Supporting information

CK2 inhibition, lipophilicity and anticancer activity of substituted tetrabromobenzotriazole regioisomers

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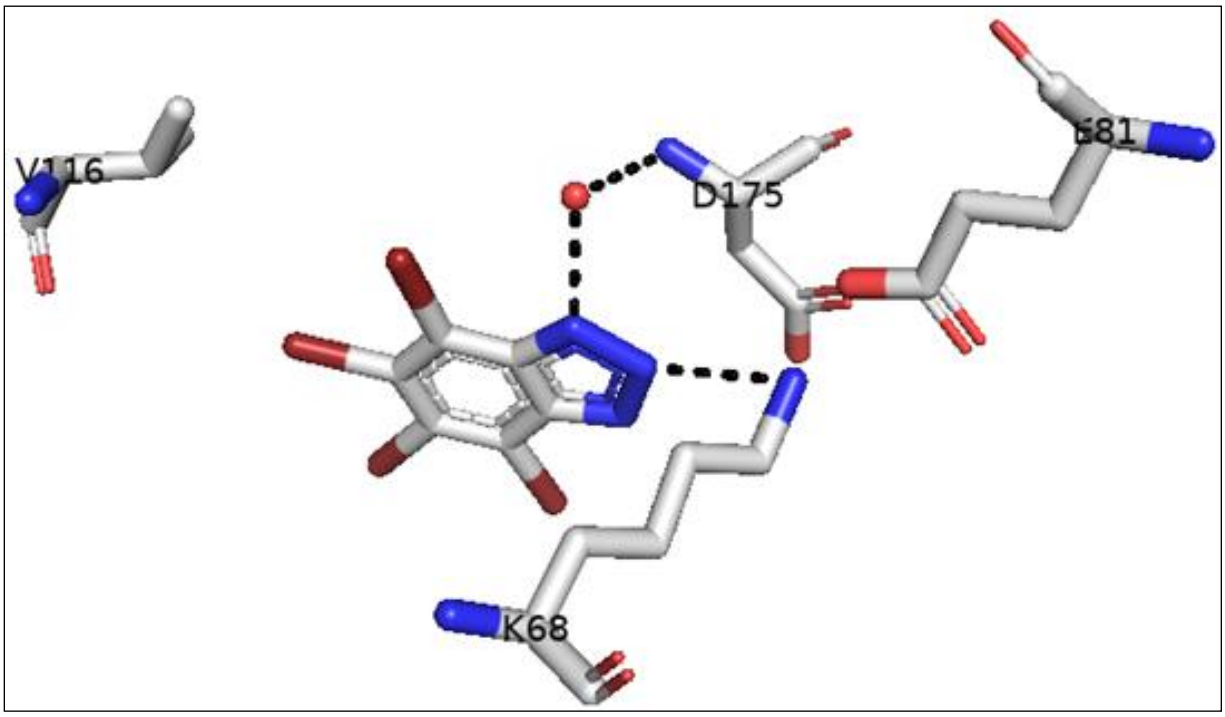
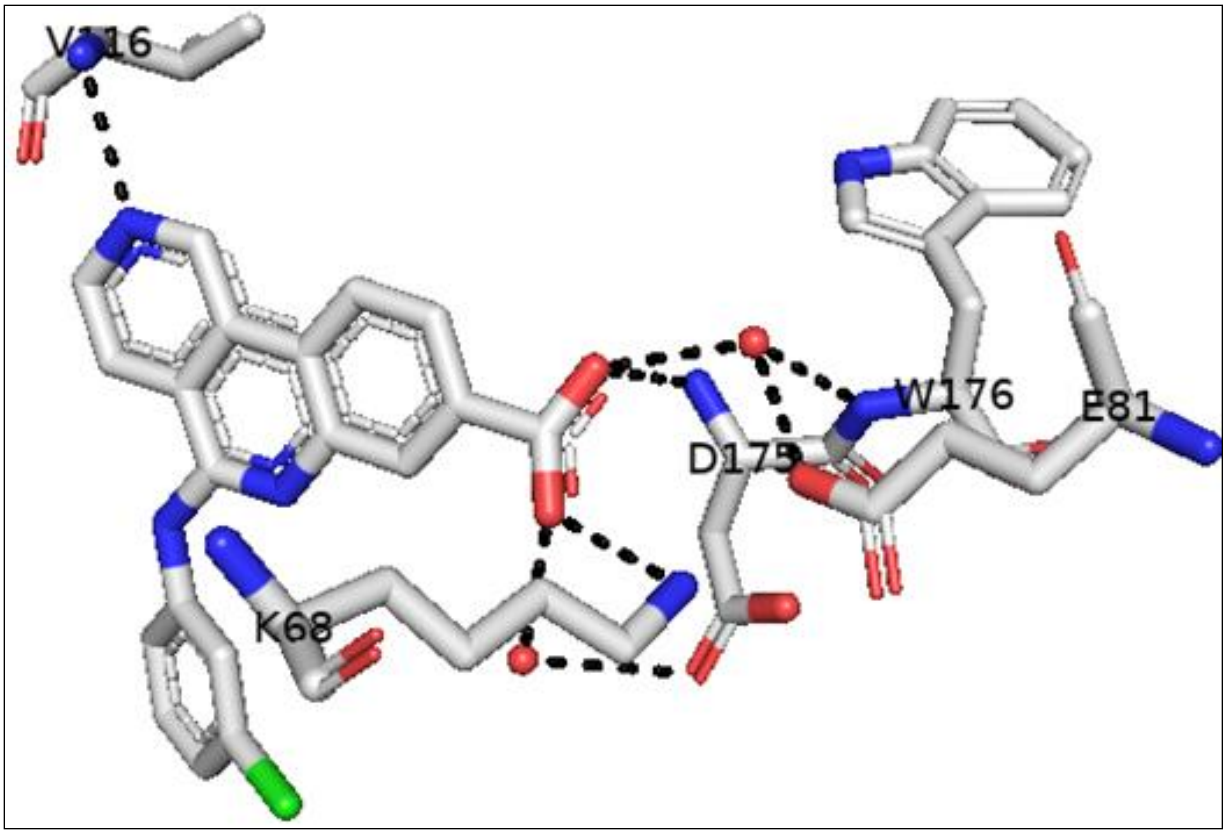
Supporting Information S1: Molecular modelling and docking with CK2α

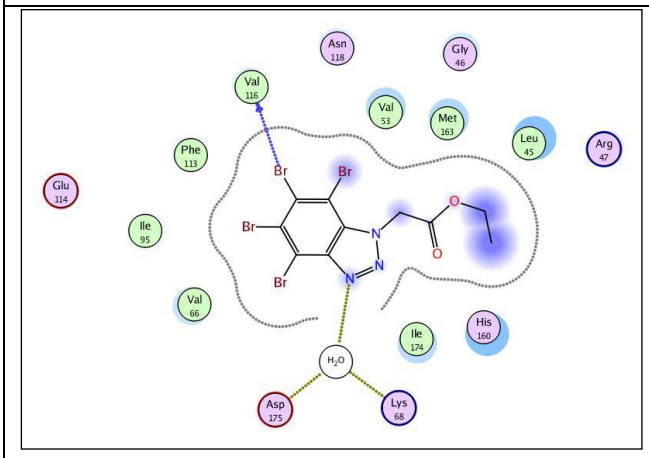
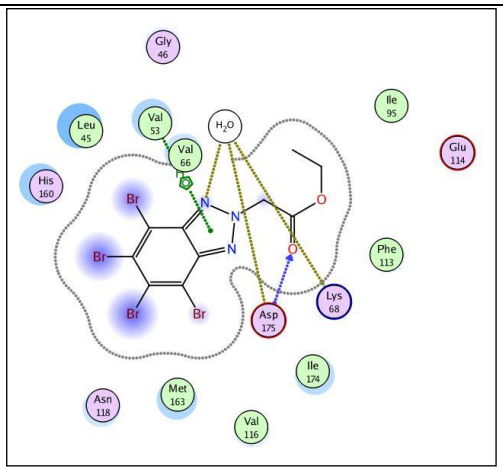
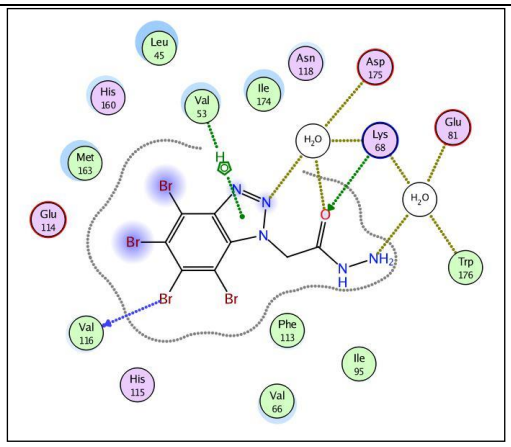
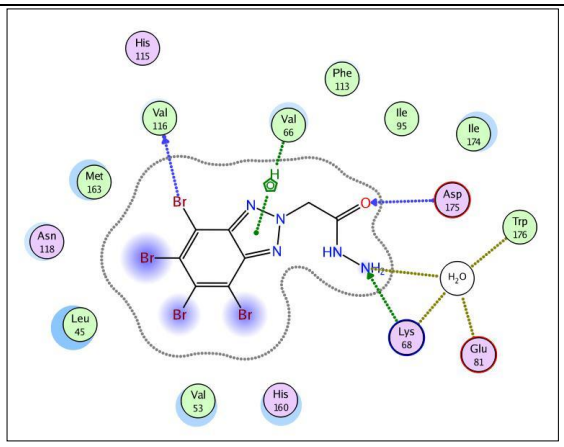
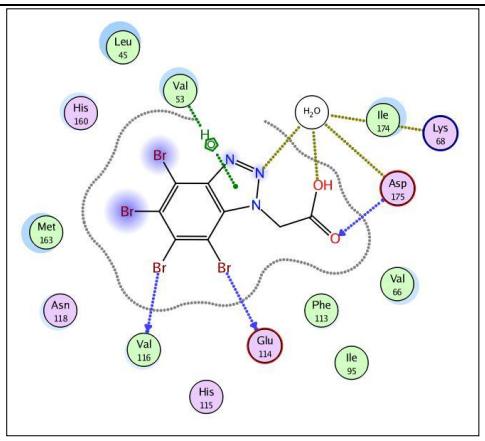
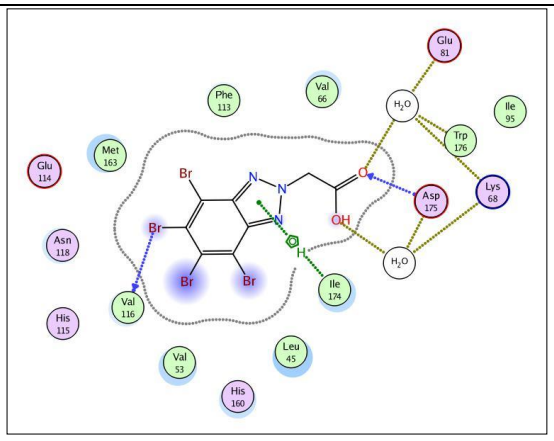
a) Binding of TBBt to CK2α of Zea mays PDB code 1j91[1]



b) Binding of CX-4945 to human CK2α PDB code 3pe1[2]

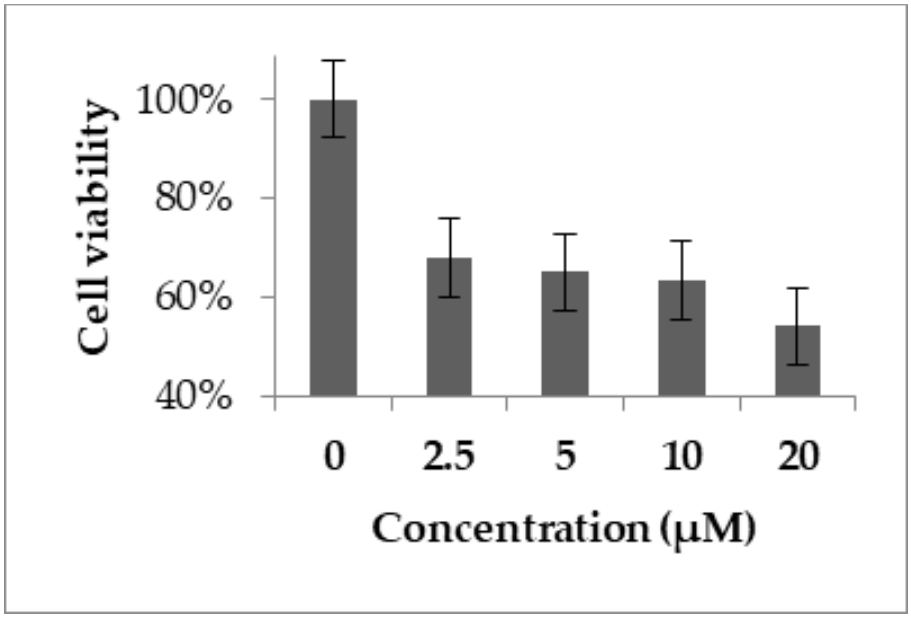


c) 2D representation of docked compounds **2-7** into human CK2α PDB code 3pe1.

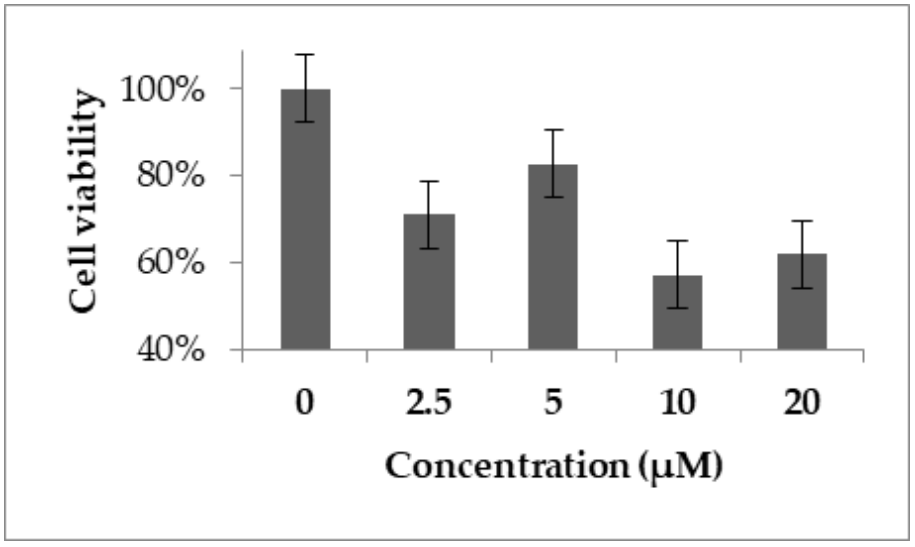
Compound 2	**Compound 3**
	
Compound 4	**Compound 5**
	
Compound 6	**Compound 7**
	

Supporting Information S2: Cell viability of adrenocarcinoma cell lines treated with 0-20 µM of compound **5** for 48 h:

a) Lung cancer celline (A549)



b) Breast cancer cell line (MCF-7)



Supporting Information S3: The R_{M0} Values, C (the percentage of **ACN**), b (Slope) and r (Correlation Coefficient) of the Equation $R_M = R_{M0} + bC$ for the studied compounds

Compound	$R_{M0} \pm SD^a$	$b \pm SD^a$	r	C_0	C range (%)
1	2.0204 ± 0.04229	-0.0268 ± 0.0006	-0.9984	75.3635	50-85
2	3.4181 ± 0.0930	-0.0404 ± 0.0012	-0.9980	84.5186	65-95
3	3.6425 ± 0.1131	-0.0417 ± 0.0015	-0.9976	87.2663	65-90
4	1.3857 ± 0.0761	-0.0240 ± 0.0011	-0.9960	57.7832	55-85
5	2.5564 ± 0.0529	-0.0326 ± 0.0007	-0.9985	78.4901	55-90
6	1.451 ± 0.0795	-0.0304 ± 0.0013	-0.9972	47.7918	50-70
7	1.2837 ± 0.0552	-0.0258 ± 0.0009	-0.9978	49.8534	50-80

Supporting Information S4: The R_{M0} Values, C (the percentage of **acetone**), b (Slope) and r (Correlation Coefficient) of the Equation $R_M = R_{M0} + bC$ for the studied compounds

Compound	$R_{M0} \pm SD^a$	$b \pm SD^a$	r	C_0	C range (%)
1	3.1568 ± 0.1224	-0.0428 ± 0.0017	-0.9944	73.7719	50-90
2	4.1509 ± 0.1421	-0.0512 ± 0.0018	-0.9970	81.1310	65-95
3	4.5127 ± 0.1152	-0.0543 ± 0.0015	-0.9980	83.0556	60-90
4	3.0668 ± 0.1654	-0.0412 ± 0.0023	-0.9911	74.5265	55-90

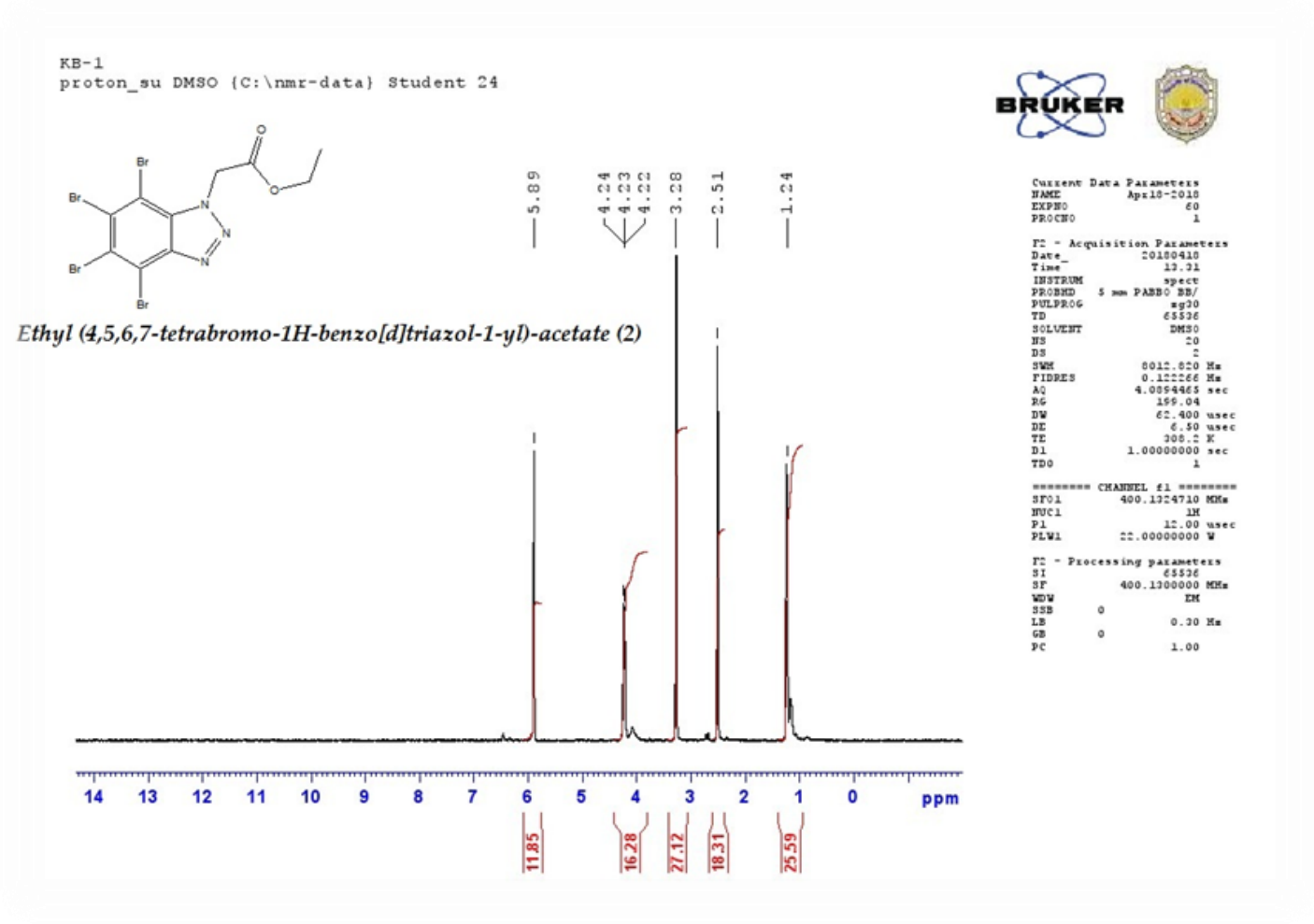
5	3.6874 ± 0.1008	-0.0470 ± 0.0013	-0.9972	78.4945	55-95
6	1.5057 ± 0.0560	-0.0271 ± 0.0008	-0.9971	55.6552	50-95
7	2.1072 ± 0.0630	-0.0351 ± 0.0009	-0.9980	60.0623	50-90

Supporting Information S5: The R_{M0} Values, C (the percentage of **MeOH**), b (Slope) and r (Correlation Coefficient) of the Equation $R_M = R_{M0} + bC$ for the studied compounds

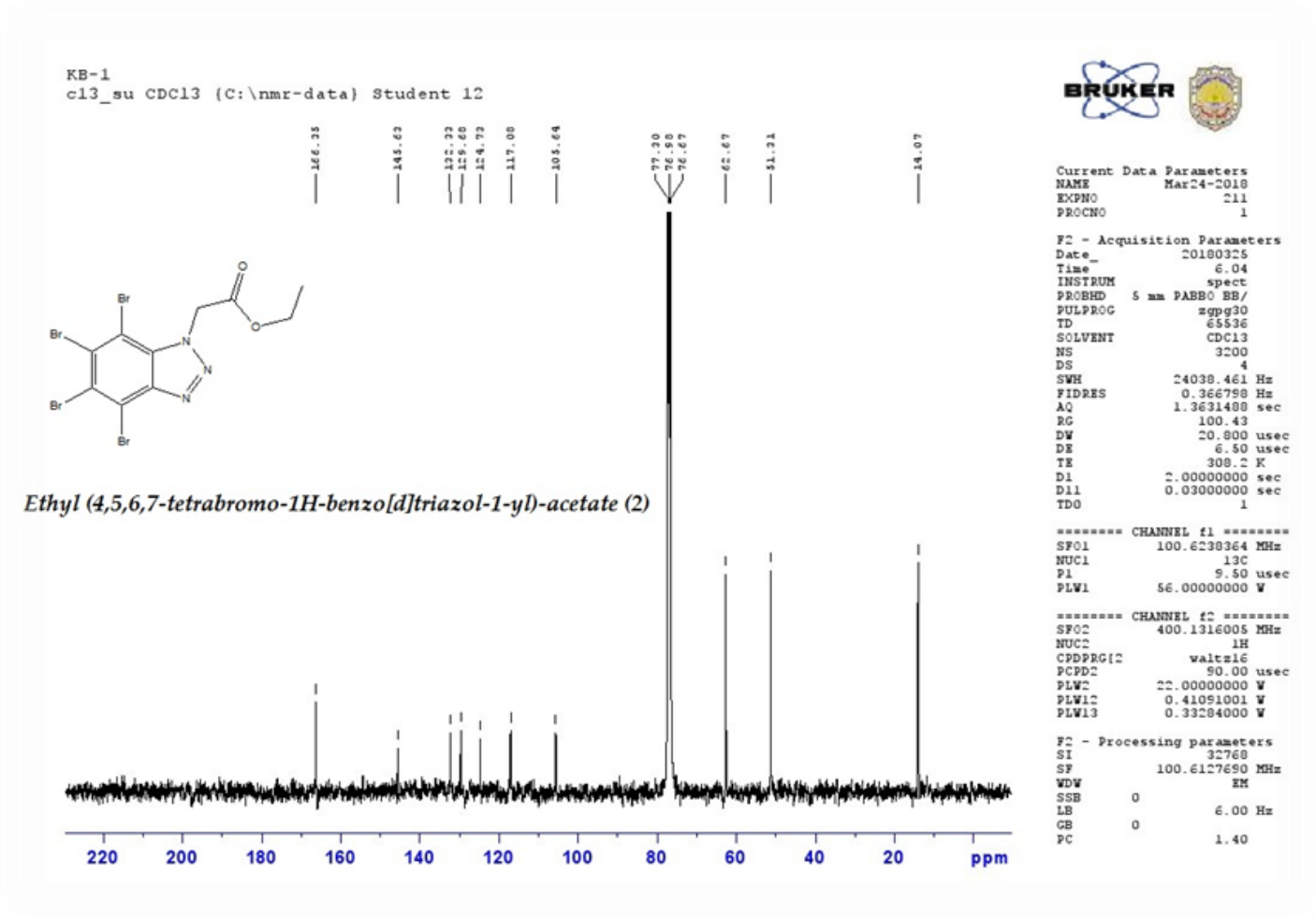
Compound	**R_{M0} ± SD[a]**	**b ± SD[a]**	**r**	**C_0**	**C range (%)**
1	3.5926 ± 0.0943	-0.0416 ± 0.0015	-0.9978	86.3354	60-90
2	4.0138 ± 0.1422	-0.0429 ± 0.0018	-0.9975	93.6391	70-90
3	4.6874 ± 0.5309	-0.0488 ± 0.006	-0.9831	96.1315	75-90
4	2.2595 ± 0.1136	-0.0286 ± 0.0015	-0.9932	79.0453	60-90
5	3.8767 ± 0.1883	-0.0438 ± 0.0023	-0.9957	88.5524	70-90
6	2.6161 ± 0.0908	-0.0397 ± 0.0013	-0.9974	65.8293	50-85
7	3.3822 ± 0.1099	-0.0465 ± 0.0015	-0.9979	72.6917	60-85

Supporting Information S6: IR, H-NMR and C13-NMR spectra

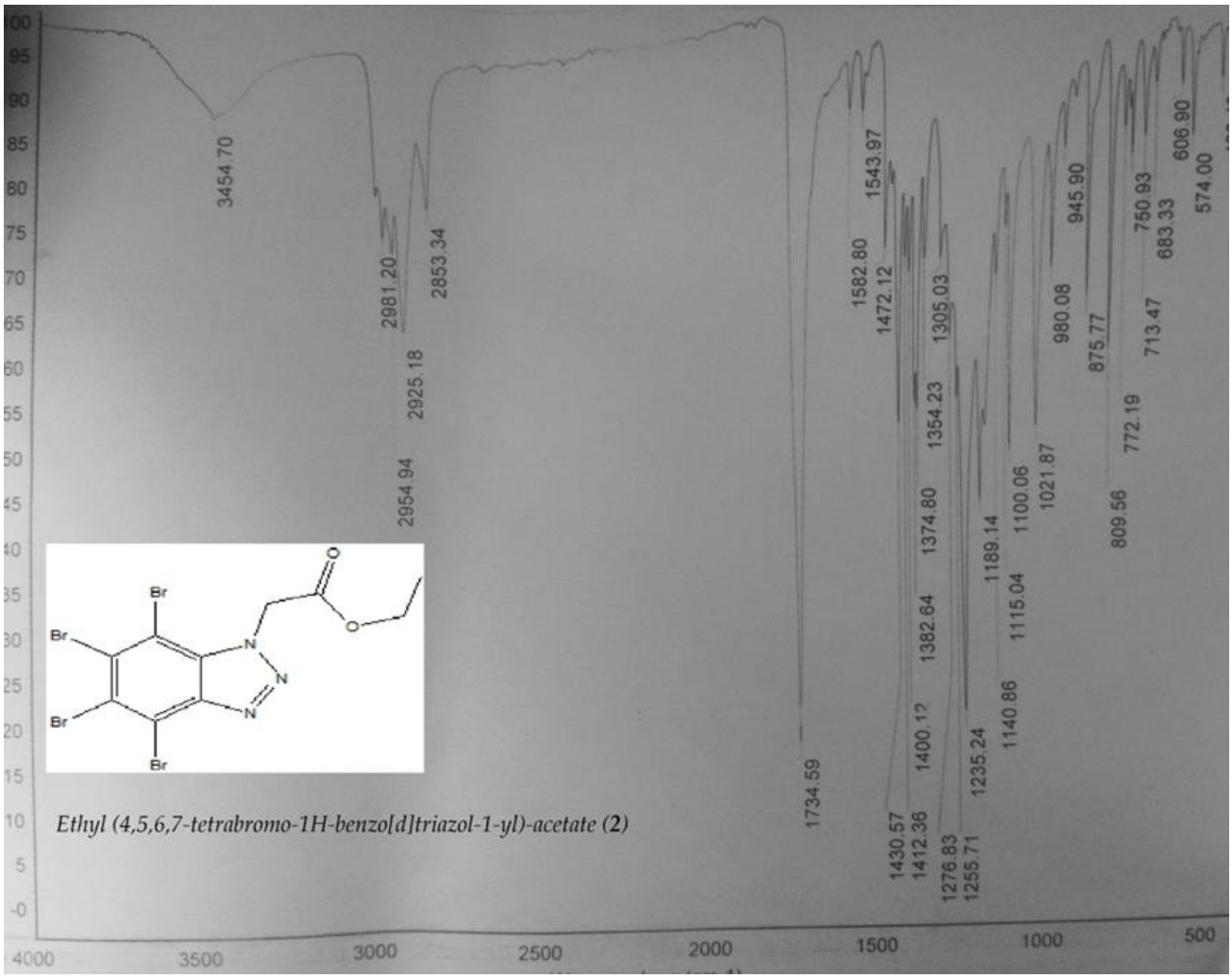
1- H^1-NMR spectrum of compound 2



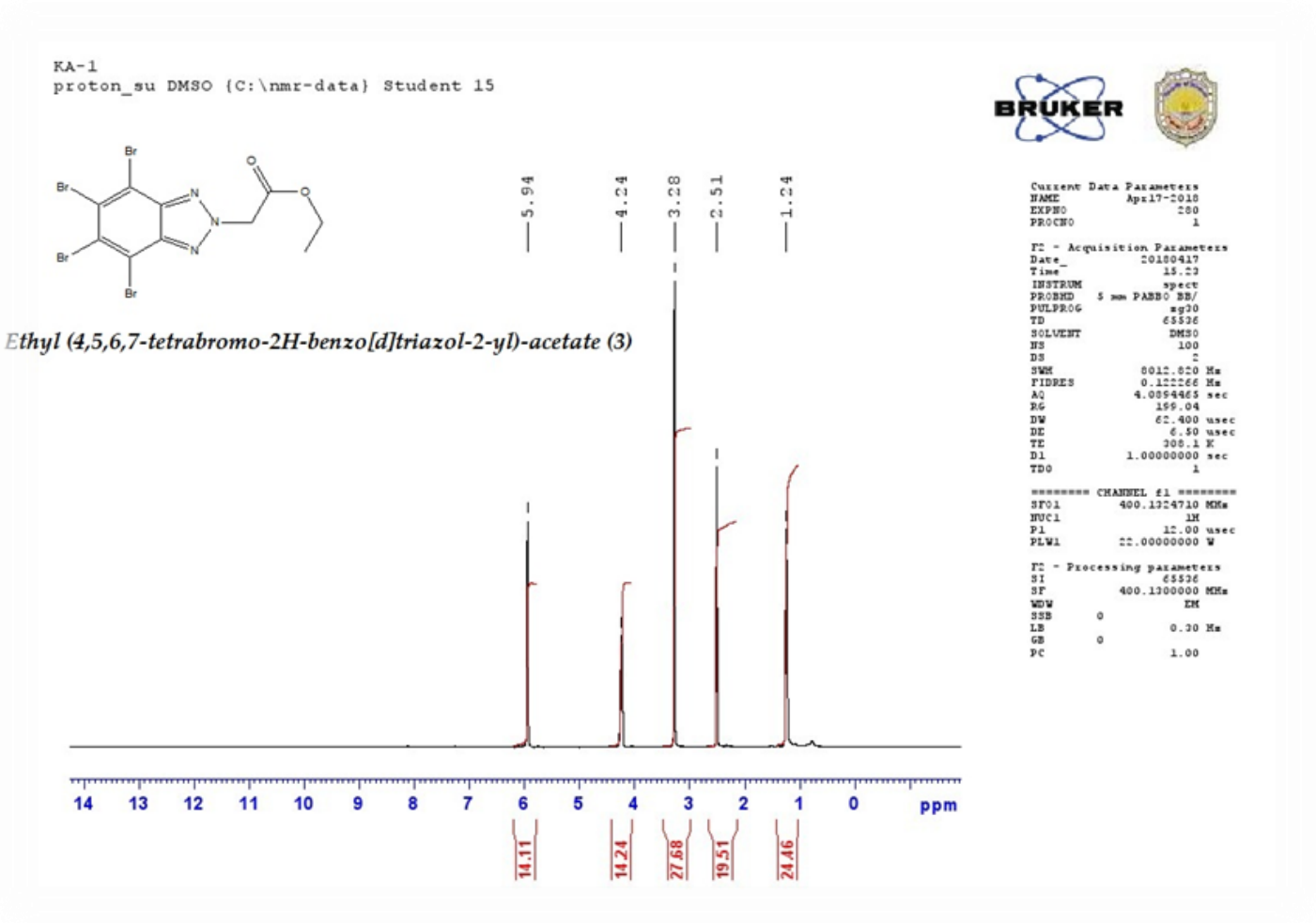
2- C^{13}-NMR spectrum of compound 2



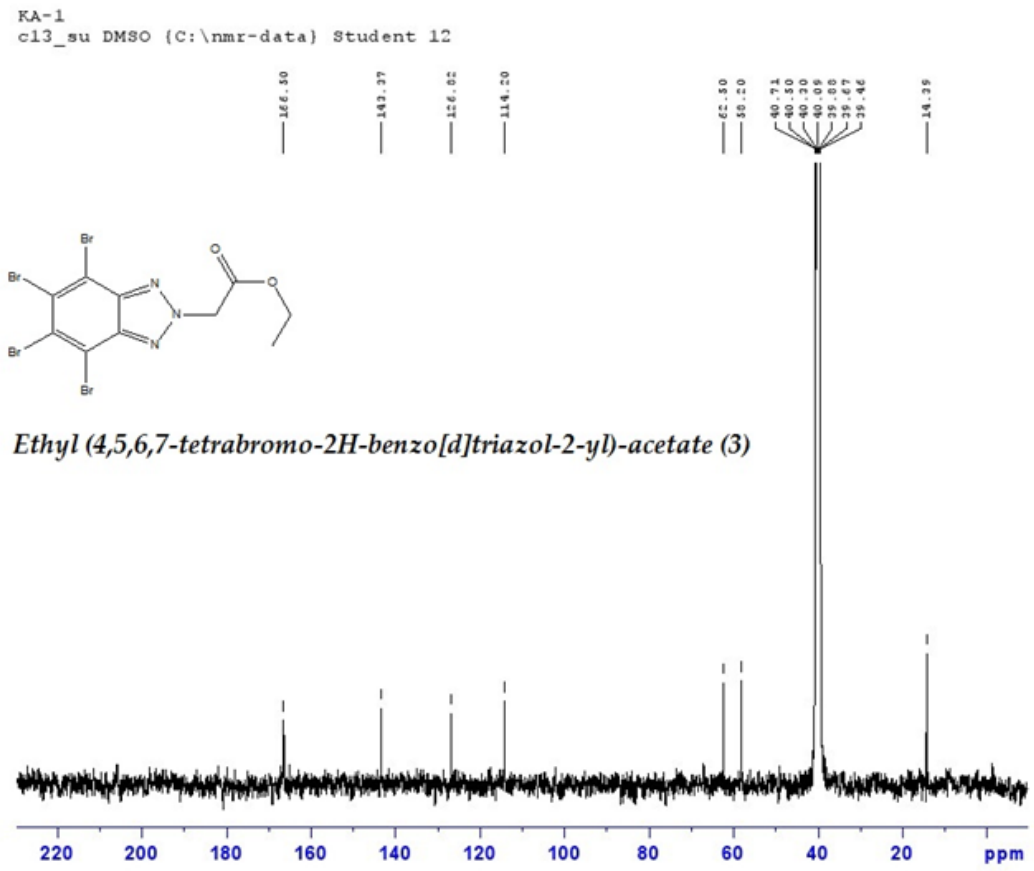
4- IR spectrum of compound 2



5- H[1]-NMR spectrum of compound 3



6- C^{13}-NMR spectrum of compound 3

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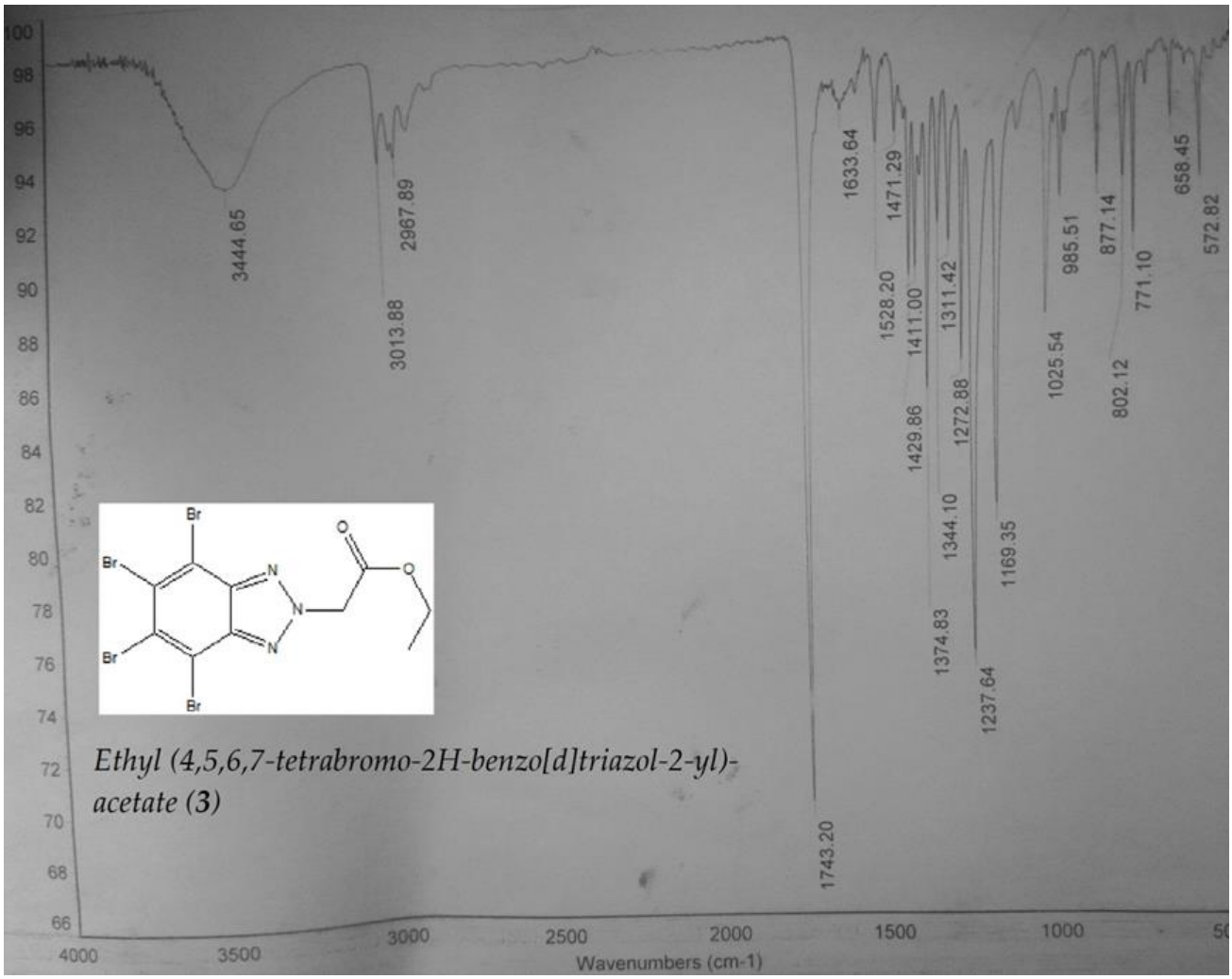
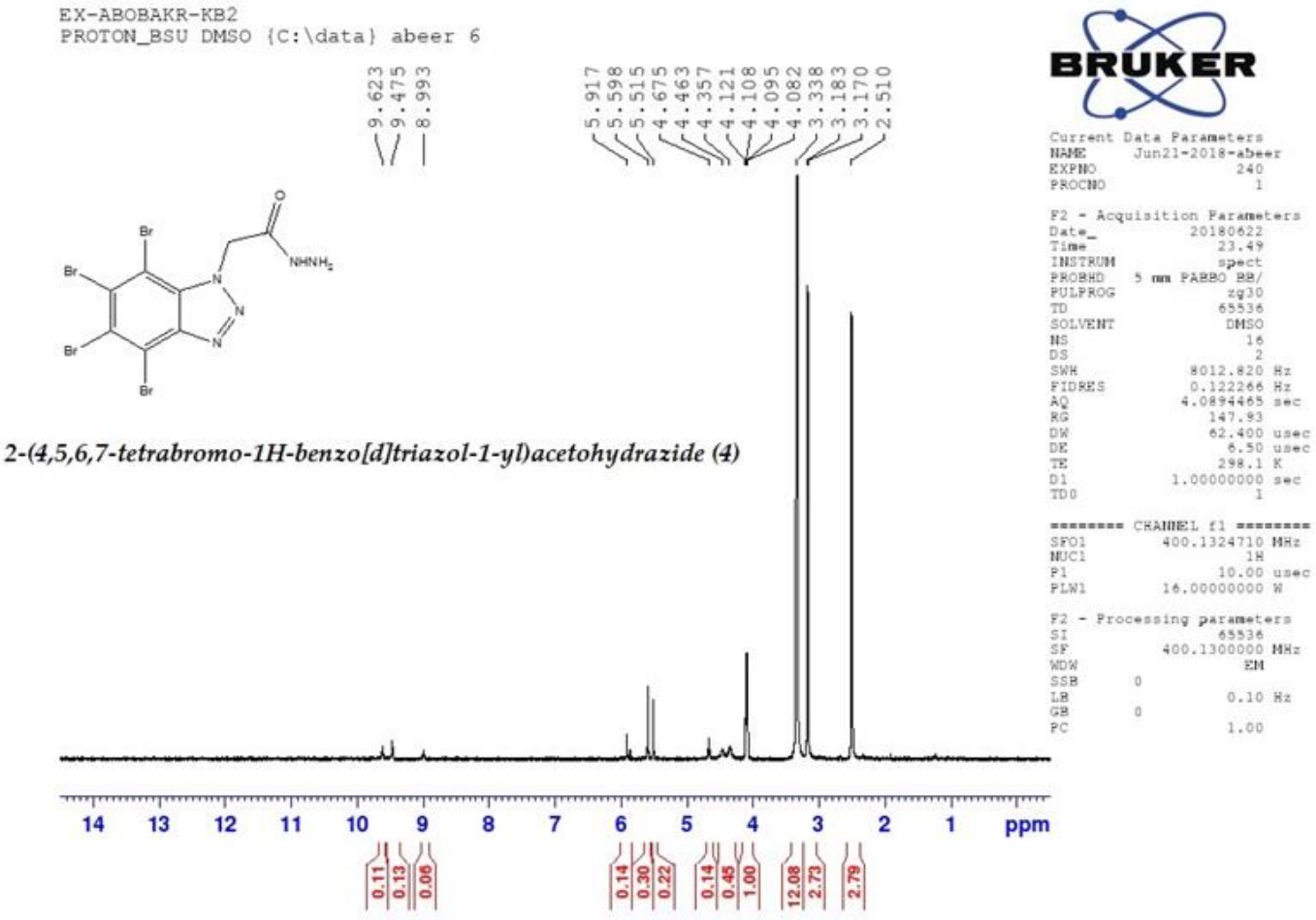


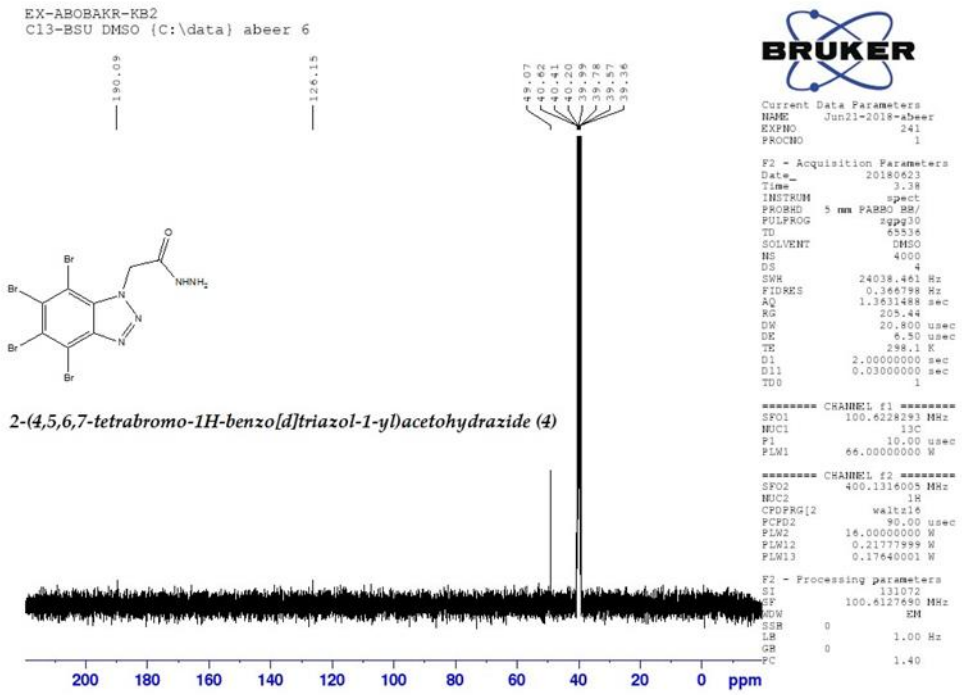
*Ethyl (4,5,6,7-tetrabromo-2H-benzo[d]triazol-2-yl)-acetate (**3**)*

8- H^1-NMR spectrum of compound 4

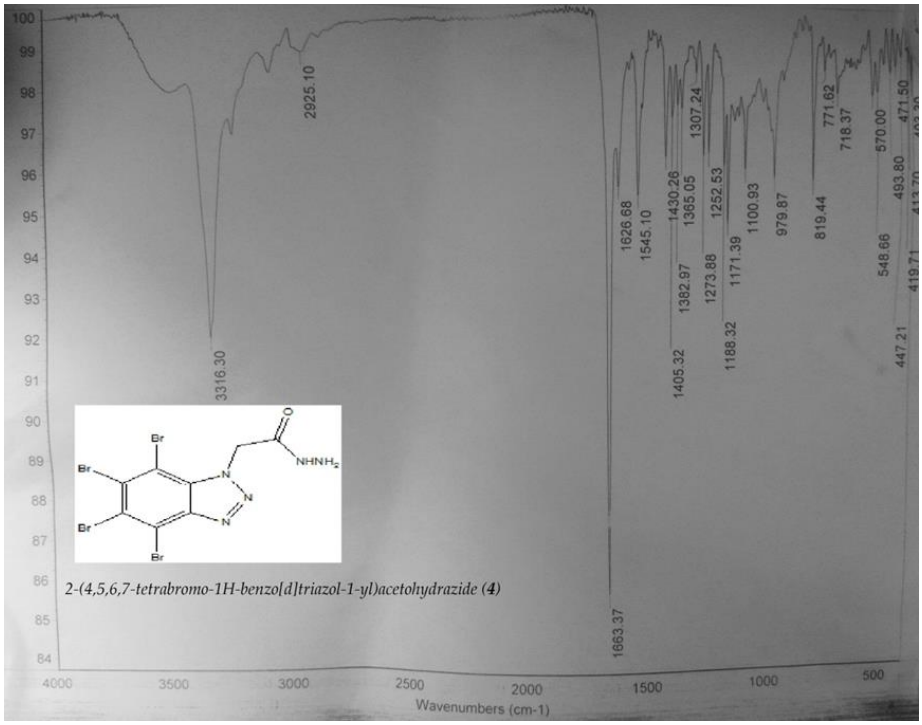


2-(4,5,6,7-tetrabromo-1H-benzo[d]triazol-1-yl)acetohydrazide (4)

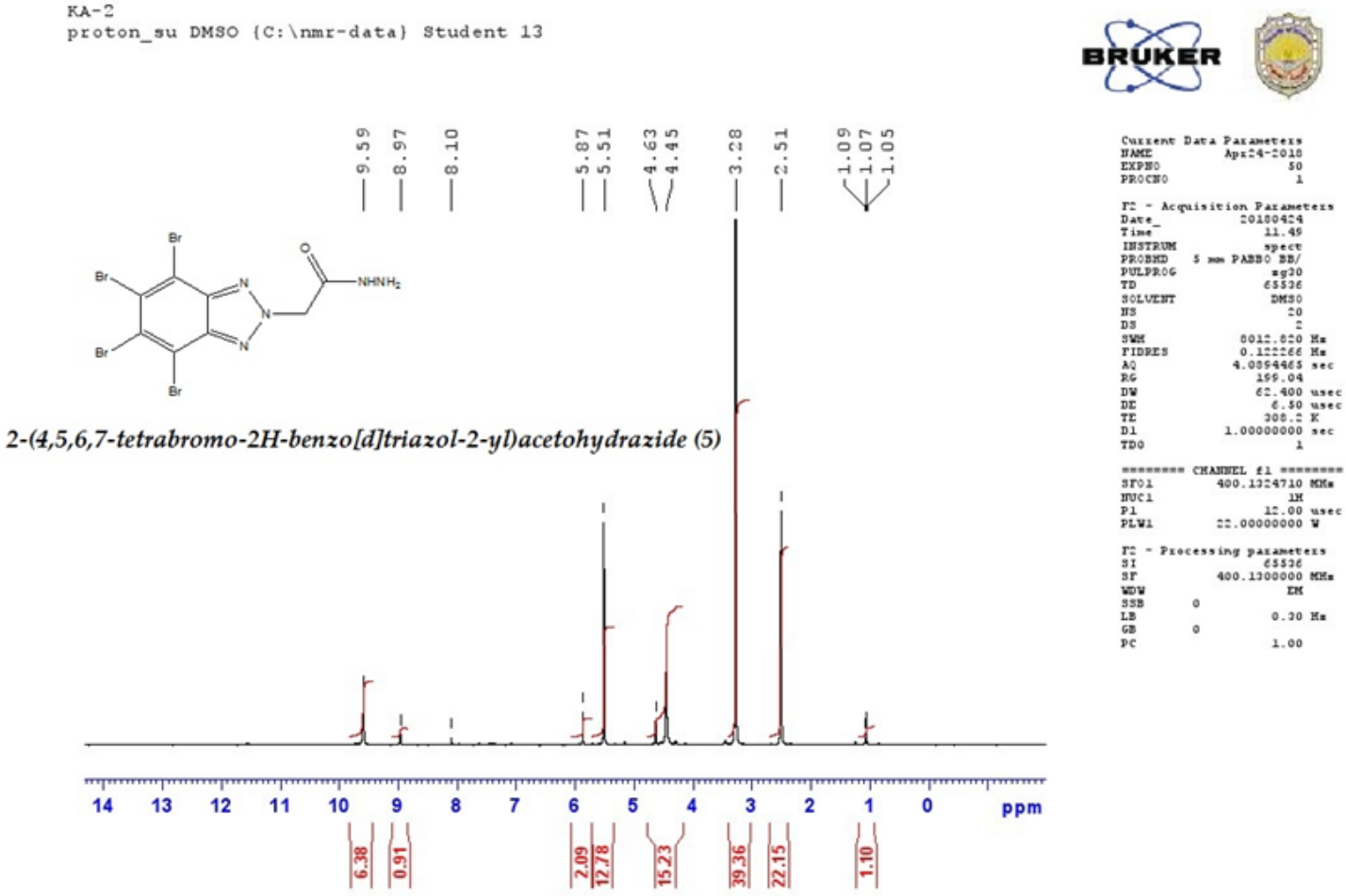
9- C13-NMR spectrum of compound 4



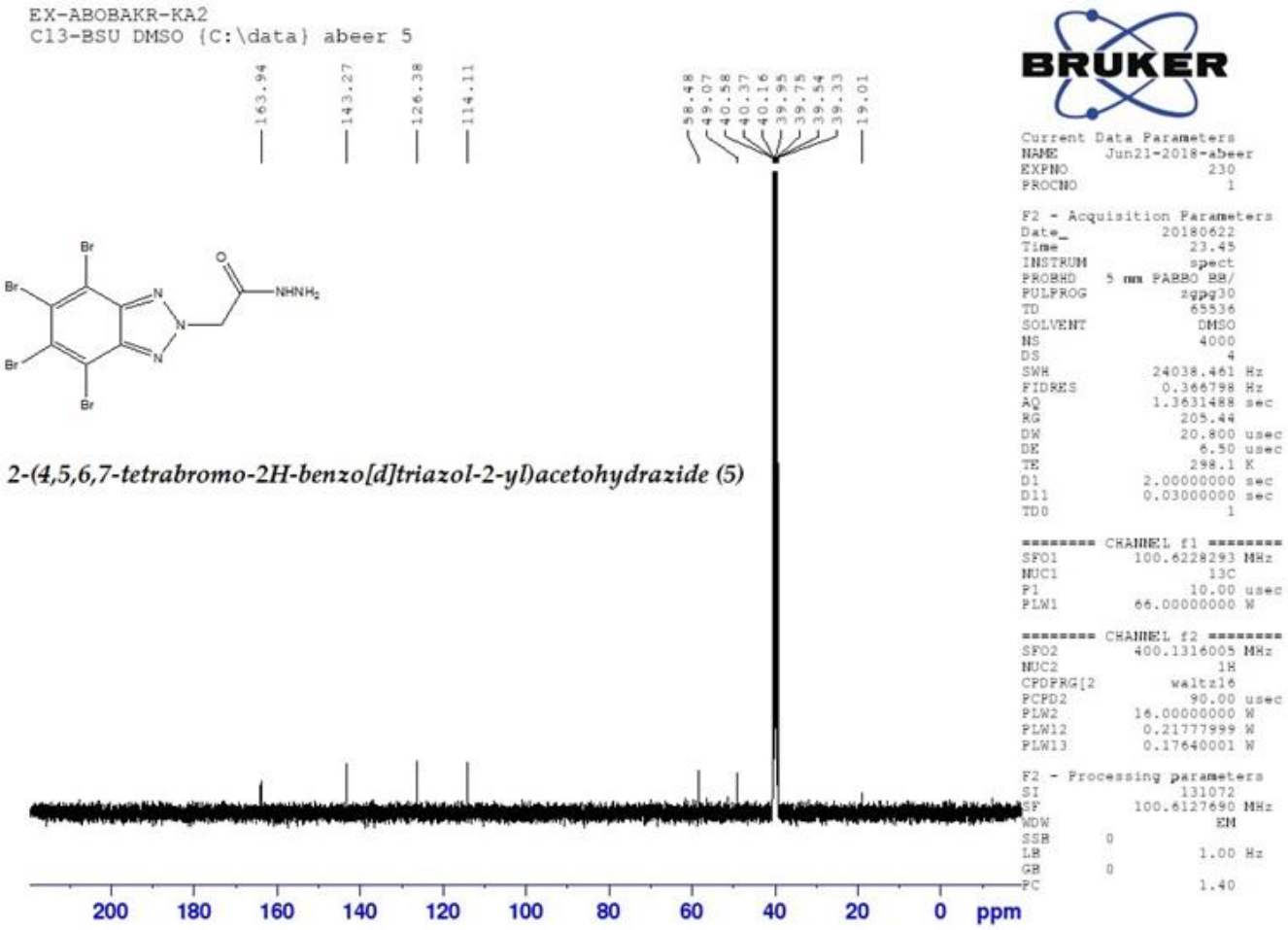
10- IR spectrum of compound 4



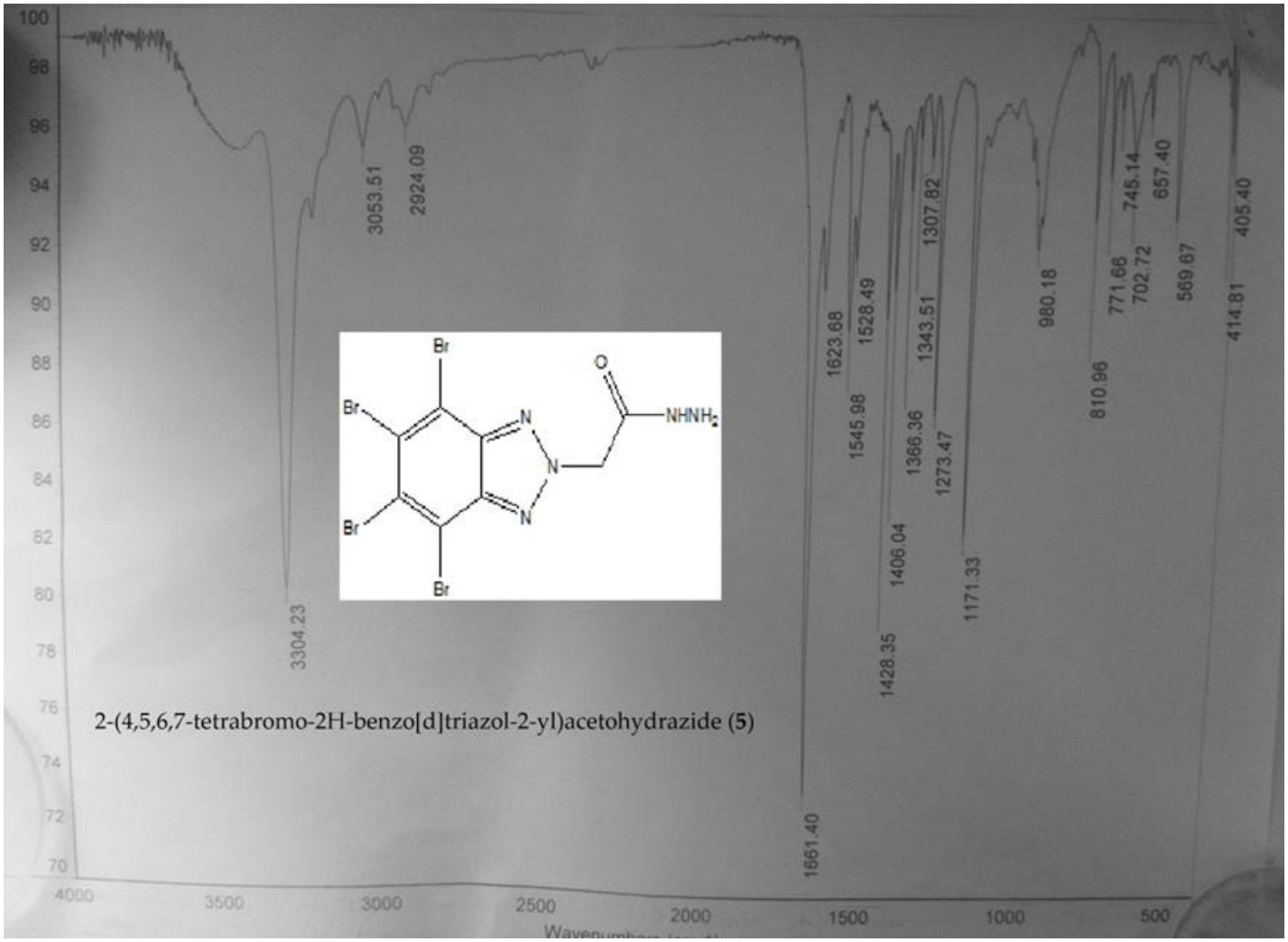
11- H[1]-NMR spectrum of compound 5



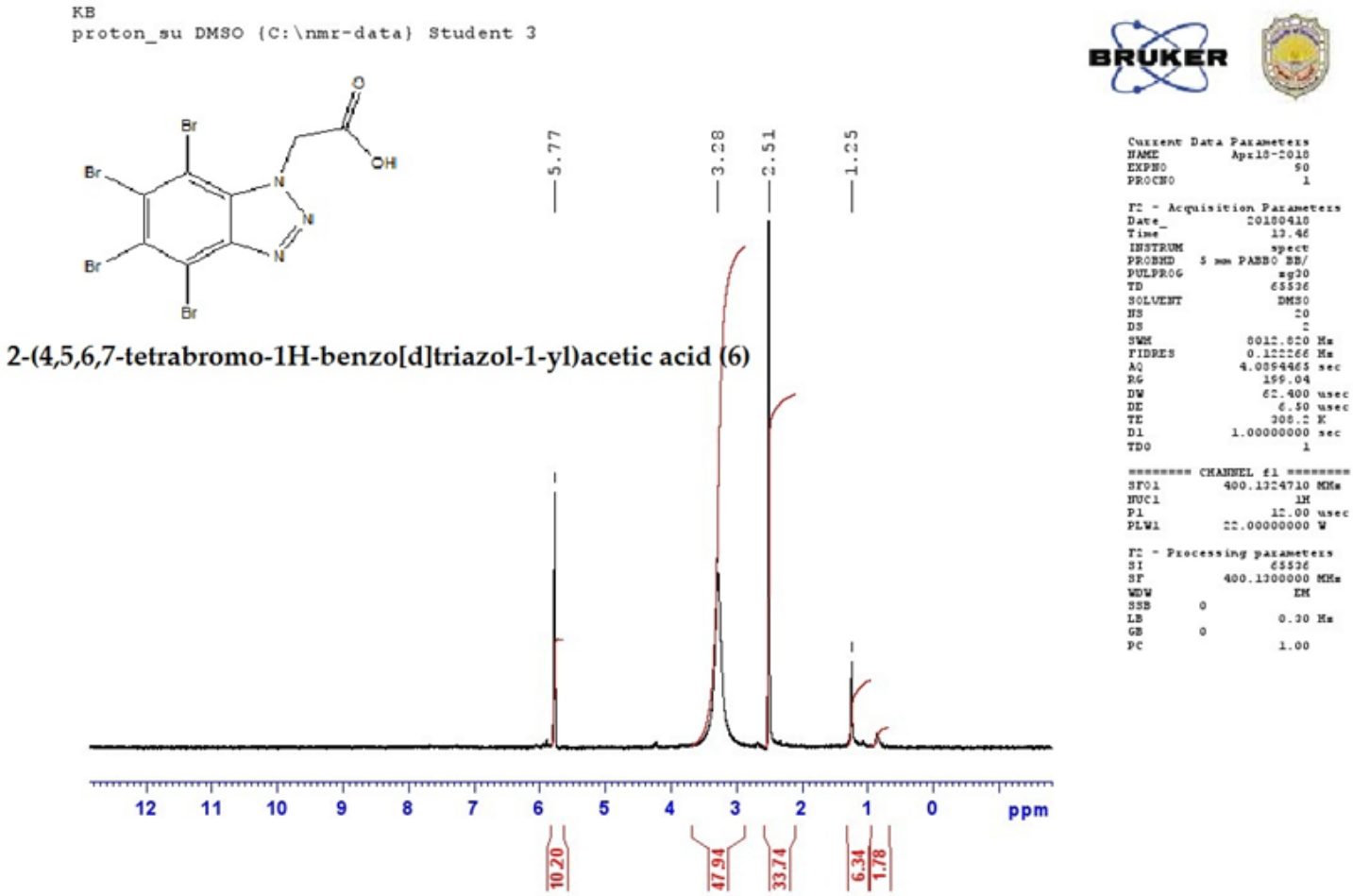
12- C13-NMR spectrum of compound 5



13- IR spectrum of compound 5

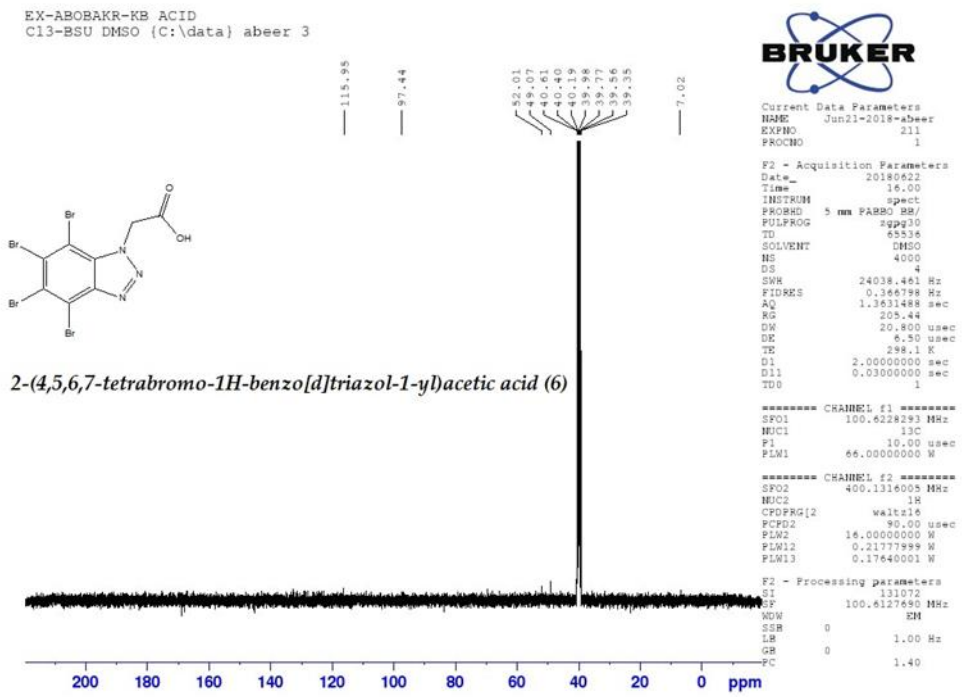


14- H1-NMR spectrum of compound 6



2-(4,5,6,7-tetrabromo-1H-benzo[d]triazol-1-yl)acetic acid (6)

15- C13-NMR spectrum



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TE 298.1 K
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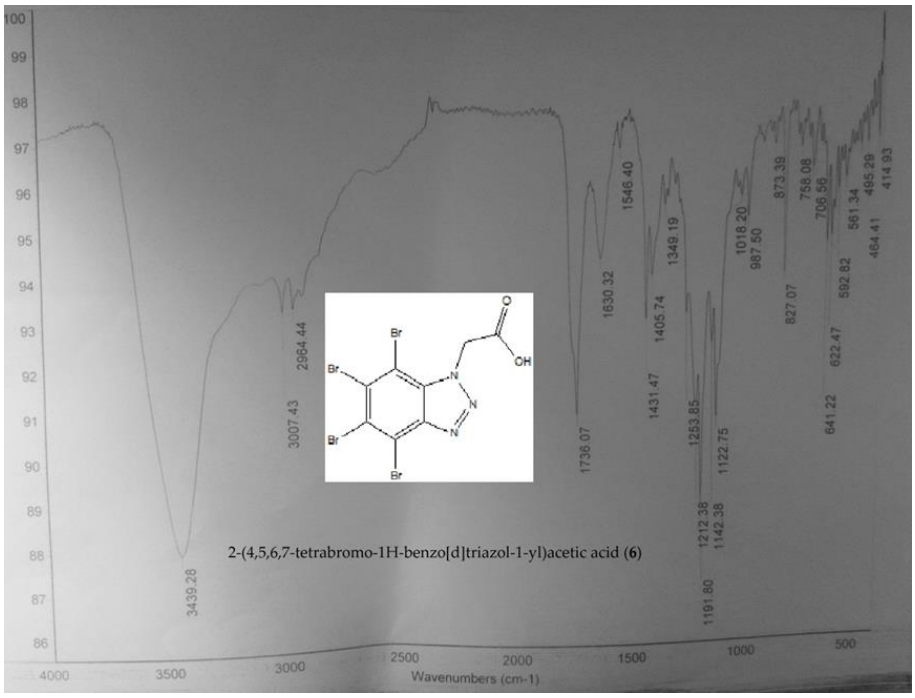
16- IR spectrum of compound 6



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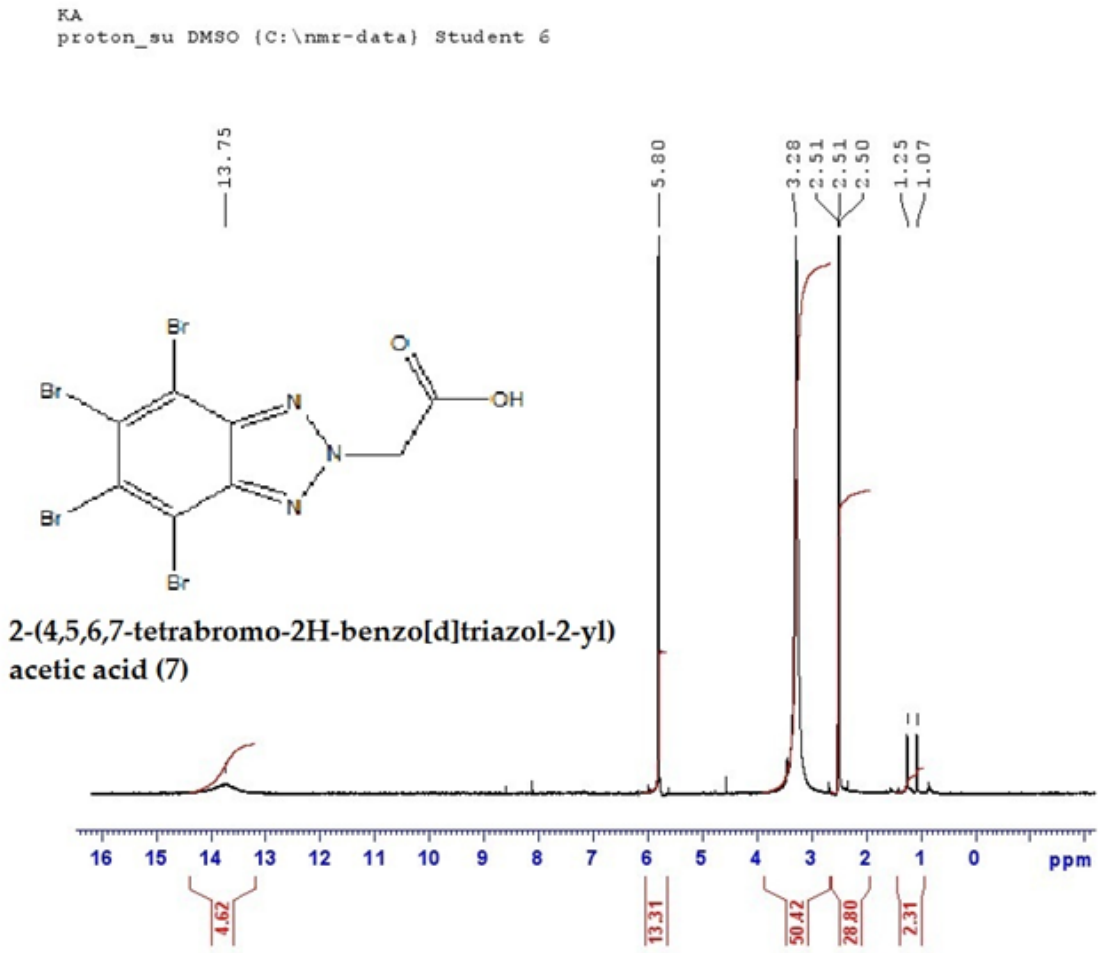
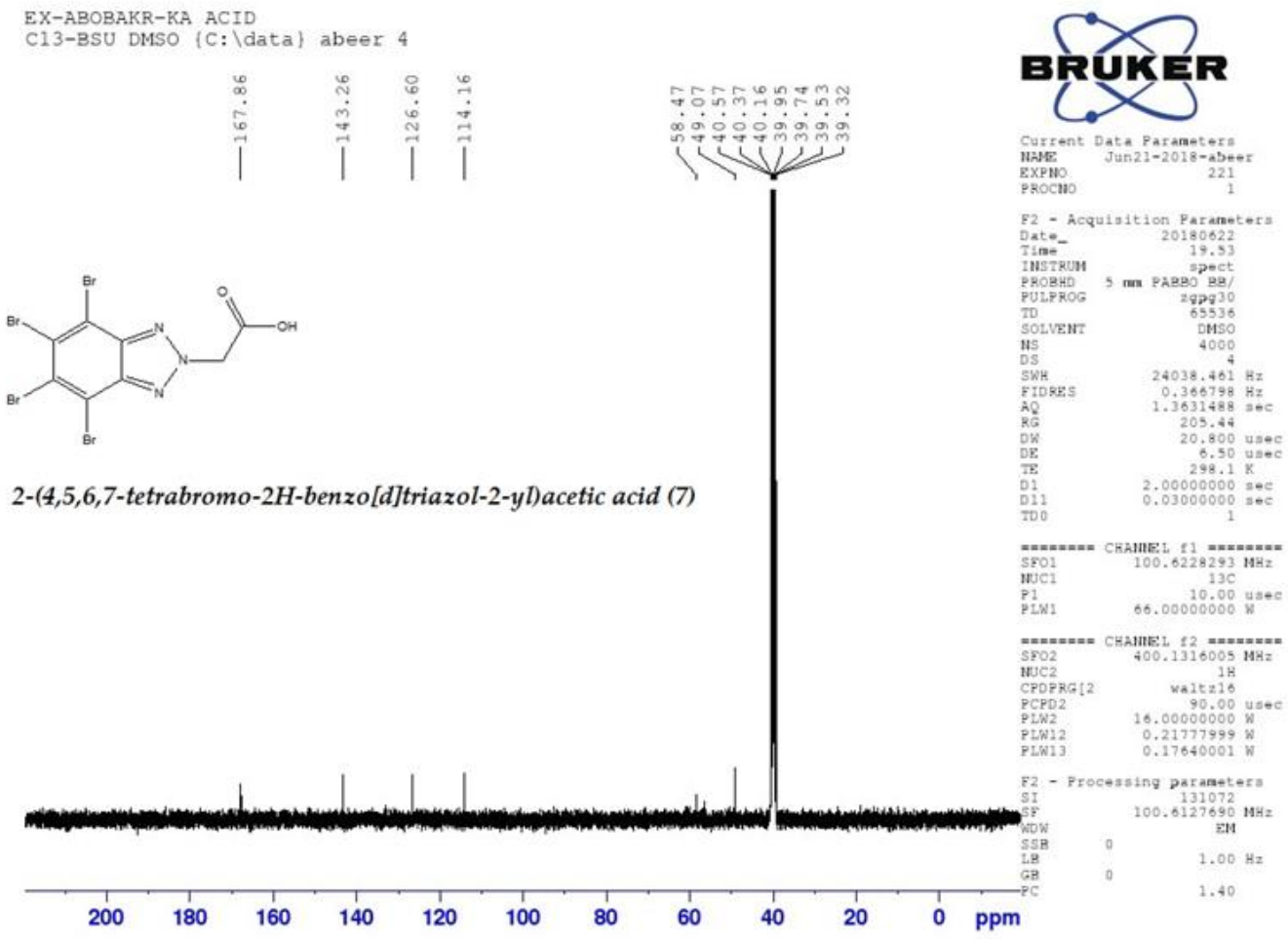
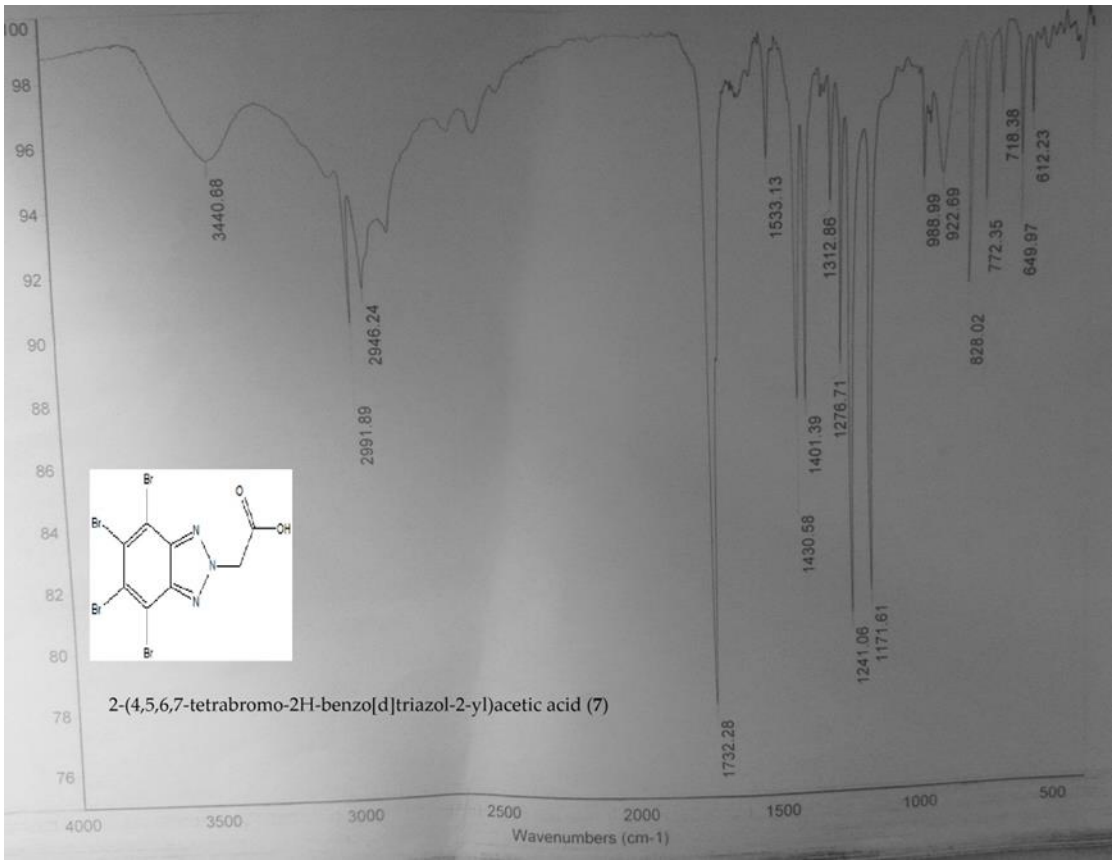


2-(4,5,6,7-tetrabromo-2H-benzo[d]triazol-2-yl) acetic acid (7)

18- C13-NMR spectrum of compound 7



19- IR spectrum of compound 7



References

1. Battistutta, R.; De Moliner, E.; Sarno, S.; Zanotti, G.; Pinna, L. A., Structural features underlying selective inhibition of protein kinase CK2 by ATP site-directed tetrabromo-2-benzotriazole. *Protein science : a publication of the Protein Society* **2001,** *10* (11), 2200-6.
2. Battistutta, R.; Cozza, G.; Pierre, F.; Papinutto, E.; Lolli, G.; Sarno, S.; O'Brien, S. E.; Siddiqui-Jain, A.; Haddach, M.; Anderes, K.; Ryckman, D. M.; Meggio, F.; Pinna, L. A., Unprecedented selectivity and structural determinants of a new class of protein kinase CK2 inhibitors in clinical trials for the treatment of cancer. *Biochemistry* **2011,** *50* (39), 8478-88.