Formula weight	302.23	
Density (calculated)	1.865 Mg/m^3	
Absorption coefficient	0.354 mm^{-1}	
F(000)	608	

Data collection:

Diffractometer type	Bruker D8 QUEST area detector
Wavelength	0.71073 Å
Temperature	100(2) K
Theta range for data collection	2.248 to 25.294°.
Index ranges	-7<=h<=7, -15<=k<=15, -14<=l<=14
Data collection software	APEX3 (Bruker AXS Inc., 2015)[1]
Cell refinement software	SAINT V8.35A (Bruker AXS Inc., 2015)[2]
Data reduction software	SAINT V8.35A (Bruker AXS Inc., 2015)

Solution and refinement:

Reflections collected	29062
Independent reflections	1953 [R(int) = 0.0424]
Completeness to theta = 25.242°	100.0 %
Observed reflections	1760[I > 2σ(I)]
Reflections used for refinement	1953
Absorption correction	Semi-empirical from equivalents[3]
Max. and min. transmission	0.99 and 0.93
Largest diff. peak and hole	0.308 and -0.300 e.$Å^{-3}$
Solution	Direct methods
Refinement	Full-matrix least-squares on F^2
Treatment of hydrogen atoms	CH calculated positions, constr. ref., OH located, isotr. ref.
Programs used	XT V2014/1 (Bruker AXS Inc., 2014)[4]
	SHELXL-2014/7 (Sheldrick, 2014)[5]
	DIAMOND (Crystal Impact)[6]
	ShelXle (Hübschle, Sheldrick, Dittrich, 2011)[7]
Data / restraints / parameters	1953 / 0 / 189
Goodness-of-fit on F^2	1.048
R index (all data)	wR2 = 0.0691
R index conventional [I>2sigma(I)]	R1 = 0.0280
CCDC No.	1514742

Table 2. Atomic coordinates and equivalent isotropic displacement parameters ($Å^2$) for **3f**. U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

	x	y	z	U(eq)	Occupancy
F1	0.78608(16)	0.65192(8)	0.86814(9)	0.0199(3)	1
O1	0.23096(17)	0.53534(9)	0.60531(9)	0.0111(3)	1
C1	0.3956(2)	0.50284(12)	0.64513(13)	0.0091(3)	1
C2	0.5208(3)	0.54876(13)	0.72984(13)	0.0103(4)	1
F2	0.79903(18)	0.50637(9)	0.94638(8)	0.0260(3)	1
F3	1.03202(15)	0.54882(8)	0.84132(8)	0.0171(2)	1
C3	0.7034(3)	0.50762(13)	0.76242(13)	0.0099(3)	1
N4	0.7843(2)	0.42160(11)	0.72207(11)	0.0101(3)	1
C5	0.6681(2)	0.37833(12)	0.64534(13)	0.0088(3)	1
S6	0.74119(6)	0.26897(3)	0.58172(3)	0.01109(13)	1
C7	0.5157(3)	0.27104(13)	0.49852(13)	0.0096(3)	1
C8	0.4574(3)	0.20102(13)	0.41909(13)	0.0108(3)	1
O9	0.2100(2)	0.14335(9)	0.28729(10)	0.0134(3)	1
C9	0.2659(3)	0.21057(13)	0.36611(13)	0.0099(3)	1
O10	-0.05252(18)	0.29090(10)	0.33795(10)	0.0123(3)	1
C10	0.1337(3)	0.28956(13)	0.39319(13)	0.0093(3)	1
C11	0.1931(3)	0.36061(12)	0.47093(13)	0.0088(3)	1
C12	0.3873(3)	0.35064(12)	0.52288(13)	0.0085(3)	1
N13	0.4795(2)	0.41299(10)	0.60534(11)	0.0077(3)	1
C14	0.8325(3)	0.55413(14)	0.85443(14)	0.0131(4)	1

Table 3. Bond lengths [Å] and angles [°] for **3f**.

F1-C14	1.338(2)	C7-C8	1.390(2)
O1-C1	1.233(2)	C7-C12	1.391(2)
C1-N13	1.411(2)	C8-C9	1.383(2)
C1-C2	1.429(2)	C8-H8	0.9500
C2-C3	1.350(2)	O9-C9	1.357(2)
C2-H2	0.9500	O9-H9	0.82(2)
F2-C14	1.342(2)	C9-C10	1.410(2)
F3-C14	1.328(2)	O10-C10	1.359(2)
C3-N4	1.363(2)	O10-H10	0.83(3)
C3-C14	1.511(2)	C10-C11	1.387(2)
N4-C5	1.314(2)	C11-C12	1.394(2)
C5-N13	1.378(2)	C11-H11	0.9500
C5-S6	1.7298(17)	C12-N13	1.421(2)
S6-C7	1.7473(17)		
O1-C1-N13	120.01(15)	O9-C9-C10	121.12(15)
O1-C1-C2	126.41(15)	C8-C9-C10	120.21(15)
N13-C1-C2	113.56(14)	C10-O10-H10	109.1(17)
C3-C2-C1	120.32(16)	O10-C10-C11	123.16(15)
C3-C2-H2	119.8	O10-C10-C9	115.54(15)
C1-C2-H2	119.8	C11-C10-C9	121.30(15)
C2-C3-N4	125.51(16)	C10-C11-C12	117.83(15)
C2-C3-C14	120.33(15)	C10-C11-H11	121.1
N4-C3-C14	114.10(15)	C12-C11-H11	121.1
C5-N4-C3	114.31(14)	C7-C12-C11	120.95(15)
N4-C5-N13	125.52(15)	C7-C12-N13	111.24(14)
N4-C5-S6	122.25(13)	C11-C12-N13	127.78(15)
N13-C5-S6	112.23(12)	C5-N13-C1	120.74(14)
C5-S6-C7	90.69(8)	C5-N13-C12	113.42(14)
C8-C7-C12	121.11(16)	C1-N13-C12	125.83(14)
C8-C7-S6	126.51(13)	F3-C14-F1	107.47(14)
C12-C7-S6	112.32(13)	F3-C14-F2	107.43(14)
C9-C8-C7	118.55(15)	F1-C14-F2	106.91(14)
C9-C8-H8	120.7	F3-C14-C3	112.87(14)
C7-C8-H8	120.7	F1-C14-C3	111.61(14)
C9-O9-H9	111.3(16)	F2-C14-C3	110.27(14)
O9-C9-C8	118.66(15)		

Symmetry transformations used to generate equivalent atoms:

Table 4. Anisotropic displacement parameters ($Å^2$) for **3f**.
The anisotropic displacement factor exponent takes the form: $-2\pi^2[h^2 a^{*2}U^{11} + ... + 2 h k a^* b^* U^{12}]$

	U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}
F1	0.0193(6)	0.0174(6)	0.0224(6)	-0.0111(5)	-0.0037(4)	0.0025(4)
O1	0.0102(6)	0.0099(6)	0.0128(6)	-0.0014(5)	-0.0017(5)	0.0031(5)
C1	0.0100(8)	0.0080(8)	0.0095(8)	0.0027(6)	0.0032(6)	-0.0011(7)
C2	0.0135(9)	0.0086(8)	0.0089(8)	-0.0004(6)	0.0019(7)	-0.0011(7)
F2	0.0294(6)	0.0372(7)	0.0106(5)	0.0068(5)	-0.0055(5)	-0.0140(5)
F3	0.0104(5)	0.0188(6)	0.0215(6)	-0.0049(4)	-0.0029(4)	-0.0014(4)
C3	0.0117(8)	0.0100(8)	0.0081(8)	0.0024(6)	0.0025(6)	-0.0024(7)
N4	0.0102(7)	0.0098(7)	0.0102(7)	0.0023(6)	-0.0004(6)	-0.0006(6)
C5	0.0084(8)	0.0078(8)	0.0102(8)	0.0030(6)	0.0017(6)	0.0002(6)
S6	0.0087(2)	0.0099(2)	0.0144(2)	-0.00193(16)	-0.00161(16)	0.00288(16)
C7	0.0075(8)	0.0102(8)	0.0113(8)	0.0020(7)	0.0010(6)	0.0006(6)
C8	0.0104(8)	0.0084(8)	0.0139(8)	-0.0009(7)	0.0029(7)	0.0019(7)
O9	0.0101(7)	0.0134(6)	0.0162(6)	-0.0078(5)	-0.0013(5)	0.0011(5)
C9	0.0130(8)	0.0090(8)	0.0082(8)	-0.0007(6)	0.0035(6)	-0.0023(7)
O10	0.0102(6)	0.0117(6)	0.0144(6)	-0.0048(5)	-0.0033(5)	0.0045(5)
C10	0.0082(8)	0.0101(8)	0.0096(8)	0.0027(6)	0.0013(6)	-0.0003(6)
C11	0.0100(8)	0.0072(8)	0.0094(8)	0.0008(6)	0.0031(6)	0.0013(6)
C12	0.0100(8)	0.0071(8)	0.0087(8)	0.0009(6)	0.0025(6)	-0.0020(6)
N13	0.0073(7)	0.0078(7)	0.0080(7)	0.0005(5)	0.0006(5)	-0.0001(5)
C14	0.0136(9)	0.0144(9)	0.0111(8)	0.0004(7)	0.0000(7)	-0.0015(7)

Table 5. Hydrogen coordinates and isotropic displacement parameters ($Å^2$) for **3f**.

	x	y	z	U(eq)	Occupancy
H2	0.4753	0.6085	0.7634	0.012	1
H8	0.5471	0.1478	0.4016	0.013	1
H9	0.086(4)	0.1426(17)	0.2747(18)	0.024(6)	1
H10	-0.114(4)	0.3434(19)	0.3529(19)	0.029(6)	1
H11	0.1044	0.4143	0.4882	0.011	1

Table 6. Torsion angles [°] for **3f**.

O1-C1-C2-C3	-176.67(16)
N13-C1-C2-C3	1.9(2)
C1-C2-C3-N4	-1.0(3)
C1-C2-C3-C14	-177.95(15)
C2-C3-N4-C5	0.0(2)
C14-C3-N4-C5	177.04(14)
C3-N4-C5-N13	0.1(2)
C3-N4-C5-S6	179.43(12)
N4-C5-S6-C7	-178.61(14)
N13-C5-S6-C7	0.83(13)
C5-S6-C7-C8	-179.59(16)
C5-S6-C7-C12	-2.47(13)
C12-C7-C8-C9	-1.6(2)
S6-C7-C8-C9	175.34(13)
C7-C8-C9-O9	178.89(15)
C7-C8-C9-C10	-0.4(2)
O9-C9-C10-O10	2.1(2)
C8-C9-C10-O10	-178.64(15)
O9-C9-C10-C11	-177.59(15)
C8-C9-C10-C11	1.6(2)
O10-C10-C11-C12	179.35(15)
C9-C10-C11-C12	-1.0(2)
C8-C7-C12-C11	2.3(2)
S6-C7-C12-C11	-175.04(13)
C8-C7-C12-N13	-179.27(15)
S6-C7-C12-N13	3.43(17)
C10-C11-C12-C7	-1.0(2)
C10-C11-C12-N13	-179.15(15)
N4-C5-N13-C1	1.0(2)
S6-C5-N13-C1	-178.42(11)
N4-C5-N13-C12	-179.60(15)
S6-C5-N13-C12	0.97(17)
O1-C1-N13-C5	176.78(15)
C2-C1-N13-C5	-1.9(2)
O1-C1-N13-C12	-2.5(2)
C2-C1-N13-C12	178.79(14)
C7-C12-N13-C5	-2.84(19)
C11-C12-N13-C5	175.50(16)
C7-C12-N13-C1	176.51(14)
C11-C12-N13-C1	-5.1(3)
C2-C3-C14-F3	-144.19(16)
N4-C3-C14-F3	38.6(2)
C2-C3-C14-F1	-23.0(2)
N4-C3-C14-F1	159.73(14)
C2-C3-C14-F2	95.66(19)
N4-C3-C14-F2	-81.60(18)

Symmetry transformations used to generate equivalent atoms:

Table 7. Hydrogen bonds for **3f** [Å and °].

D-H...A	d(D-H)	d(H...A)	d(D...A)	<(DHA)
O9-H9...N4#1	0.82(2)	2.21(2)	2.9755(19)	157(2)
O10-H10...O1#2	0.83(3)	1.86(3)	2.6884(17)	175(2)

Symmetry transformations used to generate equivalent atoms:
#1 x-1,-y+1/2,z-1/2 #2 -x,-y+1,-z+1

2.2. Molecular structure and crystal data of 2-(4,5-dihydroxy-2-methylphenylthio)-6-methylpyrimidin-4(3*H*)-one (5a)

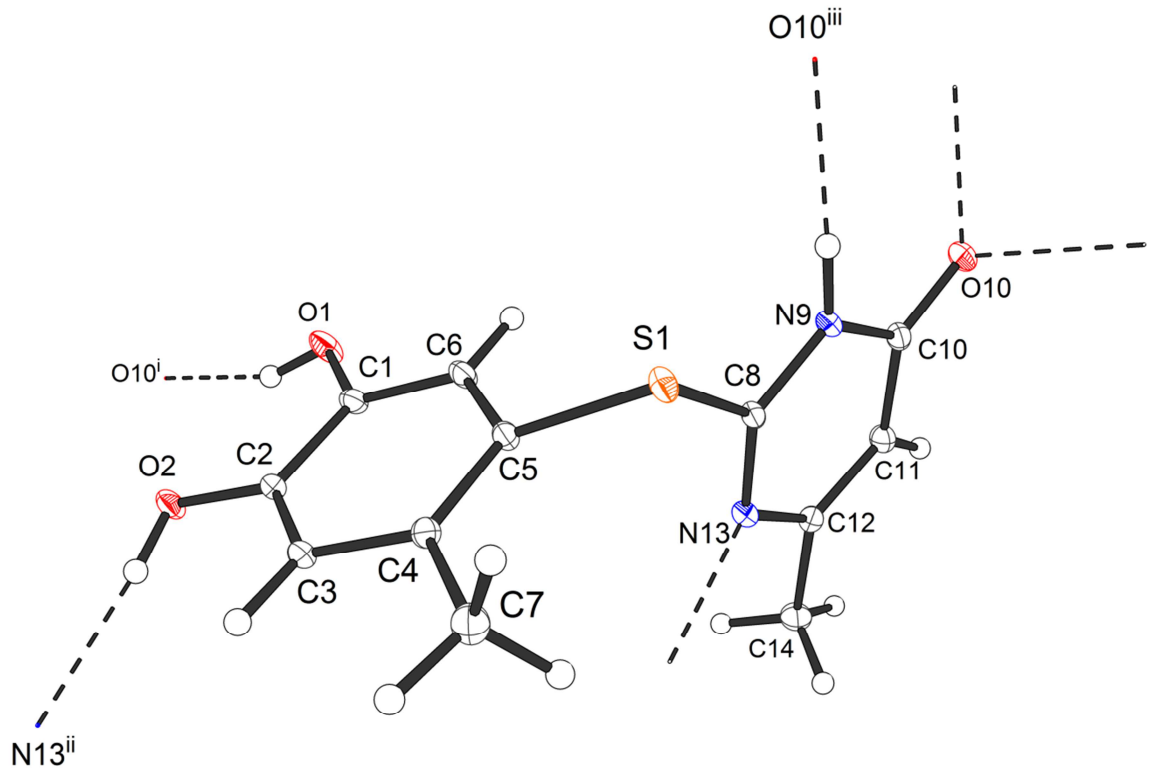


Fig. 31 Molecular structure of 2-(4,5-dihydroxy-2-methylphenylthio)-6-methylpyrimidin-4(3*H*)-one (**5a**), derived from X-ray crystal structure analysis. Thermal ellipsoids are drawn at the 50% level. Broken lines indicate hydrogen bond contacts.

Table 8. Crystal data and structure refinement for **5a**.

Crystal data

Identification code	**5a**	
Habitus, colour	block, colourless	
Crystal size	0.40 x 0.26 x 0.20 mm^3	
Crystal system	Triclinic	
Space group	P-1	Z = 2
Unit cell dimensions	a = 8.4440(3) Å	α= 68.729(1)°.
	b = 8.4669(3) Å	β= 86.391(1)°.
	c = 9.6137(4) Å	γ = 64.332(1)°.
Volume	573.66(4) $Å^3$	
Cell determination	9901 peaks with Theta 2.7 to 33.2°.	
Empirical formula	C_{12} H_{12} N_2 O_3 S	
Moiety formula	C_{12} H_{12} N_2 O_3 S	
Formula weight	264.30	
Density (calculated)	1.530 Mg/m^3	
Absorption coefficient	0.284 mm^{-1}	
F(000)	276	

Data collection:

Diffractometer type	Bruker D8 QUEST area detector
Wavelength	0.71073 Å
Temperature	100(2) K
Theta range for data collection	2.288 to 33.229°.
Index ranges	-13<=h<=13, -13<=k<=13, -14<=l<=14
Data collection software	APEX3 (Bruker AXS Inc., 2015)[1]
Cell refinement software	SAINT V8.35A (Bruker AXS Inc., 2015)[2,2]
Data reduction software	SAINT V8.35A (Bruker AXS Inc., 2015)

Solution and refinement:

Reflections collected	22332
Independent reflections	4377 [R(int) = 0.0224]
Completeness to theta = 25.242°	99.9 %
Observed reflections	3984[I > 2σ(I)]
Reflections used for refinement	4377
Absorption correction	Semi-empirical from equivalents[3]
Max. and min. transmission	0.95 and 0.91
Largest diff. peak and hole	0.539 and -0.299 e.$Å^{-3}$
Solution	Direct methods
Refinement	Full-matrix least-squares on F^2
Treatment of hydrogen atoms	CH calc. positions, “riding”, NH, OH located, isotr. ref.
Programs used	XT V2014/1 (Bruker AXS Inc., 2014)[4]
	SHELXL-2014/7 (Sheldrick, 2014)[5]
	DIAMOND (Crystal Impact)[6]
	ShelXle (Hübschle, Sheldrick, Dittrich, 2011)[7]
Data / restraints / parameters	4377 / 0 / 177
Goodness-of-fit on F^2	1.054
R index (all data)	wR2 = 0.0824
R index conventional [I>2sigma(I)]	R1 = 0.0298
CCDC No.	1514742

Table 9. Atomic coordinates and equivalent isotropic displacement parameters ($Å^2$) for **5a**. U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

	x	y	z	U(eq)	Occupancy
S1	0.41233(3)	0.54768(3)	0.28578(2)	0.01138(6)	1
C1	0.23770(11)	0.20824(12)	0.59525(10)	0.01045(14)	1
O1	0.07620(9)	0.21856(10)	0.63398(8)	0.01579(13)	1
C2	0.39471(11)	0.05585(11)	0.67876(9)	0.00933(14)	1
O2	0.37530(9)	-0.08080(9)	0.79761(7)	0.01249(12)	1
C3	0.55632(11)	0.05345(11)	0.63915(9)	0.00990(14)	1
C4	0.56848(11)	0.19758(12)	0.51462(9)	0.00965(14)	1
C5	0.41022(11)	0.34745(11)	0.43296(9)	0.00955(14)	1
C6	0.24593(11)	0.35417(12)	0.47391(10)	0.01108(14)	1
C7	0.74732(11)	0.18589(13)	0.47509(10)	0.01395(16)	1
C8	0.29570(11)	0.56442(11)	0.13139(9)	0.00953(14)	1
N9	0.17496(10)	0.74255(10)	0.04995(8)	0.01037(13)	1
O10	-0.02494(9)	0.95533(9)	-0.15454(7)	0.01350(12)	1
C10	0.08040(11)	0.78815(12)	-0.08284(9)	0.01079(14)	1
C11	0.11335(12)	0.63007(12)	-0.12192(10)	0.01218(15)	1
C12	0.23319(11)	0.45298(12)	-0.03297(9)	0.01093(14)	1
N13	0.32850(10)	0.41954(10)	0.09437(8)	0.01066(13)	1
C14	0.27281(13)	0.28265(13)	-0.06889(11)	0.01555(16)	1

Table 10. Bond lengths [Å] and angles [°] for **5a**.

S1-C8	1.7617(9)	C7-H7B	0.9800
S1-C5	1.7774(8)	C7-H7C	0.9800
C1-O1	1.3647(10)	C8-N13	1.3108(11)
C1-C6	1.3823(12)	C8-N9	1.3560(10)
C1-C2	1.4027(11)	N9-C10	1.3856(11)
O1-H1	0.831(19)	N9-H9	0.912(16)
C2-O2	1.3611(10)	O10-C10	1.2444(10)
C2-C3	1.3870(11)	C10-C11	1.4280(12)
O2-H2	0.840(19)	C11-C12	1.3666(12)
C3-C4	1.4022(11)	C11-H11	0.9500
C3-H3	0.9500	C12-N13	1.3790(11)
C4-C5	1.3973(11)	C12-C14	1.4966(12)
C4-C7	1.5034(12)	C14-H14A	0.9800
C5-C6	1.4007(12)	C14-H14B	0.9800
C6-H6	0.9500	C14-H14C	0.9800
C7-H7A	0.9800		

C8-S1-C5	102.94(4)	H7A-C7-H7C	109.5
O1-C1-C6	119.07(7)	H7B-C7-H7C	109.5
O1-C1-C2	121.36(7)	N13-C8-N9	123.01(8)
C6-C1-C2	119.54(8)	N13-C8-S1	123.08(6)
C1-O1-H1	109.2(13)	N9-C8-S1	113.84(6)
O2-C2-C3	124.31(7)	C8-N9-C10	122.76(8)
O2-C2-C1	115.95(7)	C8-N9-H9	119.8(10)
C3-C2-C1	119.73(7)	C10-N9-H9	117.3(10)
C2-O2-H2	110.0(13)	O10-C10-N9	119.63(8)
C2-C3-C4	121.86(7)	O10-C10-C11	126.06(8)
C2-C3-H3	119.1	N9-C10-C11	114.29(7)
C4-C3-H3	119.1	C12-C11-C10	120.14(8)
C5-C4-C3	117.29(7)	C12-C11-H11	119.9
C5-C4-C7	123.11(7)	C10-C11-H11	119.9
C3-C4-C7	119.61(7)	C11-C12-N13	122.28(8)
C4-C5-C6	121.48(7)	C11-C12-C14	122.29(8)
C4-C5-S1	120.24(6)	N13-C12-C14	115.41(8)
C6-C5-S1	117.95(6)	C8-N13-C12	117.41(7)
C1-C6-C5	120.07(7)	C12-C14-H14A	109.5
C1-C6-H6	120.0	C12-C14-H14B	109.5
C5-C6-H6	120.0	H14A-C14-H14B	109.5
C4-C7-H7A	109.5	C12-C14-H14C	109.5
C4-C7-H7B	109.5	H14A-C14-H14C	109.5
H7A-C7-H7B	109.5	H14B-C14-H14C	109.5
C4-C7-H7C	109.5		

Symmetry transformations used to generate equivalent atoms:

Table 11. Anisotropic displacement parameters ($Å^2$) for **5a**.
The anisotropic displacement factor exponent takes the form: $-2\pi^2[h^2 a^{*2}U^{11} + ... + 2 h k a^* b^* U^{12}]$

	U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}
S1	0.01445(10)	0.00889(9)	0.01001(9)	-0.00115(7)	-0.00091(7)	-0.00610(7)
C1	0.0088(3)	0.0084(3)	0.0116(3)	-0.0029(3)	0.0011(3)	-0.0023(3)
O1	0.0092(3)	0.0103(3)	0.0199(3)	0.0006(2)	0.0022(2)	-0.0025(2)
C2	0.0102(3)	0.0071(3)	0.0084(3)	-0.0022(3)	0.0004(2)	-0.0023(3)
O2	0.0115(3)	0.0079(3)	0.0114(3)	0.0007(2)	0.0013(2)	-0.0019(2)
C3	0.0088(3)	0.0086(3)	0.0090(3)	-0.0024(3)	-0.0008(2)	-0.0015(3)
C4	0.0094(3)	0.0101(3)	0.0087(3)	-0.0036(3)	0.0001(3)	-0.0035(3)
C5	0.0106(3)	0.0076(3)	0.0087(3)	-0.0019(3)	0.0001(3)	-0.0034(3)
C6	0.0094(3)	0.0078(3)	0.0115(3)	-0.0013(3)	-0.0004(3)	-0.0015(3)
C7	0.0104(3)	0.0161(4)	0.0133(4)	-0.0034(3)	0.0008(3)	-0.0056(3)
C8	0.0100(3)	0.0074(3)	0.0089(3)	-0.0013(3)	0.0015(2)	-0.0032(3)
N9	0.0117(3)	0.0066(3)	0.0096(3)	-0.0016(2)	0.0004(2)	-0.0023(2)
O10	0.0135(3)	0.0082(3)	0.0120(3)	-0.0003(2)	-0.0007(2)	-0.0014(2)
C10	0.0105(3)	0.0095(3)	0.0095(3)	-0.0015(3)	0.0013(3)	-0.0035(3)
C11	0.0141(3)	0.0110(3)	0.0095(3)	-0.0029(3)	0.0003(3)	-0.0046(3)
C12	0.0127(3)	0.0101(3)	0.0097(3)	-0.0037(3)	0.0027(3)	-0.0050(3)
N13	0.0119(3)	0.0077(3)	0.0102(3)	-0.0026(2)	0.0011(2)	-0.0030(2)
C14	0.0201(4)	0.0125(4)	0.0157(4)	-0.0077(3)	0.0030(3)	-0.0067(3)

Table 12. Hydrogen coordinates and isotropic displacement parameters ($Å^2$) for **5a**.

	x	y	z	U(eq)	Occupancy
H1	0.090(2)	0.116(3)	0.700(2)	0.038(5)	1
H2	0.471(3)	-0.180(3)	0.825(2)	0.038(5)	1
H3	0.6617	-0.0487	0.6981	0.012	1
H6	0.1400	0.4591	0.4182	0.013	1
H7A	0.7616	0.1864	0.3730	0.021	1
H7B	0.8395	0.0690	0.5461	0.021	1
H7C	0.7575	0.2946	0.4804	0.021	1
H9	0.148(2)	0.836(2)	0.0855(18)	0.027(4)	1
H11	0.0518	0.6480	-0.2099	0.015	1
H14A	0.2587	0.1861	0.0184	0.023	1
H14B	0.3946	0.2323	-0.0938	0.023	1
H14C	0.1911	0.3180	-0.1547	0.023	1

Table 13. Torsion angles [°] for **5a**.

O1-C1-C2-O2	1.19(12)
C6-C1-C2-O2	179.16(8)
O1-C1-C2-C3	-177.74(8)
C6-C1-C2-C3	0.23(13)
O2-C2-C3-C4	179.49(8)
C1-C2-C3-C4	-1.67(13)
C2-C3-C4-C5	1.42(12)
C2-C3-C4-C7	-178.82(8)
C3-C4-C5-C6	0.24(12)
C7-C4-C5-C6	-179.50(8)
C3-C4-C5-S1	173.42(6)
C7-C4-C5-S1	-6.33(12)
C8-S1-C5-C4	125.72(7)
C8-S1-C5-C6	-60.87(8)
O1-C1-C6-C5	179.41(8)
C2-C1-C6-C5	1.40(13)
C4-C5-C6-C1	-1.65(13)
S1-C5-C6-C1	-174.98(7)
C5-S1-C8-N13	-47.87(8)
C5-S1-C8-N9	135.24(6)
N13-C8-N9-C10	-2.52(13)
S1-C8-N9-C10	174.37(6)
C8-N9-C10-O10	-177.97(8)
C8-N9-C10-C11	3.47(12)
O10-C10-C11-C12	179.95(8)
N9-C10-C11-C12	-1.60(12)
C10-C11-C12-N13	-1.27(13)
C10-C11-C12-C14	-179.92(8)
N9-C8-N13-C12	-0.56(12)
S1-C8-N13-C12	-177.16(6)
C11-C12-N13-C8	2.41(12)
C14-C12-N13-C8	-178.85(8)

Symmetry transformations used to generate equivalent atoms:

Table 14. Hydrogen bonds for **5a** [Å and °].

D-H...A	d(D-H)	d(H...A)	d(D...A)	<(DHA)
O1-H1...O2	0.831(19)	2.269(18)	2.7091(9)	113.4(15)
O1-H1...O10#1	0.831(19)	2.083(19)	2.8236(9)	148.2(17)
O2-H2...N13#2	0.840(19)	1.906(19)	2.7407(10)	172.4(18)
N9-H9...O10#3	0.912(16)	1.950(17)	2.8386(10)	164.3(15)

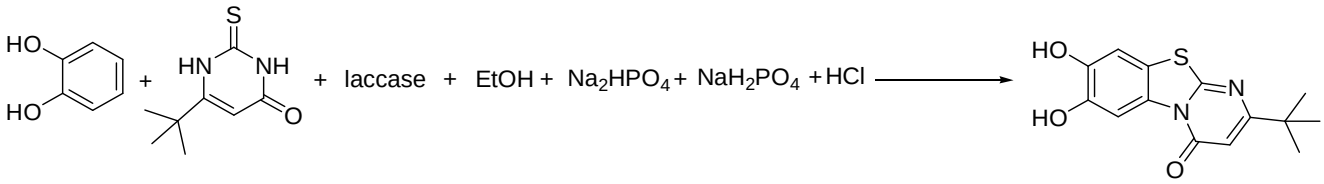
Symmetry transformations used to generate equivalent atoms:
#1 x,y-1,z+1 #2 -x+1,-y,-z+1 #3 -x,-y+2,-z

3. Greenness of the laccase-catalyzed reactions

3.1. Calculation of the E-factor, atom economy, TON and TOF of the laccase-catalyzed domino reaction between 1a and 2e

Yield of **3e** = 97%

Calculation of the E-factor[8]



1a 64 mg; **2e** 92 mg; laccase 10 mg; EtOH 157.8 mg; Na_2HPO_4 62.86 mg; NaH_2PO_4 378.8 mg; HCl 14.4 mg; **3e** 140 mg

Total amount of the reactants (taking into account a loss of 10% of the solvent used) = 64 mg + 92 mg + 10 mg + 157.8 mg + 62.86 mg + 378.8 mg + 14.4 mg = 779.86 mg.

Amount of the final product = 140 mg.

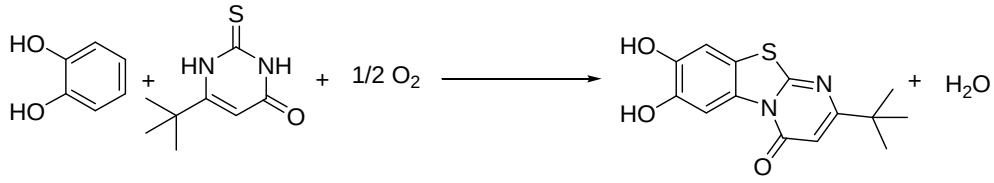
Amount of waste = 779.86 - 140 = 639.86 mg

E-factor = Amount of waste [kg]/Amount of product [kg] = 639.86/140 = **4.57 kg kg^{-1}**.

Calculation of the atom economy[9]

The atom economy of the reaction was calculated according to the following equation:

$$\%\ \text{Atom economy} = 100 \times \frac{\text{Molecular weight of the desired product}}{\text{Molecular weight of all reactants}}.$$



1a 110 g/mol; **2e** 184 g/mol; 1/2 O_2 16 g/mol; **3e** 290 g/mol

Atom economy = 100 × 290/310 = **94%**

Calculation of TON

Molecular weight of laccase from *A. bisporus* = 96 000 g/mol.[10]

Specific activity of the laccase = 1.2 U/mg.

12 U Laccase corresponds to 10 mg, ie 1.0417×10^{-7} mol = 1.0417×10^{-4} mmol.

TON = Amount of the substrate consumed [mmol] / Amount of catalyst [mmol].

TON = 0.485 [mmol] / 1.0417×10^{-4} [mmol] = **4656**.

Calculation of TOF

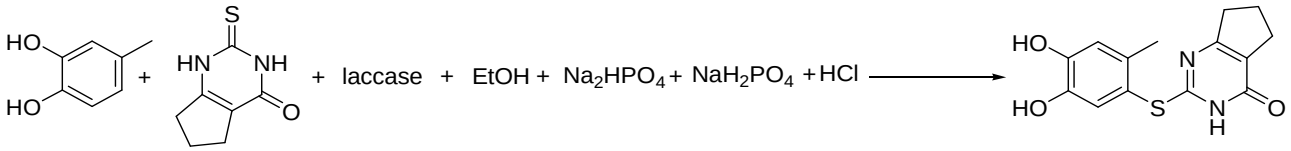
$$\mathrm{TOF} = \frac{\mathrm{TON}}{\mathrm{Time}}.$$

TOF = 4656 / 16 h = **291 h^{-1}**.

3.2. Calculation of the E-factor, atom economy, TON and TOF of the laccase-catalyzed domino reaction between 1b and 2i

Yield of **5j** = 90%

Calculation of the E-factor[8]



1b 72 mg | **2i** 84 mg | laccase 10 mg | EtOH 157.8 mg | Na_2HPO_4 62.86 mg | NaH_2PO_4 378.8 mg | HCl 14.4 mg | **5j** 130 mg

Total amount of the reactants (taking into account a loss of 10% of the solvent used) = 72 mg + 84 mg + 10 mg + 157.8 mg + 62.86 mg + 378.8 mg + 14.4 mg = 779.86 mg.

Amount of the final product = 130 mg.

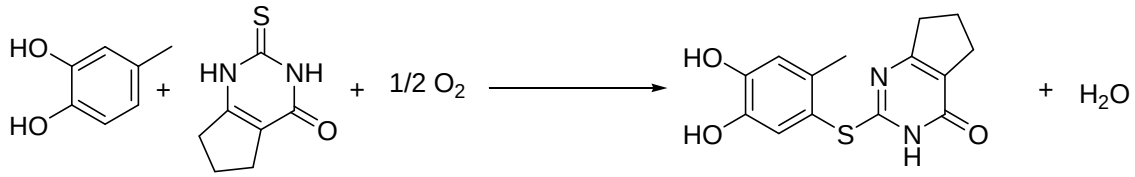
Amount of waste = 779.86 - 130 = 649.86 mg

E-factor = Amount of waste [kg]/Amount of product [kg] = 649.86/130 = **5.00 kg kg^{-1}**.

Calculation of the atom economy[9]

The atom economy of the reaction was calculated according to the following equation:

$$\%\ \text{Atom economy} = 100 \times \frac{\text{Molecular weight of the desired product}}{\text{Molecular weight of all reactants}}.$$



1b **2i** **5j**

124 g/mol 168 g/mol 16 g/mol 290 g/mol

Atom economy = 100 × 290/308 = **94%**

Calculation of TON

Molecular weight of laccase from *A. bisporus* = 96 000 g/mol.[10]

Specific activity of the laccase = 1.2 U/mg.

12 U Laccase corresponds to 10 mg, ie 1.0417×10^{-7} mol = 1.0417×10^{-4} mmol.

TON = Amount of the substrate consumed [mmol] / Amount of catalyst [mmol].

TON = 0.45 [mmol] / 1.0417×10^{-4} [mmol] = **4320**.

Calculation of TOF

$$\text{TOF} = \frac{\text{TON}}{\text{Time}}.$$

TOF = 4320 / 14 h = **308.57 h^{-1}**.

4. Determination of the activity of laccase from *Agaricus bisporus*

According to ref. 11 a 0.1 M solution of ABTS (0.3 mL) in 0.2 M phosphate buffer (pH 6) was diluted with 0.2 M phosphate buffer (2.6 mL, pH 6) and treated with a solution of laccase in the same buffer (0.1 mL). The change in absorption was followed *via* UV-spectroscopy (λ = 414 nm). One unit was defined as the amount of laccase that converts 1 μmol of ABTS per minute at pH 6 at rt.

5. IC_{50} graphs of the tested compounds against HepG2 cell line (Figures 32-46)

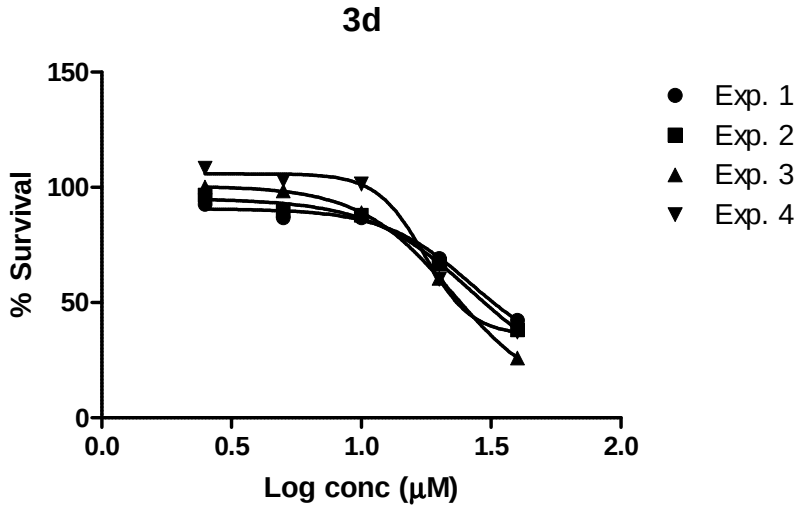


Fig. 32 IC_{50} graph of **3d**

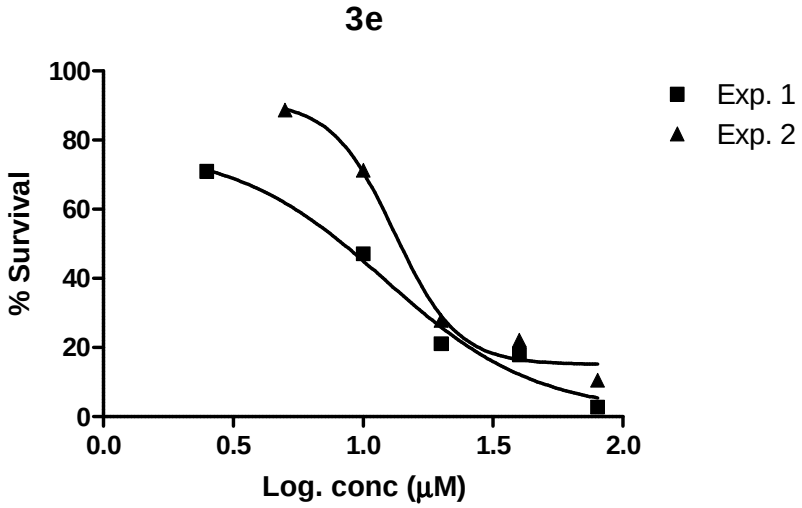


Fig. 33 IC_{50} graph of **3e**

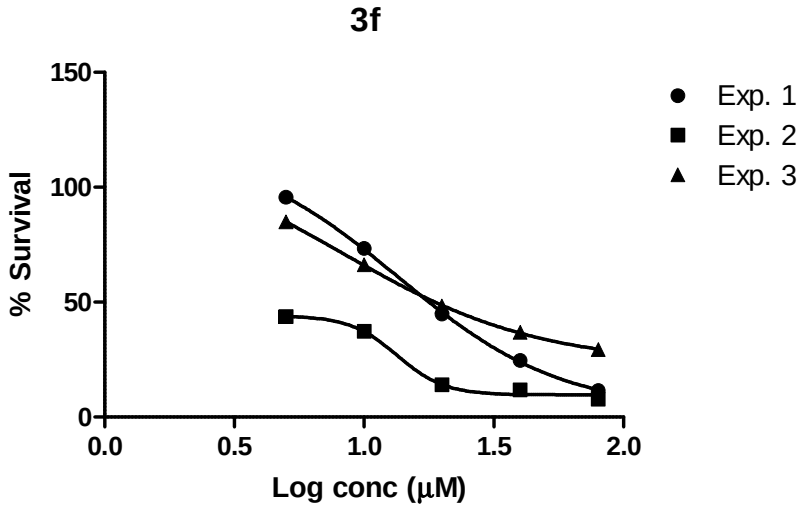


Fig. 34 IC_{50} graph of **3f**

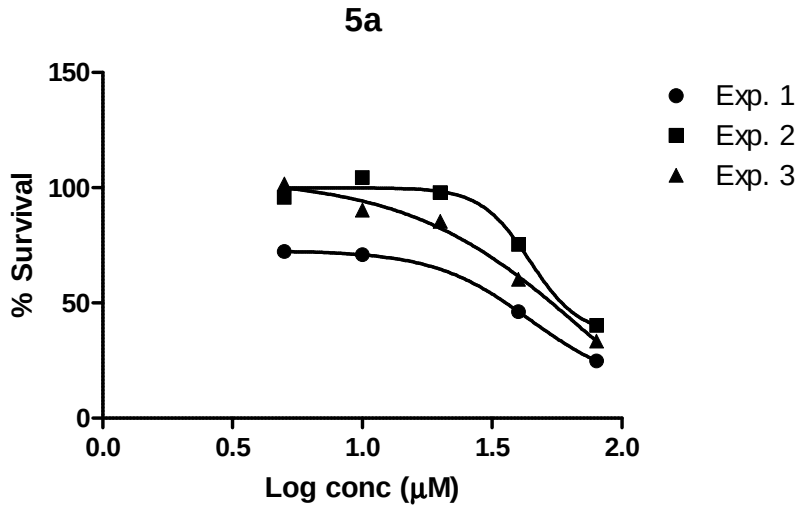


Fig. 35 IC_{50} graph of **5a**

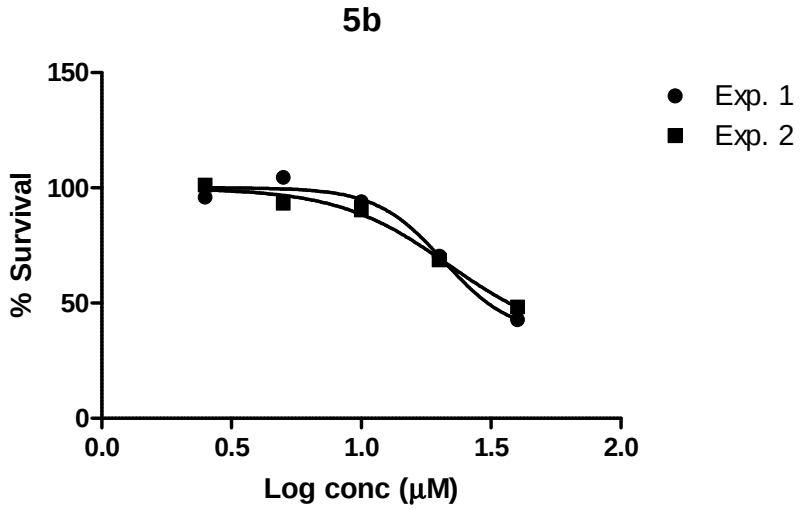


Fig. 36 IC_{50} graph of **5b**

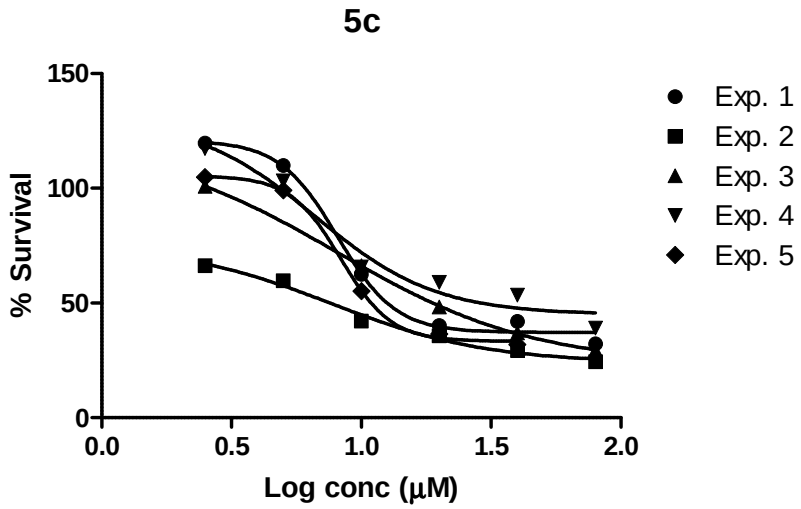


Fig. 37 IC_{50} graph of **5c**

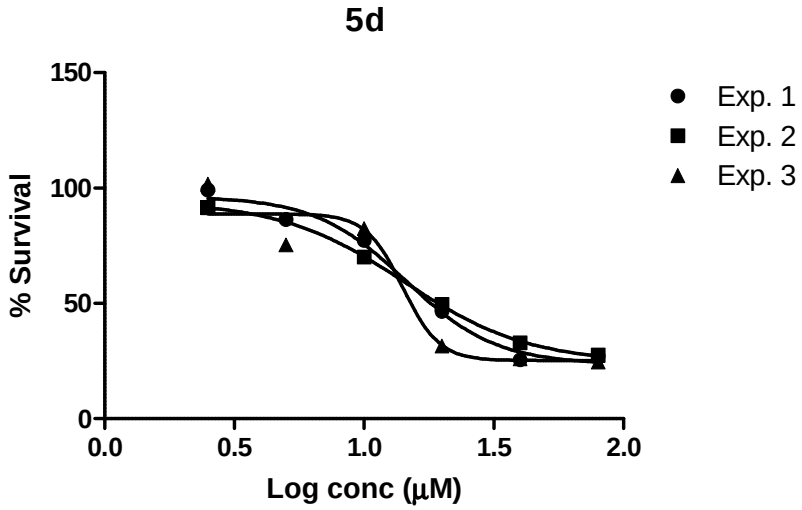


Fig. 38 IC_{50} graph of **5d**

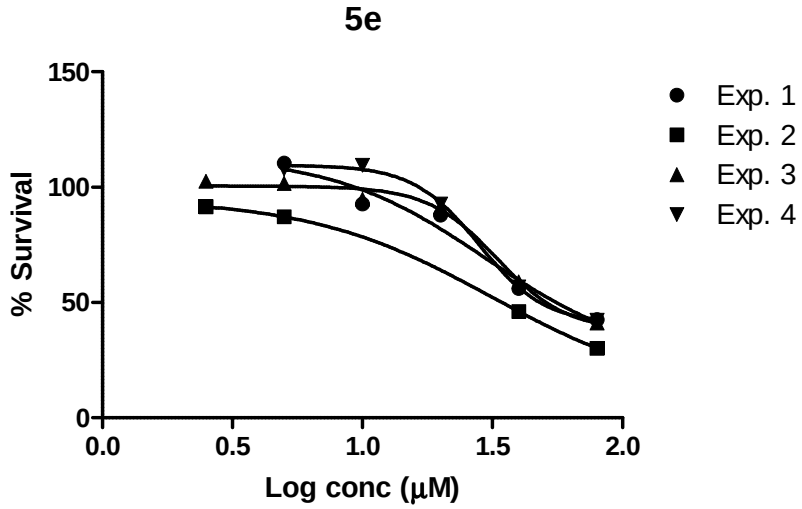


Fig. 39 IC_{50} graph of **5e**

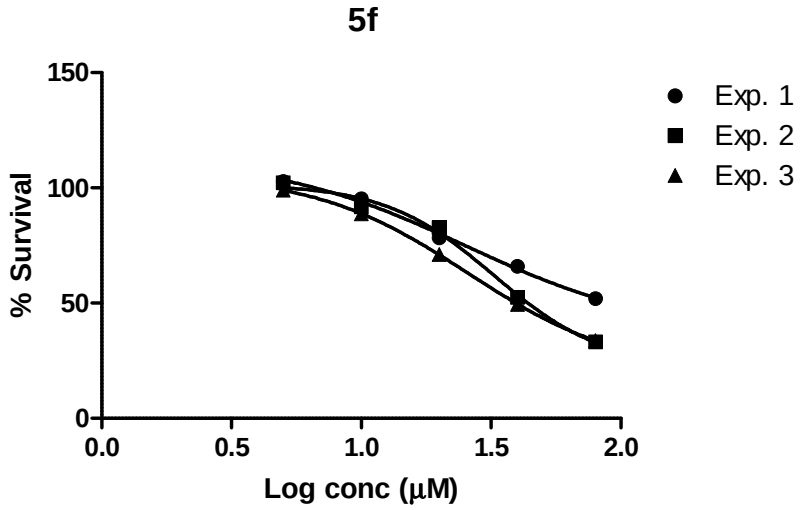


Fig. 40 IC_{50} graph of **5f**

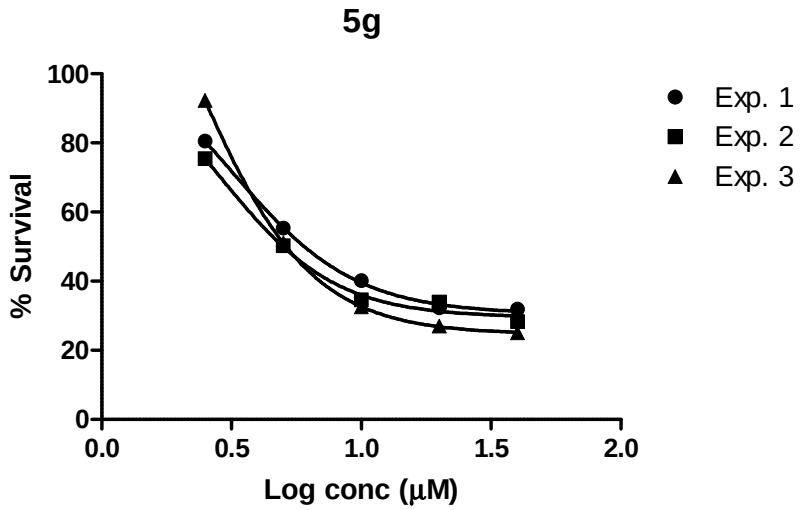


Fig. 41 IC_{50} graph of **5g**

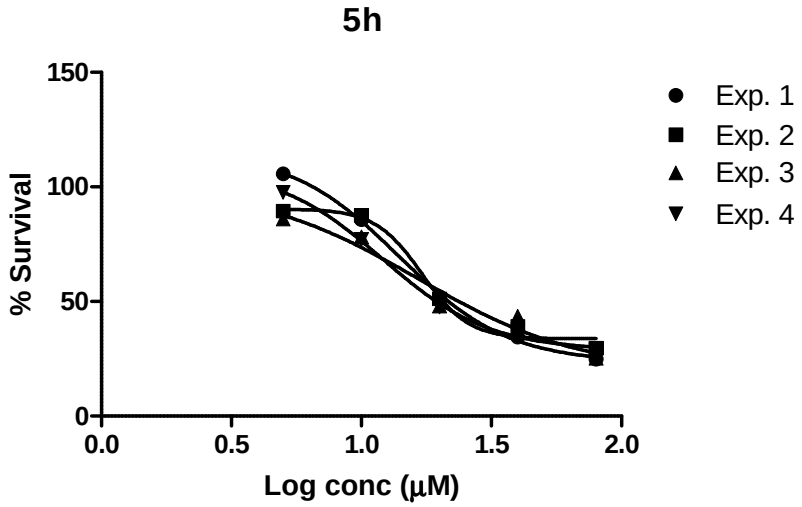


Fig. 42 IC_{50} graph of **5h**

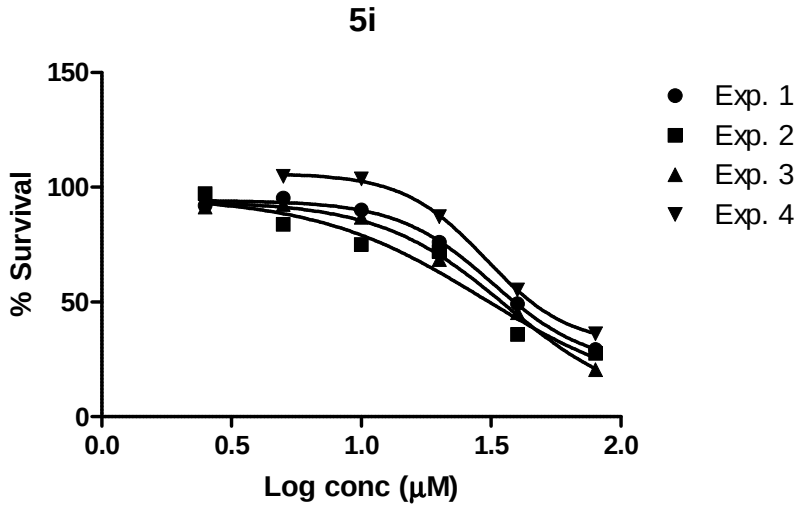


Fig. 43 IC_{50} graph of **5i**

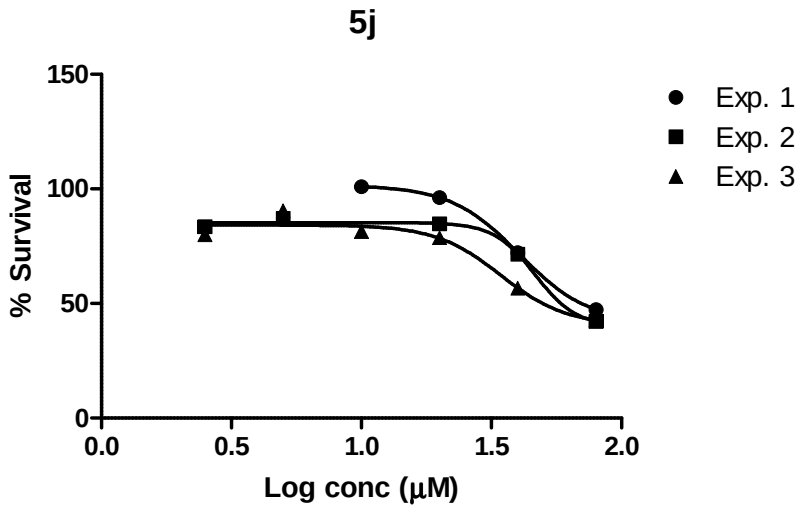


Fig. 44 IC_{50} graph of **5j**

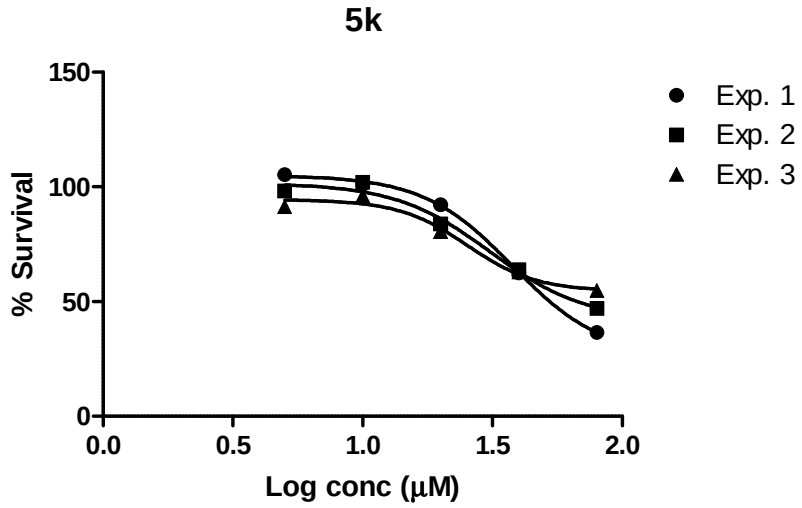


Fig. 45 IC_{50} graph of **5k**

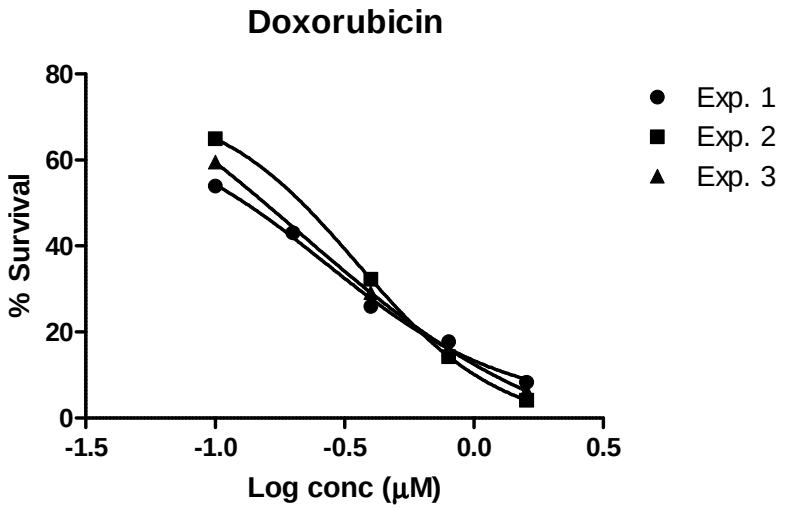


Fig. 46 IC_{50} graph of **Doxorubicin**

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