Supporting Information

Synthesis, anticancer activity and molecular docking simulation of *N*-(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)thioureas containing pyrimidine ring

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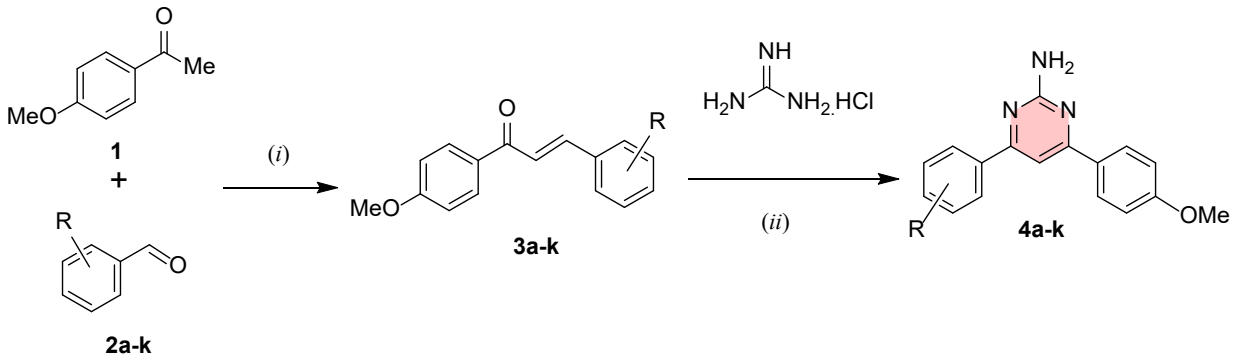
1. Synthesis of 2-(4-methoxyphenyl)-6-(R-phenyl)-2-aminopyrrimidines (5a-k)



1.1. Synthesis of Substituted chalcones 3a-k

4-Methoxyacetophenone (**1**, 0.02 mol, 2.40 g) was added to a solution of appropriate substituted benzaldehyde (**2a-k**, 0.02 mol) in 96% ethanol (5 mL). Next, KOH solution (prepared from 0.84 g of KOH in 4 mL of water) was slowly dropped into the reaction mixture while temperature of the mixture was always kept at about 15–20°C. The reaction was stirred for 4–5 h more. The solid product filtered and washed thoroughly with water to neutral. The raw product of yellow or light-yellow colour was obtained. After crystallization from 96% ethanol or solvent mixture of ethanol-toluene (1:1 in volume) appropriate α,β-unsaturated ketone was obtained as needle crystals.

3-(4-Methoxyphenyl)-1-phenylprop-2-en-1-one (3a)

From 4-methoxybenzaldehyde **2a** (2.72 g). M.p.: 61–62°C (from 96% ethanol). Yield: 3.76 (79%).

3-(4-Fluorophenyl)-1-(4-methoxyphenyl)prop-2-en-1-one (3b)

From 4-fluorobenzaldehyde **2b** (2.48 g). M.p.: 90–91°C (from 96% ethanol). Yield: 3.62 g (80%).

3-(4-Chlorophenyl)-1-(4-methoxyphenyl)prop-2-en-1-one (3c)

From 4-chlorobenzaldehyde (2.81 g). M.p.: 142–143°C (from 96% ethanol). Yield: 4.02 g (83%).

3-(3-Chlorophenyl)-1-(4-methoxyphenyl)prop-2-en-1-one (3d)

From 3-chlorobenzaldehyde (2.81 g). M.p.: 127–128°C (from 96% ethanol). Yield: 3.93 g (81%).

3-(2-Chlorophenyl)-1-(4-methoxyphenyl)prop-2-en-1-one (3e)

From 2-chlorobenzaldehyde (2.81 g). M.p.: 133–134°C (from 96% ethanol). Yield: 3.59 g (74%).

3-(4-Bromophenyl)-1-(4-methoxyphenyl)prop-2-en-1-one (3f)

From bromobenzaldehyde (3.70 g). M.p.: 154–155°C (from 96% ethanol). Yield: 4.48 g (78%).

1-(4-Methoxyphenyl)-3-(4-methylphenyl)prop-2-en-1-one (3g)

From 4-methylbenzaldehyde (2.40 g). M.p.: 99–100°C (from 96% ethanol-toluene, 1:1 in volume). Yield: 4.03 g (80%).

1-(4-Methoxyphenyl)-3-[4-(propan-2-yl)phenyl]prop-2-en-1-one (3h)

From 4-isopropylbenzaldehyde (2.96 g). M.p.: 72-73°C (from 96% ethanol-toluene, 1:1 in volume). Yield: 4.20 g (75%).

1,3-Bis(4-methoxyphenyl)prop-2-en-1-one (3i)

From 4-methoxybenzaldehyde (2.72 g). M.p.: 154-155°C (from 96% ethanol-toluene, 1:1 in volume). Yield: 4.13 g (77%).

3-(3-Methoxyphenyl)-1-(4-methoxyphenyl)prop-2-en-1-one (3j)

From 3-methoxybenzaldehyde (2.72 g). M.p.: 111-112°C (from 96% ethanol-toluene, 1:1 in volume). Yield: 4.02 g (75%).

3-[4-(Dimethylamino)phenyl]-1-(4-methoxyphenyl)prop-2-en-1-one (3k)

From 4-dimethylaminobenzaldehyde (2.98 g). M.p.: 124-124°C (from 96% ethanol-toluene, 1:1 in volume). Yield: 4.61 g (82%).

1.2. Synthesis of 2-(4-methoxyphenyl)-6-(R-phenyl)-2-aminopyrrimidines (5a-k)

Substituted chalcones **3a-k** were prepared by reaction between 4-methoxyacetophenone (**1**) and the corresponding substituted benzaldehydes (**2a-k**) according to literature procedure.[1, 2] Appropriate chalcone (0.005 mol) was dissolved in 96% ethanol (20 mL) then guanidine hydrochloride (0.0075 mol, 0.7 g) and solid NaOH (0.0225 mol, 0.9 g) were added to this solution. The reaction mixture was heated under reflux for 10–14 h. The solvent was removed under reduced pressure and cool for 3-4 hours. The separated product as a solid was filtered washed with water to a neutral reaction. Recrystallization from ethanol/toluene mixture (1:1 by volume) to afford the titled 2-(4-bromophenyl)-6-(R-phenyl)-2-aminopyrrimidines (**5a-k**).[3]

2-Amino-4-phenyl-6-(4-methoxyphenyl)pyrimidine (5a)

From benzylidene-(4-methoxyacetphonene **3a** (0.005 mol, 1.19 g). M.p.: 151–152ºC. Yield: 0.93 g (67%).

2-Amino-4-(4-methoxyphenyl)-6-(4- fluorophenyl)pyrimidine (5b)

From (4-fluorobenzylidene)-4-methoxyacetophenone **3b** (0.005 mol, 1.34 g). M.p.: 189–190ºC. Yield: 0.96 g (65%).

2-Amino-4-(4-methoxyphenyl)-6-(4-chlorophenyl)pyrimidine (5c)

From (4-chlorobenzyliden)-4-methoxyacetophenone **3c** (0.005 mol, 1.36 g). M.p.: 156–157ºC. Yield: 1.06 g (75%).

2-Amino-4-(4-methoxyphenyl)-6-(3-chlorophenyl)pyrimidine (5d)

From (3-chlorobenzyliden)-4-methoxyacetophenone **3d** (0.005 mol, 1.36 g). M.p.: 130–131ºC. Yield: 1.04 g (74%).

2-Amino-4-(4-methoxyphenyl)-6-(2-chlorophenyl)pyrimidine (5e)

From (2-chlorobenzyliden)-4-methoxyacetophenone **3e** ((0.005 mol, 1.36 g). M.p.: 161–162ºC. Yield: 0.77 g (55%).

2-Amino-4-(4-methoxyphenyl)-6-(4-bromophenyl)pyrimidine (5f)

From (4-bromobenzyliden)-4-methoxyacetophenone **3f** (0.005 mol, 1.59 g). M.p.: 170–171ºC. Yield: 1.35 g (76%).

2-Amino-4-(4-methoxyphenyl)-6-(4-methylphenyl)-pyrimidine (5g)

From (4-methylbenzyliden)-4-methoxyacetophenone **3g** (0.005 mol, 1.26 g). M.p.: 125–126ºC. Yield: 1.00 g (68%).

2-Amino-4-(4-methoxyphenyl)-6-(4-isopropylphenyl)pyrimidine (5h)

From (4-isopropylbenzyliden)-4-methoxyacetophenone **3h** (0.005 mol, 1.40 g). M.p.: 126-127ºC. Yield: 1.13 g (71%).

2-Amino-4,6-bis(4-methoxyphenyl)pyrimidine (5i)

From 1,3-bis(4-methoxyphenyl)prop-2-en-1-one **3i** (0.005 mol, 1.23 g). M.p.: 179–180ºC. Yield: 1.23 g (80%).

2-Amino-4-(4-methoxyphenyl)-6-(3-methoxyphenyl)pyrimidine (5j)

From (3-methoxybenzyliden)-4-methoxyacetophenone **3j** (0.005 mol, 1.23 g). M.p.: 140–141ºC. Yield: 1.21 g (79%).

2-Amino-4-(4-methoxyphenyl)-6-(4-dimethylaminophenyl)pyrimidine (5k)

From (4-dimethylaminobenzyliden)-4-methoxyacetophenone **3k** (0.005 mol, 1.41 g). M.p.: 175–176ºC. Yield: 1.25 g (78%).

2. Synthesis of *N*-(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl isothiocyanate

N-(2,3,4,6-Tetra-*O*-acetyl-β-D-glucopyranosyl isothiocyanate **6** was prepared from D-glucose **5** through bromide derivative according to literature procedures.[4,5]

References

1. M. C. Almeida, D. I. S. P. Resende, P. M. da Costa, M. M. M. Pinto and E. Sousa, *European Journal of Medicinal Chemistry*, 2021, **209**, 112945.
2. E. V. Filho, E. M. C. Pinheiro, S. Pinheiro and S. J. Greco, *Tetrahedron*, 2021, **92**, 132256.

3. N. F. Nadur, L. L. de Azevedo, L. Caruso, C. S. Graebin, R. B. Lacerda and A. E. Kümmerle, *European Journal of Medicinal Chemistry*, 2021, **212**, 113123.
4. A. Tashpulatov, V. Afanas' ev, M. Y. Lidak, N. Sukhova, Y. Y. Popelis and I. Rakhmatullaev, *Chemistry of Heterocyclic Compounds*, 1983, **19**, 137-141.

3. Molecular docking study

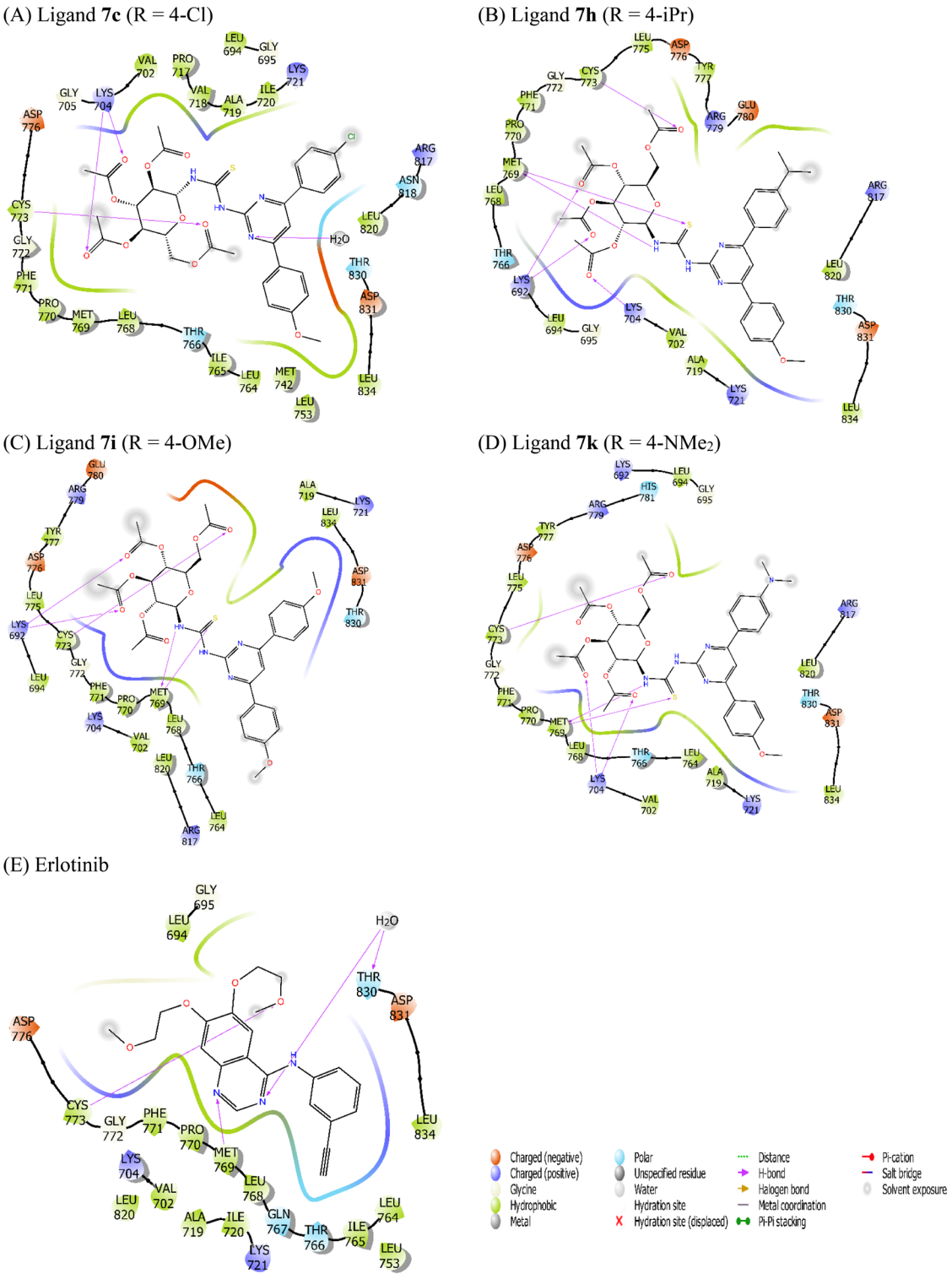


Figure 1S. The mode of interaction in the 2D-presentation of ligands **7c** (A), **7h** (B), **7i** (C), **7k** (D), and cocrystal erlotinib (E) in the active pocket on enzyme 4HJO indicated the H bonds and lipophilic interactions of each ligand corresponding to the residues.

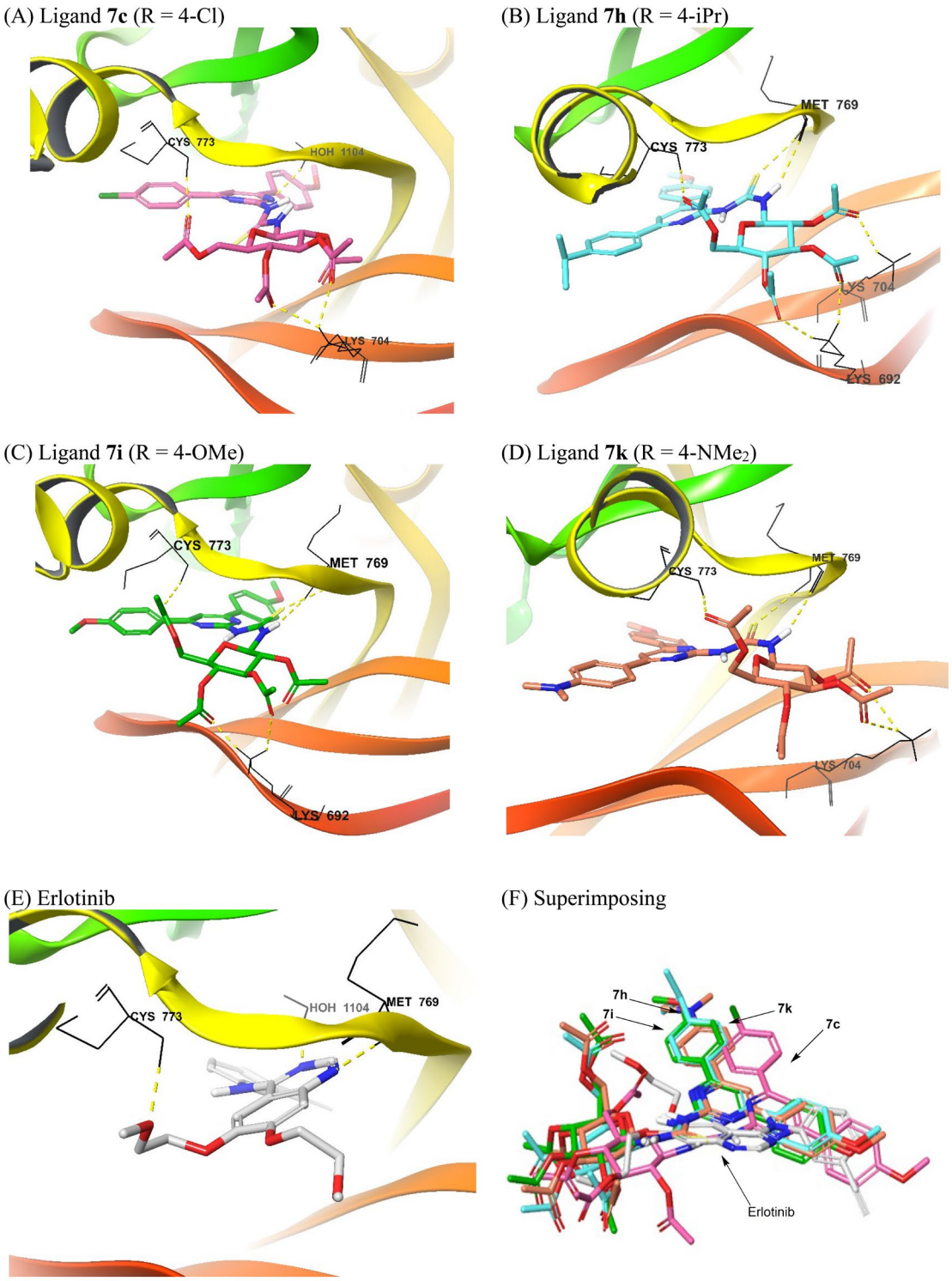
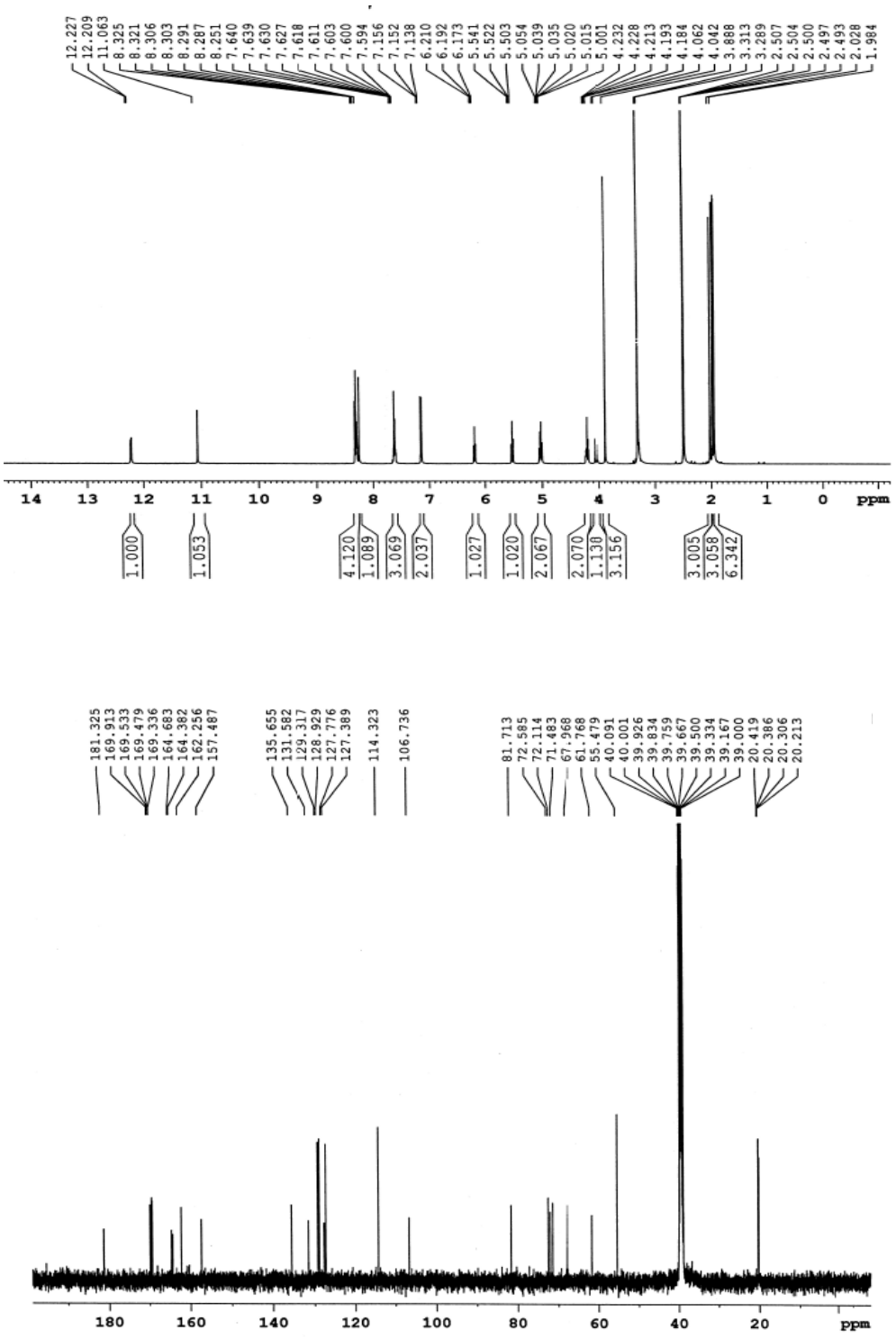
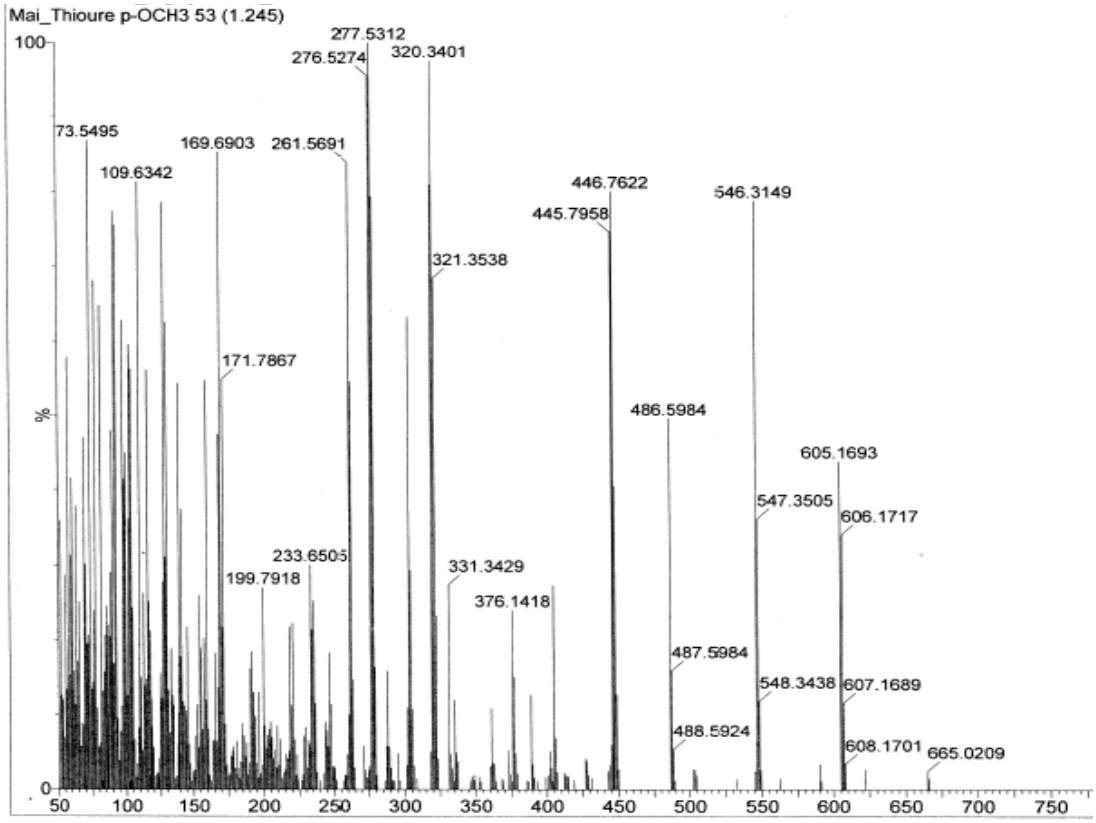


FIGURE 2S The mode of interaction in the 3D-presentation of ligands **7c** (A), **7h** (B), **7i** (C), **7k** (D), and cocrystal erlotinib (E) in the active pocket on enzyme 4HJO indicated the H bonds and lipophilic interactions of each ligand corresponding to the residues. (F) The superimposed positions of selected ligands **7c** (in magenta color),**7h** (in cyan color),**7i**, (in green color) and **7k** (in orange color) with erlotinib (in grey color) in active site of 4HJO.

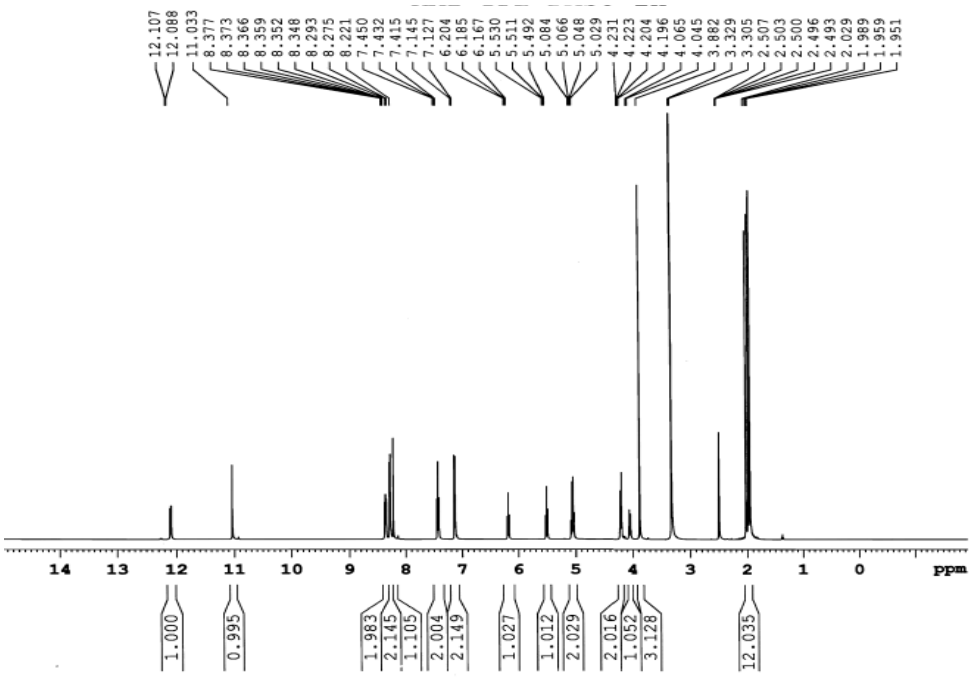
3. NMR & MS Spectra

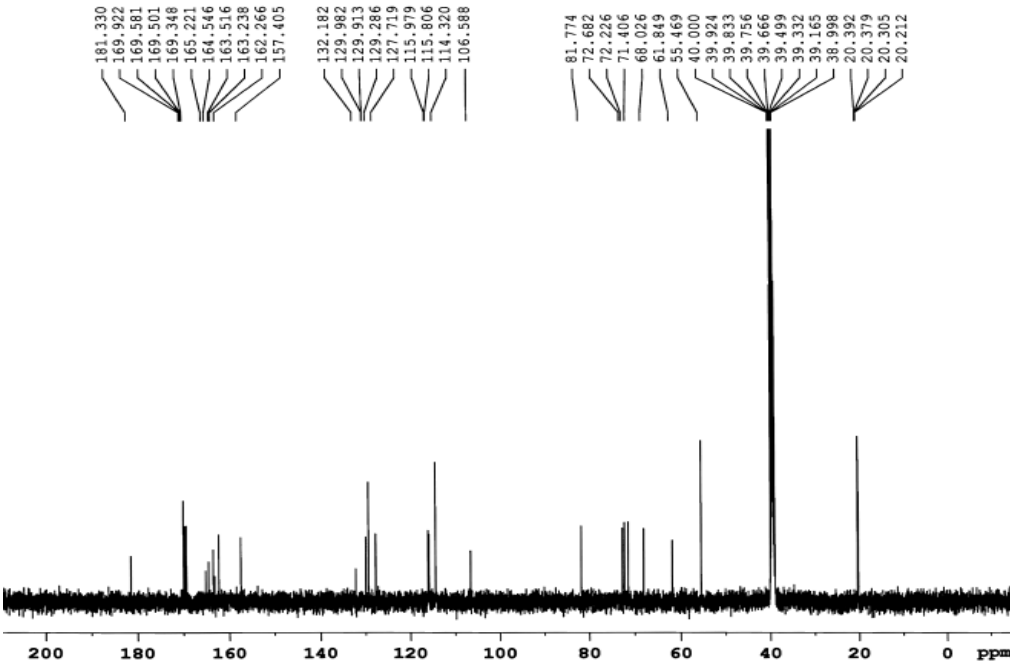
NMR & MS Spectra of N-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-N-[4-phenyl-6-(4-methoxyphenyl)pyrimidine-2'-yl]thiourea (7a)





NMR & MS Spectra of N-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-N-[4-(4-fluorophenyl)-6-(4-methoxyphenyl)pyrimidine-2-yl]thiourea (7b)

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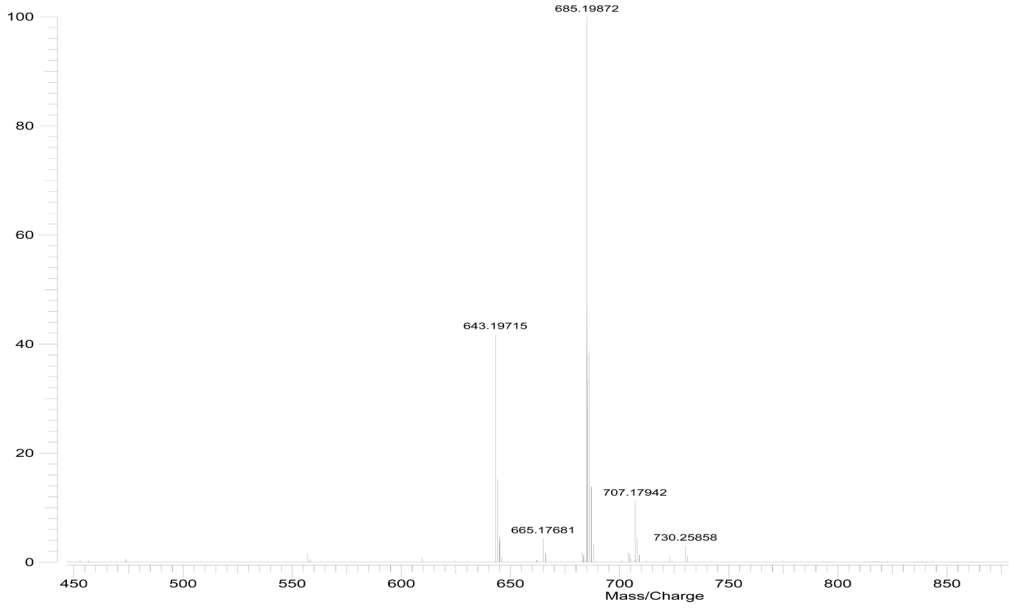
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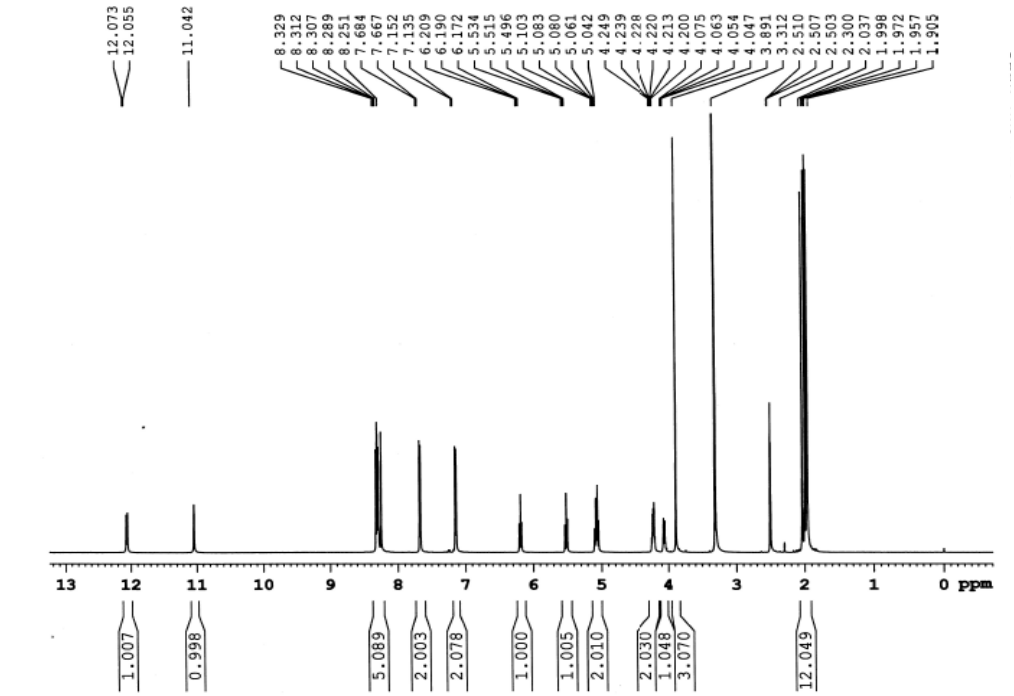
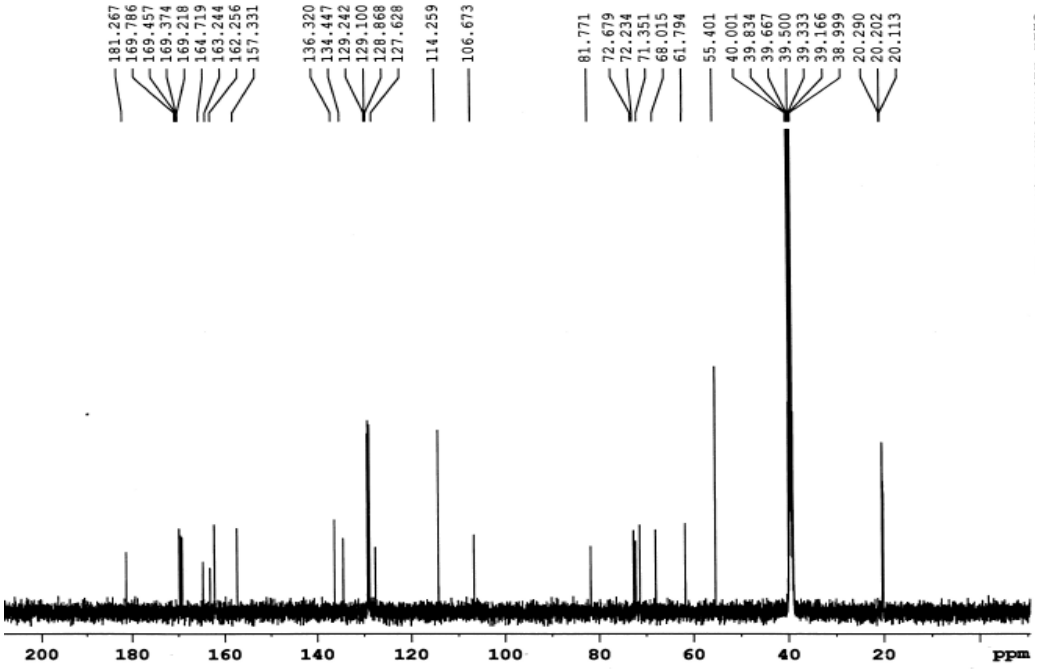
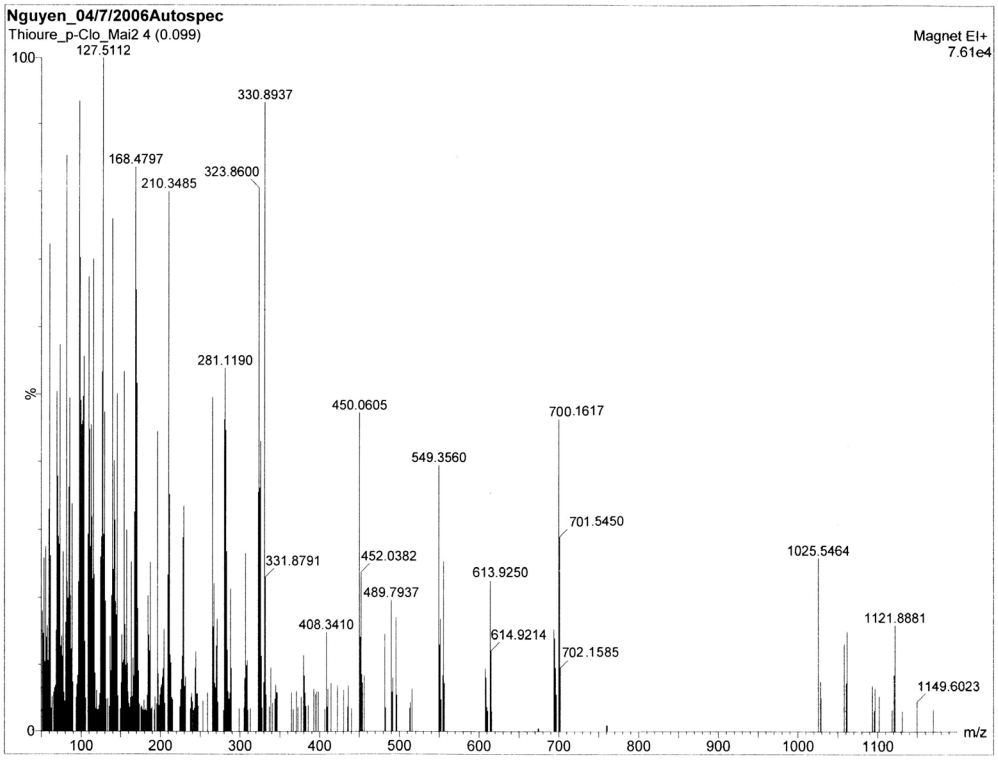
Mass/Charge

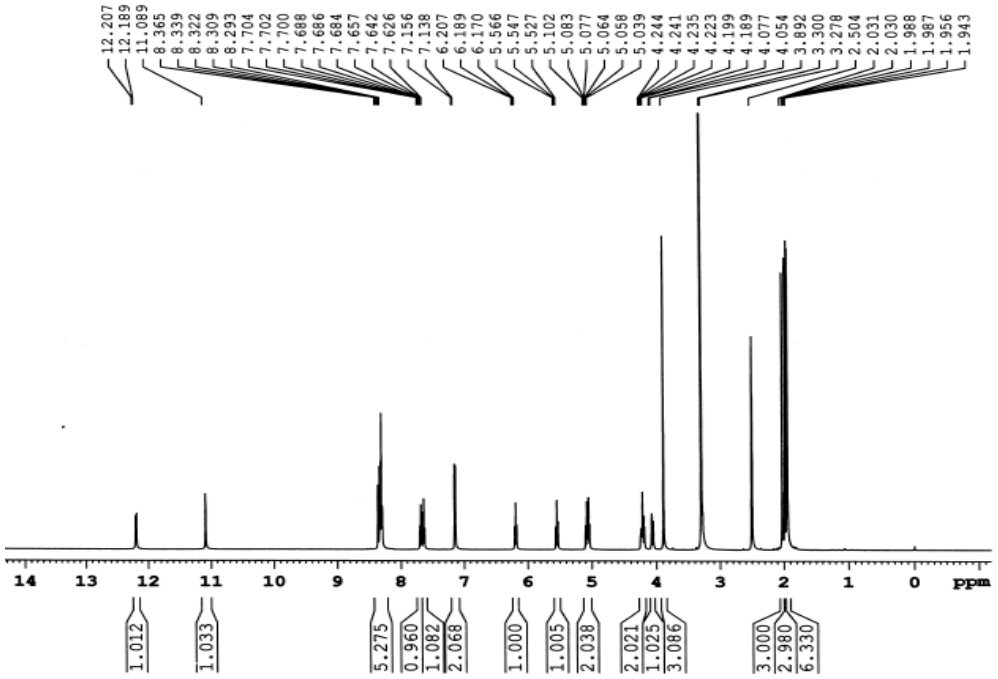
NMR & MS Spectra of N-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-N-[4-(4-chlorophenyl)-6-(4-methoxyphenyl)pyrimidine-2-yl]thiourea (7c)

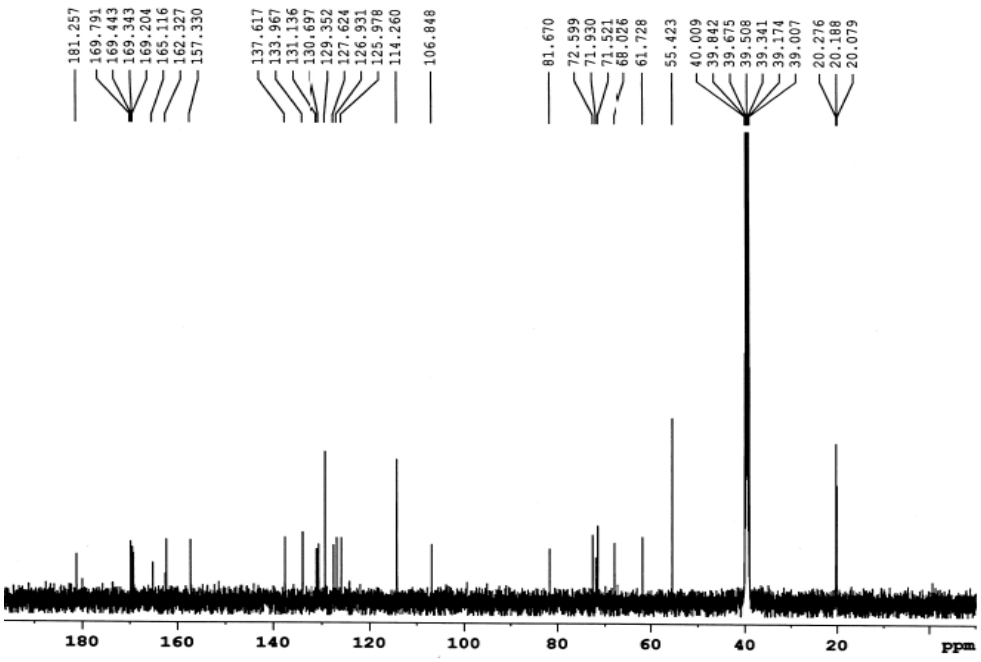






NMR & MS Spectra of N-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-N-[4-(3-chlorophenyl)-6-(4-methoxyphenyl)pyrimidine-2-yl]thiourea (7d)

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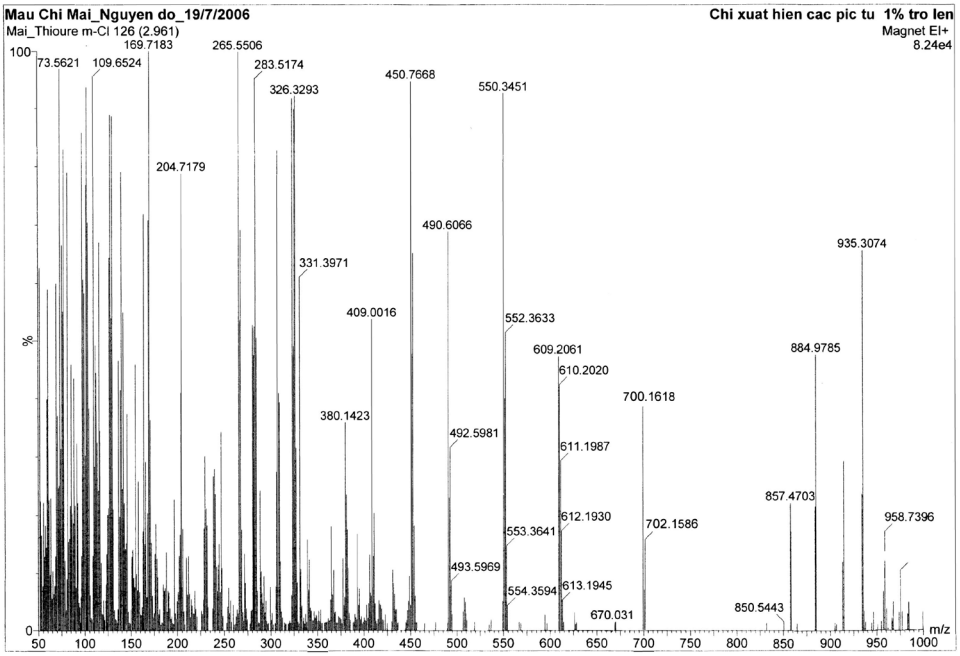
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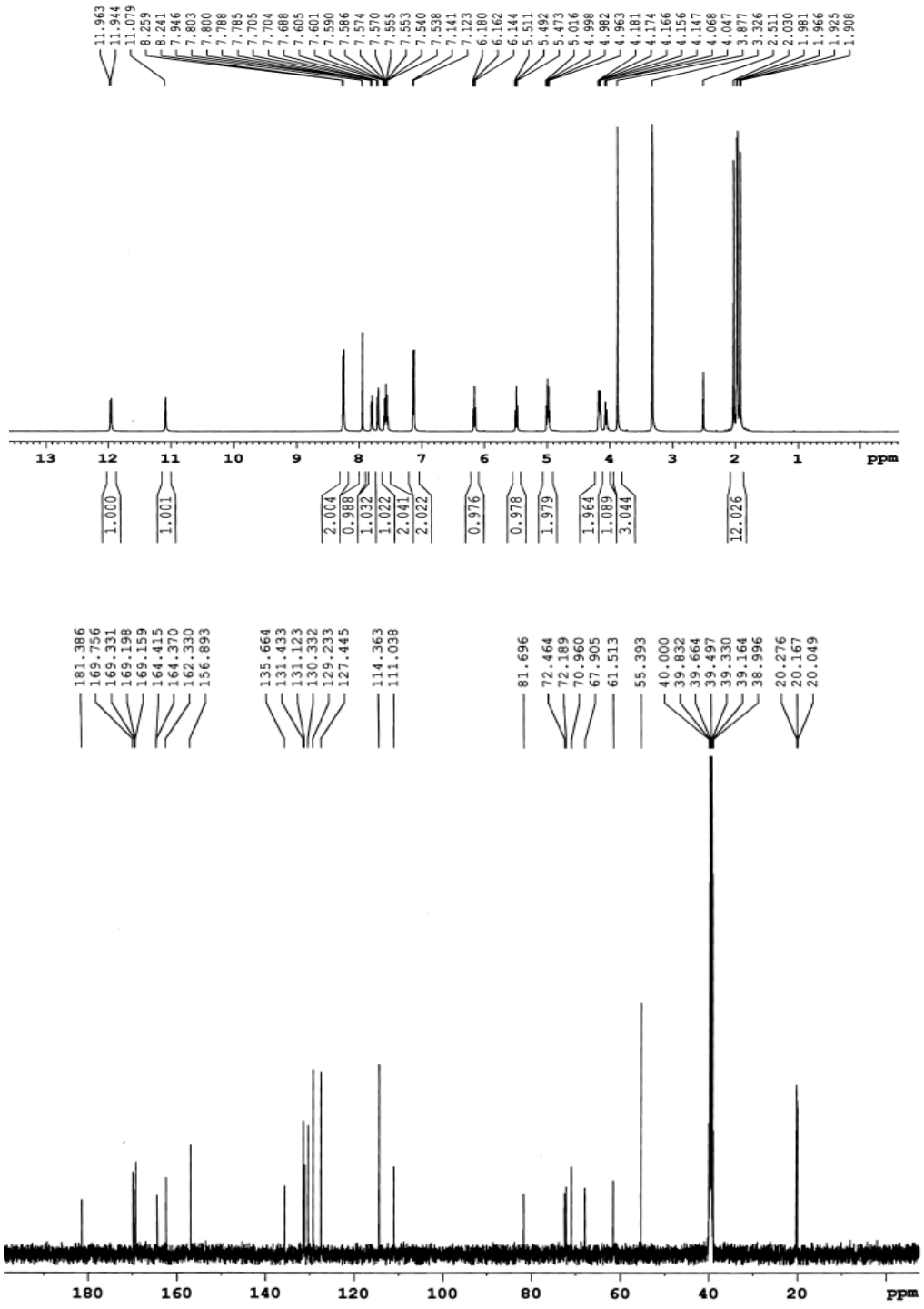
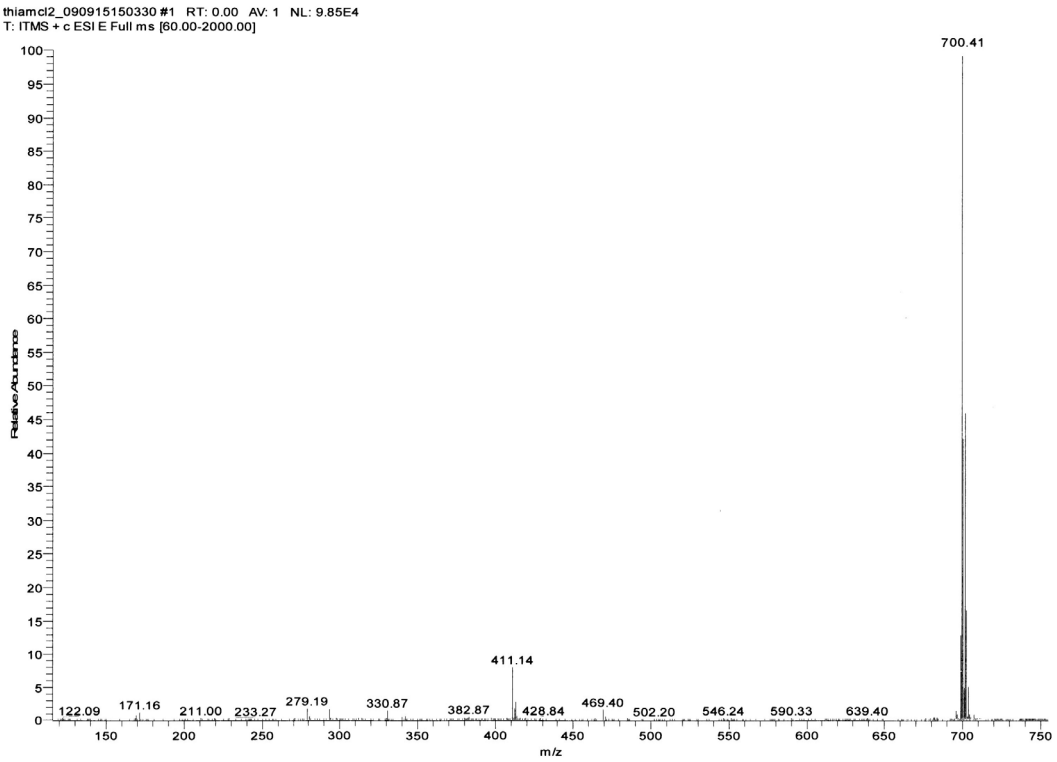
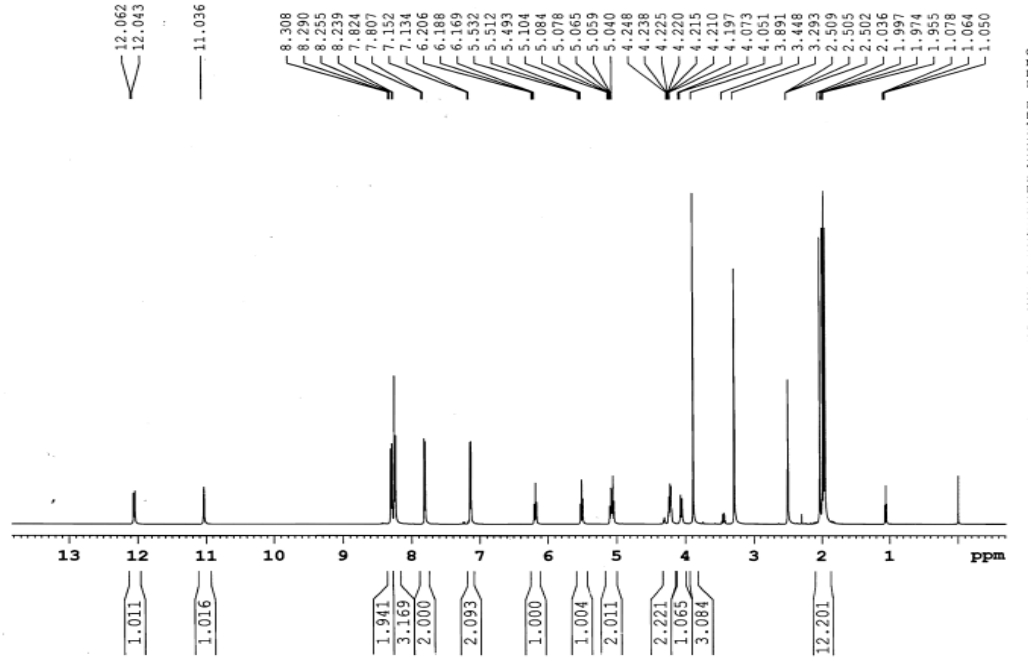
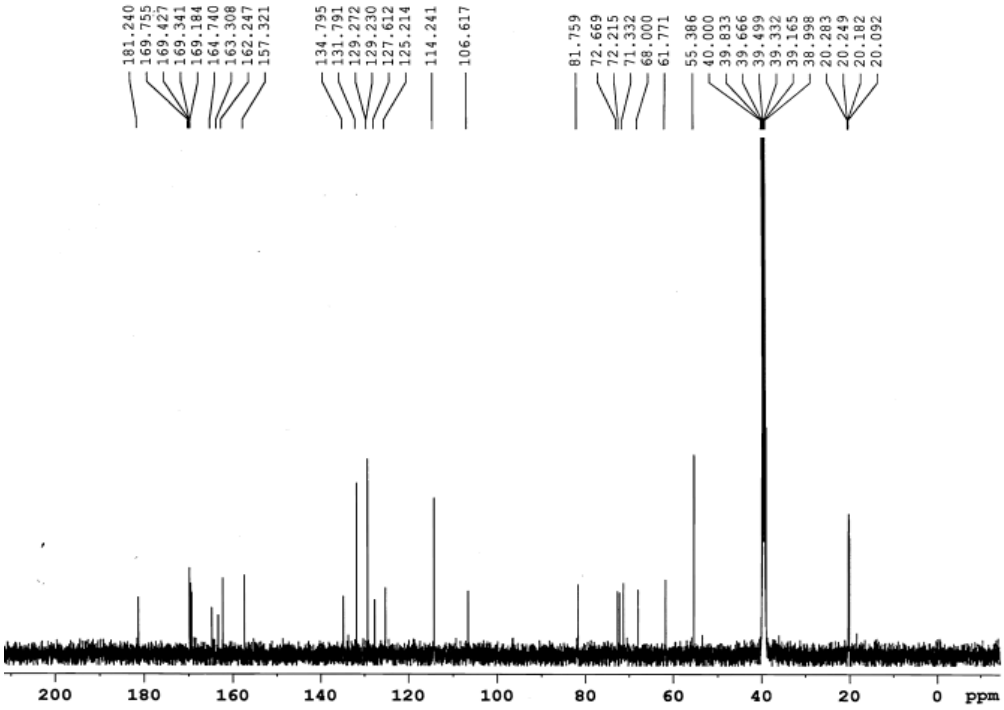
NMR & MS Spectra of N-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-N-[4-(2-chlorophenyl)-6-(4-methoxyphenyl)pyrimidine-2-yl]thiourea (7e)





NMR & MS Spectra of N-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-N-[4-(4-bromophenyl)-6-(4-methoxyphenyl)pyrimidine-2-yl]thiourea (7f)

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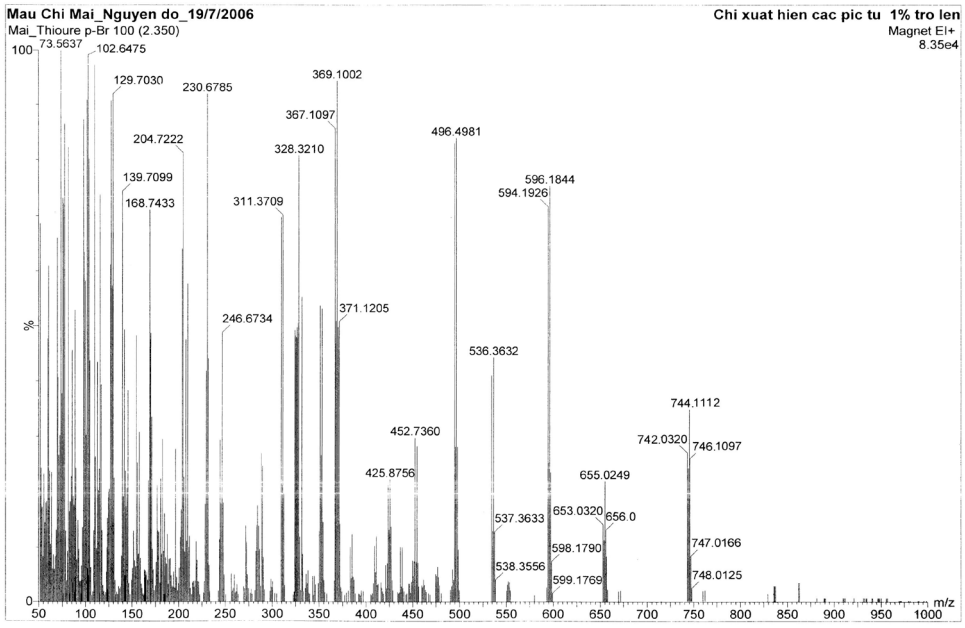
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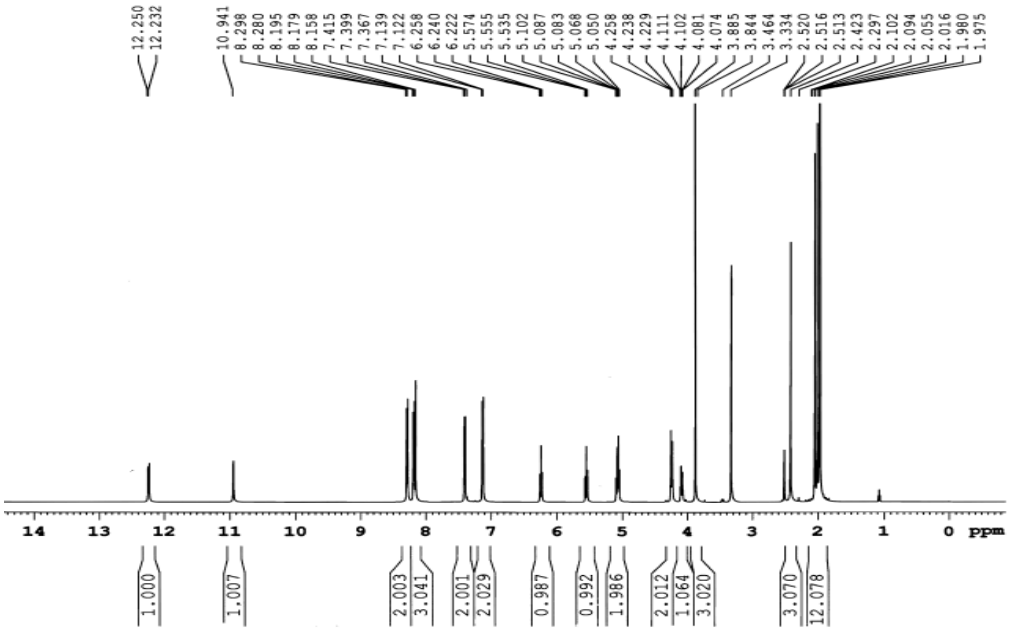
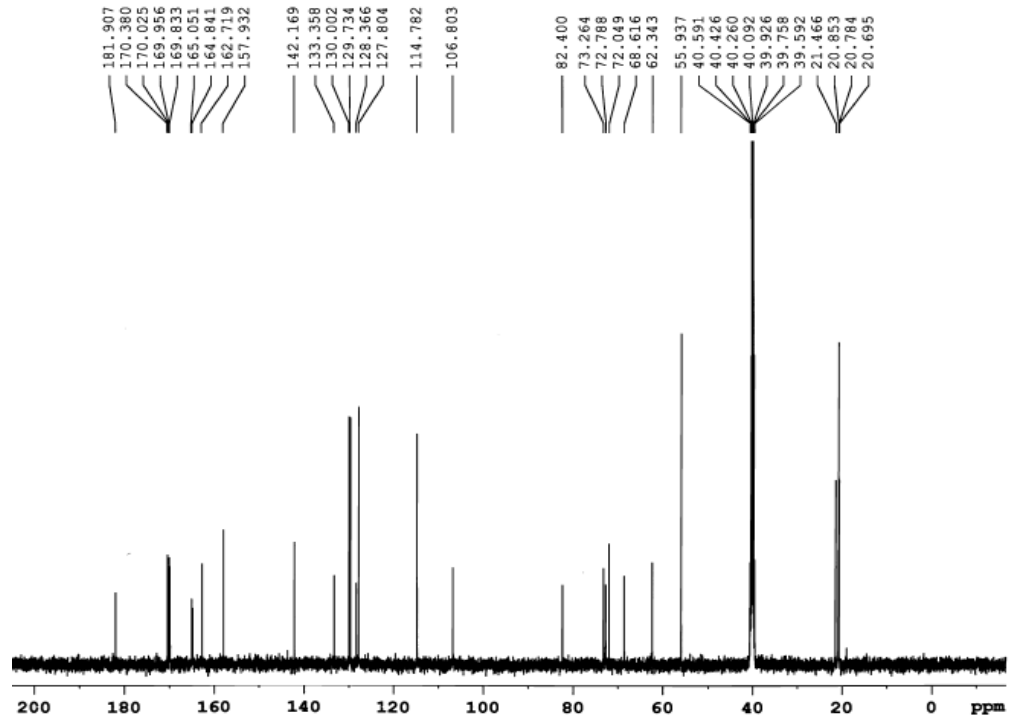
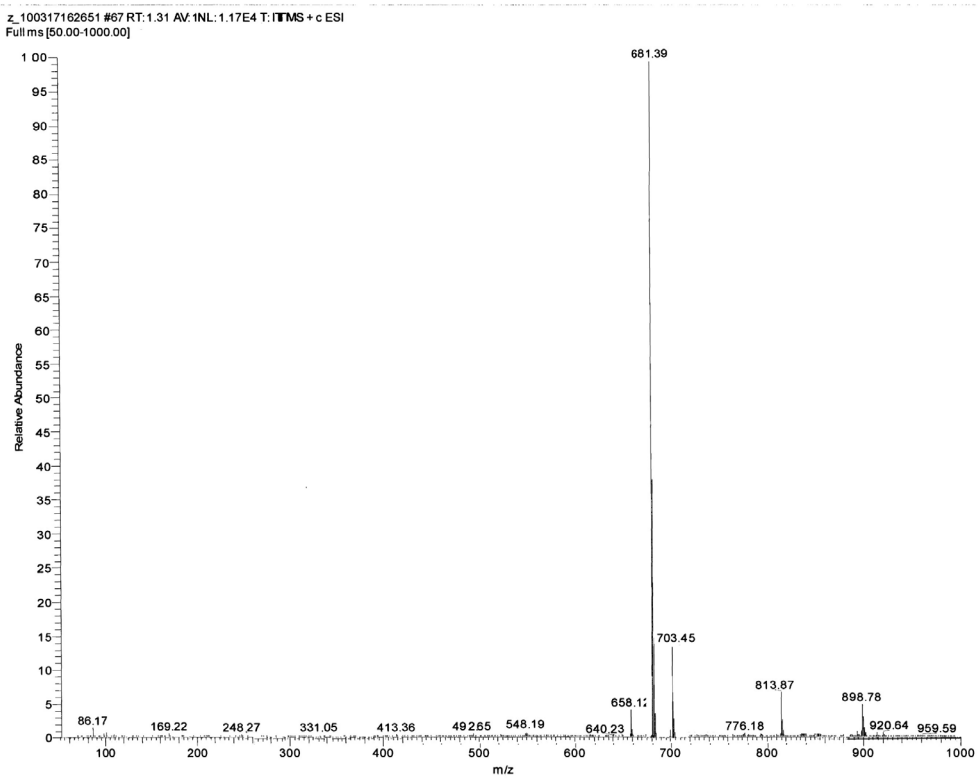
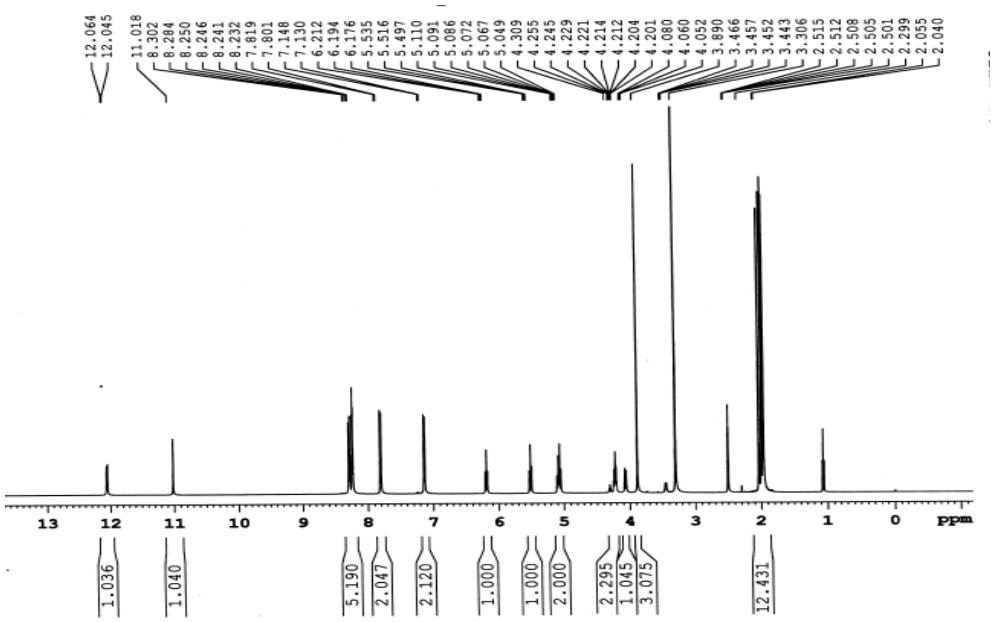
NMR & MS Spectra of N-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-N-[4-(4-methylphenyl)-6-(4-methoxyphenyl)pyrimidine-2-yl]thiourea (7g)

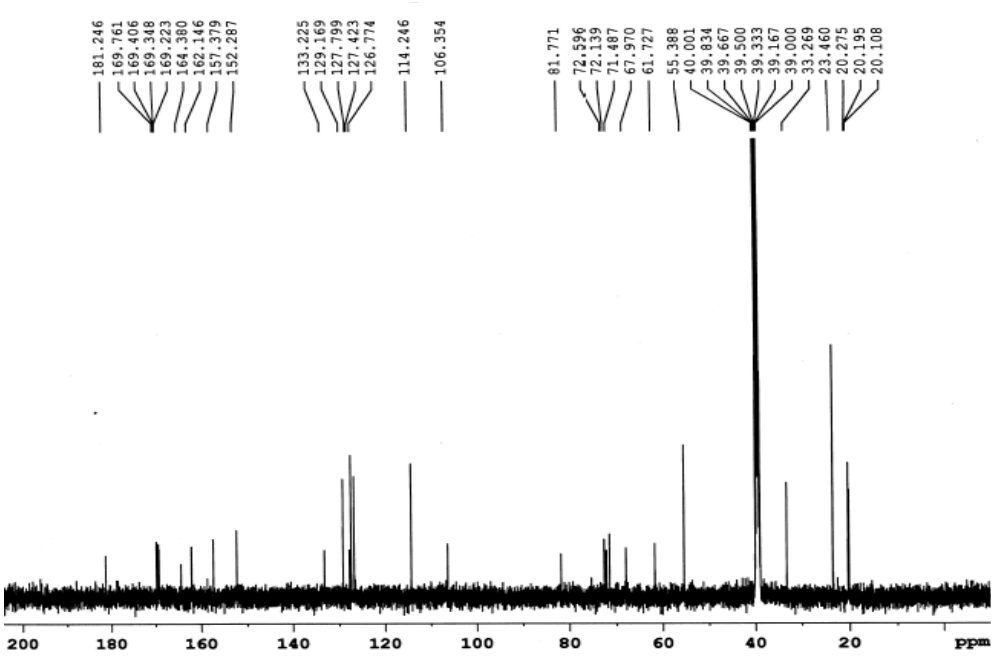






NMR & MS Spectra of N-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-N-[4-(4-isopropylphenyl)-6-(4-methoxyphenyl)pyrimidine-2-yl]thiourea (7h)

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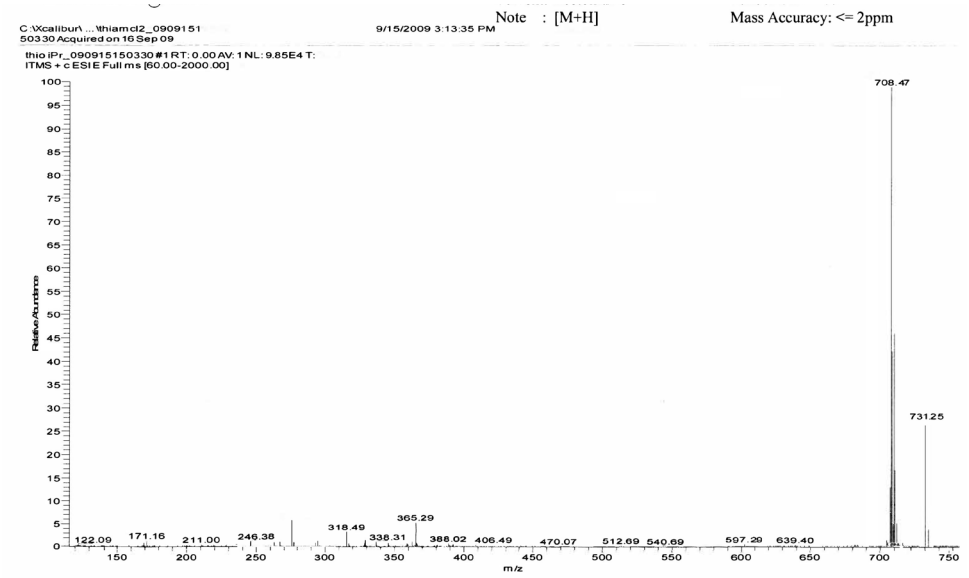
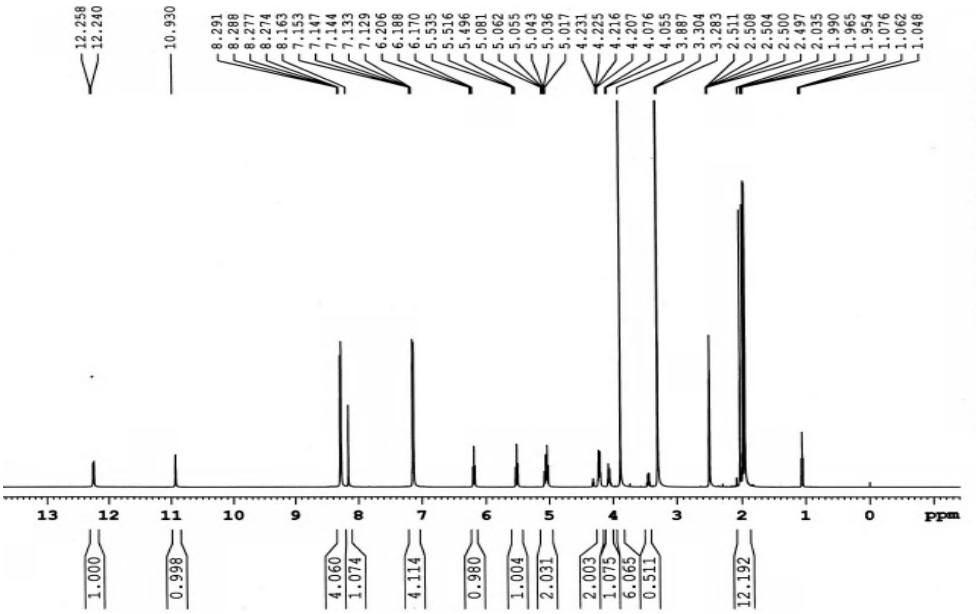
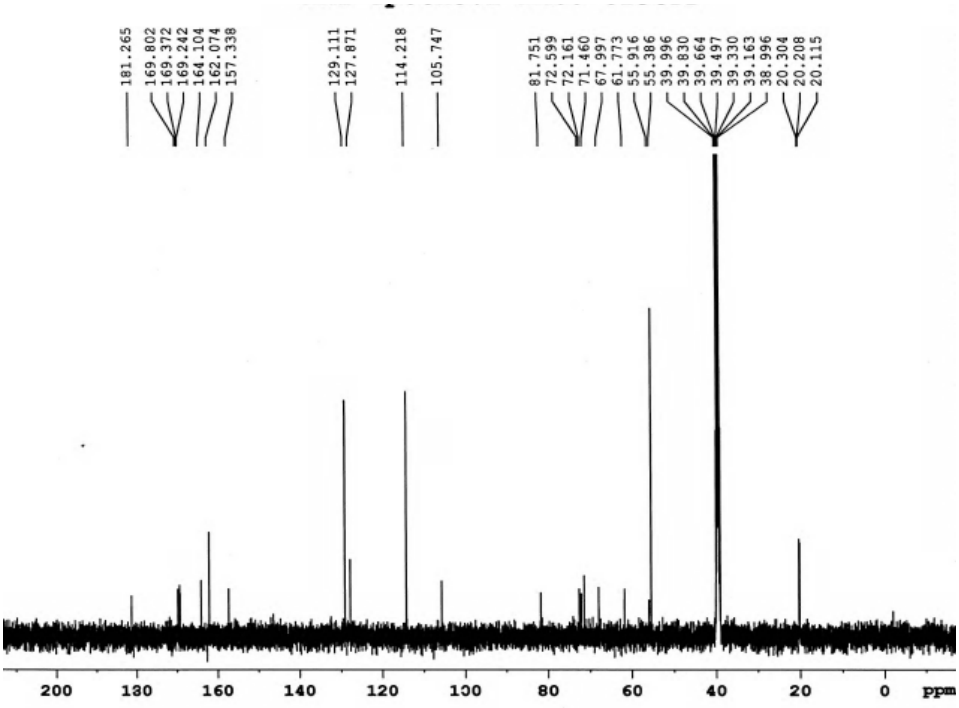
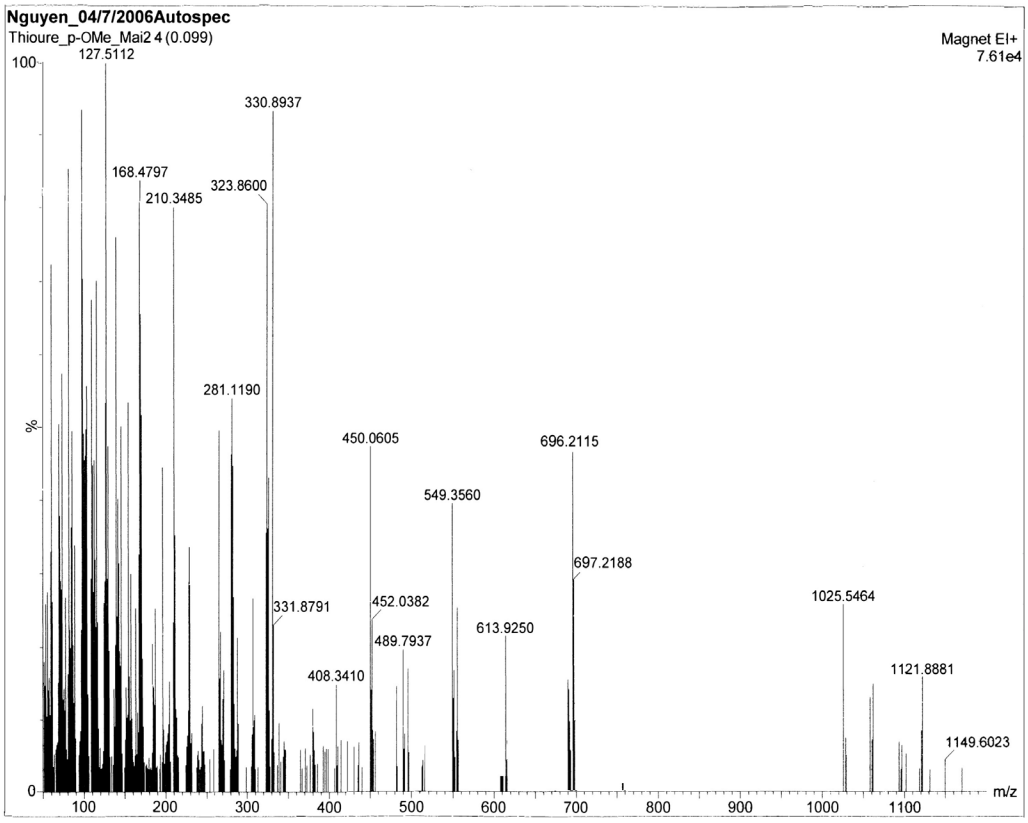
169.406
169.348
169.223
164.380
162.146
157.379
152.287
133.225
129.169
127.799
127.423
126.774
114.246
106.354
81.771
72.596
72.139
71.487
67.970
61.727
55.388
40.001
39.834
39.667
39.500
39.333
39.167
39.000
33.269
23.460
20.275
20.195
20.108
200
180
160
140
120
100
80
60
40
20
ppm


C:\Xcalibur\ ...\thiamcl2_0909151
50330 Acquired on 16 Sep 09
9/15/2009 3:13:35 PM
Note : [M+H]
Mass Accuracy: <= 2ppm
thio iPr_090915150330 #1 RT: 0.00 AV: 1 NL: 9.85E4 T:
ITMS + c ESI E Full ms [60.00-2000.00]
Relative Abundance
m/z
708.47
731.25
365.29
318.49
338.31
388.02
406.49
470.07
512.69
540.69
597.29
639.40
122.09
171.16
211.00
246.38

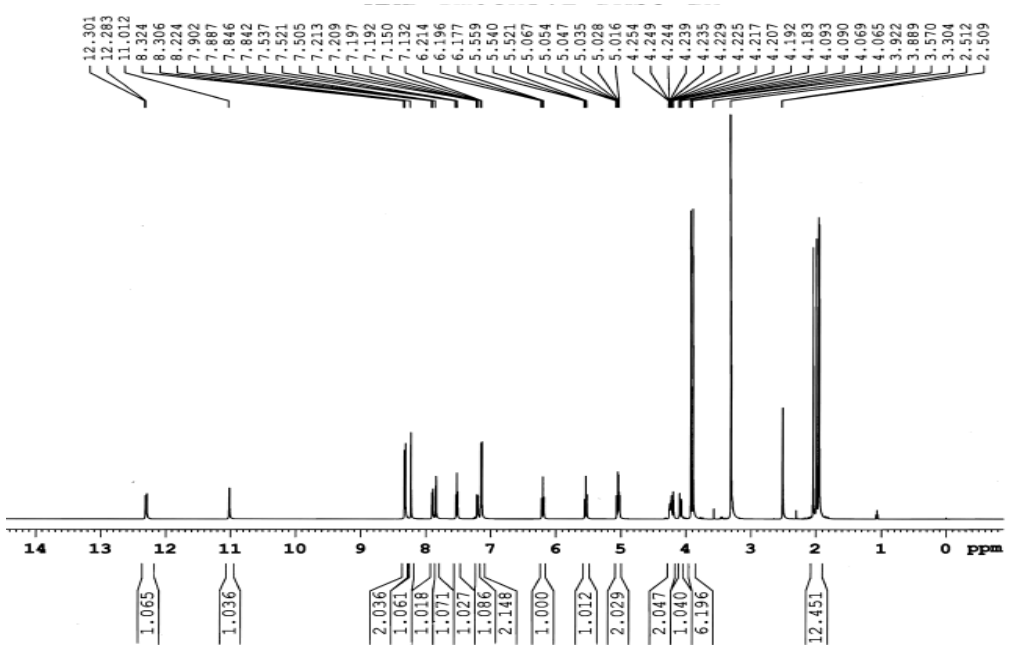
NMR & MS Spectra of N-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-N-[4-(4-methoxyphenyl)-6-(4-methoxyphenyl)pyrimidine-2-yl]thiourea (7i)

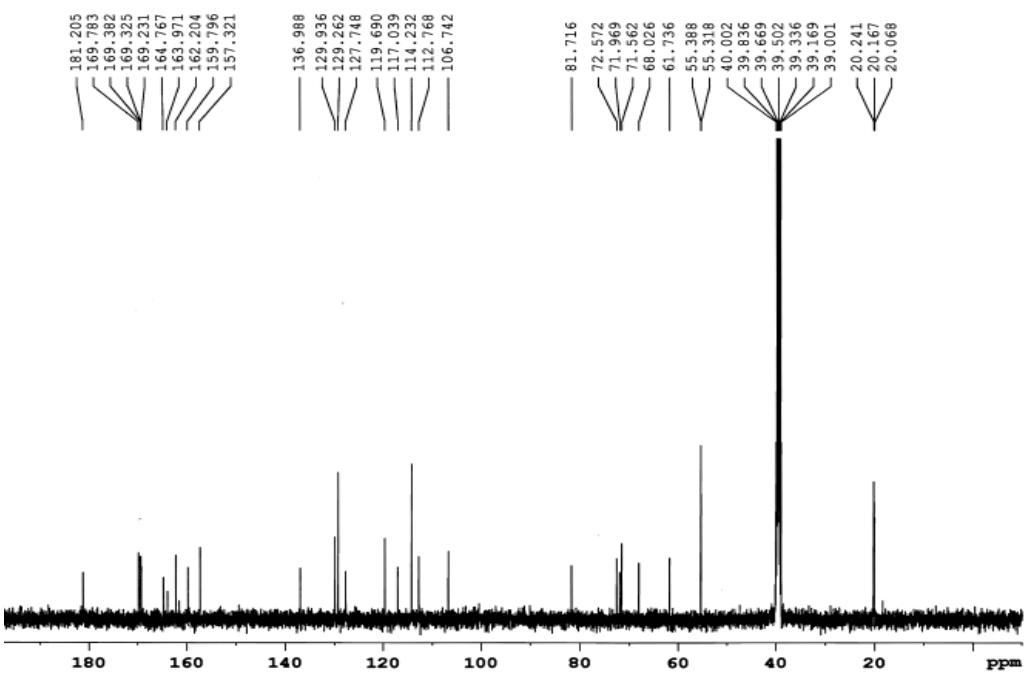






NMR & MS Spectra of N-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-N-[4-(3-methoxyphenyl)-6-(4-methoxyphenyl)pyrimidine-2-yl]thiourea (7j)

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169.382
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169.231
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163.971
162.204
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157.321
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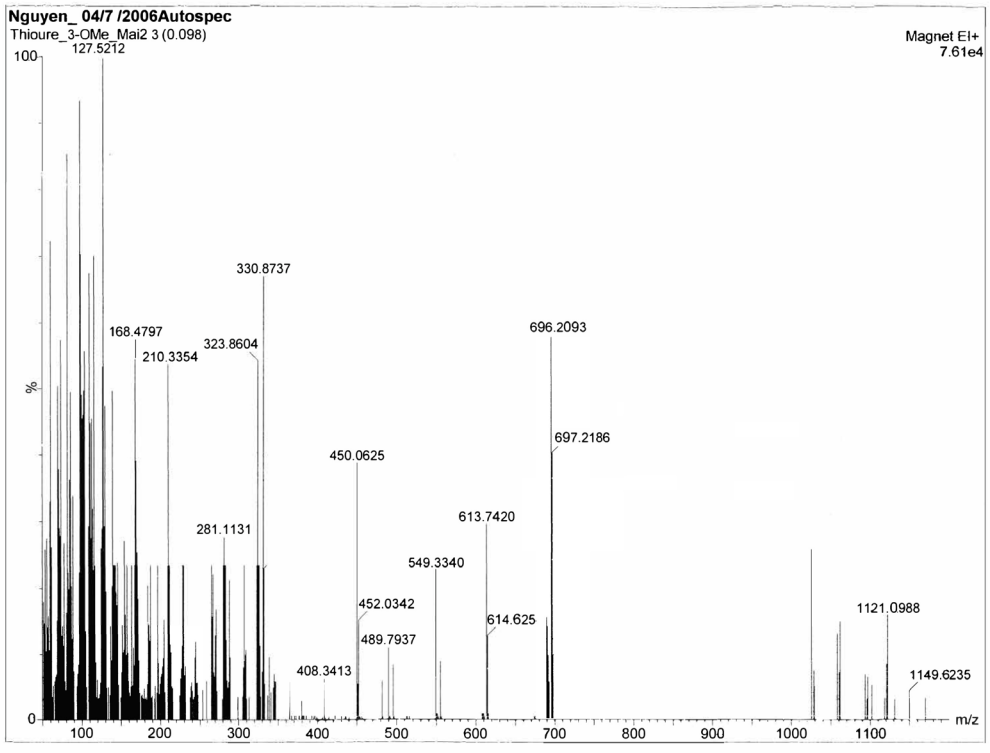
129.262
127.748
119.690
117.039
114.232
112.768
106.742
81.716
72.572
71.969
71.562
68.026
61.736
55.388
55.318
40.002
39.836
39.669
39.502
39.336
39.169
39.001
20.241
20.167
20.068
180
160
140
120
100
80
60
40
20
ppm


Nguyen_ 04/7 /2006Autospec
Thioure_3-OMe_Mai2 3 (0.098)
Magnet EI+
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100
%
0
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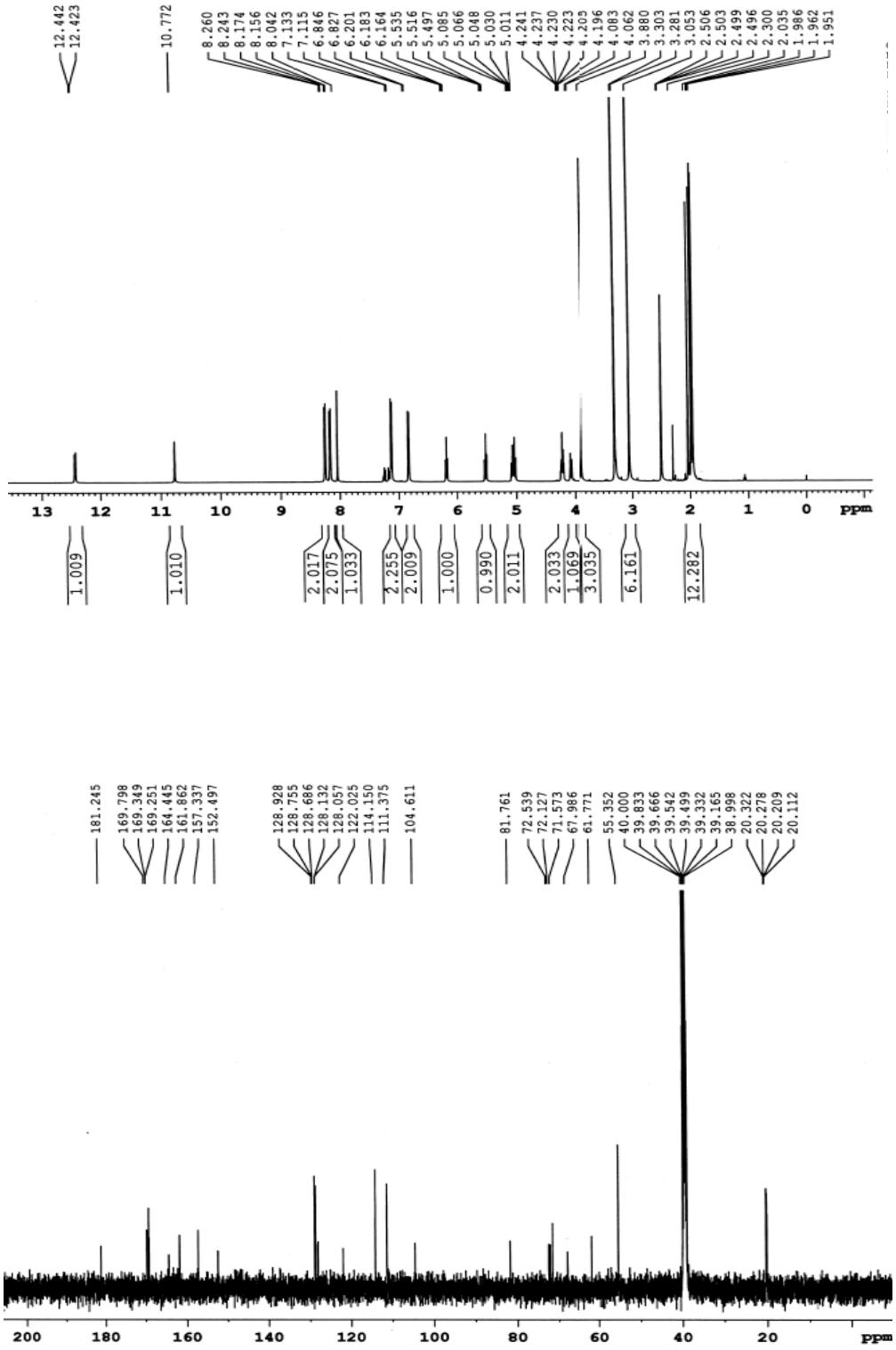
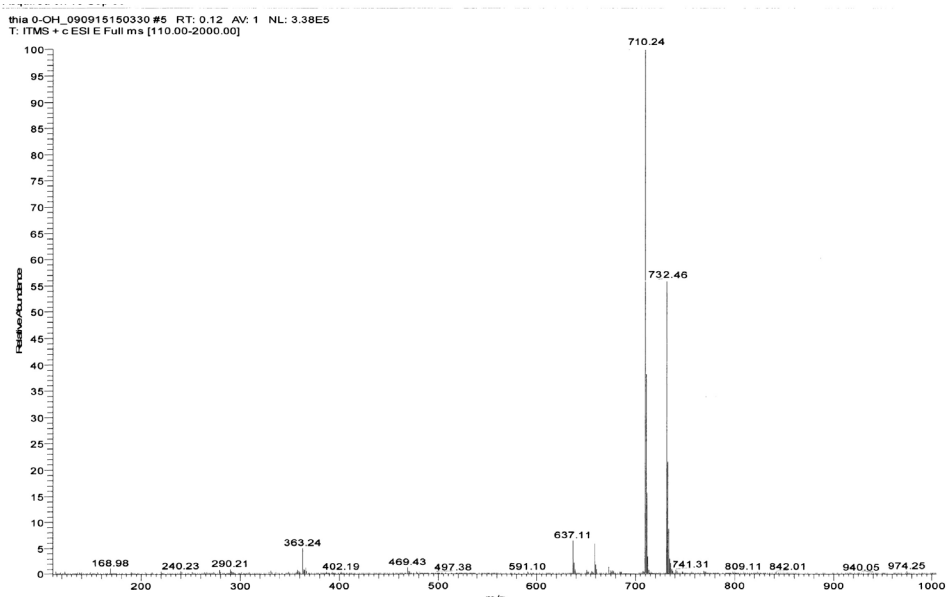
330.8737
168.4797
323.8604
210.3354
696.2093
697.2186
450.0625
281.1131
613.7420
549.3340
452.0342
614.625
489.7937
408.3413
1121.0988
1149.6235
100
200
300
400
500
600
700
800
900
1000
1100
m/z

NMR & MS Spectra of N-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-N-[4-(4-dimethylaminophenyl)-6-(4-methoxyphenyl)pyrimidine-2-yl]thiourea (7k)




thia 0-OH_090915150330 #5 RT: 0.12 AV: 1 NL: 3.38E5
T: ITMS + c ESI E Full ms [110.00-2000.00]
Relative Abundance
710.24
732.46
637.11
363.24
168.98
240.23
290.21
402.19
469.43
497.38
591.10
741.31
809.11
842.01
940.05
974.25