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SYNTHESIS, BIOLOGICAL EVALUATION OF SOME NEW THIOPHENE DERIVATIVES

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Abstract – The reaction of 2-pyridylacetophenone (**2**) with phenyl isothiocyanate gave thiocarbamoyl derivative **4** which on reaction with α-halocarbonyl compounds in a mixture of ethanol and *N*,*N*-dimethylformamide in the presence of triethylamine afforded thiophenes **6**, **8**, **10**, **12** and **14** derivatives. While, when the same reaction was carried out in ethanol without *N*,*N*-dimethylformamide, it afforded the corresponding acyclic compounds **5**, **7**, **9**, **11** and **13** which on reflux in *N*,*N*-dimethylformamide in the presence of triethylamine gave the corresponding thiophene derivatives. The newly synthesized compounds were characterized by analytical, spectral data and evaluated their antimicrobial activities. Compounds **6**, **8**, **10**, **12**, **13** and **14** were found to have high antimicrobial activities.

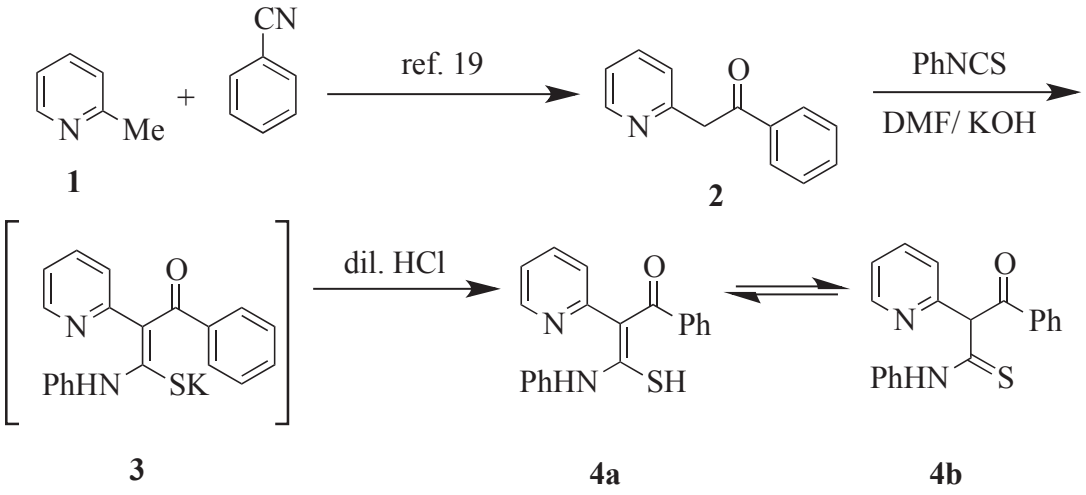
Aryl isothiocyanates are versatile reagents that have been used as synthetic intermediates to prepare biologically active heterocyclic compounds.[1] The diversity of biological and physiological activities of several organic sulfur heterocycles may be attributed to the presence of the N=C=S fragment, characteristics of thiazoles, thiazolines and thiazolidines.[2] These are known to exhibit pesticidal,[3] anticonvulsant,[4] herbicidal, antiviral, fungicidal, bactericidal, antiprotozoal[5-11] and hypoglycemic activity.[12] They also act as chemotherapeutic agents.

We have been particularly interested to study if reactions of such thiocarbamoyl might be extended to include a more general synthesis of other classes of organic compounds and its utility as synthetic intermediate for the synthesis of new heterocyclic compounds. The present work reports on the synthesis of several new thiophene derivatives by the reaction of thiocarbamoyl **4** with compounds containing an active methylene group in the presence of a base. Reaction of such thiocarbamoyl **4** has not been reported before, but we found to give products in excellent yields under very mild conditions.

Moreover, in continuation of the previously reported work (from our lab)[13-15] the resulting thiazole and

thiophene derivatives have latent functional substituents, which have potential for further chemical transformation and new routes for the preparation of substituted thiazole and thiophene derivatives. Now, we have extended our synthetic program to the synthesis of otherwise inaccessible heterocyclic ring system, utilizing phenyl isothiocyanate as a key starting material and examined their activities as antimicrobial agents.

As a part of reported research on drug discovery program,[16-18] we needed to describe facile and rapid procedure for construction of drug like small organic molecules using various reagents. Firstly, the acidic methylene compound **2** was formed by reaction of α-picoline (**1**) with benzonitrile in the presence of *n*-butyllithium at -40 °C according to the reported method.[19] Thus, the base catalyzed reaction of compound **2** with phenyl isothiocyanate in dry dimethylformamide (DMF) in the presence of sodium ethoxide or potassium hydroxide at room temperature led to the formation of the non-isolable intermediate **3** which gave thiocarbamoyl derivative **4** in two tautomeric structures upon treatment with dil. HCl (**Scheme 1**).[20-22]



1 **2** **3** **4a** **4b**

Scheme 1

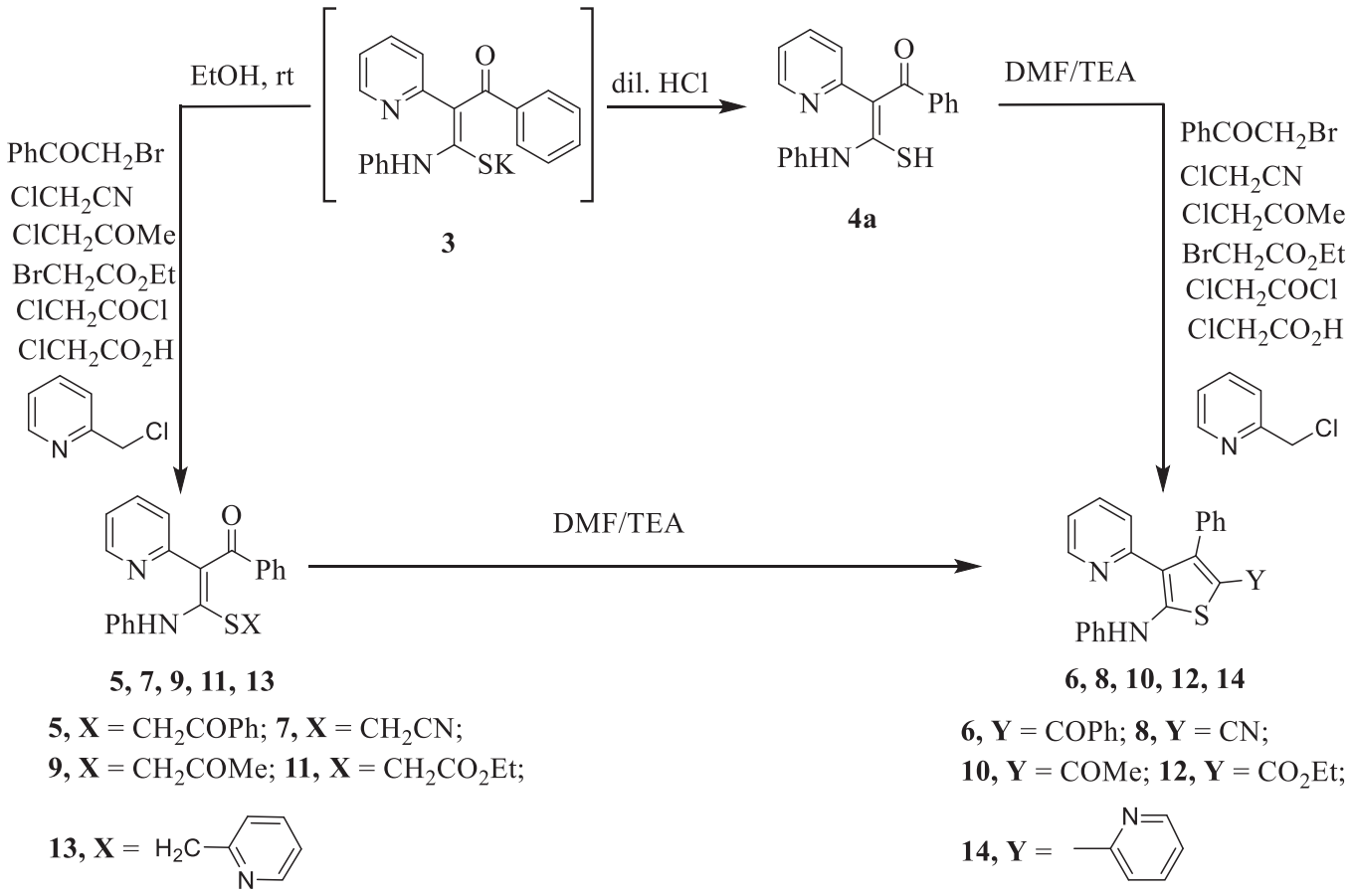
Assignment of compound **4** was based on elemental analysis, IR, ^{1}H NMR spectral data. The IR spectrum showed absorption bands at 3350 cm^{-1} (NH), 1710 cm^{-1} (CO) and 1290 cm^{-1} (C=S) functions, respectively. ^{1}H NMR spectrum of **4** displayed a singlet signal at δ 1.65 ppm, singlet signal for SH proton and singlet signal at δ 10.30 corresponding to NH proton. It also showed multiplet signals at δ 7.30-8.51 ppm for aromatic and pyridine protons. Its mass spectrum showed the molecular ion peak at *m/z* = 332 (M^+, 100%) corresponding to the molecular formula $C_{20}H_{16}N_2OS$. ^{1}H NMR spectrum of compound **4a** showed singlet signal at δ 1.65 ppm due to SH proton, which can be only attributed for the thiole form **4a**. Also, the same ^{1}H NMR spectrum revealed the absence of any singlet signal in the region at δ 4.50-6.00 ppm due to the CH proton in structure **4b** which confirm that the thiole tautomer is present in very poor

extent. The IR measurement showed band at 1290 cm^{-1} due to C=S, in the solid state, indicating its presence in thiole-thione tautomeric mixture as well.[23]

Compound **4** also undergoes cyclization[13-15,24-26] upon the reaction with equimolar amount of phenacyl bromide in boiling DMF in the presence of a catalytic amount of TEA to yield a product **6**, which was analyzed precisely for $C_{28}H_{20}N_2OS$.

The structure of compound **6** was inferred from its spectral data. Thus, the IR spectrum showed absorption bands at 3351, 1700 and 1599 cm^{-1} corresponding to NH, CO and C=C functions, respectively. Its ^1H NMR spectrum showed singlet signal at δ 9.69 ppm due to NH proton and multiplet signals integrated for 19H centered at 7.41-8.56 ppm (aromatic and pyridine protons). On shaking **6** with D_2O, the signal at δ 9.60 disappeared.

Moreover, compound **6** was confirmed by its mass spectrum which showed the molecular ion at *m/z* = 432 (M^+, 69%) corresponding to the molecular formula $C_{28}H_{20}N_2OS$. Structure **6** was further proved by alternative synthesis. Thus, it was found that, stirring of **3** with phenacyl bromide in ethanol at room temperature afforded the acyclic intermediate **5** by hydrogen bromide (HBr) elimination.



3 **4a**

5, 7, 9, 11, 13 **6, 8, 10, 12, 14**

5, X = CH_2COPh; **7, X** = CH_2CN;
9, X = CH_2COMe; **11, X** = CH_2CO_2Et;

13, X =

6, Y = COPh; **8, Y** = CN;
10, Y = COMe; **12, Y** = CO_2Et;

14, Y =

Scheme 2

Structure of **5** was suggested for the reaction product on the basis of both elemental and spectral data. The IR spectrum showed absorption bands at 3371, 1700 and 1695 cm^{-1} corresponding to NH, two CO functions, respectively. The configuration of compound **5** is *Z* isomer which is proved by intramolecular condensation to form the corresponding thiophene derivatives which cannot be formed from *E* isomer.

Refluxing of compound **5** in DMF with few drops of TEA led to the formation of a product identical on all respects (mp, mixed mp, and IR) to **6** (**Scheme 2**).

Similarly, when the intermediate potassium salt **3** was stirred with chloroacetonitrile in ethanol at room temperature, the corresponding acyclic intermediate **7** is exclusively isolated in good yield. The structure of **7** has been confirmed on the basis of elemental and spectral data. The IR spectrum exhibits bands at 3330 cm^{-1} (NH), 2220 cm^{-1} (CN), 1700 cm^{-1} (CO) and 1605 cm^{-1} (C=C). Its ^{1}H NMR spectrum reveals a singlet signal at δ 4.38 ppm due to CH_2 protons, multiplet signals at δ 7.31-8.44 ppm for aromatic and pyridine protons and singlet signal at δ 9.61 ppm corresponding to NH proton. The configuration of compound **7** is *Z* isomer which is proved by intramolecular condensation to form the corresponding thiophene derivatives which cannot be formed from *E* isomer.

Furthermore, refluxing of the acylic intermediate **7** in DMF containing a catalytic amount of TEA afforded the thiophene derivative **8**. Thiophene **8** was established based on its elemental and spectral analyses. On the other hand, it has been found that compound **8** is directly formed by refluxing **4** with chloroacetonitrile in DMF in the presence of catalytic amount of TEA (**Scheme 2**).

In order to extend the utility of thiocarbamoyl derivative **4** as a building block for preparation of sulfur containing heterocycles, the intermediate potassium salt **3** reacted readily with chloroacetone in the presence of ethanol at room temperature to afford the acyclic intermediate **9** by potassium chloride (KCl) elimination.

The acyclic intermediate **9** was established based on its IR spectrum which showed bands at 3410, 1700, 1695 and 1620 cm^{-1} corresponding to NH, two CO and C=C functions, respectively. Also, its ^{1}H NMR spectrum showed multiplet signals at δ 7.31-8.51 ppm for aromatic and pyridine protons. The configuration of compound 9 is *Z* isomer which is proved by intramolecular condensation to form the corresponding thiophene derivatives which cannot be formed from *E* isomer. Refluxing of compound **9** in DMF in the presence of a catalytic amount of TEA gave thiophene **10**. Structure **10** was confirmed by its alternative synthesis. Also, refluxing of compound **4** with chloroacetone in DMF in the presence of catalytic amount of TEA afforded the thiophene derivative **10** in good yield (**Scheme 2**).

The ^{1}H NMR spectrum of thiophene **10** reveals two characteristic singlet signals at δ 2.15 and 9.95 ppm due to CH_3 and NH protons.

Interestingly, when compound **4** was treated with an equimolar amount of ethyl chloroacetate, ethyl bromoacetate, chloroacetic acid or with chloroacetyl chloride in a mixture of DMF and ethanol in the

presence of a catalytic amount of TEA, thiophene **12** analyzed for $C_{24}H_{20}N_2O_2S$ was isolated in each case in good yield (**Scheme 2**). Moreover, the reaction of the intermediate **3** with an equimolar amount of ethyl chloroacetate, ethyl bromoacetate or with chloroacetic acid in ethanol led to the formation of compound **11**. The acyclic structure **11** was established based on its IR spectrum that showed absorption bands at 3350, 1730, 1700 and 1617 cm^{-1} attributable to the NH, two CO and C=C functions, respectively. Its ^1H NMR spectrum reveals a triplet signal at δ 1.20 (3H, CH_3), quartet at δ 4.18 (2H, CH_2) and a D_2O exchangeable NH at δ 9.91 ppm. The structure of **11** was confirmed by its mass spectrum which showed a peak at *m/z* = 418 (M^+, 41%). The configuration of compound **11** is *Z* isomer which is proved by intramolecular condensation to form the corresponding thiophene derivatives which cannot be formed from *E* isomer. Refluxing of **11** in DMF with a catalytic amount of TEA afforded the corresponding thiophene derivative **12**.

Finally, when compound **4** was treated with 2-chloromethylpyridine in a mixture of DMF and EtOH in the presence of a catalytic amount of TEA afforded thiophene **14** (**Scheme 2**). Alternatively, compound **14** was obtained by refluxing the acyclic intermediate **13** in DMF in the presence of a catalytic amount of TEA. The acyclic structure **13** was established based on its IR spectrum that showed absorption bands at 3355 and 1700 cm^{-1} corresponding to NH and CO groups. Its ^1H NMR spectrum reveals two singlet signals at δ 5.18 and 9.90 ppm due to CH_2 and NH protons. The configuration of compound **13** is *Z* isomer which is proved by intramolecular condensation to form the corresponding thiophene derivatives which cannot be formed from *E* isomer.

BIOLOGICAL EVALUATION

Twelve of new synthesized target compounds **2**, **4-14** were evaluated for their *in vitro* antibacterial activity against *Bacillus subtilis* and *Bacillus thuringiensis* as examples of Gram-positive bacteria and *Escherichia coli* and *Pseudomonas aeruginosa* as examples of Gram-negative bacteria. They were also evaluated for their in vitro antifungal potential against *Fusarium oxysporum* and *Botrytis fabae* fungal strains.

Agar-diffusion method was used for the determination of the antibacterial and antifungal activity. Chloramphenicol, Cephalothin and Cycloheximide were used as reference drugs. The results were recorded for each tested compound as the average diameter of inhibition zones (IZ) of bacterial or fungal growth around the disks in mm. The minimum inhibitory concentration (MIC) measurement was determined for compounds showed significant growth inhibition zones (> 14 mm) using two fold serial dilution method. The MIC (μg/mL) and inhibition zone diameter values are recorded in **Table 1**.

Table 1. Minimal inhibitory concentration (MIC[a], μg/mL) and inhibition zone (mm) of some new synthesized compounds

Compound No.	MIC in μg/mL , and inhibition zone (mm)					
	Bacteria				Fungi	
	Gram-positive bacteria		Gram-negative bacteria			
	B. subtilis	*B. thuringiensis*	*E. coli*	*P. aerugunosa*	*F. oxysporum*	*B. fab*ae
2	50 (18)	50 (19)	100 (15)	100 (16)	100 (15)	50 (20)
4	25 (26)	50 (20)	100 (16)	50 (20)	50 (20)	50 (20)
5	6.25 (38)	12.5 (30)	25 (25)	25 (26)	50 (18)	50 (21)
6	6.25 (37)	6.25 (37)	12.5 (30)	25 (25)	12.5 (30)	12.5 (28)
7	6.25 (38)	6.25 (37)	12.5 (28)	12.5 (27)	25 (25)	12.5 (26)
8	3.125 (41)	6.25 (38)	6.25 (36)	12.5 (29)	12.5 (29)	12.5 (28)
9	6.25 (37)	6.25 (39)	12.5 (29)	12.5 (30)	50 (21)	50 (21)
10	6.25 (37)	6.25 (37)	12.5 (31)	12.5 (31)	12.5 (28)	12.5 (27)
11	6.25 (38)	6.25 (38)	12.5 (30)	12.5 (30)	25 (26)	25 (25)
12	3.125 (43)	3.125 (42)	6.25 (37)	12.5 (29)	12.5 (29)	12.5 (28)
13	3.125(33)	6.25 (37)	6.25 (37)	6.25 (37)	12.5 (50)	6.25 (38)
14	3.125 (41)	3.125 (43)	6.25 (38)	6.25 (36)	6.25(30)	6.25 (37)
Reference drugs						
Chloramphenicol	3.125 (43)	3.125 (44)	6.25 (37)	6.25 (38)	NT[b]	NT
Cephalothin	6.25 (36)	6.25 (36)	6.25 (35)	6.25 (37)	NT	NT
Cycloheximide	NT	NT	NT	NT	3.125(43)	3.125(42)

[a]MIC: Minimal inhibitory concentration values with SEM = 0.02.
[b]NT: Not tested

Compounds **6**, **8**, **10**, **12**, **13** and **14**, in general, possess good antibacterial activity due to the presence of thiophene ring (except compound **13**).

It was observed that compounds **6** and **10** exhibited nearly the same activity against *B. subtilis*, *B. thuringiensis* and *E. coli.* So, when benzoyl group in thiophene ring in compound **6** was replaced by acetyl group (compound **10**) the activity remained unchanged (MIC= 6.25 μg/mL) against *B. subtilis* (MIC= 6.25 μg/mL) against *P. aerugunosa* and (MIC= 12.5 μg/mL) against *E. coli*, respectively.

On the other hand, compound **12** which contains more electron withdrawing group attached to thiophene ring showed a better activity against the same organisms, while the presence of two pyridine rings in

compounds **13** and **14** seems to be responsible for its highest activity against the tested organisms.

From **Table 1**, it was observed that compound **14** exhibited equipotent activity to Chloramphenicol in inhibiting the growth of both Gram-positive and -negative bacteria.

In conclusion, the objective of the present study was to synthesize and investigate the antimicrobial activities of some new thiocarbamoyl and thiophene derivatives with the hope of discovering new structure leds serving as antimicrobial agents.

Our aim has been verified by the synthesis of two different groups of structures comprising basically sulfur compounds.

The obtained results revealed that compounds of thiophene derivative, in general, exhibited antimicrobial activity than compounds containing thiocarbamoyl moiety (except compound **13**).

EXPERIMENTAL

All melting points are incorrect in degree centigrade and determined on Gallenkamp electric melting point apparatus. The infrared (IR) spectra were recorded (KBr disk) on a Mattson 5000 FTIR spectrometer at the Faculty of Science, Mansoura University, Egypt. The ^{1}H-NMR spectra were determined on a Bruker WPSY 200 MHz spectrometer with tetramethylsilane (TMS) as an internal standard and the chemical shifts are in δ ppm using dimethylsulfoxide (DMSO-d_6) as a solvent. The mass spectra were recorded at 70 eV with Varian MAT 311 at the Micro analytical Center, Faculty of Science, Cairo University. Elemental analyses (C, H and N) were carried out at the Faculty of Science, Cairo University. The results were found to be in a good agreement (± 0.03) with the calculated values.

Chemistry

Synthesis of 2-pyridylacetophenone (2)

It was prepared according to the reported method.[19] Yield 75%; mp 40-42 °C (Lit. mp 39-44 °C[19]).

Synthesis of 3-mercapto-1-phenyl-3-(phenylamino)-2-(pyridin-2-yl)prop-2-en-1-one (4)

A mixture of compound **2** (1.97 g, 0.01 mol) and phenyl isothiocyanate (1.35 mL, 0.01 mol) was stirred overnight in DMF in the presence of anhydrous potassium hydroxide (0.008 g, 0.015 mol). The reaction mixture was poured onto ice water then acidified with dilute HCl. The resulting white crystals were filtered and washed with cold water. The filtered product was crystallized from MeOH.

Yield 65%; mp 210 °C; IR (KBr): ν/cm^{-1}= 3350 (NH), 1710 (CO), 1290 (C=S); ^{1}H-NMR (DMSO-d_6) δ (ppm) = 1.65 (s, 1H, SH), 7.30-8.51 (m, 14H, Ar-H), 10.30 (s, 1H, NH); MS (EI, 70 eV): *m/z* (%) = 332 (M^+, 100); Anal. Calcd for $C_{20}H_{16}N_2OS$ (332.42): C, 72.26; H, 4.85; N, 8.43. Found: C, 72.15; H, 4.79; N, 8.40%.

General procedure for the synthesis of compounds 5, 7, 9, 11 and 13

A mixture of compound **3** (5.55 g, 0.015 mol) and α-haloketones namely (phenacyl bromide (1.99 g, 0.01 mol), chloroacetonitrile (0.75 g, 0.01 mol), chloroacetone (0.92 g, 0.01 mol), ethyl bromoacetate (1.65 g, 0.01 mol) or ethyl chloroacetate (1.21 g, 0.01 mol) and 2-chloromethylpyridine (1.27 g, 0.01 mol), respectively) was stirred in EtOH at room temperature for 6 h. The resulting white crystals were collected by filtration and washed with EtOH. The separated solid product was crystallized from MeOH to give compounds **5**, **7**, **9**, **11** and **13**, respectively.

3-((2-Oxo-2-phenylethyl)thio)-1-phenyl-3-(phenylamino)-2-(pyridin-2-yl)prop-2-en-1-one (5)

Yield 71%; mp 261 °C; IR (KBr): ν/cm^{-1}= 3371 (NH), 1700, 1695 (2CO); ^{1}H-NMR (DMSO-*d*$_6$) δ (ppm) = 4.89 (s, 2H, CH_2), 7.51-8.56 (m, 19H, Ar-H), 10.71 (s, 1H, NH); MS (EI, 70 eV): *m/z* (%) = 450 (M^+, 100); Anal. Calcd for $C_{28}H_{22}N_2O_2S$ (450.56): C, 74.64; H, 4.92; N, 6.22. Found: C, 74.58; H, 4.90; N, 6.01%.

((3-Oxo-3-phenyl-1-(phenylamino)-2-(pyridin-2-yl)prop-1-en-1-yl)thio)acetonitrile (7)

Yield 75%; mp 197 °C; IR (KBr): ν/cm^{-1}= 3330 (NH), 2220 (CN), 1700 (CO), 1605 (C=C); ^{1}H-NMR (DMSO-*d*$_6$) δ (ppm) = 4.38 (s, 2H, CH_2), 7.31-8.44 (m, 14H, Ar-H), 9.61 (s, 1H, NH); MS (EI, 70 eV): *m/z* (%) = 371 (M^+, 100); Anal. Calcd for $C_{22}H_{17}N_3OS$ (371.46) C, 71.14; H, 4.61; N, 11.31. Found: C, 71.00; H, 4.58; N, 11.27%.

3-((2-Oxopropyl)thio)-1-phenyl-3-(phenylamino)-2-(pyridin-2-yl)prop-2-en-1-one (9)

Yield 70%; mp 209 °C; IR (KBr): ν/cm^{-1}= 3410 (NH), 1700 (CO), 1695 (CO), 1620 (C=C); ^{1}H-NMR (DMSO-*d*$_6$) δ (ppm) = 2.30 (s, 3H, CH_3), 4.31 (s, 2H, CH_2), 7.31-8.51 (m, 14H, Ar-H), 9.21 (s, 1H, NH); MS (EI, 70 eV): *m/z* (%) = 388 (M^+, 47); Anal. Calcd for $C_{23}H_{20}N_2O_2S$ (388.49) C, 71.11; H, 5.19; N, 7.21. Found: C, 71.00; H, 5.18; N, 7.20%.

Ethyl 2-((3-oxo-3-phenyl-1-(phenylamino)-2-(pyridin-2-yl)prop-1-en-1-yl)thio)acetate (11)

Yield 67%; mp 185 °C; IR (KBr): ν/cm^{-1}= 3350 (NH), 1730, 1700 (2CO), 1617 (C=C); ^{1}H-NMR (DMSO-*d*$_6$) δ (ppm) = 1.20 (t, 3H, CH_3), 3.81 (s, 2H, CH_2), 4.18 (q, 2H, CH_2), 7.34-8.53 (m, 14H, Ar-H), 9.91 (s, 1H, NH); MS (EI, 70 eV): *m/z* (%) = 418 (M^+, 41); Anal. Calcd for $C_{24}H_{22}N_2O_3S$ (418.51) C, 68.88; H, 5.31; N, 6.69. Found: C, 68.81; H, 5.29; N, 6.63%.

1-Phenyl-3-(phenylamino)-2-(pyridin-2-yl)-3-((pyridin-2-ylmethyl)thio)prop-2-en-1-one (13)

Yield 69%; mp 215 °C; IR (KBr): ν/cm^{-1}= 3355 (NH), 1700 (CO); ^{1}H-NMR (DMSO-*d*$_6$) δ (ppm) = 5.18 (s, 2H, CH_2), 7.34-8.56 (m, 18H, Ar-H), 9.90 (s, 1H, NH); MS (EI, 70 eV): *m/z* (%) = 423 (M^+, 32); Anal. Calcd for $C_{26}H_{21}N_3OS$ (423.53) C, 73.73; H, 5.00; N, 9.92. Found: C, 73.69; H, 5.00; N, 9.89%.

General procedure for the synthesis of thiophene derivatives 6, 8, 10, 12 and 14

Method A

A mixture of compound **4** (4.98 g, 0.015 mol) and α-haloketone (0.01 mol) was heated under reflux for 4

h in DMF containing a catalytic amount of triethylamine. The reaction mixture was poured onto cold water then acidified by dil. HCl. The solid material was collected by filtration then dried and recrystallized from EtOH to give the targeted compounds.

Method B

A solution of compound **5**, **7**, **9**, **11** and **13** in DMF was refluxed for about 3 h in the presence of few drops of triethylamine. The reaction mixture was poured onto cold water then acidified by dil. HCl. The solid material was collected by filtration then dried and recrystallized from EtOH to give the targeted compounds.

Phenyl (3-phenyl-5-(phenylamino)-4-(pyridin-2-yl)thiophen-2-yl)methanone (6)

Yield 71%; mp 281 °C; IR (KBr): ν/cm^{-1}= 3351 (NH), 1700 (CO), 1599 (C=C); ^{1}H-NMR (DMSO-d_6) δ (ppm) = 7.41-8.56 (m, 19H, Ar-H), 9.69 (s, 1H, NH); MS (EI, 70 eV): *m/z* (%) = 432 (M$^+$, 69); Anal. Calcd for $C_{28}H_{20}N_2OS$ (432.54) C, 77.75; H, 4.66; N, 6.48. Found: C, 77.71; H, 4.63; N, 6.40%.

3-Phenyl-5-(phenylamino)-4-(pyridin-2-yl)thiophene-2-carbonitrile (8)

Yield 71%; mp 270 °C; IR (KBr): ν/cm^{-1}= 3330 (NH), 2220 (CN); ^{1}H-NMR (DMSO-d_6) δ (ppm) = 7.31-8.56 (m, 14H, Ar-H), 10.10 (s, 1H, NH); MS (EI, 70 eV): *m/z* (%) = 353 (M$^+$, 35); Anal. Calcd for $C_{22}H_{15}N_3S$ (353.44) C, 74.76; H, 4.28; N, 11.29. Found: C, 74.71; H, 4.22; N, 11.23%.

1-(3-Phenyl-5-(phenylamino)-4-(pyridin-2-yl)thiophen-2-yl)ethan-1-one (10)

Yield 73%; mp 189 °C; IR (KBr): ν/cm^{-1}= 3330 (NH), 1700 (CO); ^{1}H-NMR (DMSO-d_6) δ (ppm) = 2.15 (s, 3H, CH_3), 7.34-8.57 (m, 14H, Ar-H), 9.95 (s, 1H, NH); MS (EI, 70 eV): *m/z* (%) = 370 (M$^+$, 90); Anal. Calcd for $C_{23}H_{18}N_2OS$ (370.47) C, 74.57; H, 4.90; N, 7.56. Found: C, 74.56; H, 4.87; N, 7.53%.

Ethyl 3-phenyl-5-(phenylamino)-4-(pyridin-2-yl)thiophene-2-carboxylate (12)

Yield 69%; mp 212 °C; IR (KBr): ν/cm^{-1}= 3350 (NH), 1720 (CO); ^{1}H-NMR (DMSO-d_6) δ (ppm) = 1.33 (t, 3H, CH_3), 4.32 (q, 2H, CH_2), 7.31-8.55 (m, 14H, Ar-H), 10.10 (s, 1H, NH); MS (EI, 70 eV): *m/z* (%) = 400 (M$^+$, 80); Anal. Calcd for $C_{24}H_{20}N_2O_2S$ (400.50) C, 71.98; H, 5.03; N, 6.99. Found: C, 71.96; H, 5.00; N, 6.97%.

***N*,4-Diphenyl-3,5-di(pyridin-2-yl)thiophen-2-amine (14)**

Yield 73%; mp 215 °C; IR (KBr): ν/cm^{-1}= 3350 (NH); ^{1}H-NMR (DMSO-d_6) δ (ppm) = 7.33-8.55 (m, 18H, Ar-H), 10.10 (s, 1H, NH); MS (EI, 70 eV): *m/z* (%) = 405 (M$^+$, 100); Anal. Calcd for $C_{26}H_{19}N_3S$ (405.50) C, 77.01; H, 4.72; N, 10.36. Found: C, 76.98; H, 4.70; N, 10.34%.

Antimicrobial studies

It was carried out according to the reported method.[27]

Minimal Inhibitory Concentration (MIC) measurement

It was carried out according to the reported method.[27]

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REFERENCES

1. A. K. Mukerjee and R. Ashare, *Chem. Rev.*, 1991, **91**, 1.
2. H. A. Ead, S. O. Abdelah, and N. A. Kassab, *Arch. Pharm.*, 1997, **320**, 1227.
3. N. C. Misra and K. K. Patnaik, *Indian J. Appl. Chem.*, 1971, **34**, 148.
4. R. B. Rao and S. R. Singh, *J. Indian Chem. Soc.*, 1973, **50**, 492.
5. S. S. Parmer and T. K. Gupta, *J. Med. Chem.*, 1972, **15**, 99.
6. A. F. Pavlenko and S. D. Moshchitiskii, *Chem. Heterocycl. Compd.*, 1967, **3**, 195 (*Chem. Abstr.*, 1967, **68**, 114479).
7. M. Tisler and A. Andolsek, *J. Med. Chem.*, 1971, **14**, 53.
8. S. R. Singh, *J. Indian Chem. Soc.*, 1975, **52**, 734.
9. H. S. Chaudhary and H. K. Pujari, *Indian J. Chem.*, 1972, **10**, 764.
10. P. N. Dhal, T. E. Ashary, and A. Nayak, *Indian J. Chem.*, 1975, **13**, 46.
11. S. K. Mallick and A. R. Martin, *J. Med. Chem.*, 1971, **14**, 528.
12. W. H. Burton and W. L. Buddle, *J. Med. Chem.*, 1970, **13**, 1009.
13. A. A. Fadda, H. M. Refat, and M. E. A. Zaki, *Molecules*, 1999, **5**, 701.
14. A. A. Fadda, M. A. Metwaly, and S. B. Bondock, *Sulfur Lett.*, 2002, **25**, 199.
15. A. A. Fadda, E. Abdel-Latif, and R. El-Mekawy, *Phosphorus, Sulfur Silicon Relat. Elem.*, 2008, **183**, 1940.
16. A. El-Shafei, A. A. Fadda, A. M. Khalil, and T. A. E. Amen, *Bioorg. Med. Chem.*, 2009, **17**, 5096.
17. A. A. Fadda and S. B. Bondock, *Eur. J. Med. Chem.*, 2011, **46**, 2555.
18. A. A. Fadda, E. Abdel-Latif, and R. El-Mekawy, *Eur. J. Med. Chem.*, 2009, **44**, 1250.
19. M. Graser and H. Kopacka, *Inorg. Chim. Acta*, 2013, **401**, 38.
20. H. Minegishi, Y. Futamura, S. Fukashiro, M. Muroi, M. Kawatani, H. Osada, and H. Nakamura, *J. Med. Chem.*, 2015, **58**, 4230.
21. X.-M. Zeng, C.-Y. Meng, J.-X. Bao, D.-C. Xu, J.-W. Xie, and W.-D. Zhu, *J. Org. Chem.*, 2015, **80**, 11521.
22. A. M. Malla, M. Parveen, F. Ahmad, S. Azaz, and M. Alam, *RSC Adv.*, 2015, **5**, 19552.
23. P. J. Daries and J. M. Muchowski, *J. Heterocycl. Chem.*, 1975, **12**, 761.
24. K. A. Ali, H. S. Abdalghfar, K. Mahmoud, and E. A. Ragab, *J. Heterocycl. Chem.*, 2013, **50**, 1157.

25. K. A. Ali, H. M. Hosni, D. H. Elngar, and A. El-Galil E. Amr, *Phosphorus, Sulfur Silicon Relat. Elem.*, 2014, **189**, 1831.
26. A. Fadda, H. Refat, and Sh. Kamal, *Eur. J. Chem.*, 2014, **5**, 296.
27. A. A. Fadda, E. S. M. Afsah, and R. S. Awad, *Eur. J. Med. Chem.*, 2013, **60**, 421.