Synthesis, characterization of a new series of thiohydantoin derivatives and study their biological activity against prostate cancer and microbes.

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ABSTRACT

Prostate cancer is one of the most common malignant diseases in men, and the design of new chemical compounds against prostate cancer is one of the challenges faced by modern medicine. In this study, we prepared and characterized a new series of thiohydantoin derivatives by reacting maleimide derivatives (1C-4) with cyclohexyl isothiocyanate.The prepared compounds were diagnosed using (FT-IR ,^{1}H-NMR ,^{13}C-NMR).MTT assay was used to determine the activity of the prepared compounds against prostate cancer cell lines, as the data indicated, based on IC50 values, that the prepared compounds do not have anti-prostate cancer activity. The prepared compounds were also studied against selected types of bacteria and fungi, as the data indicated that some of the prepared series compounds had anti-bacterial and anti-fungal activity under study, as the compound 4C showed a high anti-*staph* activity.

Key words: Thiohydantoin, antibacterial activity, antifungal activity, prostate cancer.

الخلاصة :

يعد سرطان البروستات من أكثر الأمراض الخبيثة شيوعاً لدى الرجال كما ان تصميم مركبات كيميائية جديدة مضادة لسرطان البروستات أحدى التحديات التي يواجهها الطب الحديث . في هذه الدراسة قمنا بتحضير وتشخيص سلسلة جديدة من مشتقات الثيوهيدانتوين وذلك من خلال تفاعل مشتقات الماليماید (1C-4) مع cyclohexyl isothiocyanate.

تم تشخيص المركبات المحضرة باستخدام (^{13}C-NMR, ^{1}H-NMR, FT-IR).

تم استخدام فحص MTT لتحديد نشاط المركبات المحضرة ضد خطوط خلايا سرطان البروستات حيث أشارت البيانات وبالاعتماد على قيم IC50 أن المركبات المحضرة ليس لها فعالية مضادة لخطوط خلايا سرطان البروستات . كذلك تم دراسة المركبات المحضرة ضد أنواع مختارة من البكتريا والفطريات حيث تشير البيانات أن لبعض مركبات السلسلة المحضرة فعالية مضادة للبكتريا والفطريات قيد الدراسة حيث أظهر المركب 4C فعالية عالية مضادة لبكتريا *Staph* .

INTRODUCTION

Hydantoin was first isolated by Bayer in 1861 in the course of her research on uric acid [1]. The data indicate that the nucleus of thiohydantoin is one of the important tributaries of the medical and pharmaceutical aspects, and the first compound that was prepared for this type of compound was by Peter Klason in 1890[2]. 2-thiohydantoin is a biologically active molecule with diverse activities to support the medical and pharmaceutical aspects of facing the challenges faced by the scientific community in combating drug-resistant diseases that are spreading alarmingly where it has been used as an anti-cancer [3-8],anti-mutagenic [9], an antiviral [10], anti-tuberculosis [11], antibacterial [12] [13][14]also showed anti-HIV activity [15] and anti-Alzheimer's disease [16]. One of the most important things that drew the attention of researchers to the thiohydantoin nucleus recently is that it is the nucleus of the parent compound in the anti-prostate cancer drug (enzalutamide, apalutamide) [17]. Recently, some prostate cancer patients have acquired resistance to the drugs prescribed for the treatment of prostate cancer patients (enzalutamide, apalutamide), and the reason is attributed to the emergence of the F876L mutation[18]. Data show that many major corporations have abandoned their support for the discovery of new antibiotics over the last two decades, owing to the economic challenges that come with this aspect[19] [20]. In this study, we prepared a new series of thiohydantoin derivatives and was studied against prostate cancer cell lines. The prepared compounds were also studied against specific types of bacteria and fungi, as the data indicated that some of the prepared compounds had anti-bacterial and anti-fungal activity under study.

Experimental

Chemistry

Infrared spectra of the prepared compounds were recorded using a Japanese-made Shimadzu 8400 FT.IR device and in the form of potassium bromide tablets in the (400–4000) cm^{-1} region at room temperature in the laboratories of the Department of Chemistry - College of Education for Pure Sciences - University of Basra. ^{1}H and ^{13}C-NMR spectra were recorded using a BRUCKER-500MHz and 125MHz instrument, DMSO- d6 as a solvent , and TMS as an internal reference. All chemical displacements were measured in ppm. ^{1}H and ^{13}C-NMR spectra were recorded at University of Tehran-Iran

Synthesis of compounds (1C-4) [21] [22]

0.01mol of maleimide (1A-4) with 0.01mol of Isoniazid(isonicotinohydrazide) (B) was dissolved in 30ml of absolute ethanol in a round flask equipped with a magnetic stirrer and leaving the mixture under reflux

for (6-32)hrs . The reaction was monitored by TLC(thin layer chromatography). After the reaction is completed, the filtrate is separated and recrystallized or washed in absolute ethanol.

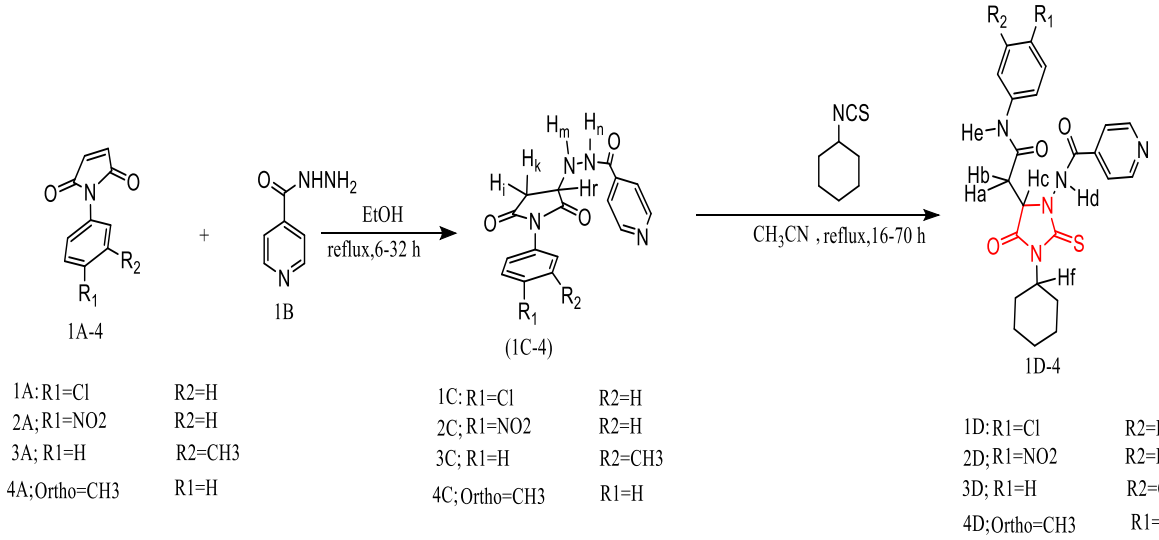
Synthesis of compounds (1D-4) [21] [22]

0.02 mol of maleimide derivatives (1C-4) were reacted with 0.02 mol of cyclohexyl isothiocyanate in the presence few drops of glacial acetic acid as a catalyst in 30 ml of acetonitrile in a round flask equipped with a magnetic stirrer. The mixture was heated by reflux for 18-70 hrs. The reaction was monitored by TLC(thin layer chromatography). After the reaction is completed, the solvent is evaporated and then recrystallized by a mixture of acetone and hexane.

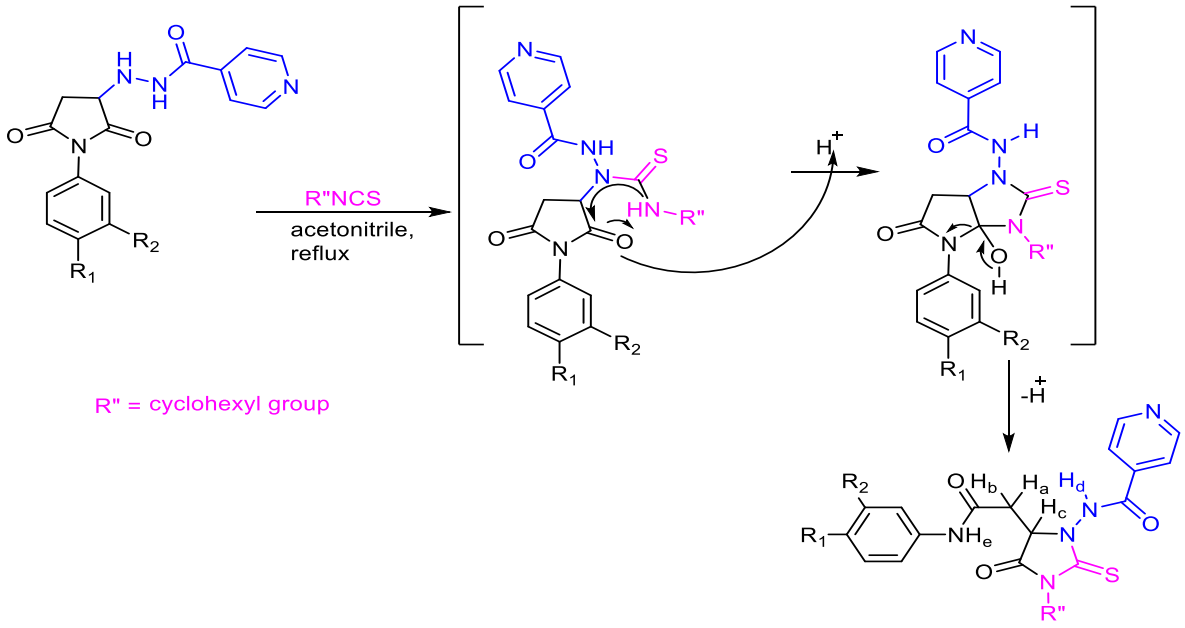
RESULTS AND DISCUSSION

In this study, we prepared a new series of thiohydantoin derivatives, where the reaction included two steps. In the first step, the maleimide derivatives (1C-4) were prepared by reacting different maleimides with Isonazid (isonicotinohydrazide) (B) in absolute ethanol, and the time required for the reaction to occur was (6-32) hrs. The second step included the preparation of thiohydantoin derivatives (1D-4) through the reaction of maleamide derivatives (1C-4) with cyclohexyl isothiocyanate in acetonitrile as a solvent and in the presence of drops of glacial acetic acid as a catalyst,Scheme 1. The prepared series compounds were diagnosed by (FT-IR, ^{1}HNMR, ^{13}C-NMR). The obtained spectra data indicate that the structures of the assumed compounds are correct. The most important bands shown by the FT-IR spectrum, wide band at 3300-3200 belong to the (NH group) It is also noticed that a wide band appears at 3479-3300 belongs to the (NH amide group). The weak band at 3186-3000 is due to (CH arom.). Another band appears at 2951-2900 due to (CH-aliph). The strong and distinct sharp band at 1766-1650 is attributed to the (C=O) group, while the group of bands that appear at 1615-1450 belongs to (C=C arom). . Also, the recorded spectrum of the prepared compounds showed two bands, one at 1449-1400 attributed to the (C = S) group, and the other at 1350-1300 belonged to the (C-N) group. Among the most important signals that were shown by the recorded ^{1}HNMR spectrum of maleimide derivatives (1C-4). doublet of doublet signal at 3.0 ppm with a coupling constant *J = (18.0, 4.1 Hz)* attributed to Hi. doublet of doublet signal at 2.8 ppm with a coupling constant *J = (18.0, 4.0 Hz)* is attributed to Hk .Also the recorded ^{1}HNMR spectrum of thiohydantoin derivatives (1D-4) showed a triplet signal at 4.63ppm with a coupling constant *J = 4.3 Hz* due to Hc, multiple signal at 4.63ppm

attributed to Hf, multiple signal at (3-2.7)ppm attributed to HaHb. The recorded 13C-NMR spectrum of maleimide derivatives (1C-4) and thiohydantoin (1D-4) derivatives showed signs consistent with the structures of the putative compounds. The most important signals showed by the ^{13}C-NMR spectrum, a signal at 174ppm is attributed to the carbonyl group in (imidazolidine-2,4-dione), a signal at 164ppm is attributed to a carbonyl group in (Isonicotinohydrazide), a signal at 171ppm belongs to a carbonyl group in (thioimidazole), a signal at 174ppm attributed to the thiocarbonyl group (C=S). A signal at 164ppm attributed to the carbonyl group appears in (NNHC=O) 'signal at 166ppm is attributed to the carbonyl group in (ArNHC=O). Figures 1 to 14 shows the spectra that were recorded for the prepared compounds.



Scheme (1) shows the general method for preparing the prepared compounds



Scheme 2 shows the Mechanism of preparing thiohydantoins (1D-4).

Structure analysis data of the prepared compounds;

N'-(1-(4-chlorophenyl)-2,5-dioxopyrrolidin-3-yl)isonicotinohydrazide(1C):

White solid powder; yield 89%; mp= 218-222 ºC;IR (KBr, cm^{-1}) 3408 (NH amide), 3317(NH), 3061 (CH arom), 2993,2937 (CH-aliph), 1784 (C=O imidazolidine-2,4-dione), 1710 (C=O imidazolidine-2,4-dione),1639(C=O Isonicotinohydrazide), 1546, 1498 (C=C arom), 1406 (C-N).^{1}H NMR (500 MHz, DMSO) δ 10.46 (s, 1H,H_n), 8.74(d , *J* = 5.0,2H,H-Ar), 7.73(d , *J* = 5.0 Hz ,2H,H-Ar), 7.58 (d, *J* = 8.6 Hz , 2H,H-Ar), 7.33 (d, *J* = 8.5 Hz, 2H,H-Ar) ,6.14 (s, 1H,H_m), 4.31-4.21 (m, 1H,H_r), 3.09 (dd, *J* = 17.9, 8.6Hz, 1H,H_i), 2.80 (dd, *J* = 18.0, 3.9 Hz, 1H,H_k) , ^{13}C NMR (126 MHz, DMSO) δ175 (C=O imidazolidine-2,4-dione), 174 (C=O imidazolidine-2,4-dione), 164.39(C=O Isonicotinohydrazide), 150, 139, 132, 131, 128.95, 128.63,121 (C-Ar), 57(CH_r), 34(CH_iH_k).

N'-(1-(4-nitrophenyl)-2,5-dioxopyrrolidin-3-yl)isonicotinohydrazide(2C):

White solid powder; yield 49%; mp= 203-205 ºC ; IR (KBr, cm-1) 3415 (NH amide), 3304(NH), 1788(C=O imidazolidine-2,4-dione), 1720 (C=O imidazolidine-2,4-dione), 1641, 1527,1469, (C=C arom), 1350 (C-N).^{1}H NMR (500 MHz, DMSO) δ 10.53 (d, *J* = 6.1 Hz , 1H,H_n), 8.74(d , *J* = 5.1,2H,H-Ar), 8.38(dd , *J* = 7.6 ,4.5Hz ,2H,H-Ar), 7.75 (d, *J* = 5.0 Hz , 2H,H-Ar), 7.64 (d, *J* = 8.5 Hz, 2H,H-Ar) ,6.24 (d, *J* = 6.0 Hz, 1H,H_m), 4.32 (dt, *J* = 5.9, 2.9 Hz, 1H,H_r), 3.16 (ddd, *J* = 17.9, 8.6,1.9 Hz, 1H,H_i), 2.80 (dt, *J* = 18.0, 3.0 Hz, 1H,H_k) ^{13}C NMR (126 MHz, DMSO) δ 174.75(C=O imidazolidine-2,4-dione), 174.24(C=O

imidazolidine-2,4-dione), 164(C=O Isonicotinohydrazide), 150, 146, 139, 137, 127, 124, 121 ,124 , 121 (C-Ar), 57(CH_r), 34(CH_iH_k).

N'-(2,5-dioxo-1-(m-tolyl)pyrrolidin-3-yl)isonicotinohydrazide(3C):

White solid powder; yield 31%; mp= 200-202 ºC;IR (KBr, cm-1) 3477 (NH amide), 3325(NH), 1708(C=O imidazolidine-2,4-dione), 1643 (C=O Isonicotinohydrazide), 1537, 1492,1471, (C=C arom), 1340 C-N), ^{1}H NMR (500 MHz, DMSO) δ 10.47 (d, *J* = 6.0 Hz , 1H,H_n), 8.81 – 8.69 (m , 2H,H-Ar), 7.74 (dd, *J* = 4.5, 1.8 Hz , 2H,H-Ar), 7.37 (t, *J* = 7.6 Hz, 1H,H-Ar), 7.23 (d, *J* = 7.6 Hz, 1H , H-Ar), 7.07 (d, *J* = 8.9 Hz, 2H, H-Ar) 6.12 (t, *J* = 5.5 Hz, 1H,H_m), 4.28 (dt, *J* = 8.7, 4.3 Hz, 1H,H_r), 3.09 (dd, *J* = 17.9, 8.5 Hz, 1H,H_i), 2.80 (dd, *J* = 17.9, 3.7 Hz, 1H,H_k) , 2.33 (s, 3H,CH_3).

N'-(2,5-dioxo-1-(o-tolyl)pyrrolidin-3-yl)isonicotinohydrazide(4C):

White solid powder; yield 47%; mp= 184-186 ºC;IR (KBr, cm-1) IR (KBr, cm-1) 3410 (NH amide), 3327 (NH) ,3041 (CH arom), 2987,2933 (CH-aliph), 1784(C=O imidazolidine-2,4-dione), 1712 (C=O imidazolidine-2,4-dione), 1643, 1552,1529,1469, (C=C arom.), 1390 (C-N).^{1}H NMR (500 MHz, DMSO) δ 10.47 (d, *J* = 6.2 Hz , 1H,H_n), 8.74 (d , *J* = 4.4 Hz, 2H,H-Ar), 7.76-7.75 (m, *J* = 2H,H-Ar), 7.34 (dd, *J* = 4.5,1.2 Hz, 2H,H-Ar), 7.34 (dd, *J* = 4.5,1.2 Hz, 2H , H-Ar), 7.30-7.28 (m, 1H, H-Ar) 7.15 (dd, *J* = 8.2,2.7 Hz, 1H,H-Ar), 7.76-7.75 (m, *J* = 2H,H-Ar), 6.15-6.13 (m, 1H,Hm), ,4.38-4.35 (m,1H,H_r), 3.20 (dd, *J* = 17.8, 8.6 Hz, 1H,H_i), 3.11 (dd, *J* = 18.0, 8.6 Hz, 1H,H_k) , 2.04 (s,CH_3).

N-(5-(2-((4-chlorophenyl)amino)-2-oxoethyl)-3-cyclohexyl-4-oxo-2-thioxoimidazolidin-1-yl)isonicotinamide(1D):

White solid powder; yield 42 %, mp.= 140-143 ºC; IR (KBr, cm-1): 3479 (NH amide), 3415 (NH), 3118 ,3053 (CH arom.), 2995 (CH-aliph.), 1749 (C=O thioimidazole), 1710 (C=O amide),1678, 1616,1533 (C=C arom.), 1490 (C=S), 1311 (C-N); ^{1}H NMR (500 MHz, DMSO-d6) δ 11.65(s, 1H, Hd), 10.24 (s, 1H, He), 8.78 (d, *J* = 6.1 Hz, 2H, H-Ar), 7.78 (d, *J* = 6.2 Hz, 2H, H-Ar), 7.55 (d,*J*=8.8Hz, 2H, H-Ar),7.35 (d, *J* = 8.6 Hz, 2H, H-Ar), 4.64 (t, *J* = 4.3 Hz, 1H, Hc), 4.50-4.45 (m, 1H, Hf), 3.20- 2.98 (m, 2H, Ha, Hb), 2.18 (dt, *J*=23.9,12.0Hz ,2H, H-cyclohexyl), 1.83(d, *J*=13.1Hz ,2H, H-cyclohexyl), 1.74 (t,*J*=15.6 2H, H-cyclohexyl),1.65 (d, *J*=12.8 Hz ,1H, H-cyclohexyl)1.29 (s, 2H, H-cyclohexyl), 1.16(t,*J*=13.0,1H, H-cyclohexyl) ; ^{13}C NMR (126 MHz, DMSO-d6) δ 184 (C=S), 171 (C=O thioimidazole), 166 (ArNHC=O), 164 (NNHC=O), 150, 138, 137, 128, 126, 121,120 (C-Ar), 59 (CHc) , 55 (CHf), 34 (CHaHb), 28, 27, 25.58, 25, 24 (C-cyclohexyl).

N-(3-cyclohexyl-5-(2-((4-nitrophenyl)amino)-2-oxoethyl)-4-oxo-2-thioxoimidazolidin-1-yl)isonicotinamide(2D):

White solid powder; yield 60%; mp= 210-213 ºC ; IR (KBr, cm^{-1}) 3304 (NH amide), 3151 (NH) ,2929 (CH arom.), 2856 (CH-aliph.), 1766(C=O thioimidazole), 1749 (C=O thioimidazole), 1730 (C=O amide), 1597, 1552,1512, (C=C arom.), 1332 (C-N). ; ^{1}H NMR (500 MHz, DMSO-d6) δ 11.66 (s, 1H, Hd), 10.75 (s, 1H, He), 8.81-8.76 (m, 2H, H-Ar), 8.25-8.19 (m, 2H, H-Ar), 7.82-7.74 (m, 4H, H-Ar), 4.68 (t, *J* = 4.2 Hz, 1H, Hc), 4.49 (ddt, *J* = 12.2, 7.2, 3.8 Hz ,1H, Hf), 3.12 (d, *J* = 4.2 Hz, 2H, Ha, Hb), 2.28-2.13 (m, 2H, H-cyclohexyl), 1.84 (d, *J*=15.0 Hz ,3H, H-cyclohexyl), 1.79-1.71 (m, 2H, H-cyclohexyl),1.66 (d, *J*=12.4 Hz ,1H, H-cyclohexyl),1.29(ddd, *J* = 16.0, 10.9, 3.2 Hz,1H, H-cyclohexyl), 1.17 (ddd, *J* = 16.0, 8.3, 3.3 Hz, 1H,H- cyclohexyl).

N-(3-cyclohexyl-4-oxo-5-(2-oxo-2-(m-tolylamino)ethyl)-2-thioxoimidazolidin-1-yl)isonicotinamide(3D):

White solid powder; yield 53%; mp= 235-238 ºC; ^{1}H NMR (500 MHz, DMSO-d6) δ 11.62 (s, 1H, Hd), 9.96 (s, 1H, He), 8.79-8.74 (m, 2H, H-Ar), 7.80-7.75 (m, 2H, H-Ar), 7.35-7.28 (m, 2H, H-Ar), 7.16 (t, *J* = 7.8 Hz, 1H), 6.85 (d, *J* = 7.5 Hz, 1H), 4.64 (t, J = 4.4 Hz, 1H, Hc), 4.52 – 4.43 (m, 1H, Hf), 3.06 – 2.94 (m, 2H, Ha, Hb), 2.25 (s, 3H,CH_3), 2.19 (d, *J* = 18.1 Hz, 2H, H-cyclohexyl), 1.83 (d, *J* = 13.2 Hz, 2H, H-cyclohexyl), 1.74 (t, *J* = 14.4 Hz, 2H,H-cyclohexyl),1.65 (d, J=13.2 Hz ,1H, H-cyclohexyl), 1.30 – 1.28 (m, 3H, H-cyclohexyl), 1.17 (t, *J* = 13.0 Hz, 1H, H-cyclohexyl) .

N-(3-cyclohexyl-4-oxo-5-(2-oxo-2-(o-tolylamino)ethyl)-2-thioxoimidazolidin-1-yl)isonicotinamide (4D):

White solid powder; yield 58%; mp= 60-62 ºC; ^{1}H NMR (500 MHz, DMSO-d6) δ 11.62 (s, 1H, Hd), 9.37 (s, 1H, He), 8.84-8.72 (m, 2H, H-Ar), 7.87-7.71 (m, 2H, H-Ar), 7.37 (t, *J* = 11.0 Hz, 1H,H-Ar), 7.18 (d, *J* = 7.4 Hz, 1H, H-Ar), 7.12 (t, *J* = 7.5 Hz, 1H, H-Ar), 7.05 (t, *J* = 7.4 Hz, 1H, H-Ar), 4.67 (t, *J* = 4.5 Hz, 1H, Hc), 4.46 (tt, *J* = 12.6, 3.8 Hz, 1H, Hf), 3.05 (qd, *J* = 17.1, 4.5 Hz, 2H, Ha, Hb), 2.27-2.12 (m, 4H, H-cyclohexyl), 1.82 (s, 2H, H-cyclohexyl), 1.72 (s, 3H,CH_3) , 1.64 (d, *J*=13.1 Hz ,1H, H-cyclohexyl), 1.32 – 1.23 (m, 1H, H-cyclohexyl),1 .16 (dd, *J* = 14.3, 10.9 Hz, 1H) ; ^{13}C NMR (126 MHz, DMSO-d6) δ184 (C=S), 171(C=O thioimidazole), 166(ArNHC=O), 164 (NNHC=O), 150, 138, 136, 131, 130, 125,124,121, (C-Ar), 59 (CHc) , 55 (CHf), 34 (CHaHb), 28, 27, 25.56, 25.42, 24 (C-cyclohexyl),17(CH_3).

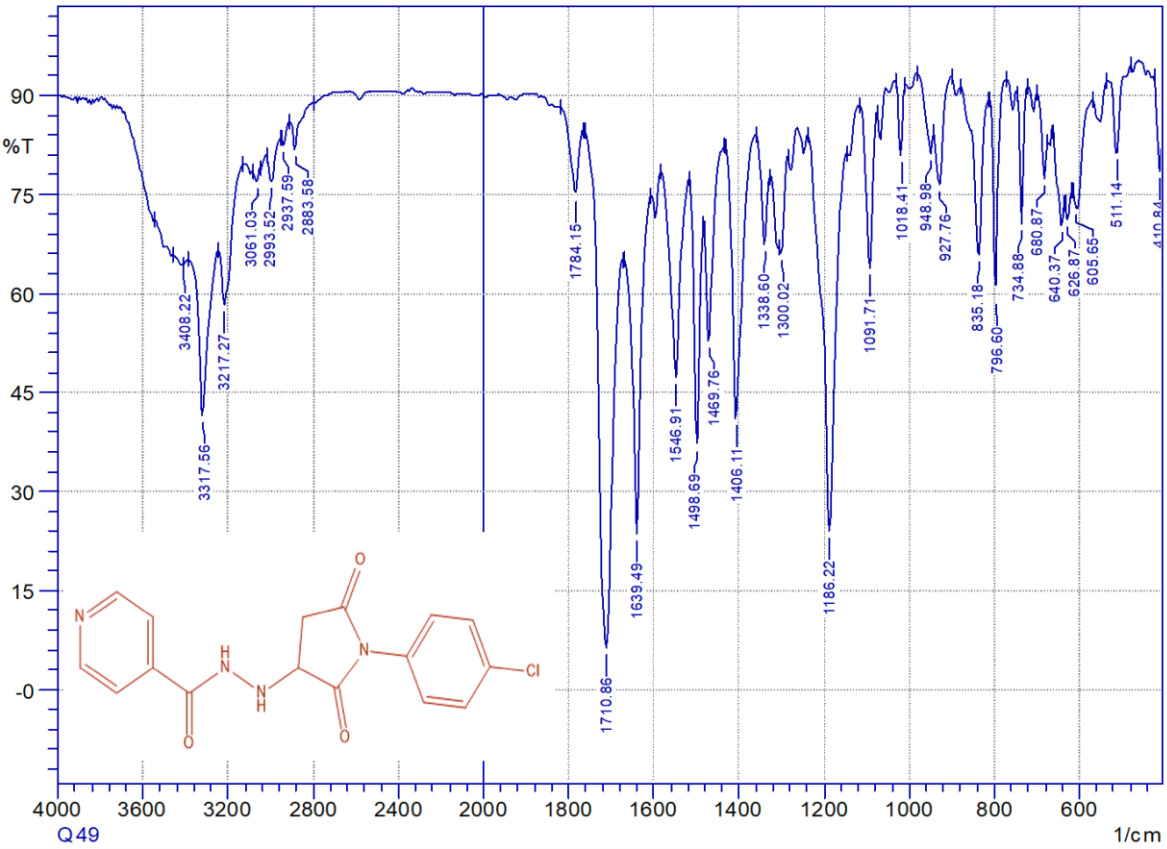


Figure (1) FT-IR spectrum of compound 1C

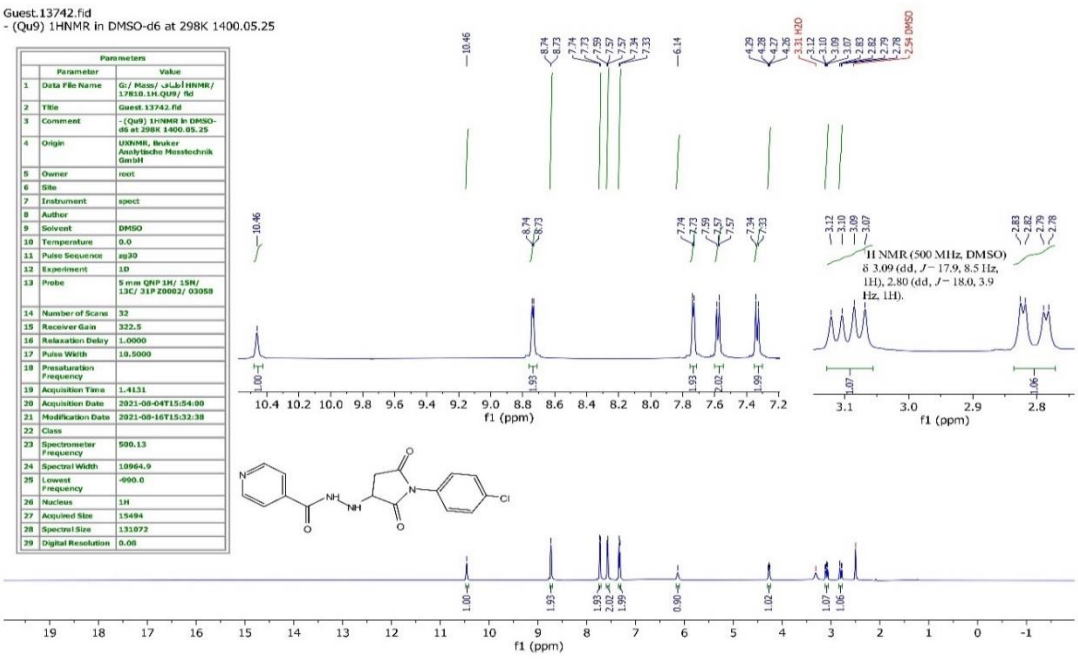


Figure (2) ^{1}H-NMR spectrum of compound 1C

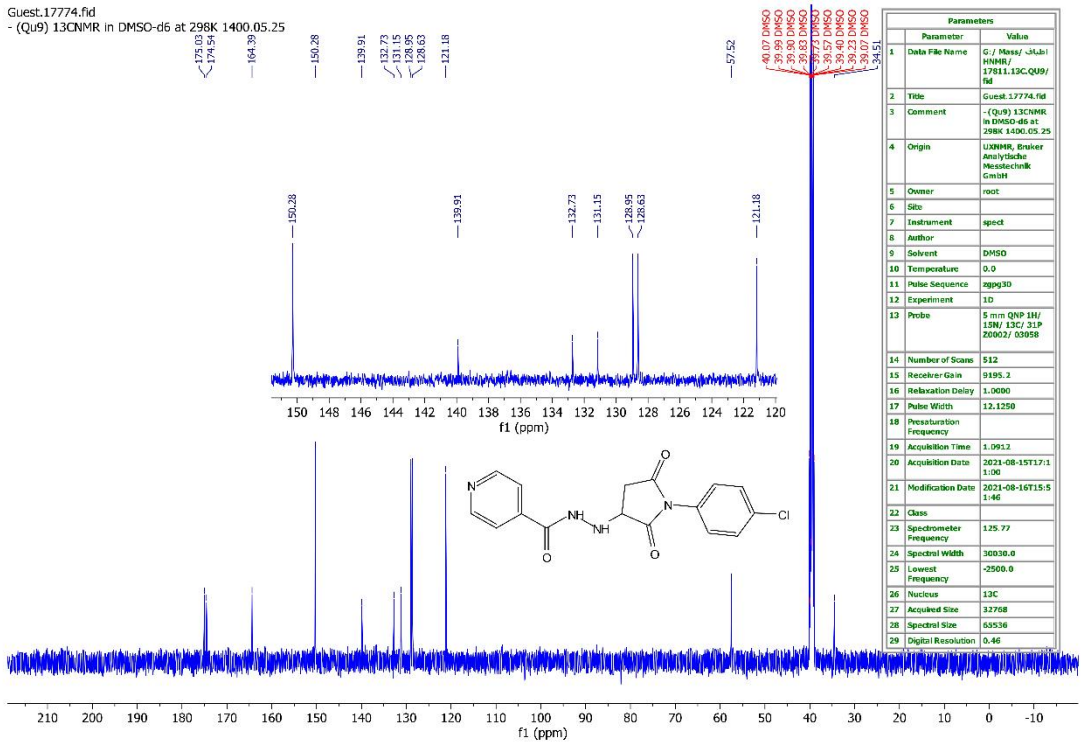


Figure (3) ^{13}C-NMR spectrum of compound 1C

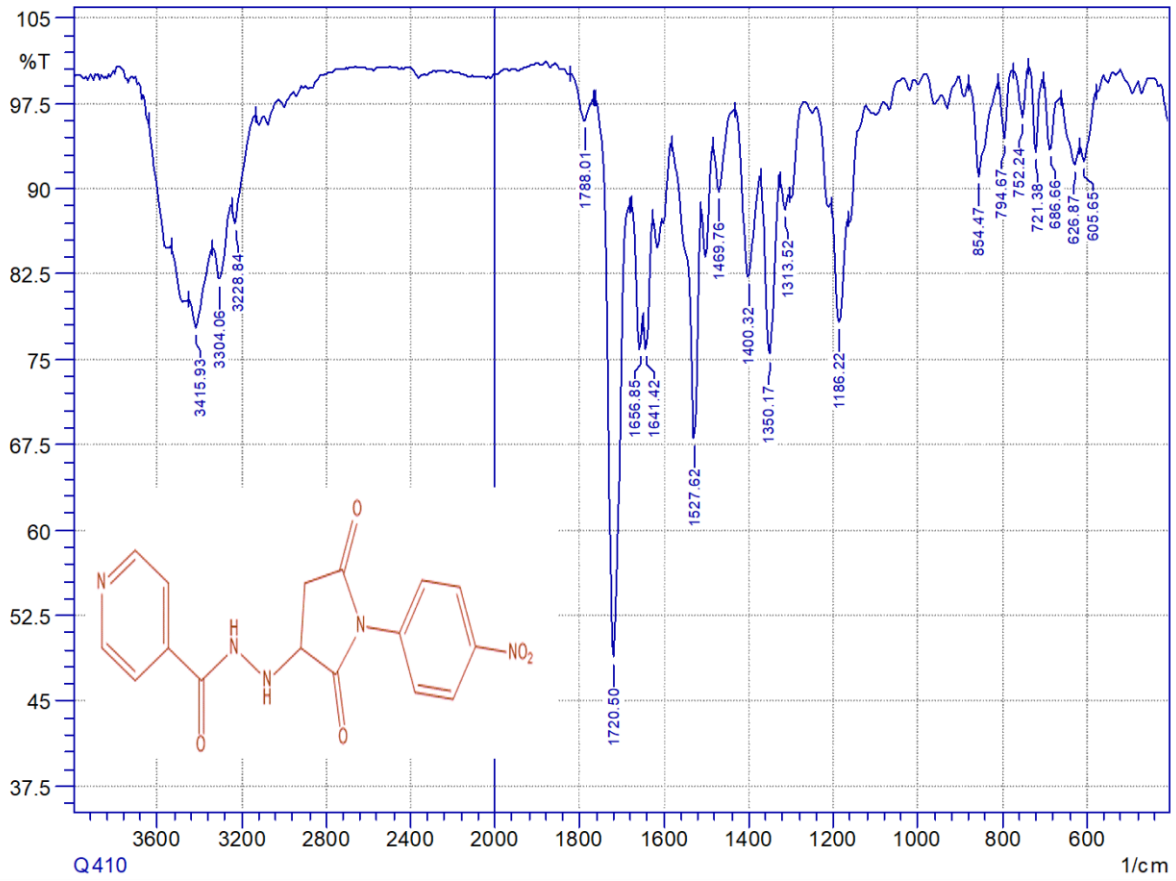


Figure (4) FT-IR spectrum of compound 2C

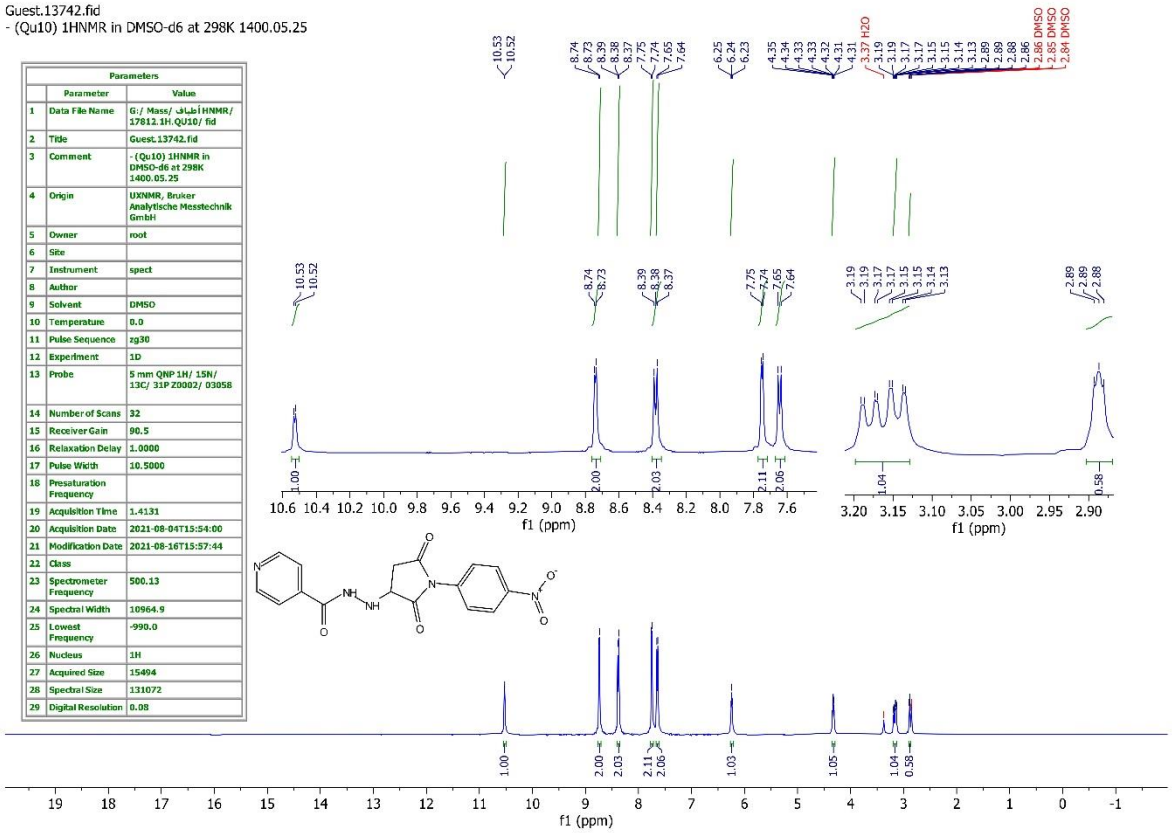


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Figure (5) ^{1}H-NMR spectrum of compound 2C

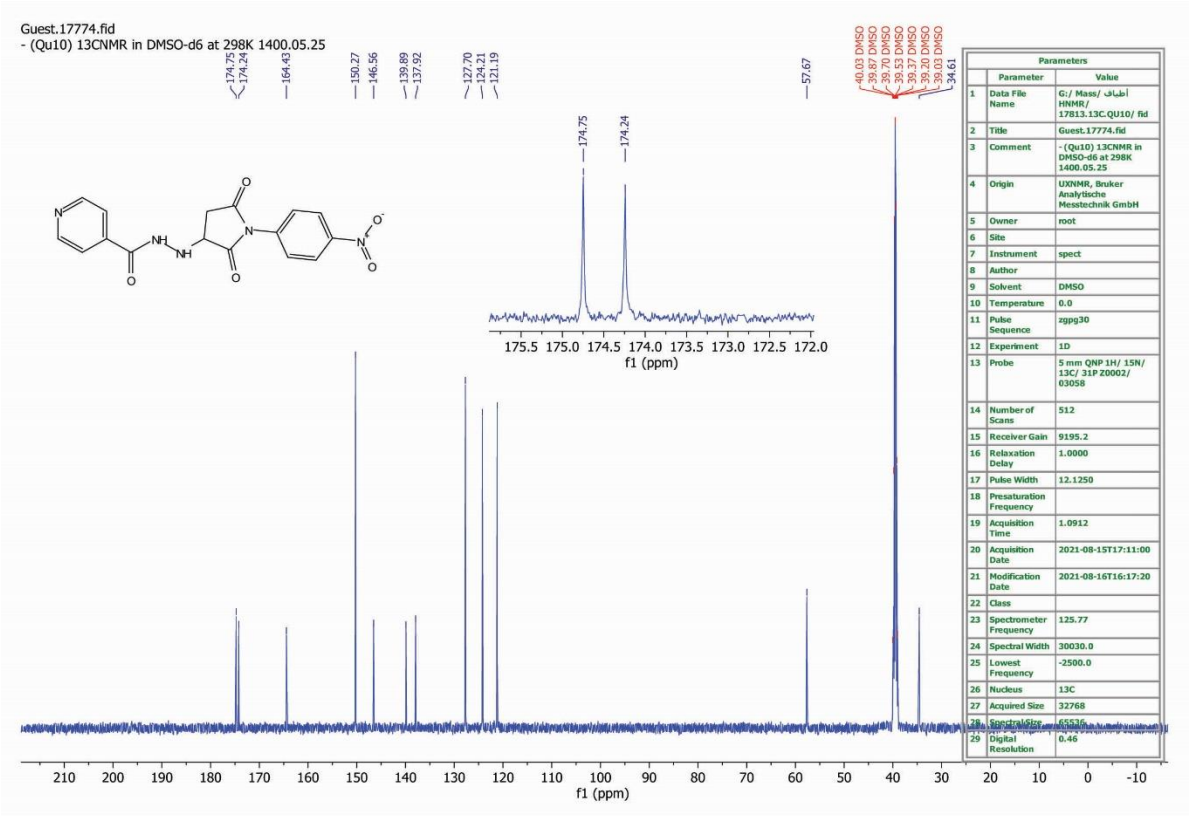

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Figure (6) ^{13}C-NMR spectrum of compound 2C

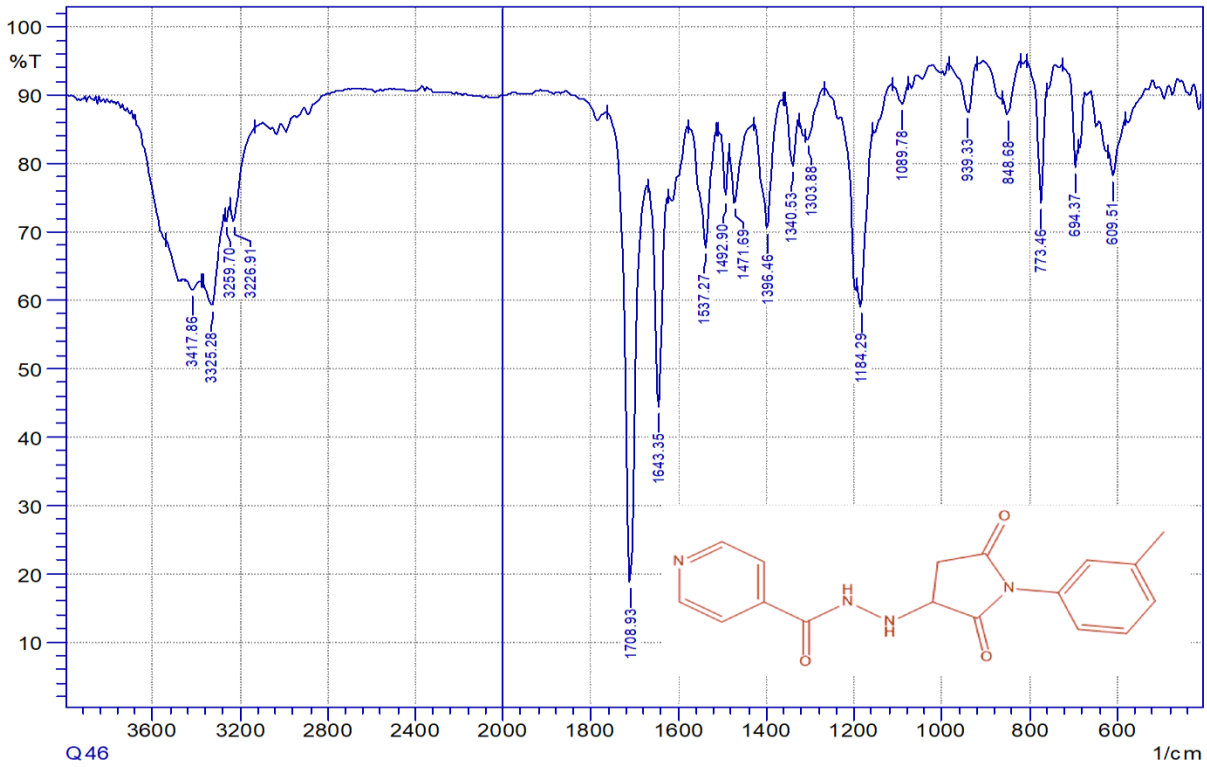

Figure (7) FT-IR spectrum of compound 3C

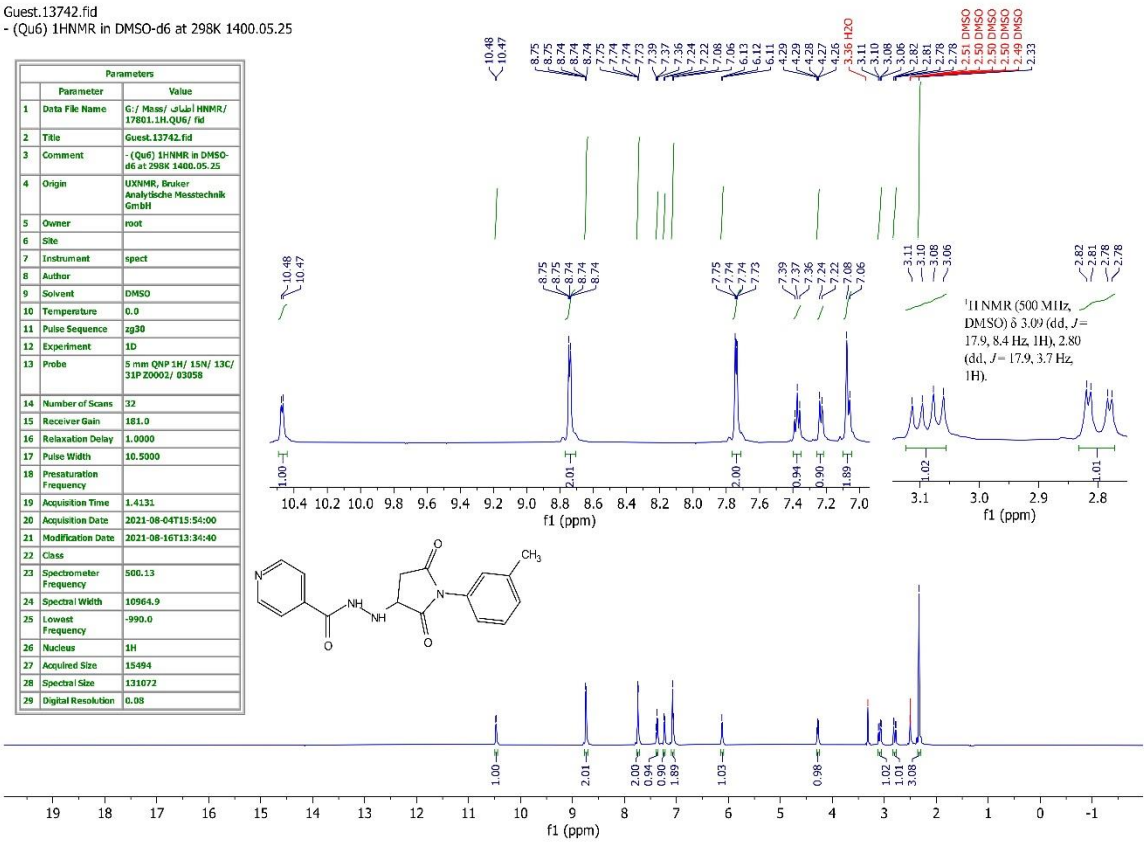

Figure (8) ^{1}H-NMR spectrum of compound 3C

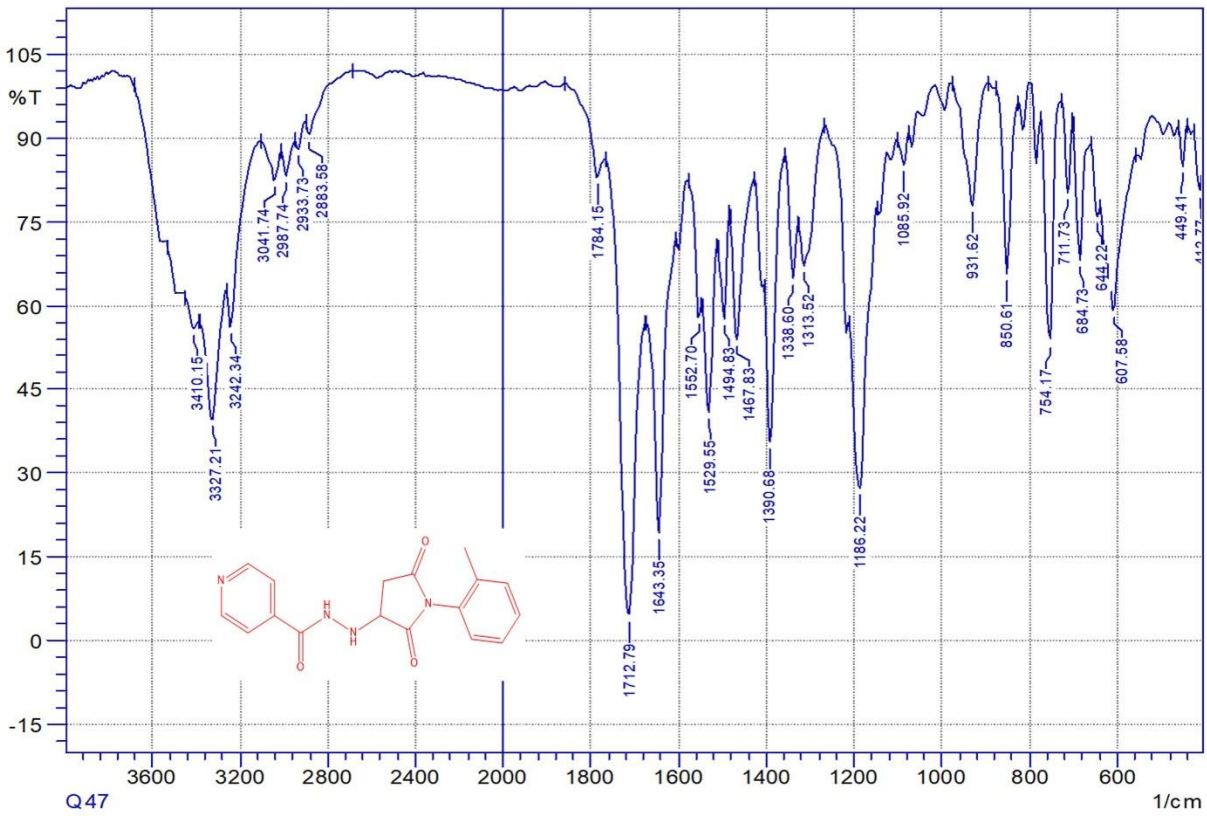

Figure (9) FT-IR spectrum of compound 4C

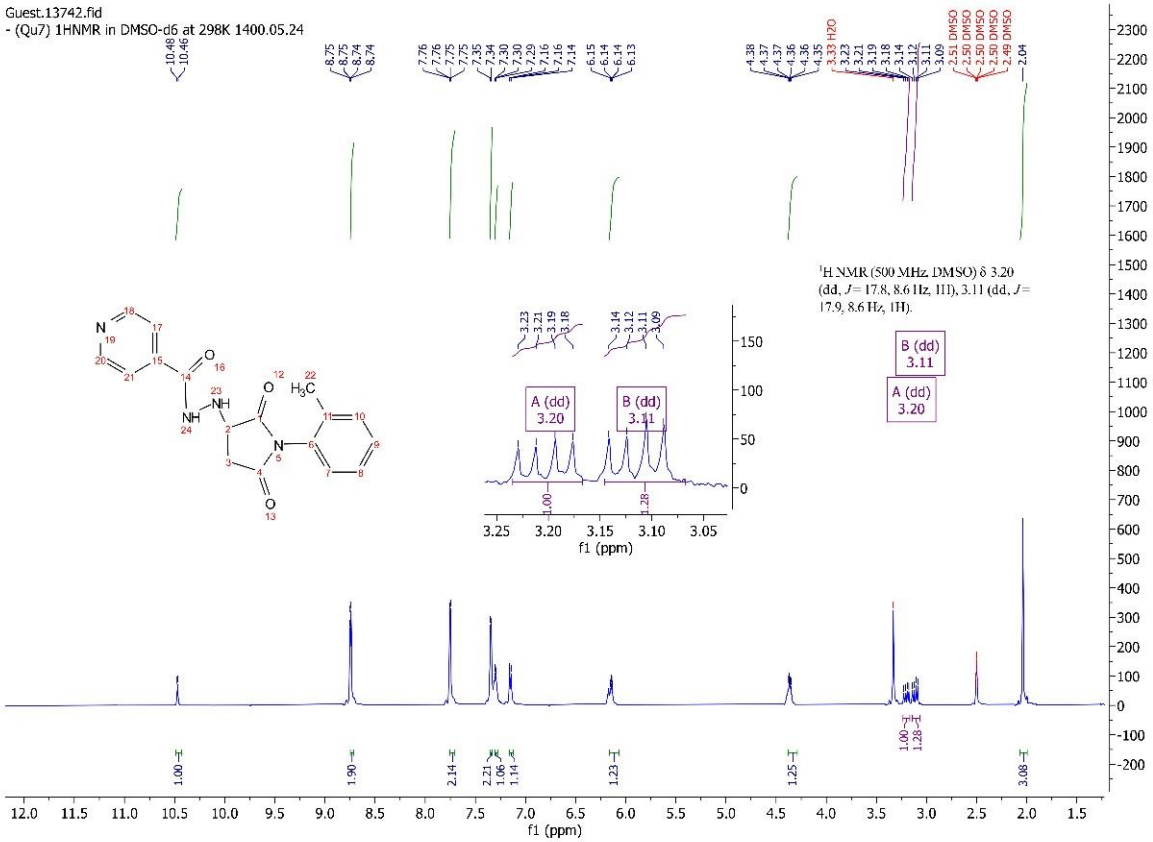

Figure (10) ^{1}H-NMR spectrum of compound 4C

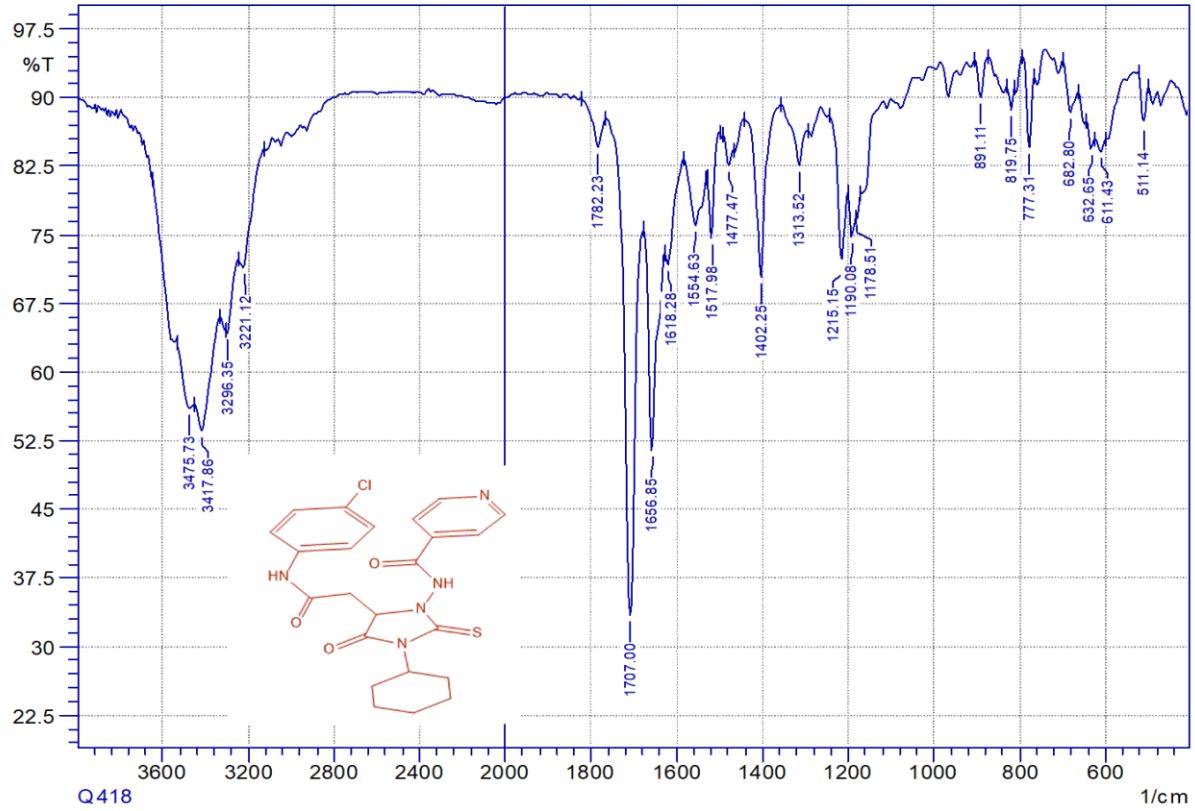

Figure (11) FT-IR spectrum of compound 1D

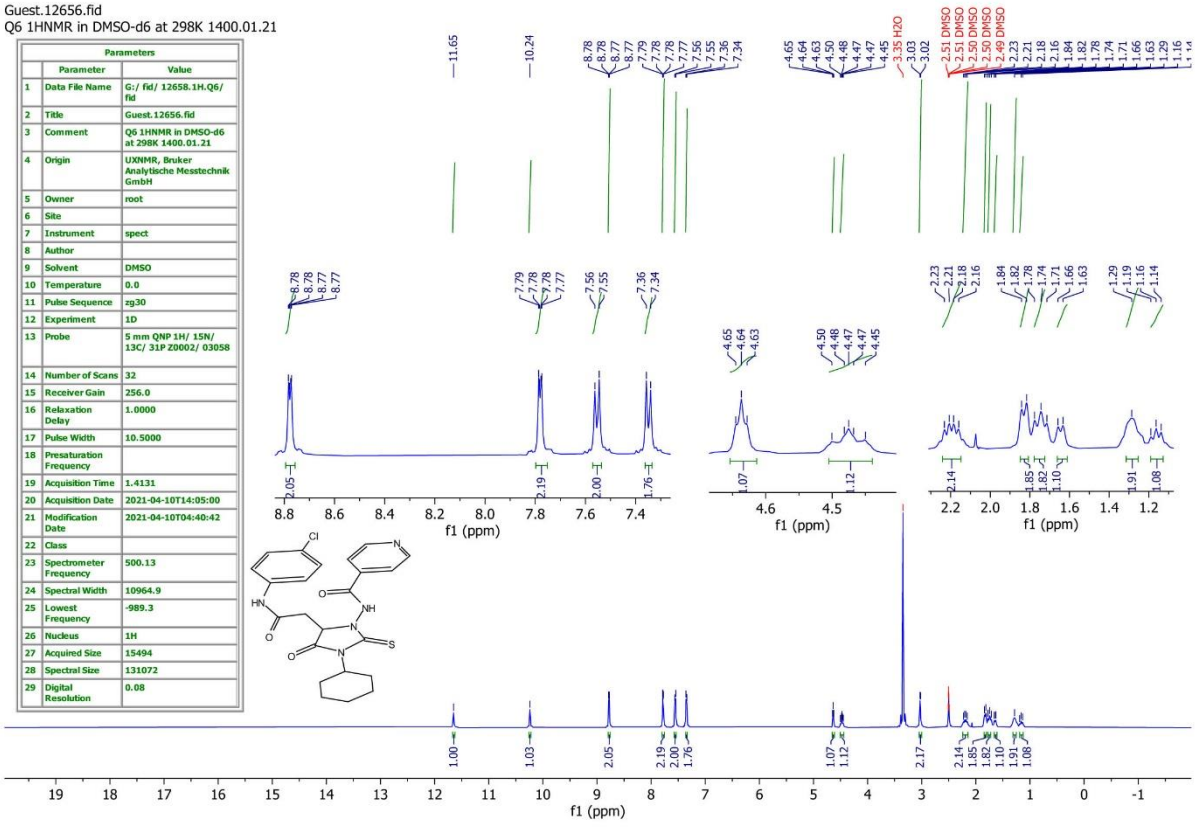

Figure (12) ^{1}H-NMR spectrum of compound 1D

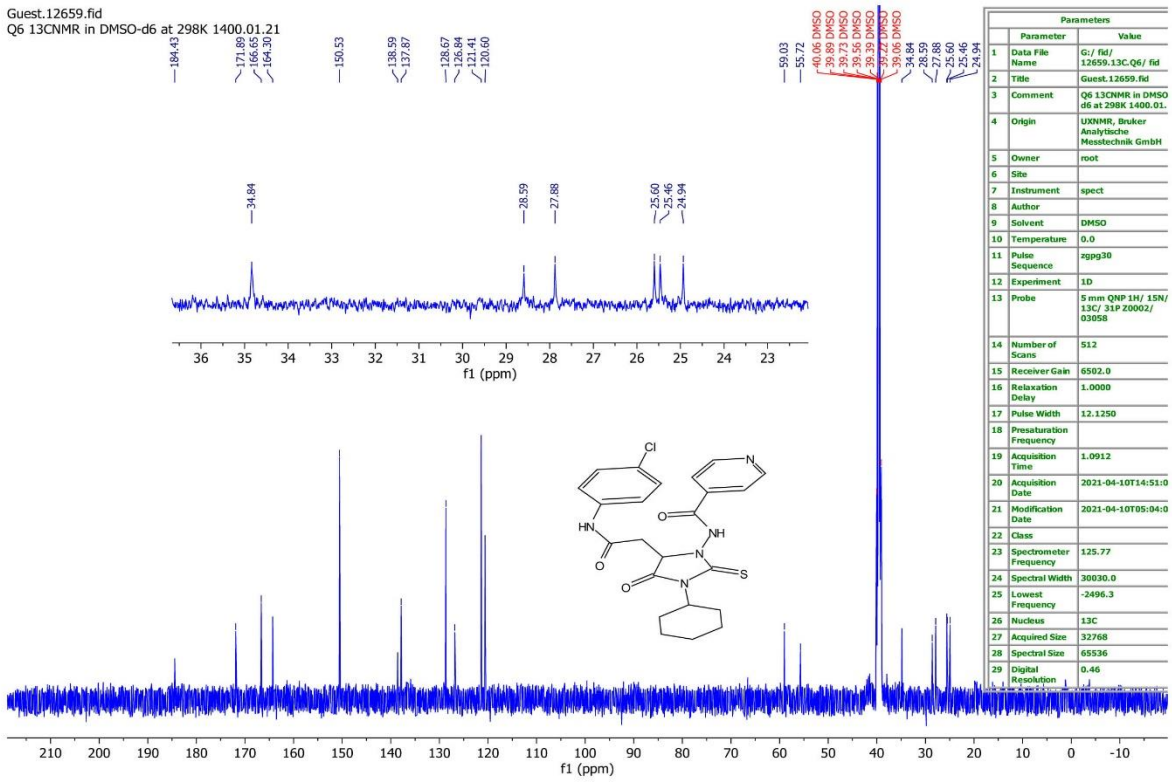

Figure (13) ^{13}C-NMR spectrum of compound 1D

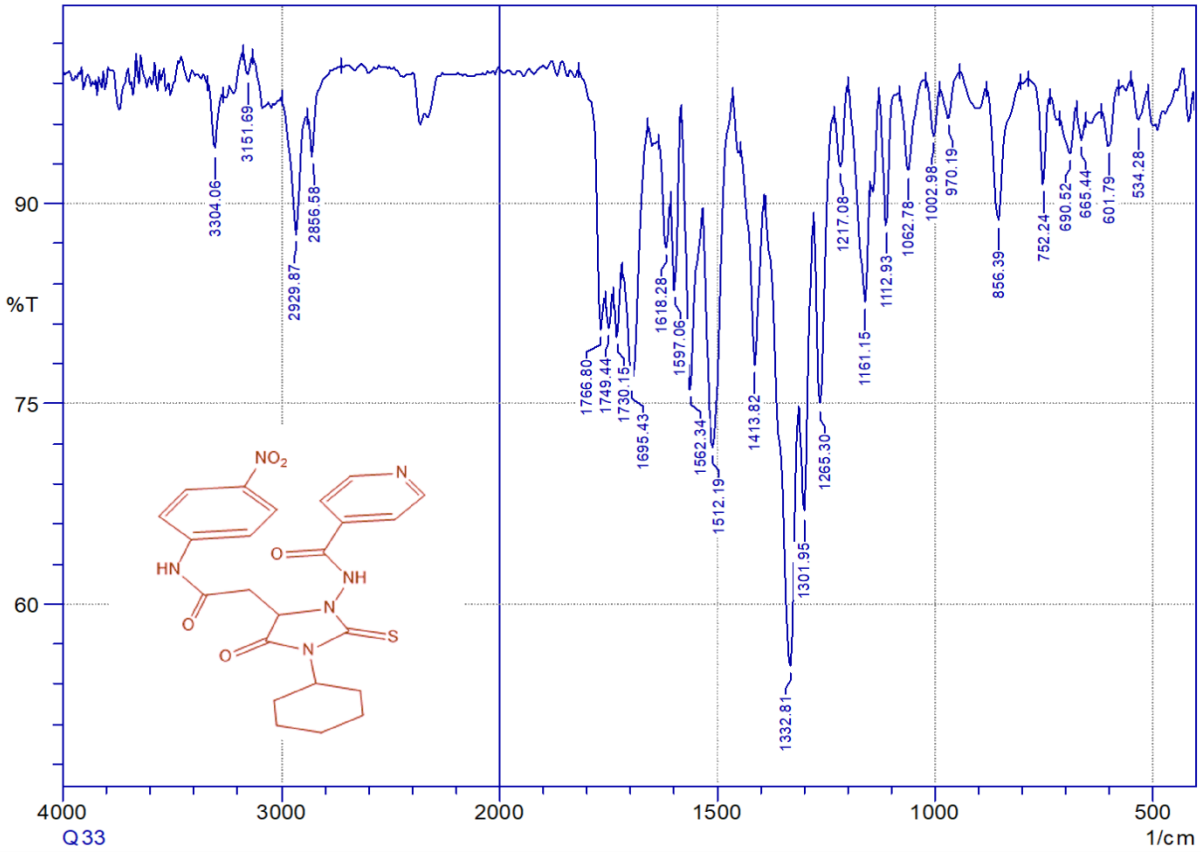

Figure (14) FT-IR spectrum of compound 2D

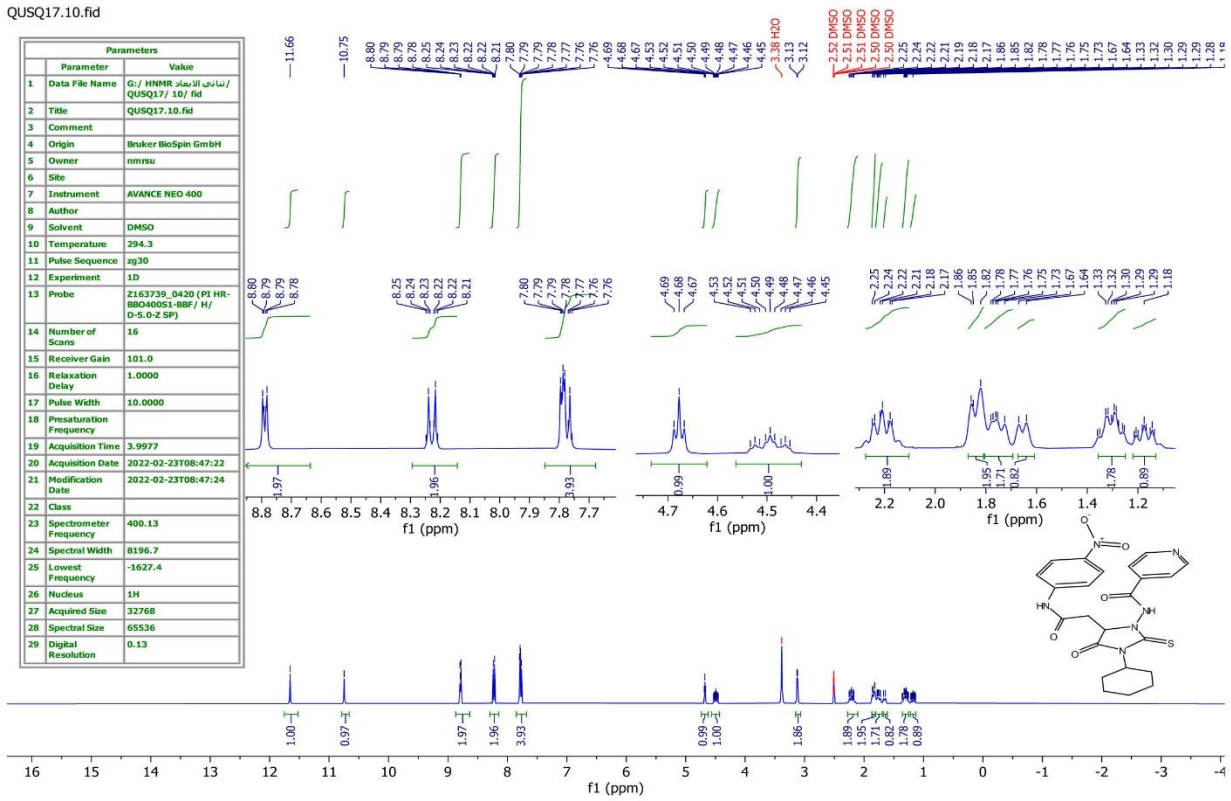

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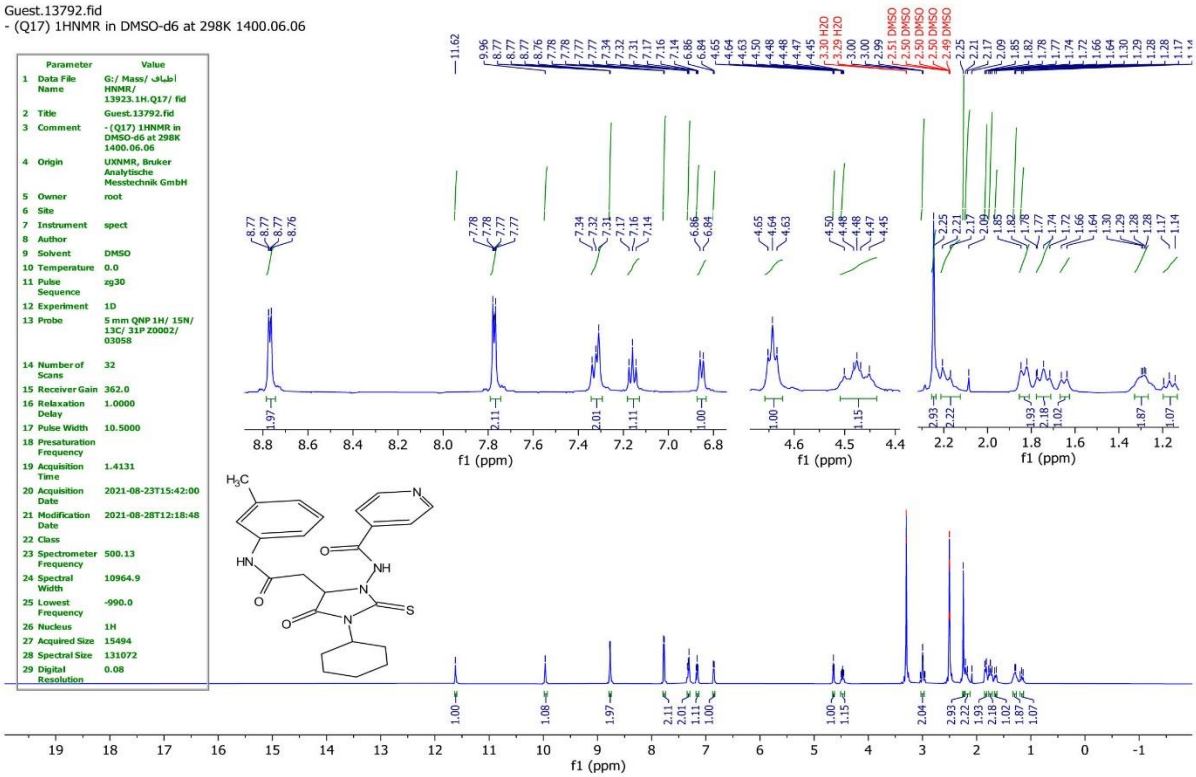
Figure (15) ^{1}H-NMR spectrum of compound 2D



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5	Owner	root
6	Site	
7	Instrument	spect
8	Author	
9	Solvent	DMSO
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Figure (16) ^{1}H-NMR spectrum of compound 3D

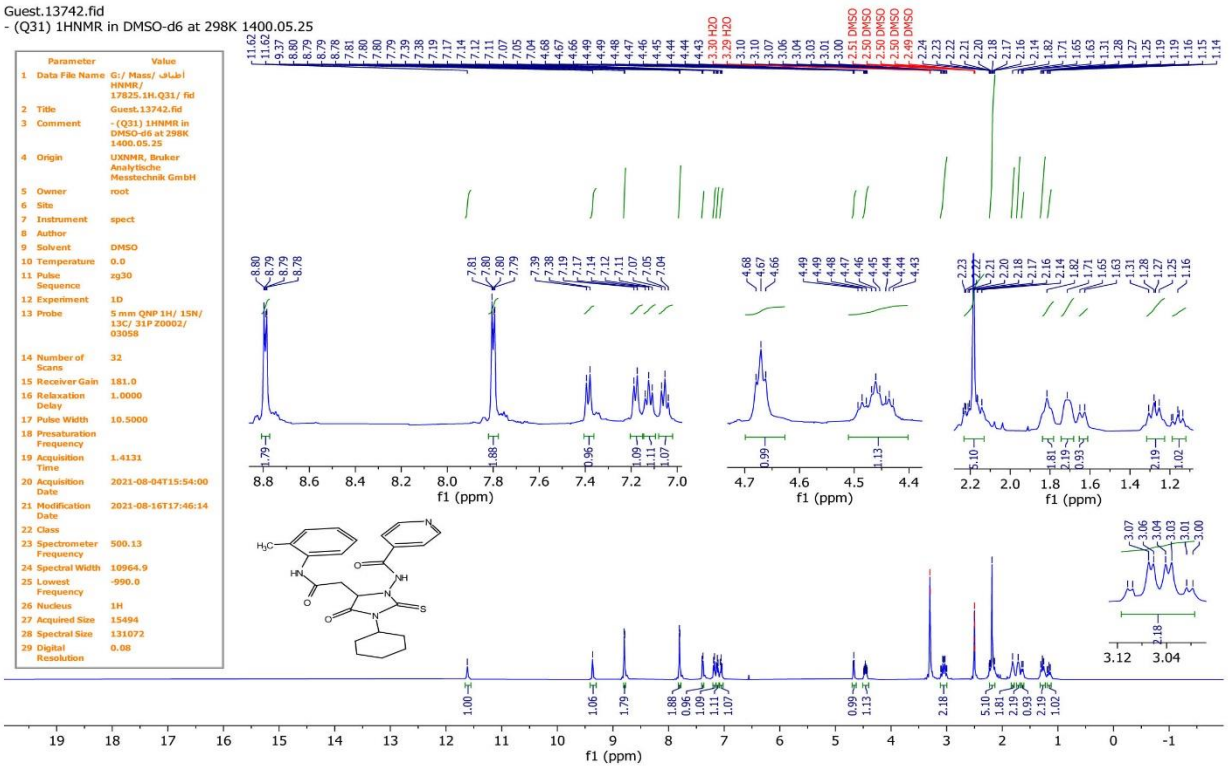


Figure (17) ^{1}H-NMR spectrum of compound 4D

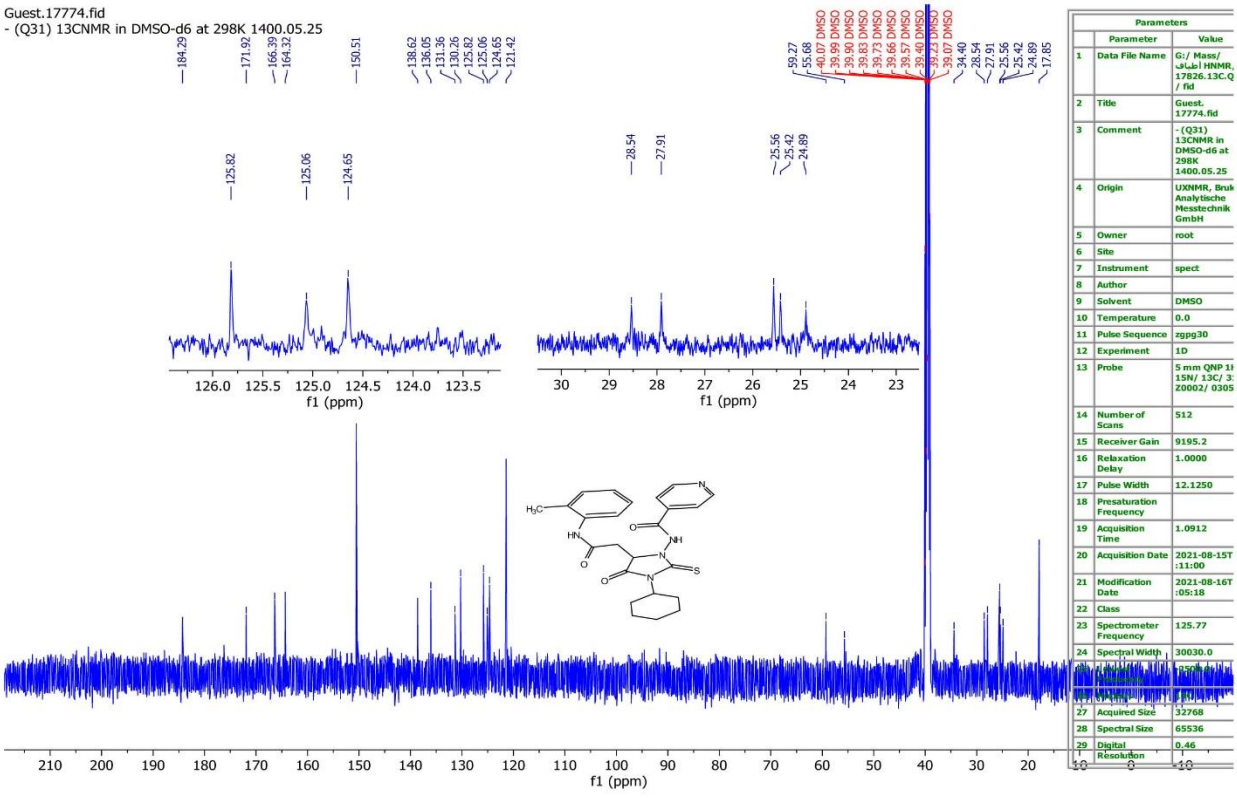
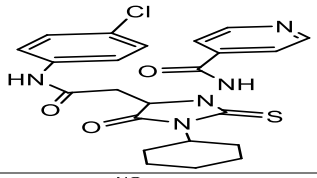
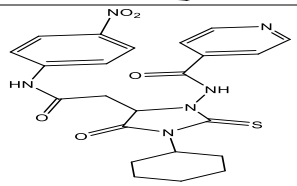


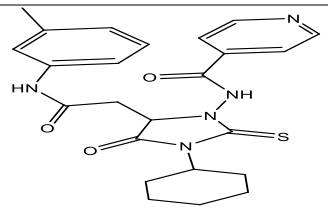
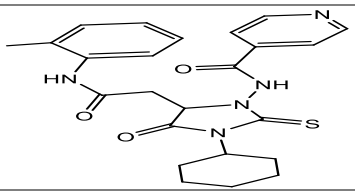
Figure (18) ^{13}C-NMR spectrum of compound 4D

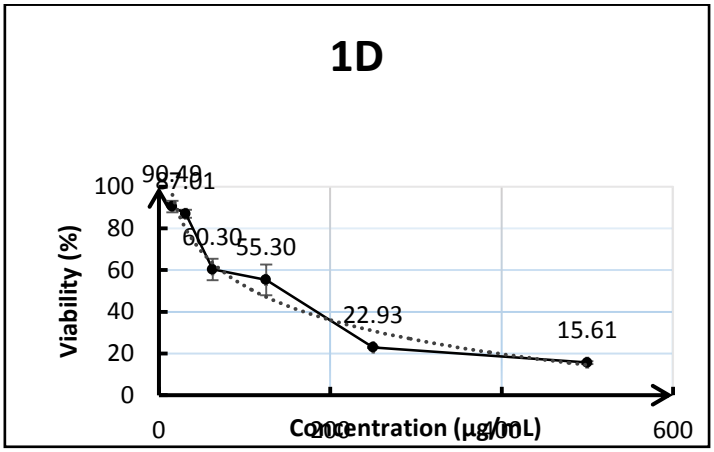
The activity of the prepared compounds against prostate cancer. [23]

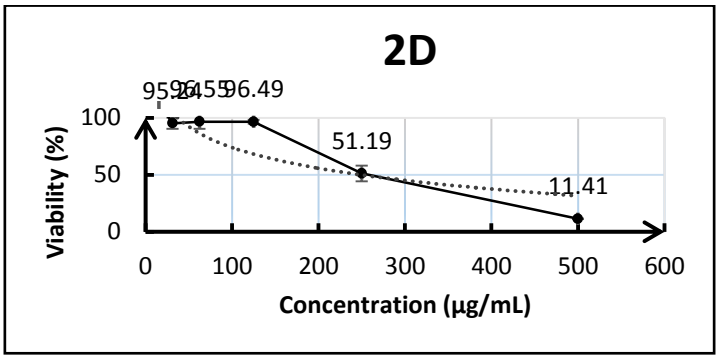
Compound symbol	Compound structure	PC_3 cells IC_{50} in µg/mL	
1D		110.41	Inactive
2D		243.053	Inactive

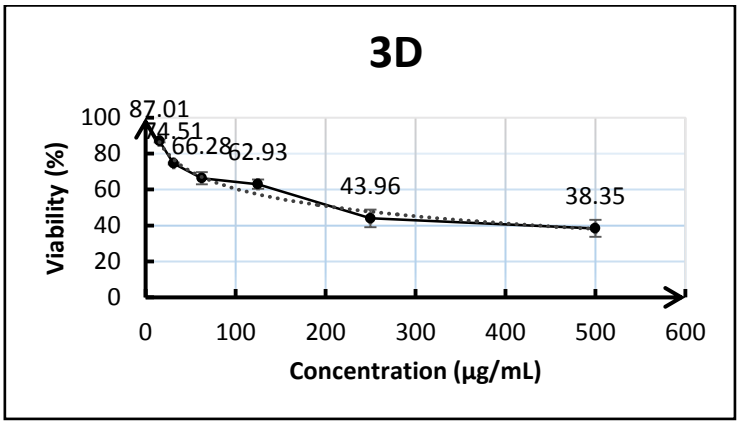
The discovery of anti-prostate cancer drugs is one of the most challenging challenges faced by modern medicine, and the reason is due to the large number of mutations that occur in the Ar (Androgen receptor). It has been found that the thiohydantoin nucleus is an excellent drug carrier for the treatment of prostate cancer. In this study, we examined a series of thiothydantoin derivatives prepared as anti-prostate cancer . Depending on the IC50 value of the compounds of the prepared series, we note that there is a noticeable difference in the activity of the compounds against prostate cancer cells. This discrepancy is attributed to the difference of functional groups on the aromatic ring in the prepared compounds. Depending on the IC50 value, none of the compounds under study showed anti-prostate cancer activity, Table 1.

Table (1) shows the IC50 values of the compounds under study versus PC3 cells.

3D		211.281	Inactive
4D		156.56	Inactive







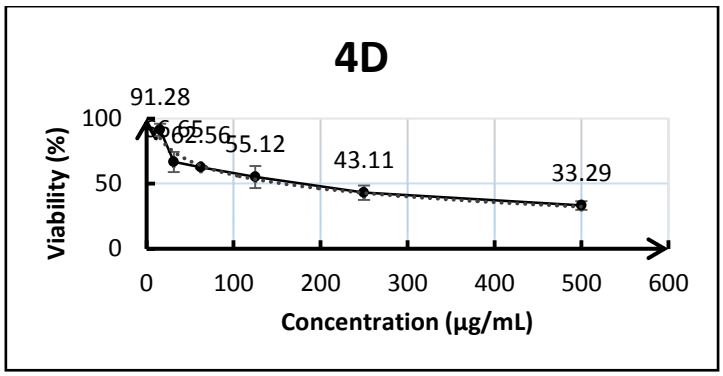


Figure 1 the graphs of the response of the compounds under study against prostate cancer cells

The activity of the prepared compounds against bacteria and fungi [24] [25]

Thiohydantoin (1D-4) and maleimide derivatives (1C-4) are found in a wide range of heterocyclic compounds, and the biological activity of this type of compound usually depends on the nature of the substituting groups on the ring. Studies confirm that the compounds that contain in their structure the nucleus of the thiohydantoin have anti-bacterial and anti-fungal properties [26] [27]. In this study, we evaluated the prepared series compounds against selected strains of bacteria and fungi. The prepared compounds were studied as antibacterials *E.Coli and Staph*, compared with Spiromycin as a positive control, and they were evaluated as against fungi on two types of fungi, *Candida albican and Aspargilus*.

The compounds (4C,1D,3D,4D) showed *anti-staph* activity, as the 4C compound showed high *anti-staph* activity. None of the studied compounds showed *anti-E.Coli* activity except for the compound 4C, which showed medium activity. The compounds (1C,4C,2D,3D) showed *anti- aspargilus* activity, while the rest of the compounds under study were not activity. The data indicate that all the compounds under study have antifungal activity for *Candida albican*, Table 2.

Table (2) shows the activity of the compounds under study against selected types of bacteria and fungi.

Compound symbol	The biological activity of the bacteria under study		The biological activity of the fungi under study	
	Staph	*E.Coli*	*Aspargilus*	*Candida albican*
1C	-	-	**12**	19
2C	-	-	-	16
3C	-	-	-	13
4C	**20**	**15**	**12**	20
1D	16	-	-	21
2D	-	-	11	25
3D	15	-	15	32
4D	15	-	-	37

Spiromycin	23	23	No tested	No tested	**Negative -:**

No activity

CONCLUSION

The advantage of preparing this series of compounds is easy access to the product and ease of purification. The graph of the response of the compounds under study against prostate cancer cells indicates that there is a significant effect of the aromatic ring functional groups in their activity against prostate cancer cell lines. Depending on the IC50 values of the compounds under study, none of the compounds showed anti-prostate cancer activity, and we need further study to increase the activity of the prepared series compounds against prostate cancer cells. The data of the compounds under study indicate that there is a significant effect of the functional groups in the aromatic ring in its anti-bacterial and anti-fungal activity. Compound 4C showed high activity against *Staph* bacteria due to the presence of the methyl group in the ortho position.

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