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Supplementary material

CARBONIC ANHYDRASE INHIBITORY ACTIVITIES OF NOVEL PROTON TRANSFER SALTS AND THEIR Cu(II) COMPLEXES

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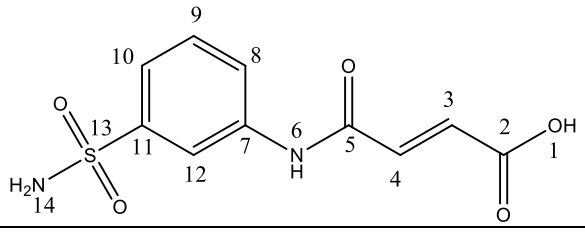
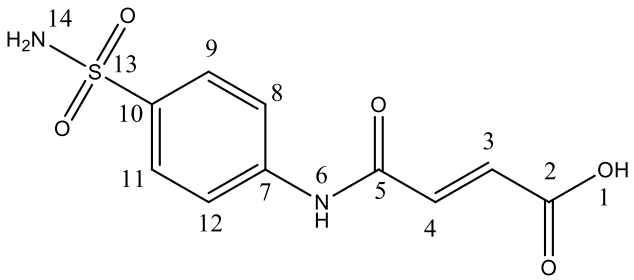
Table S1. ^{1}H-NMR and ^{13}C-NMR chemical shifts (ppm) with coupling constants and assignments for compound Hmabsmal



H^{1}	12.95 (1H s)	C_{2}	164
H^{3}	6.48 (1H, d) [$^{3}J_{H3\text{-}H4}$ = 11.98 Hz]	C_{3}	131
H^{4}	6.34 (1H, d) [$^{3}J_{H4\text{-}H3}$ = 11.96 Hz]	C_{4}	132
H^{6}	10.60 (1H, s)	C_{5}	167
H^{8}	7.55 (1H, d) [$^{3}J_{H8\text{-}H9}$ = 7.26 Hz]	C_{7}	140
H^{9}	7.55 (1H, t) [$^{3}J_{H9\text{-}H8,10}$ = 7.72 Hz]	C_{8}	121
H^{10}	7.75 (1H, d) [$^{3}J_{H10\text{-}H9}$ = 6.83 Hz]	C_{9}	117
H^{12}	8.24 (1H, s)	C_{10}	123
H^{14}	7.39 (2H, s)	C_{11}	145
		C_{12}	130

Table S2. ^{1}H-NMR and ^{13}C-NMR chemical shifts (ppm) with coupling constants and assignments for compound HSAMAL



H^{1}	12.9 (1H, s)	C^{2}	168 ppm
H^{3}	6.50 (1H, d) [$^{3}J_{H3\text{-}H4}$ = 11.96 Hz]	C^{3}	137 ppm
H^{4}	6.36 (1H, d) [$^{3}J_{H4\text{-}H3}$ = 11.95 Hz]	C^{4}	119 ppm
H^{6}	10.8 (1H, s)	C^{5}	164 ppm
H^{8}, H^{9}	7.80 (4H, s)	C^{7}	139 ppm
H^{12}	7.30 (2H, s)	C^{8}	132 ppm
		C^{9}	131 ppm
		C^{10}	142 ppm

Table S3 IR spectral data of **1-4** (cm^{-1})

	***m*absmal**	***p*absmal**	**Clabt**	**1**	**2**	**3**	**4**	**5**
ν(OH)	2900(br)	2900(br)	-	-	-	3544(br)	3453(br)	3490(br)
$\nu(NH_2)$	3315(m)	3352(m)	3453(m)	3392(m)	3388(m)	3385(m)	3391(m)	3331(m)
	3229(m)	3263(m)	3265(m)	3339(m)	3304(m)	3223(m)	3256(m)	3261(m)
	3205(m)	3212(m)		3313(m)	3261(m)	3304(m)	3240(m)	3294(m)
				3282(m)	3239(m)	3271(m)	3174(m)	3234(m)
				3187(m)	3207(m)	3244(m)		3211(m)
$\nu(NH)^+$	-	-	-	2724(w)	2759(w)	-	-	-
				2462(w)	2535(w)			
$\nu(CH)_{Ar.}$	3095(w)	3068(w)	3058(w)	3040(w)	3062(w)	3078(w)	3064(w)	3073(w)
$\nu(C\text{-}H)_{Alf.}$	3040(w)	3011(w)	-	2960(w)	2979(w)	3051(w)	2990(w)	2999(w)
	2934(w)	2952(w)		2865(w)	2921(w)	2935(w)	2976(w)	2953(w)
	2984(w)	2807(w)		2818(w)	2851(w)	2857(w)	2889(w)	2871(w)
$\nu(C{=}O)_{amit}$	1620(s)	1630(w)	-	1620(s)	1612(s)	1617(s)	1641(s)	1600(s)
$\nu(C{=}O)_{asit}$	1688(s)	1695(s)	-	1665(s)	1658(s)	1641(s)	1691(s)	1666(s)
ν(C=N)	1578(s)	1549(s)	1627(s)	1594(s)	1609(s)	1574(s)	1592(s)	1596(s)
ν(C=C)	1541(s)	1496(s)	1528(s)	1524(s)	1552(s)	1538(s)	1542(s)	1541(s)
	1492(s)	1468(s)	1441(s)	1473(s)	1496(s)	1486(s)	1529(s)	1477(s)
	1472(s)	1425(s)	1427(s)	1427(s)	1460(s)	1425(s)	1495(s)	1440(s)
	1435(s)	1401(s)			1401(s)		1443(s)	
ν(C-O)	1343(s)	1397(s)	-	1355(s)	1335(s)	1352(s)	1368(s)	1341(s)
	1211(s)	1194(s)		1210(s)	1211(s)	1266(s)	1201(s)	1278(s)
	1090(s)	1093(s)		1098(s)	1098(s)	1054(s)	1092(s)	1085(s)
ν(S=O)	1211(s)	1264(s)	-	1250(s)	1259(s)	1245(s)	1258(s)	1247(s)
	1162(s)	1153(s)		1184(s)	1152(s)	1150(s)	1150(s)	1142(s)
ν(M-N)	-	-	-	-	-	550(w)	585(w)	-
ν(M-O)	-	-	-	-	-	454(w)	498(w)	462(w)

[a] abbreviations: w, weak; m, medium; s, strong; b, broad

Table S4 Thermal analyses results of compounds **3-5**

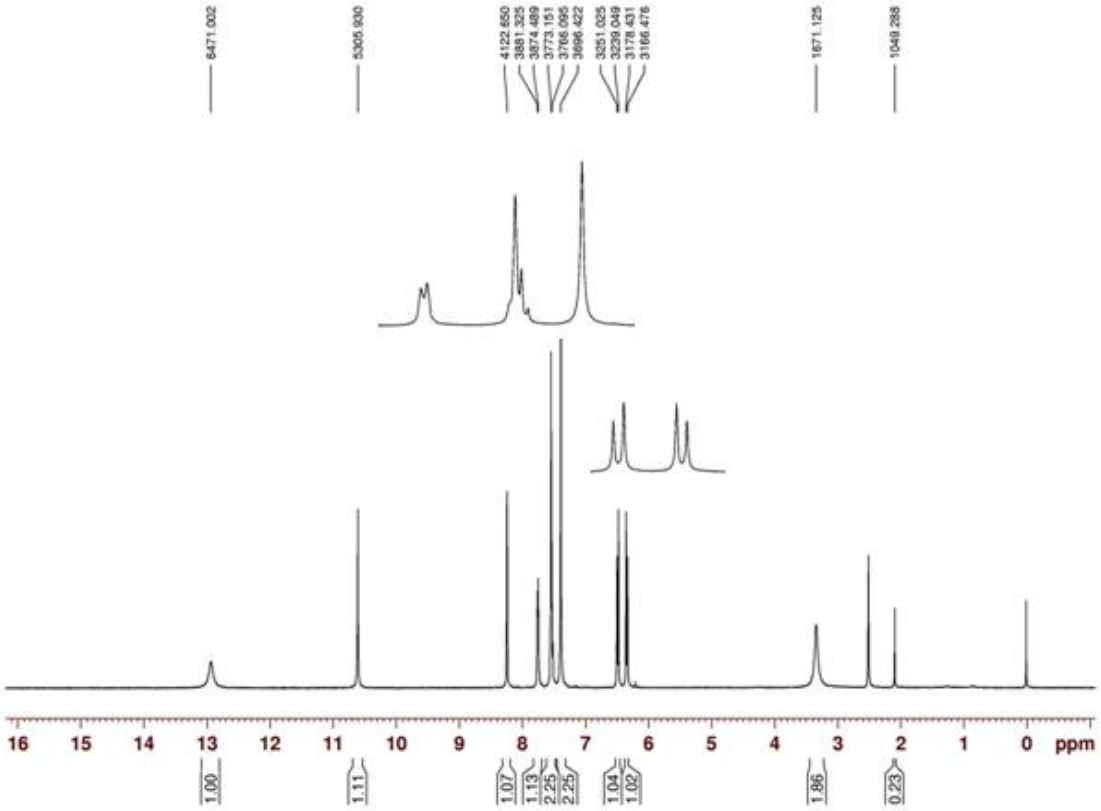
Compound	**Temperature (ºC)**	**DTG_{max} (ºC)**	**Leaving Group**	**Found (%)**	**Calculated (%)**
3	30-140	136	$3H_2O$	5.30	5.27
	140-350	140, 263, 300	$C_{18}H_{18}N_4O_6S_2$	43.70	43.93
	350-900	436, 561	$C_{16}H_{10}N_4O_4S_2Cl_2$	44.50	44.60
	-	-	Cu	6.50	6.20
4	30-135	89	H_2O, OH	11.10	11.25
	135-235	203	$C_5H_6NO_2S$	47.70	47.65
	235-900	385, 441	$C_{19}H_{14}N_5O_3S_2Cl_2$	32.57	32.48
	-	-	Cu	8.63	8.62
5	35-105	78, 90	$3H_2O$	8.50	8.23
	105-411	203, 289, 303, 312, 322	$C_{10}H_{12}N_2O_4S_2$	44.00	43.95
	411-700	477	$C_4H_2N_2O_3$	37.88	38.13
	-	-	Cu	9.62	9.69

Table S5 Optical properties for **1-5** DMSO (nm($Lmol^{-1}cm^{-1}$))

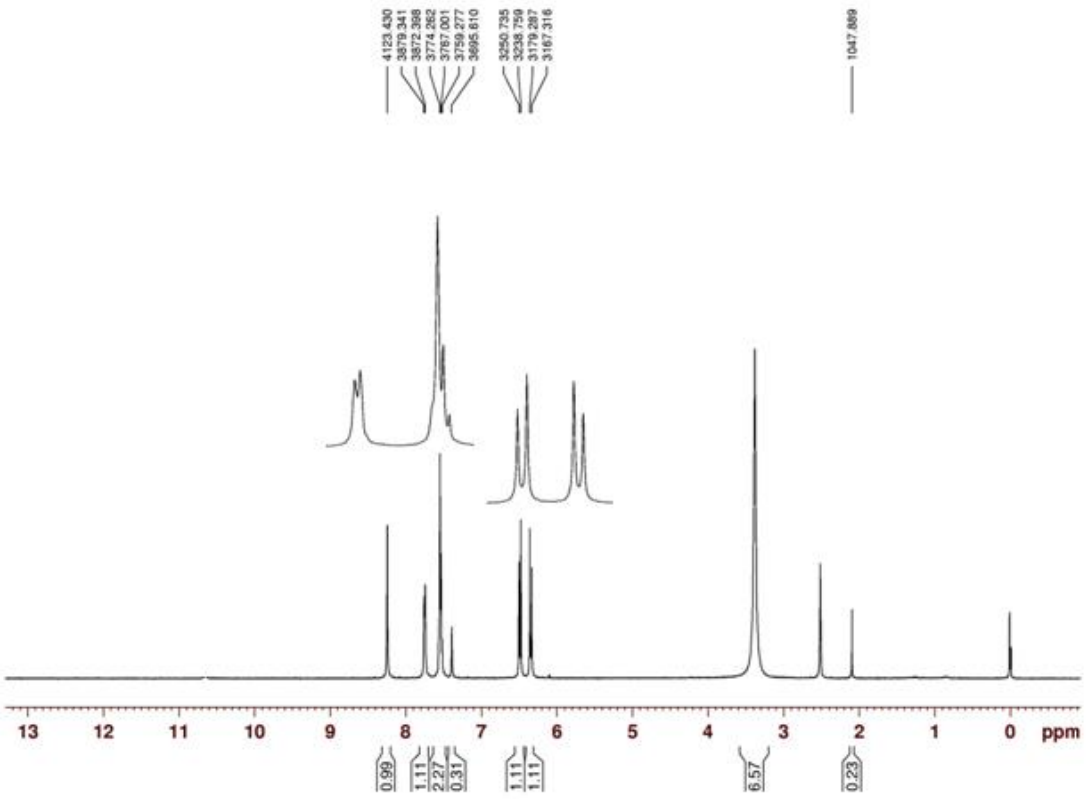
	DMSO
Clabt	287(29430)
H*m*absmal	290(24510)
H*p*absmal	301(43400) 290(33540)
1	290(29790)
2	290(30610)
3	297(39140) 290(34550) 767(13)
4	288(19990) 782(40)
5	290(26710) 763(36)

Table S6. Purification data of hCA I and hCA II isozymes

Purification Steps		**Activity (EU/mL)**	**Total Volume (mL)**	**Protein (mg/mL)**	**Total Protein (mg)**	**Total Activity (EU)**	**Specific Activity (EU/mg protein)**	**Yield (%)**	**Purification Factor**
Hemolysate		125.16	100	18.24	1824	12516	6.86	100	1
Affinity	**hCA I**	421.12	5	0.38	1.90	2105.60	1108.21	16.82	161.54
column	**hCA II**	599.95	5	0.25	1.25	2999.75	2399.80	23.97	349.83



b



a

Fig. S1 ^{1}H NMR spectra of compound H*m*absmal; a) in DMSO, b) in DMSO with D_2O

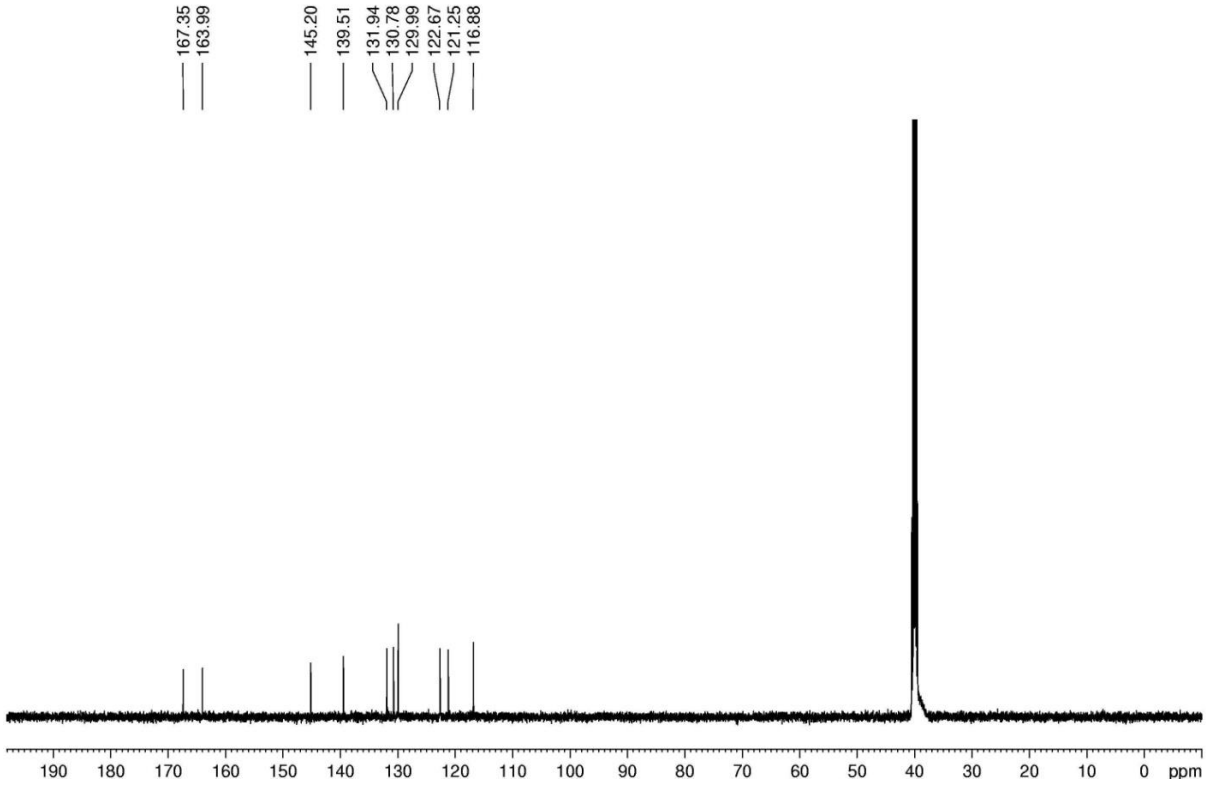
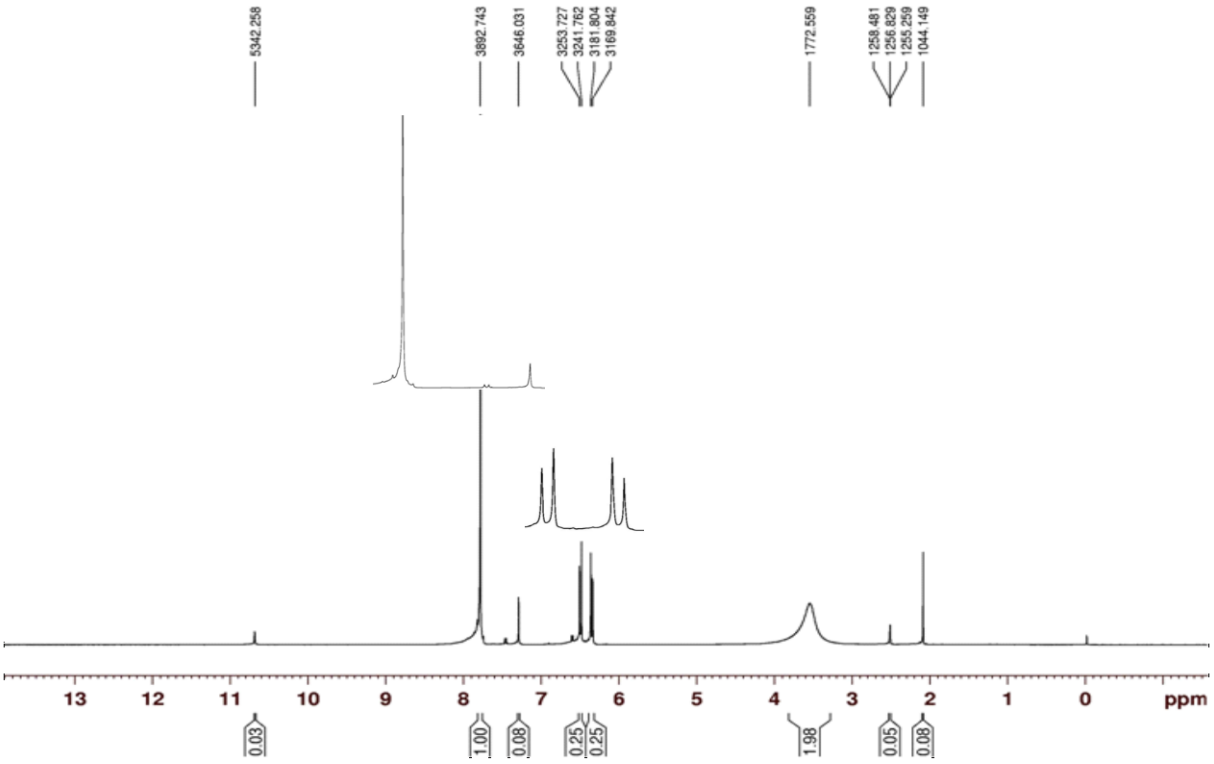


Fig. S2 ^{13}C NMR spectra of compound H*m*absmal



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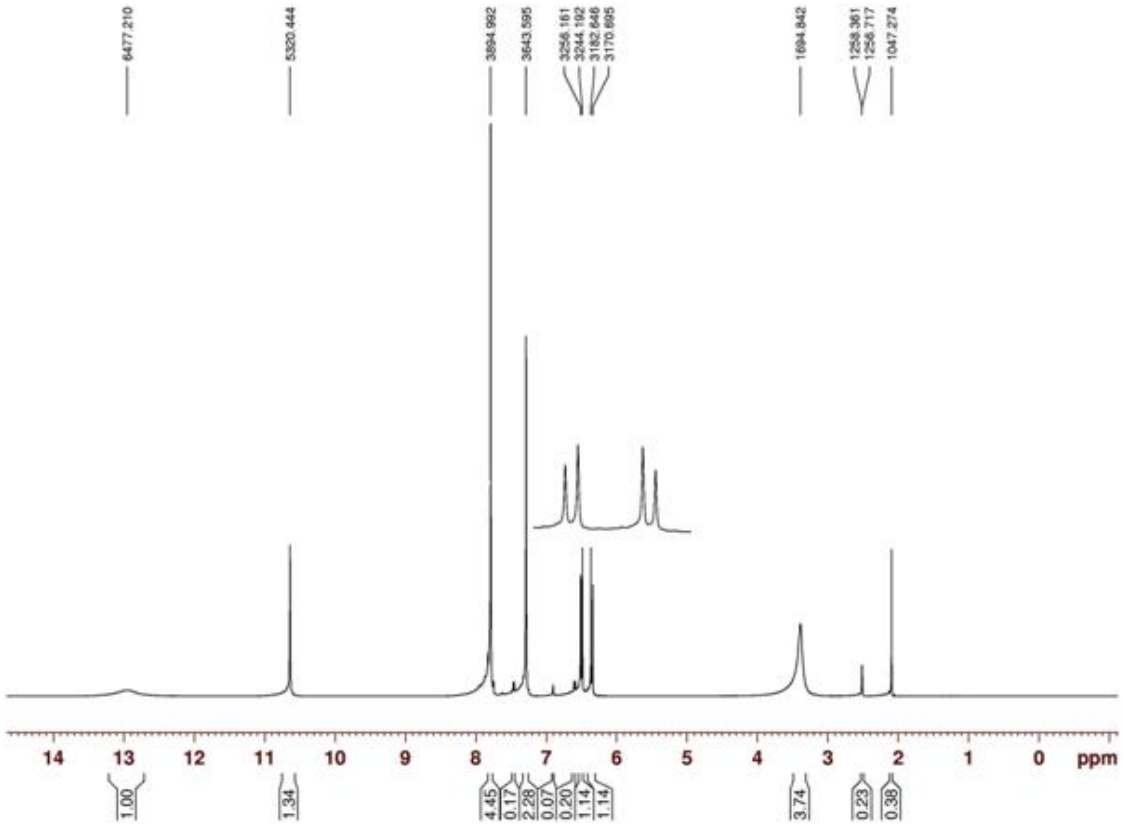


a

Fig. S3 [1]H NMR spectra of compound H*p*absmal; a) in DMSO, b) in DMSO with D_2O

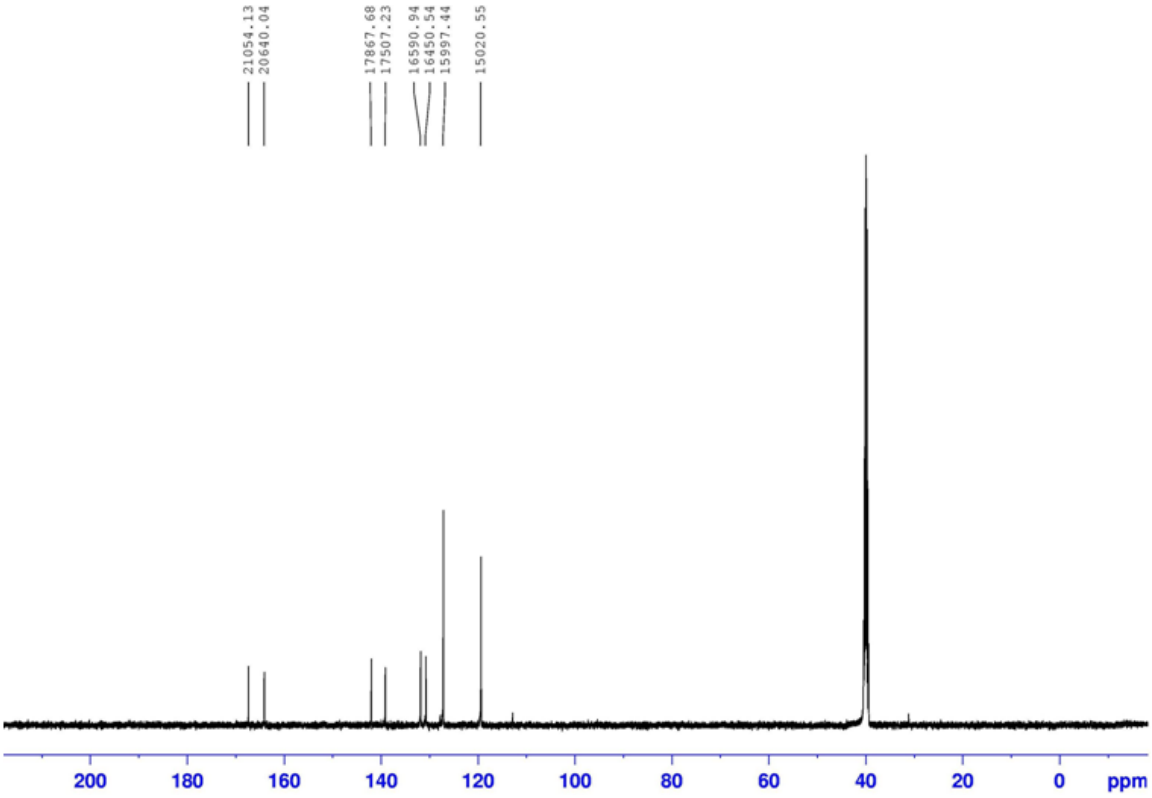


Fig. S4 ^{13}C NMR spectra of compound H*p*absmal

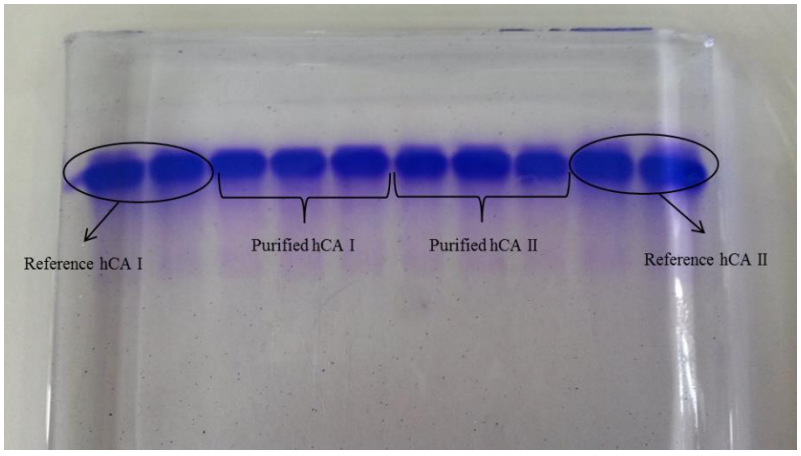
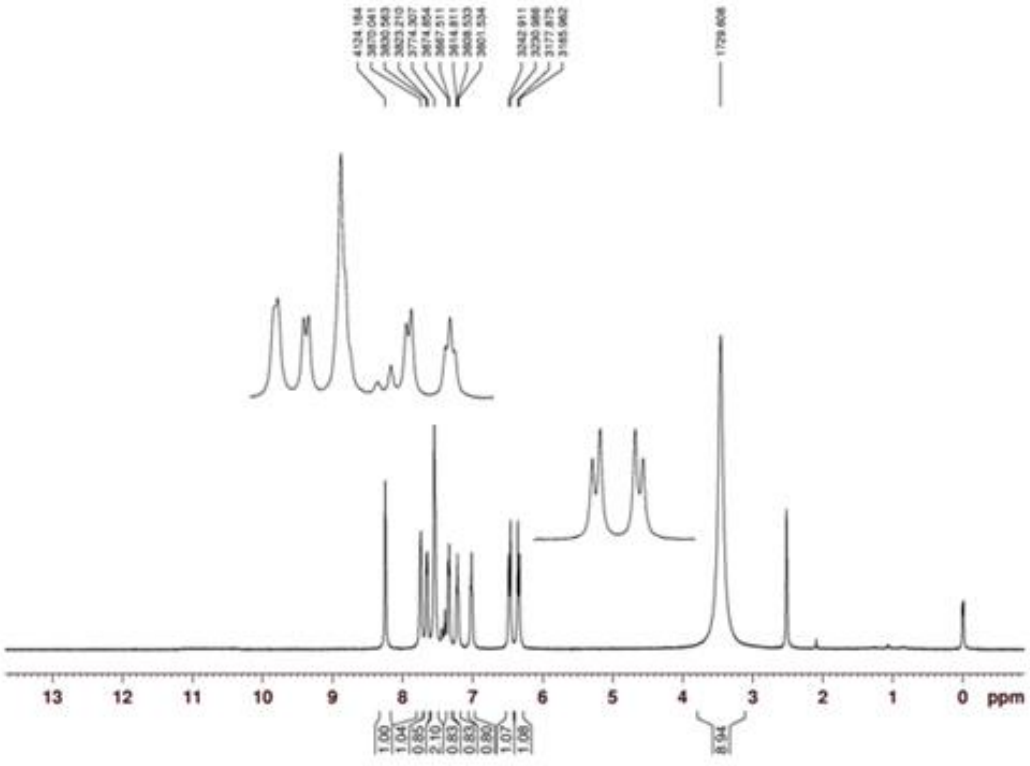
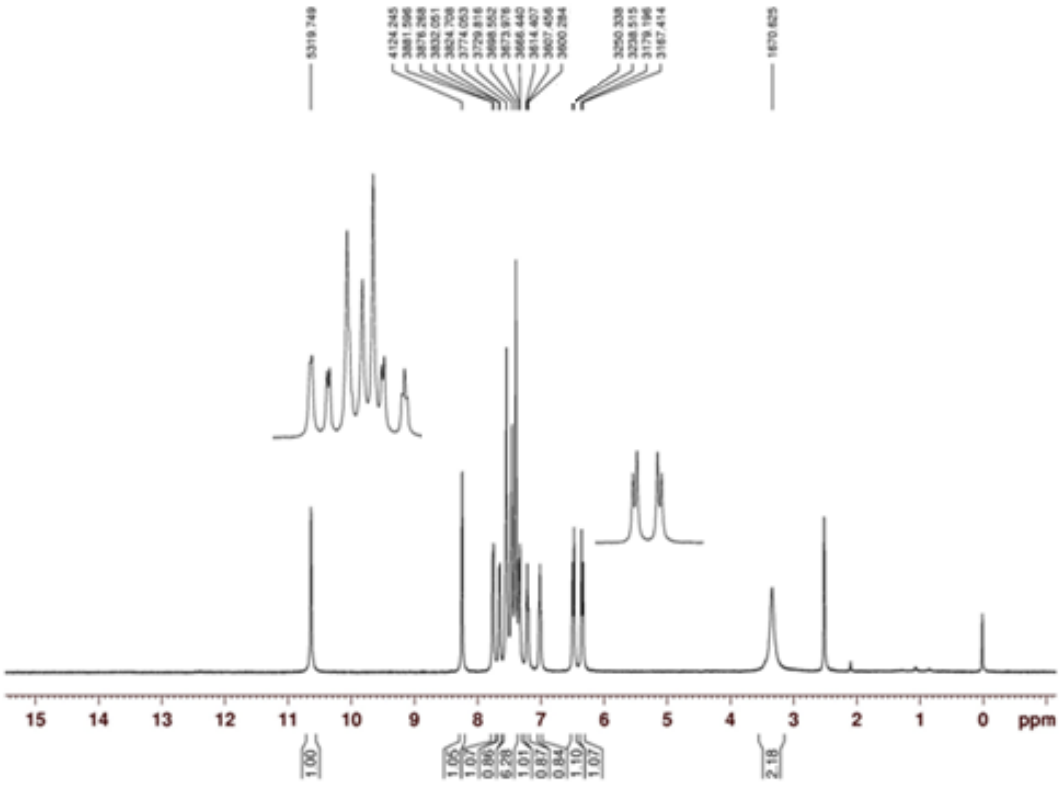


Fig. S5 SDS PAGE analysis of purified isozymes. (Reference hCA I: Sigma C4396, reference hCA II: Sigma C6165)



b



a

Fig. S6 ^{1}H NMR spectra of compound **1**; a) in DMSO, b) in DMSO with D_2O

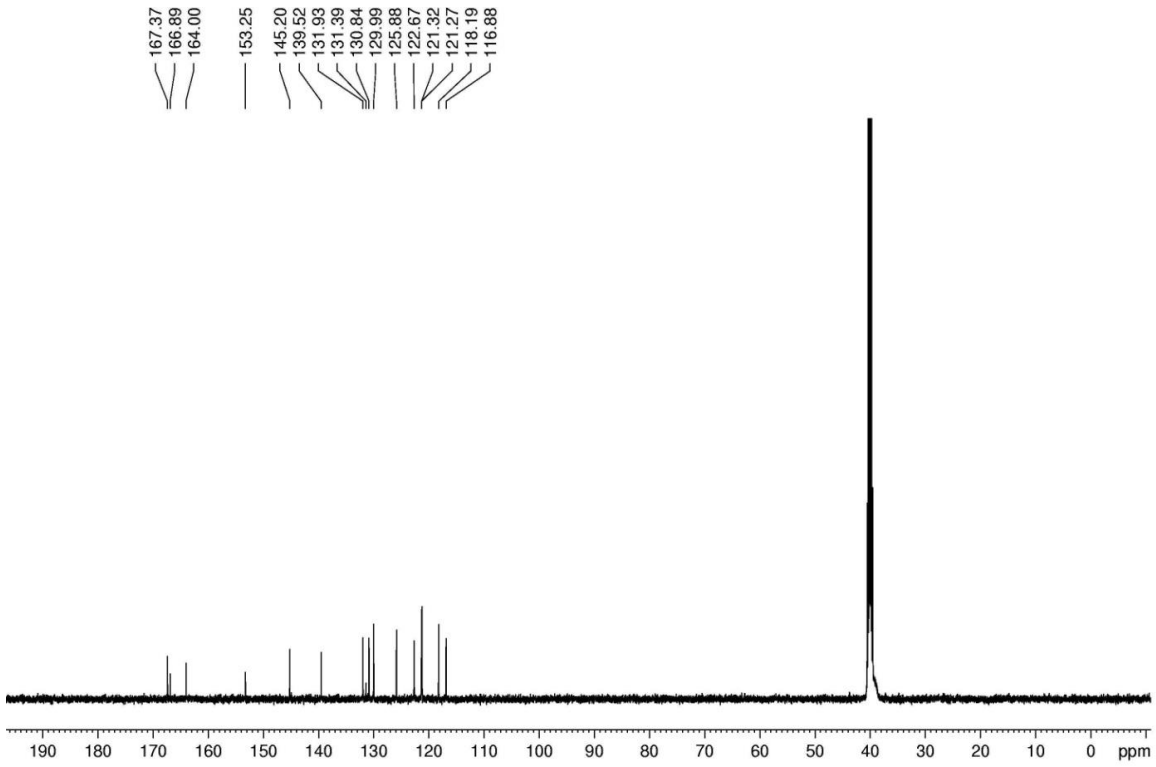
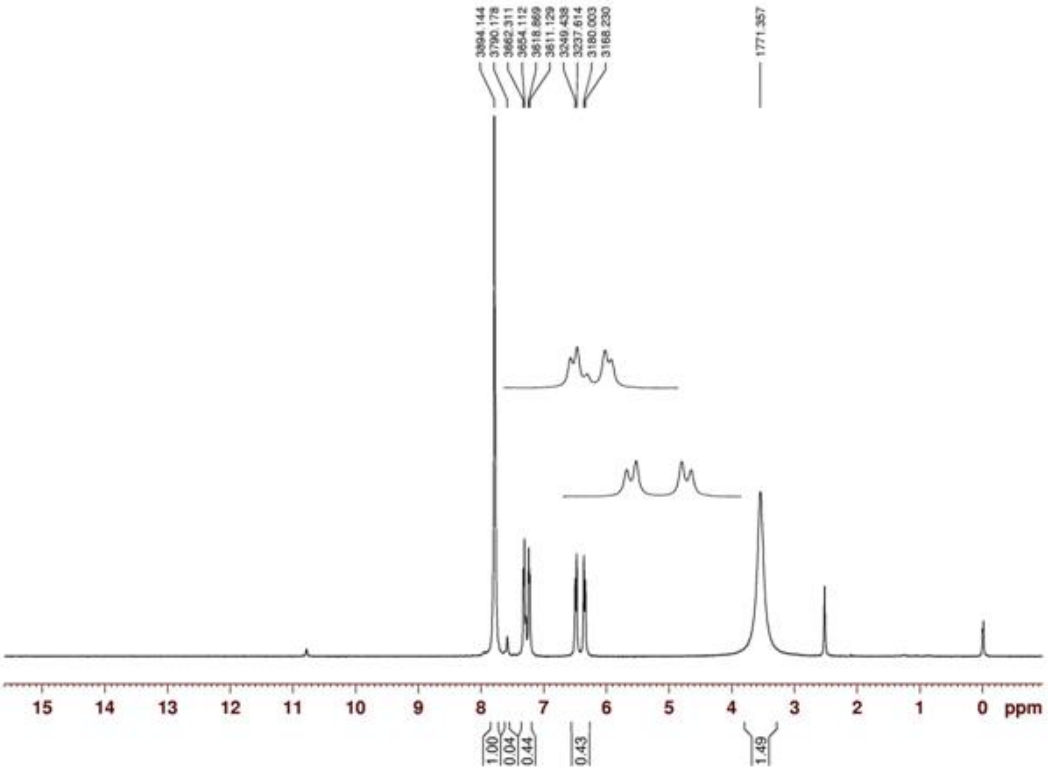
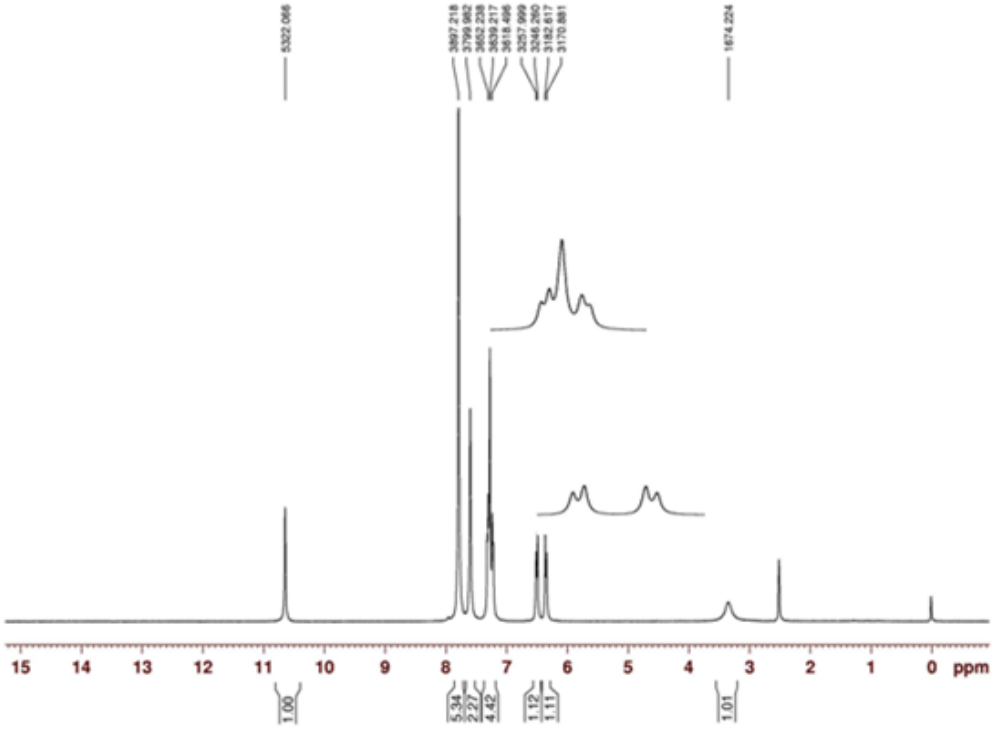


Fig. S7 ^{13}C NMR spectra of compound **1**



b



a

Fig. S8 ^{1}H NMR spectra of compound **2**; a) in DMSO, b) in DMSO with D_2O.

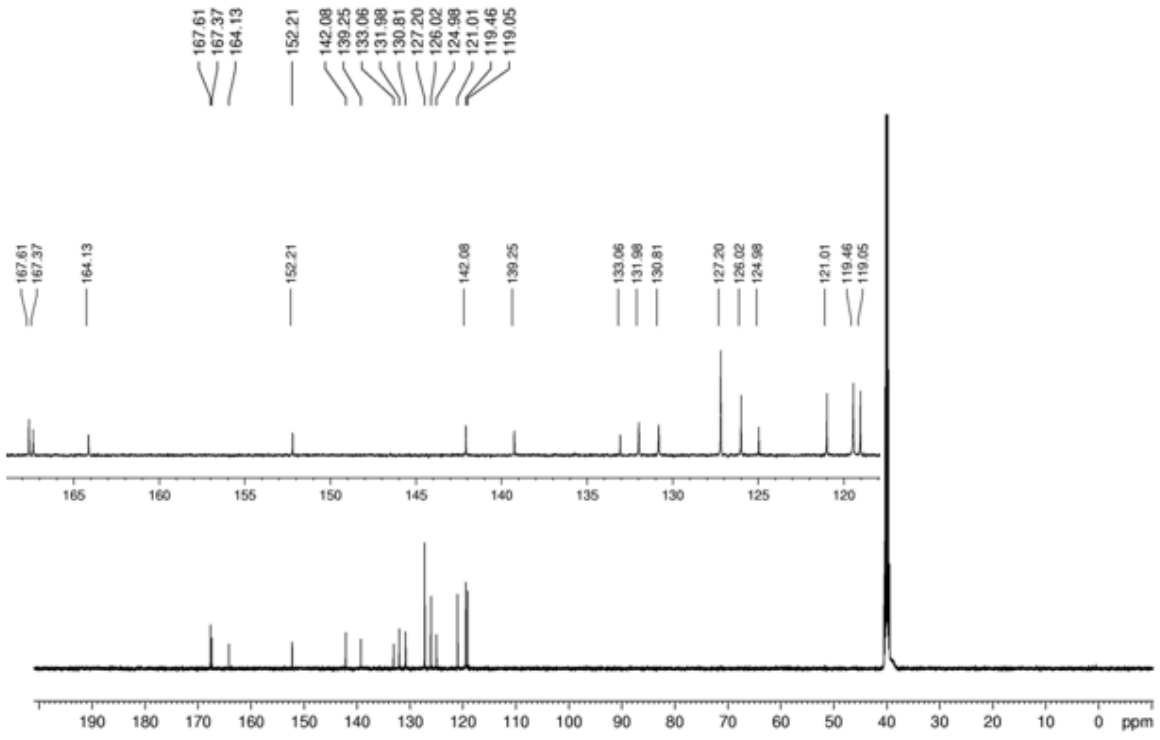


Fig. S9 ^{13}C NMR spectra of compound **2**.

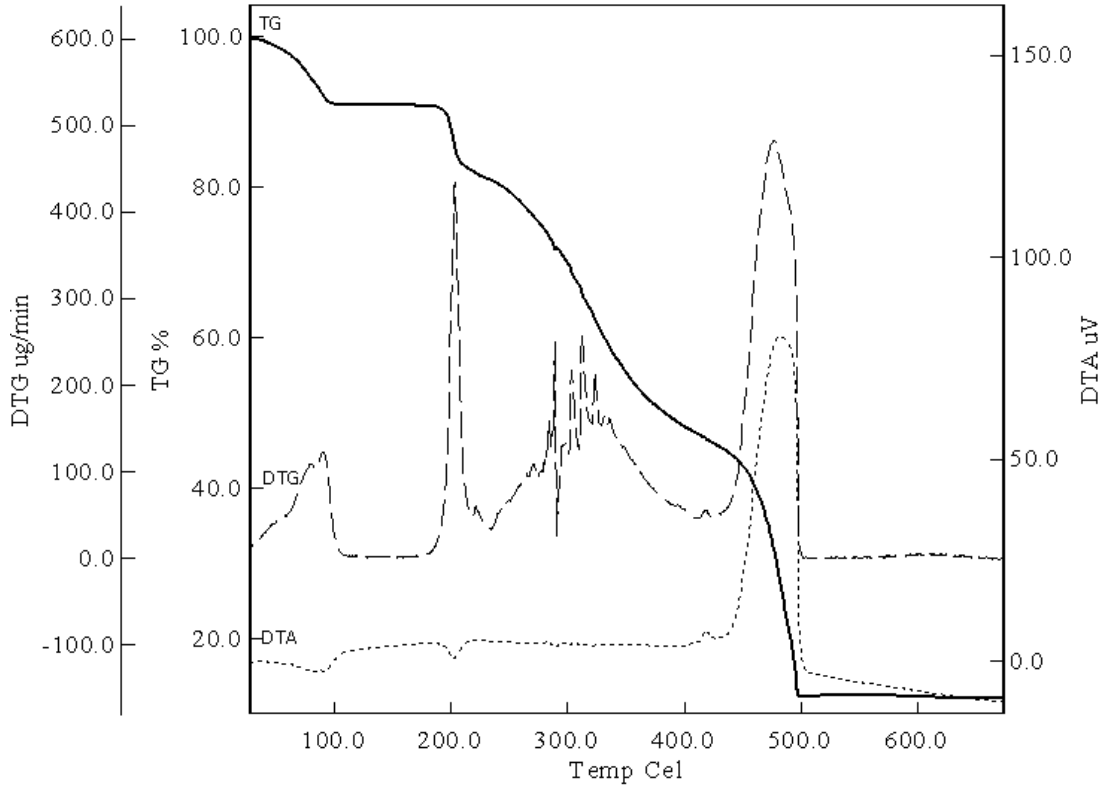


Fig. S10 TG-DTG and DTA curves of **3.**

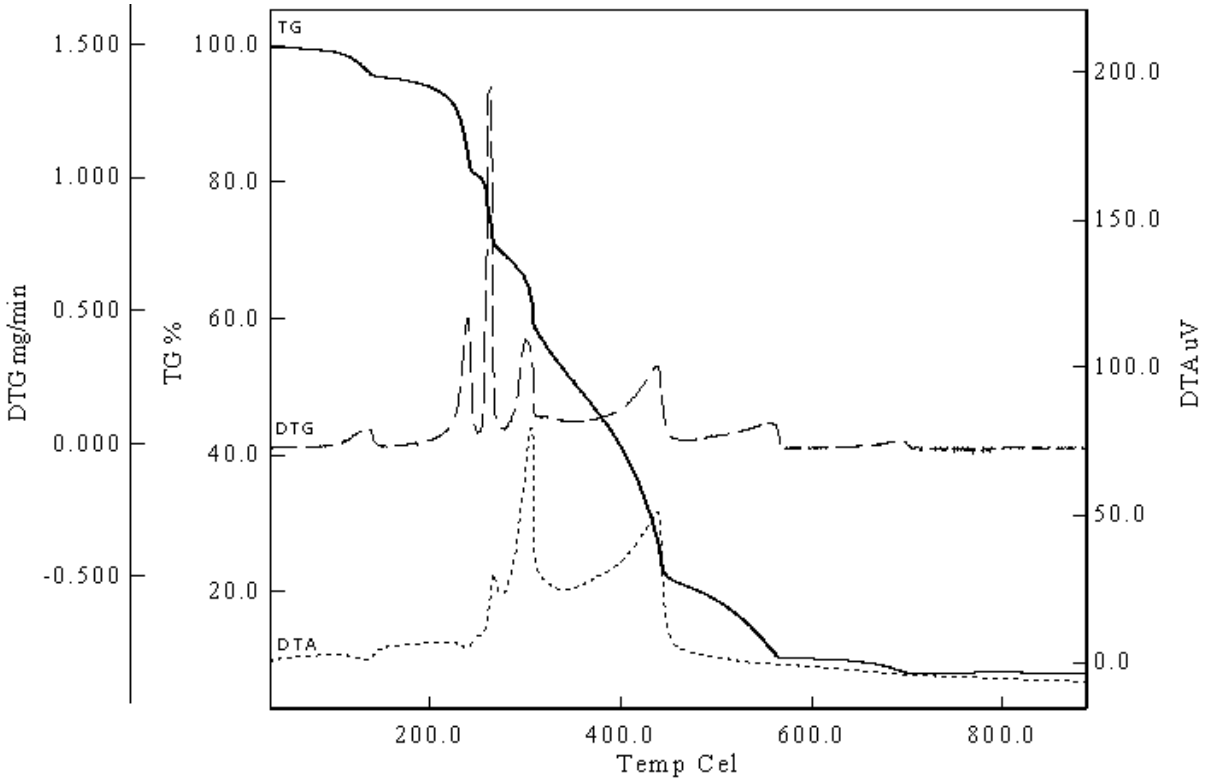


Fig. S11 TG-DTG and DTA curves of **4.**

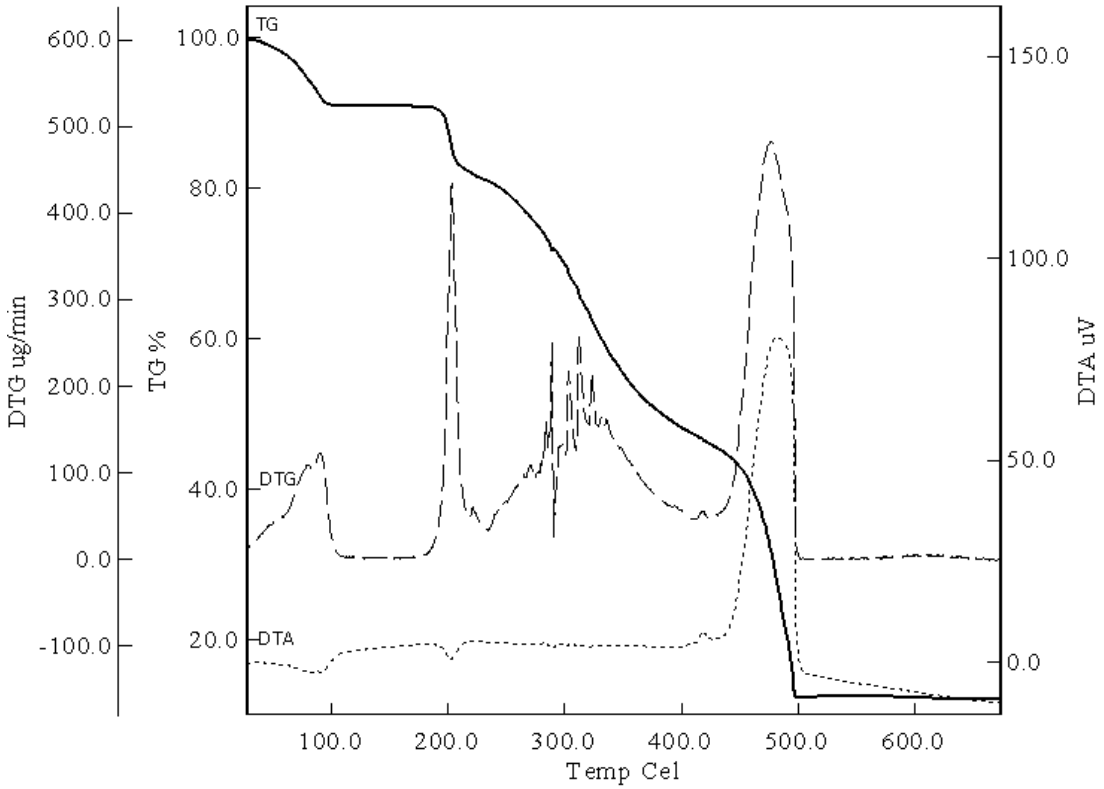


Fig. S12 TG-DTG and DTA curves of **5.**