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Original research paper

Novel thiophene derivatives with sulfonamide, isoxazole, benzothiazole, quinoline and anthracene moieties as potential anticancer agents

MOSTAFA M. GHORAB[1,2]*
MAHMOUD S. BASHANDY[3,4]
MANSOUR S. ALSAID[1]

[1]*Department of Pharmacognosy College of Pharmacy, King Saud University, P.O. Box 2457, Riyadh 11451, Kingdom of Saudi Arabia*

[2]*Department of Drug Radiation Research, National Center for Radiation Research and Technology Atomic Energy, Authority, P.O.Box 29 Nasr City, Cairo, Egypt*

[3]*Department of Chemistry, Faculty of Science (Boy's), Al-Azhar University Nasr City, Cairo, Egypt*

[4]*Department of Chemistry, University College at Al-Jomoom, Umm Al-Qura University, 2026, Makkah Saudi Arabia*

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A novel series of thiophenes having biologically active sulfonamide **2–11**, 3-methylisoxazole **12**, 4-methoxybenzo[*d*]thiazole **13**, quinoline **14, 15**, benzoylphenylamino **16**, and anthracene-9,10-dione **17** moieties were prepared. Structures of the newly synthesized compounds were established by elemental analysis and spectral data. All newly synthesized compounds were evaluated for their *in vitro* anticancer activity against human breast cancer cell line (MCF7). Most of the screened compounds showed cytotoxic activities compared to doxorubicin as a positive control. Compounds **6**, **7**, **9** and **13** (IC_{50} values 10.25, 9.70, 9.55 and 9.39 μmol L^{-1}) revealed higher cytotoxic activities than that of doxorubicin (IC_{50} = 32.00 μmol L^{-1}). Also, compounds **5**, **8** and **10** were found nearly as active as doxorubicin (IC_{50} 28.85, 23.48 and 27.51 μmol L^{-1}).

Keywords: thiophenes, sulfonamides, isoxazole, benzothiazole, quinoline, anthracene, anticancer activity

Thiophenes have been reported to possess interesting biological and pharmacological activities where several derivatives are used as antibacterial (1–3), anti-inflammatory (4), anticancer (5, 6), and antiviral agents (7). Also, sulfonamides constitute an important class of drugs, with several types of pharmacological agents possessing anticancer activity (8–12), among others. A large number of structurally novel sulfonamides have ultimately been reported to show substantial anticancer activity *in vitro* and *in vivo* (13). Several mechanisms have been reported for anticancer activity of sulfonamide compounds and the most prominent of these mechanisms was thought to be the inhibition of carbonic anhydrase (14–16). The mechanism of tumor inhibition by the sulfonamide carbonic anhydrase

* Correspondence; e-mail: mmsghorab@yahoo.com

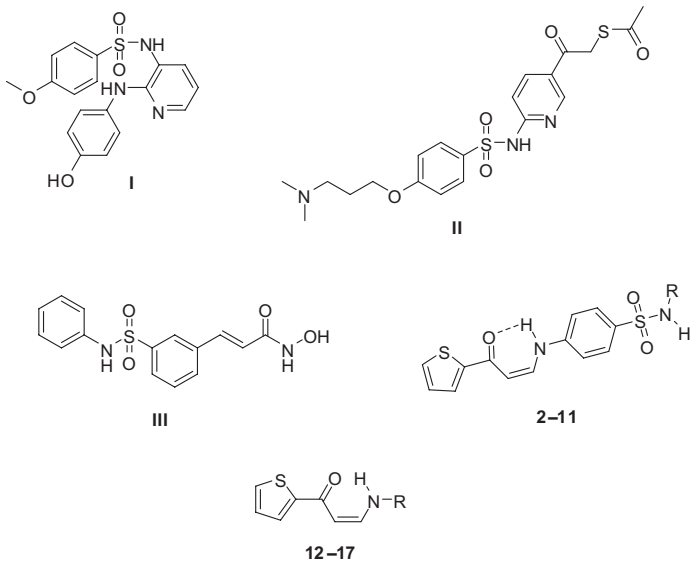


Fig. 1. Biologically active compounds **I–III** and structures of target compounds **2–17**.

inhibitor was suggested by Chegwidden and Spencer (17). These compounds may reduce the provision of bicarbonate for the synthesis of nucleotides and other cell components such as membrane lipids. Also, isoxazole, benzothiazole, quinoline and anthracene derivatives posseses a wide range of biological activities, including anti-inflammatory (18), antibacterial (19) and anticancer activity (20–22). In addition, several novel quinoline derivatives have been reported to show substantial anticancer activities (23–26). Also, a host of structurally novel sulfonamides have recently been reported to show substantial anticancer activity *in vitro* and/or *in vivo*. Since the discovery of E-7010 (**I** , 27), sulfonamides have emerged as an important class of anticancer agents, which interact with a wide range of different cellular targets such as disruption of microtubules assembly and carbonic anhydrase inhibition (17). Some of these compounds having the NH of the sulfonamide group substituted by an aryl group showed potent anticancer activity such as KD5170 (**II)** and PXD101 (**III**) (28). In continuation of our work (29–33), it seemed of interest to design and synthesize a novel series of thiophenes bearing biologically active sulfonamide, isoxazole, benzoxazole, quinoline and anthracene moieties and to evaluate their anticancer activity.

EXPERIMENTAL

Melting points (uncorrected) were determined in open capillaries on a Gallenkemp melting point apparatus (Sanyo Gallenkemp, UK). Pre-coated silica gel plates (silica gel 0.25 mm, 60 G F 254, Merck, Germany) were used for thin layer chromatography. Dichloromethane/methanol (9.5:0.5, *V*/*V*) mixture was used as a developing solvent system. IR spectra were recorded in KBr discs using an IR-470 Shimadzu spectrometer (Shimadzu,

Japan). NMR spectra in DMSO-d_6 were recorded on Bruker Ac-500 ultra shield NMR spectrometer (δ in ppm, Bruker, Switzerland) at 500 MHz, using TMS as an internal standard. Elemental analyses were performed on a Carlo Erba 1108 Elemental Analyzer (Heraeus, Germany). Elemental analyses of all compounds were within ± 0.4 % of the theoretical values. Starting compound, 1-(thiophen-2-yl)ethanone was purchased from Sigma (USA).

Syntheses

3-(Dimethylamino)-1-(thiophen-2-yl)prop-2-en-1-one (**1**) was prepared according to the reported method (34).

*(Z)-4-(3-Oxo-3-(thiophen-2-yl)prop-1-enylamino)benzenesulfonamide (***2***), (Z)*-N-*carbamimidoyl-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)benzenesulfonamide (***3***), (Z)*-N-*(3-methylisoxazol-5-yl)-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)benzenesulfonamide (***4***), (Z)*-N-*(3,4-dimethyl-isoxazol-5-yl)-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)benzene-sulfonamide (***5***), (Z)-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)*-N-*(thiazol-2-yl)benzenesulfonamide (***6***), (Z)-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)*-N-*(1-phenyl-1*H*-pyrazol-5-yl)benzenesulfonamide (***7***), (Z)-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)*-N-*(pyridin-2-yl)benzenesulfonamide (***8***), (Z)-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)*-N-*(pyrimidin-2-yl)benzenesulfonamide (***9***), (Z)*-N-*(4-methylpyrimidin-2-yl)-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)benzenesulfonamide (***10***) and (Z)*-N-*(4,6-dimethylpyrimidin-2-yl)-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)benzenesulfonamide (***11***). General procedure*. – A mixture of (**1**) (1.81 g, 0.01 mol) and sulfonamide derivatives (0.01 mol) in ethanol (20 mL) containing acetic acid (10 mL) was refluxed for 6 h. The obtained solid was recrystallized from dioxane while hot to give **2–11**.

*(Z)-3-(3-Methylisoxazol-5-ylamino)-1-(thiophen-2-yl)prop-2-en-1-one (***12***).* – A mixture of **1** (1.81 g, 0.01 mol) and 3-methyl-5-aminoisoxazole (0.98 g, 0.01 mol) in ethanol (20 mL), containing acetic acid (10 mL) was refluxed for 6 h. The obtained solid was recrystallized from dioxane to give **12**.

*(Z)-3-(4-Methoxybenzo[*d*]thiazol-2-ylamino)-1-(thiophen-2-yl)prop-2-en-1-one (***13***).* – A mixture of (**1**) (1.81 g; 0.01 mol) and 2-amino-4-methoxybenzothiazole (1.80 g, 0.01 mol) in ethanol (20 mL) containing (10 mL) acetic acid was refluxed for 8 h. The obtained solid was recrystallized from ethanol to give **13**.

*(Z)-3-(Quinolin-3-ylamino)-1-(thiophen-2-yl)prop-2-en-1-one (***14***)* and *(Z)-3-(2-methylquinolin-4-ylamino)-1-(thiophen-2-yl)prop-2-en-1-one (***15***).* – A mixture of **1** (1.81 g, 0.01 mol) and 3-aminoquinoline or 4-amino-2-methylquinoline (0.01 mol) in absolute ethanol (20 mL) glacial acetic acid (10 mL) was refluxed for 18 h. The obtained solid while hot was recrystallized from dioxane to give **14** and **15**, respectively.

*(Z)-3-(4-Benzoylphenylamino)-1-(thiophen-2-yl)prop-2-en-1-one (***16***)* and *(Z)-2-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)anthracene-9,10-dione (***17***).* – A mixture of **1** (1.81 g, 0.01 mol) and 4-aminophenyl-phenyl-methanone or 2-aminoanthracene-9,10-dione (0.01 mol) in ethanol (20 mL), containing acetic acid (10 mL) was refluxed for 8 h. The solid obtained, while hot, was recrystallized from dioxane to give (**16**) and (**17**), respectively. Physical, chemical and spectral data of newly synthesized compounds are given in Tables I and II.

Table I. Physical and analytical data of the newly synthesized compounds

Compd.	M. p. (°C)	Yield (%)	Mol. formula (M_r)	Analysis calcd./found (%)		
				C	H	N
2	254.6	93	$C_{13}H_{12}N_2O_3S_2$ (308.38)	50.63 50.33	3.92 3.62	9.08 9.31
3	265.2	90	$C_{14}H_{14}N_4O_3S_2$ (350.42)	47.99 47.60	4.03 4.28	15.99 15.68
4	239.8	89	$C_{17}H_{15}N_3O_4S_2$ (389.45)	52.43 52.13	3.88 3.57	10.79 10.43
5	238.1	86	$C_{18}H_{17}N_3O_4S_2$ (403.48)	53.58 53.23	4.25 4.55	10.41 10.08
6	269.8	79	$C_{16}H_{13}N_3O_3S_3$ (391.49)	49.09 49.33	3.35 3.71	10.73 10.42
7	242.4	74	$C_{22}H_{18}N_4O_3S_2$ (450.53)	58.65 58.96	4.03 4.29	12.44 12.70
8	193.0	80	$C_{18}H_{15}N_3O_3S_2$ (385.46)	56.09 56.33	3.92 3.71	10.90 10.58
9	301.7	78	$C_{17}H_{14}N_4O_3S_2$ (386.45)	52.84 52.49	3.65 3.31	14.50 14.77
10	276.9	77	$C_{18}H_{16}N_4O_3S_2$ (400.47)	53.98 53.62	4.03 4.29	13.99 13.69
11	262.6	83	$C_{19}H_{18}N_4O_3S_2$ (414.50)	55.05 55.32	4.38 4.09	13.52 13.84
12	147.2	61	$C_{11}H_{10}N_2O_2S$ (234.27)	56.39 56.77	4.30 4.65	11.96 11.86
13	130.8	71	$C_{15}H_{12}N_2O_2S_2$ (316.39)	56.94 56.69	3.82 3.58	8.85 8.49
14	215.8	66	$C_{16}H_{12}N_2OS$ (280.34)	68.55 68.91	4.31 4.08	9.99 10.12
15	208.8	69	$C_{17}H_{14}N_2OS$ (294.37)	69.36 69.70	4.79 4.46	9.52 9.19
16	168.9	61	$C_{20}H_{15}NO_2S$ (333.40)	72.05 72.33	4.53 4.19	4.20 4.52
17	211.8	77	$C_{21}H_{13}NO_3S$ (359.06)	70.18 70.51	3.65 3.19	3.90 3.58

Table II. Spectral data of the newly synthesized compounds

Compd.	IR (υ, cm^{-1})	^1H NMR (DMSO-d_6) (δ, ppm)	^{13}C NMR (DMSO-d_6) (δ, ppm)
2	3340, 3248, 3190 (NH, NH_2), 3101 (CH arom.), 1628 (C=O), 1360, 1157 (SO_2)	6.1, 7.1 (2d, 2H, CH=CH, *J* = 7.1, 7.6 Hz), 7.2-8.0 (m, 9H, Ar-H + SO_2NH_2), 10.4 (s, 1H, NH, D_2O exchangeable)	94.8, 115.8 (2), 128.5 (2), 128.6, 130.4, 138.0, 142.5, 144.2, 145.7, 146.4, 180.3
3	3425, 3320, 3185 (NH, NH_2), 3100 (CH arom.), 2970, 2897 (CH aliph.), 1646 (C=O), 1380, 1130 (SO_2)	6.4, 7.4 (2d, 2H, CH=CH, *J* = 7.7, 7.8 Hz), 6.7 (s, 2H, NH_2, D_2O- exchangeable), 7.5-7.9 (m, 7H, Ar-H), 8.1 (s, 1H, NH imino; D_2O- exchangeable), 10.3 (s, 1H, NH aniline; D_2O-exchangeable), 11.7 (s, 1H, SO_2NH, D_2O- exchangeable)	94.6, 115.6 (2), 128.4 (2), 128.6, 129.8, 137.7, 138.6, 144.4, 145.7, 146.5, 158.0, 180.2
4	3464, 3370 (2NH), 3066 (CH arom.), 2966, 2860 (CH aliph.), 1632 (C=O), 1589 (C=N), 1334, 1161 (SO_2)	2.3 (s, 3H, CH_3), 6.1 (d, 1H, CHCO; *J* = 7.2 Hz), 6.4 (s, 1H, CH oxazole), 7.2 (d, 1H, CH-NH; *J* = 7.5 Hz), 7.3-7.9 (m, 7H, Ar-H), 10.4 (d, 1H, NH; *J* = 7.4 Hz), 11.2 (s, 1H, SO_2NH, D_2O-exchangeable)	12.0, 95.3, 95.5, 116.0 (2), 128.6 (2), 128.8, 130.6, 132.7, 133.5, 144.2, 145.1, 146.3, 153.2, 169.8, 180.3
5	3317, 3209 (2NH), 2924, 2860 (CH aliph.), 1685 (C=O), 1635 (C=N), 1340, 1192 (SO_2)	2.1 (s, 6H, $2CH_3$), 6.1 (d, 1H, CHCO; *J* = 7.2 Hz), 7.2 (d, 1H, **CH**-NH; *J* = 7.4 Hz), 7.3-7.9 (m, 7H, Ar-H), 10.5 (d, 1H, NH; *J* = 7.8 Hz), 11.7 (s, 1H, SO_2NH, D_2O-exchangeable)	5.8, 10.3, 95.5, 100.0, 116.0 (2), 128.7 (2), 130.1, 130.6, 133.5, 142.1, 145.1, 145.6, 146.2, 155.5, 161.4, 183.4
6	3390, 3290 (2NH), 3090 (CH arom.), 2940, 2860 (CH aliph.), 1635 (C=O), 1593 (C=N), 1373, 1138 (SO_2)	6.1, 7.2 (2d, 2H, CH=CH; *J* = 7.2, 7.3 Hz), 6.4, 7.3 (2d, 2H, CH=CH thiazole; *J* = 7.7 Hz), 7.4-8.0 (m, 7H, Ar-H), 10.4 (d, 1H, NH; *J* = 7.0 Hz), 11.7 (s, 1H, SO_2NH, D_2O-exchangeable)	94.9, 108.1, 115.8 (2), 128.6 (2), 129.9, 130.4, 132.6, 133.3, 135.1, 142.4, 144.1, 146.4, 170.0, 180.3
7	3340, 3290 (2NH), 3065 (CH arom.), 2970, 2836 (CH aliph.), 1646 (C=O), 1610 (C=N), 1370, 1183 (SO_2)	6.1, 7.2 (2d, 2H, CH=CH, *J* = 7.6, 7.7 Hz), 6.4, 7.8 (2d, 2H, CH-CH pyrazole, *J* = 7.1, 7.3 Hz), 7.3-8.0 (m, 12H, Ar-H), 10.4 (d, 1H, NH, *J* = 7.6 Hz), 11.7 (s, 1H, SO_2NH, D_2O-exchangeable)	95.4, 99.9, 116.0 (2), 124.2 (2), 127.4, 128.6 (2), 128.7, 130.6 (2), 131.8, 138.2, 139.5, 142.2, 143.9, 144.0, 144.9, 145.6, 146.3, 180.4
8	3310, 3280 (2NH), 3055 (CH arom.), 2940, 2860 (CH aliph.), 1636 (C=O), 1597 (C=N), 1396, 1176 (SO_2)	6.4, 7.2 (2d, 2H, CH=CH, *J* = 7.1, 7.3 Hz), 7.3-8.1 (m, 11H, Ar-H), 10.4 (d, 1H, NH, *J* = 7.8 Hz), 11.7 (s, 1H, SO_2NH, D_2O-exchangeable)	95.0, 112.0, 113.2, 115.8 (2), 128.6 (2), 128.8, 129.9, 135.3, 139.9, 140.1, 143.2, 144.2, 145.7, 146.3, 152.8, 180.4

9	3464,3267 (2NH), 3035 (CH arom.), 2939, 2870 (CH aliph.), 1655 (C=O), 1582 (C=N), 1338, 1195 (SO_2)	6.4, 7.3 (2d, 2H, CH=CH, *J* = 7.0, 7.2 Hz), 7.4-8.5 (m, 10H, Ar-H), 10.4 (d, 1H, NH, *J* = 7.6 Hz), 11.7 (s, 1H, SO_2NH, D_2O-exchangeable)	95.3, 112.1, 115.6 (2), 128.6 (2), 130.0, 132.5, 133.7, 143.8, 144.8, 145.6, 146.3, 158.3 (2), 180.3, 183.3
10	3433, 3370 (2NH), 3060 (CH arom.), 2947, 2862 (CH aliph.), 1632 (C=O), 1593 (C=N), 1363, 1153 (SO_2)	2.3 (s, 3H, CH_3), 6.4, 7.3 (2d, 2H, CH=CH, *J* = 7.1, 7.4 Hz), 7.4-8.3 (m, 9H, Ar-H), 10.4 (d, 1H, NH, *J* = 8.1 Hz), 11.7 (s, 1H, SO_2NH, D_2O-exchangeable)	23.2, 95.2, 114.6, 115.4 (2), 128.6 (2), 129.7, 129.9, 133.4, 142.2, 144.7, 145.6, 146.3, 156.5, 168.7, 180.3, 183.6
11	3210 (2NH), 3066 (CH arom.), 2993, 2816 (CH aliph.), 1697 (C=O), 1635, 1593 (2C=N), 1373, 1157 (SO_2)	2.2 (s, 6H, $2CH_3$), 6.4, 7.2 (2d, 2H, CH=CH, *J* = 7.3, 7.5 Hz), 6.7-7.9 (m, 8H, Ar-H), 10.4 (d, 1H, NH, *J* = 8.0 Hz), 11.7 (s, 1H, SO_2NH, D_2O-exchangeable)	23.0 (2), 95.1, 99.6, 115.2 (2), 128.4 (2), 128.6, 129.9, 133.3, 134.3, 144.5, 145.6, 146.3, 167.3, 172.0, 180.3, 183.0
12	3132 (NH), 3082 (CH arom.), 2940, 2836 (CH aliph.), 1642 (C=O), 1635 (C=N)	2.3 (s, 3H, CH_3), 6.4 (s, 1H, CH isoxazole), 6.2, 7.3 (2d, 2H, CH=CH; *J* = 6.9 Hz), 7.5-7.9 (m, 3H, Ar-H), 10.3 (d, 1H, NH, *J* = 7.5 Hz)	12.1, 95.8, 100.6, 128.4, 133.7, 142.3, 145.4, 146.0, 160.3, 169.9, 180.4
13	3421 (NH), 3093 (CH arom.), 2930, 2860 (CH aliph.), 1645 (C=O), 1635 (C=N)	3.9 (s, 3H, OCH_3), 6.6, 7.2 (2d, 2H, CH=CH; *J* = 8.1, 8.3 Hz), 7.3-7.5 (m, 6H, Ar-H), 8.5 (d, 1H, NH; *J* = 6.8 Hz)	55.7, 108.1, 113.2, 113.6, 121.4, 128.8, 129.0, 136.5, 136.9, 138.1, 140.2, 142.0, 149.8, 165.1, 185.8
14	3209 (NH), 3047 (CH arom.), 2920, 2836 (CH aliph.), 1651 (C=O), 1612 (C=N)	6.2, 6.5 (2d, 2H, CH=CH; *J* = 7.2, 7.3 Hz), 7.4-8.0 (m, 8H, Ar-H), 8.9 (s, 1H, N=CH, quinoline), 11.8 (d,1H,NH, *J* = 7.5 Hz)	94.9, 127.0, 127.1, 127.2, 127.3, 128.6, 129.8, 130.2, 133.1, 134.9, 143.0, 143.1, 144.8, 145.8, 146.5, 180.2
15	3340 (NH), 3078 (CH arom.), 2970, 2830 (CH aliph.), 1643 (C=O), 1620 (C=N)	2.4 (s, 3H, CH_3), 5.9, 7.1 (2d, 2H, CH=CH; *J* = 7.0, 7.2 Hz), 6.4 (s, 1H, CH quinoline), 7.2-8.1 (m, 7H, Ar-H), 8.4 (d, 1H, NH; *J* = 7.8 Hz)	23.9, 92.3, 101.9, 122.2, 123.1, 128.2, 128.9, 129.0, 129.5, 136.9, 138.1, 140.2, 142.0, 142.6, 153.6, 160.2, 185.8
16	3433 (NH), 3046 (CH arom.), 2930, 2840 (CH aliph.), 1640 (2C=O)	6.2, 7.3 (2d, 2H, CH=CH, *J* = 7.4, 7.5 Hz), 7.2-8.0 (m, 12H, Ar-H), 10.5 (d, 1H, NH; *J* = 7.1 Hz)	113.2, 115.6 (2), 128.6 (4), 130.1 (4), 131.0, 133.8, 134.7, 148.8, 151.3, 152.8, 153.9, 163.4, 190.2
17	3444 (NH), 3070 (CH arom.), 2960, 2840 (CH aliph.), 1670, 1650 (3C=O)	6.5, 6.9 (2d, 2H, CH=CH; *J* = 7.0, 7.2 Hz), 6.9-8.2 (m, 10H, Ar-H), 10.7 (d, 1H, NH; *J* = 7.3 Hz)	100.9, 112.0, 119.6, 121.0, 126.3, 128.5 (2), 129.5, 132.9 (2), 133.3 (2), 133.6, 134.3, 134.5, 134.8, 141.4, 145.7, 180.0, 183.3 (2)

In vitro *anticancer activity*

Cytotoxic activity was measured *in vitro* for the newly synthesized compounds using the SulfoRhodamine-B stain (SRB) assay (35). Cells were plated in 96-multiwell microtiter plates (10^4 cells per well) for 24 h before treatment with the compound, to allow attachment of cells to the plate wall. Test compounds were dissolved in DMSO and diluted with saline to the appropriate concentration. Different concentrations of the compounds under test (10, 25, 50 and 100 μmol L^{-1}) were added to the cell monolayer. Triplicate wells were prepared for each individual dose. Monolayer cells were incubated with the compound(s) for 48 h at 37 °C and in an atmosphere of 5 % CO_2. After 48 h, the cells were fixed, washed, and stained for 30 min with 0.4 % (*m*/*V*) SRB in 1 % acetic acid. Excess unbound dye was removed by four washes with 1 % acetic acid and attached stain was recovered with Tris-EDTA buffer. Color intensity was measured in an enzyme-linked immunosorbent assay ELISA reader. The relation between the surviving fraction and drug concentration was plotted to get the survival curve for the breast tumor cell line after the specified time. The molar concentration required for 50 % inhibition of cell viability (IC_{50}) was calculated and the results are given in Table III.

Table III. In vitro *anticancer screening of the newly synthesized compounds against the human breast cancer cell line MCF7*

Compound	Compound concentration (μmol L^{-1})				IC_{50} (μmol L^{-1})
	10	25	50	100	
	Surviving fraction[a]				
Doxorubicin	0.551 ± 0.026	0.480 ± 0.003	0.139 ± 0.005	0.130 ± 0.016	32.00
2	0.814 ± 0.008	0.660 ± 0.025	0.419 ± 0.003	0.393 ± 0.006	38.25
3	0.792 ± 0.021	0.701 ± 0.031	0.347 ± 0.017	0.378 ± 0.018	36.24
4	0.807 ± 0.080	0.723 ± 0.031	0.293 ± 0.010	0.133 ± 0.014	47.30
5	0.845 ± 0.013	0.515 ± 0.021	0.380 ± 0.003	0.443 ± 0.017	28.85
6	0.541 ± 0.003	0.323 ± 0.020	0.360 ± 0.018	0.460 ± 0.015	10.25
7	0.480 ± 0.010	0.327 ± 0.016	0.313 ± 0.005	0.381 ± 0.007	9.70
8	0.693 ± 0.023	0.503 ± 0.033	0.377 ± 0.010	0.391± 0.015	23.48
9	0.443 ± 0.017	0.251 ± 0.012	0.355 ± 0.020	0.290 ± 0.009	9.55
10	0.810 ± 0.022	0.550 ± 0.019	0.331 ± 0.013	0.350 ± 0.015	27.51
11	0.872 ± 0.025	0.638 ± 0.016	0.370 ± 0.030	0.307 ± 0.005	34.50
12	0.825 ± 0.013	0.668 ± 0.021	0.307 ± 0.007	0.271 ± 0.017	33.55
13	0.435 ± 0.009	0.233 ± 0.006	0.371 ± 0.018	0.309 ± 0.011	9.39
14	0.792 ± 0.030	0.361 ± 0.010	0.145 ± 0.010	0.174 ± 0.010	36.40
15	0.659 ± 0.016	0.569 ± 0.011	0.137 ± 0.011	0.233 ± 0.013	39.70
16	0.920 ± 0.020	0.417 ± 0.014	0.272 ± 0.020	0.146 ± 0.012	42.50
17	0.904 ± 0.032	0.685 ± 0.011	0.355 ± 0.012	0.209 ± 0.011	53.01

[a]Mean ± S.E, $n = 3$.

RESULTS AND DISCUSSION

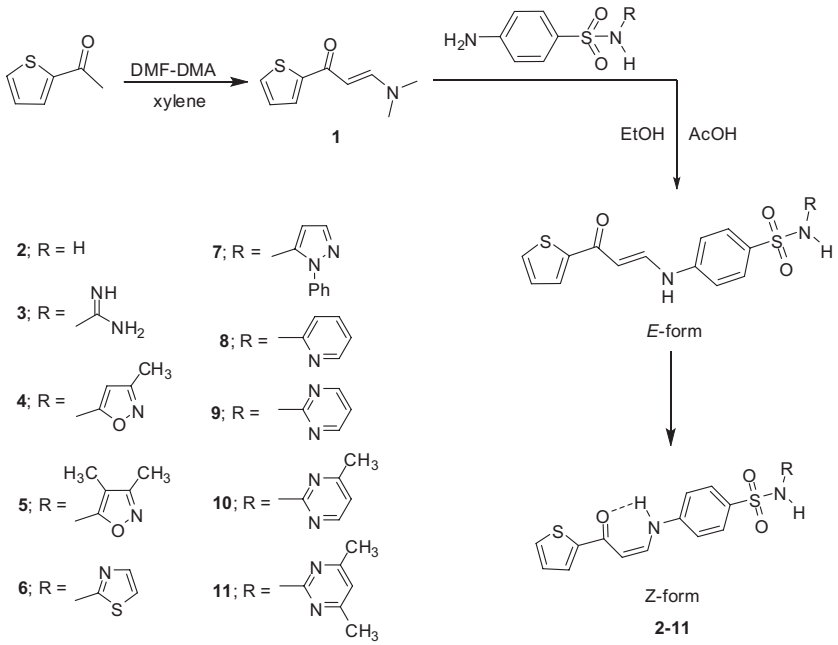
Chemistry

The current research deals with the synthesis of a new series of thiophene derivatives with sulfonamide, isoxazole, benzothiazole, quinoline and anthracene moieties through reaction of 2-acetyl thiophene with sulfa-drugs, aromatic amines and heterocyclic aromatic amines, and evaluation of their anticancer activity.

Schemes 1 and 2 display the synthesis of thiophenes having the biologically active sulfonamide **2–11**, 3-methylisoxazole **12**, 4-methoxybenzo[*d*]thiazole **13**, quinoline **14**, **15**, benzoylphenylamino **16**, and anthracene-9,10-dione **17** moieties.

Enaminone derivatives are highly reactive intermediates extensively used for the synthesis of heterocyclic compounds. Thus, treatment of 3-(dimethylamino)-1-(thiophen-2-yl) prop-2-en-1-one (**1**) with sulfonamide derivatives in refluxing ethanol/acetic acid mixture afforded the corresponding sulfonamide derivatives **2–11**. Structures of the latter products were assigned on the basis of their analytical and spectral data. ^{1}H NMR spectra of compounds **2–11** support the assumption that these structures were in *Z*-form and not in *E*-form, while the coupling constant of doublet signals for olefinic protons was equal to 7.0–7.8 Hz. Z-form was stabilized by intramolecular hydrogen bonding.

The IR spectra of the reaction products showed in each case three absorption bands corresponding to 2NH functions in the 3464–3185 cm^{-1} region, in addition to carbonyl



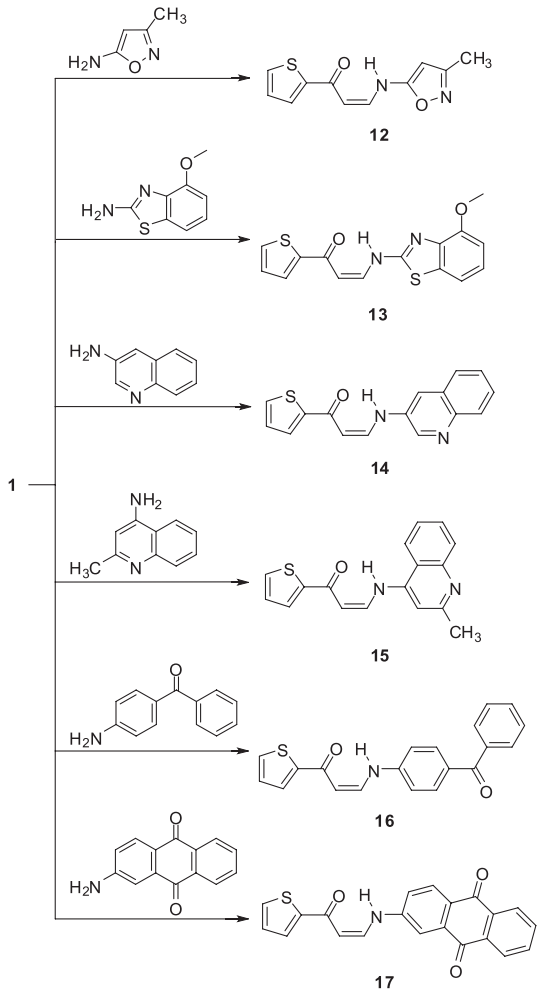
Sheme 1

absorption band in the 1697–1628 cm^{-1} region and absorption bands due to SO_2 functions in the 1396–1130 cm^{-1} region. ^1H NMR spectra of compounds **2–11** revealed a doublet in the 6.1–7.4 ppm region corresponding to the CH=CH group.

Also, interaction of compound **1** with 3-methyl-5-aminoisoxazole furnished the corresponding 3-methylisoxazole derivative **12**. (*Z*)-3-(3-Methylisoxazol-5-ylamino)-1-(thiophen-2-yl)prop-2-en-1-one (**12**) was proven on the basis of elemental analysis and spectral data. Its IR spectrum showed bands at 3132 (NH) and 1642 cm^{-1} (C=O). The ^1H NMR spectrum in DMSO-d_6 revealed signals at 2.3 ppm due to the CH_3 group and at 6.4 ppm corresponding to CH isoxazole. In addition, the corresponding benzothiazole derivative **13** was obtained in good yield *via* reaction of **1** with 4-methoxybenzo[d]thiazol-2-amine. The IR spectrum of **13** exhibited bands at 3421 (NH) and 1645 cm^{-1} (C=O). The ^1H NMR spectrum of compound **13** revealed signals at 3.9 ppm attributed to the OCH_3 group and at 8.5 ppm due to the NH group. Quinoline derivatives (**14** and **15**) were obtained *via* reaction of **1** with quinolin-3-amine or 2-methylquinolin-4-amine, respectively, in a refluxing ethanol/acetic acid mixture. Structures of compounds **14** and **15** were established on the basis of elemental analysis and spectral data. The IR spectrum of **14** showed bands at 3209 cm^{-1} (NH), 1651 cm^{-1} (C=O), and 1612 cm^{-1} (C=N). The ^1H NMR spectrum of **14** revealed signals at 8.9 ppm due to N=CH of quinoline and 11.8 ppm corresponding to the NH group. The IR spectrum of **15** exhibited bands at 3340 cm^{-1} (NH), 1643 cm^{-1} (C=O) and 1620 cm^{-1} (C=N). ^1H NMR spectrum of **15** revealed signals at 2.4 ppm attributed to the CH_3 group and at 8.4 ppm corresponding to the NH group. Interaction of **1** with 4-aminophenyl-phenyl-methanone gave the corresponding benzoylphenylamino derivative **16**. Its IR spectrum showed bands at 3433 (NH), and 1640 cm^{-1} (2C=O). The ^1H NMR spectrum of **16** showed a multiplet for aromatic protons. Finally, interaction of **1** with 2-aminoanthracene-9,10-dione yielded the corresponding anthracene-9,10-dione derivative **17**. Compound **17** was proven on the basis of elemental analysis and spectral data. Thus, its IR spectrum showed bands at 3444 cm^{-1} (NH) and 1670, 1650 cm^{-1} (3C=O). The ^1H NMR spectrum of **17** revealed the presence of a doublet at 6.5 and 6.9 ppm due to CH=CH groups, a multiplet at 6.9–8.2 ppm attributed to aromatic protons, and a signal at 10.7 ppm due to the NH group (Scheme 2).

In vitro *anticancer activity*

The newly synthesized compounds were evaluated for their *in vitro* anticancer activity against the human breast cancer cell line MCF7. Doxorubicin was used as the reference drug. Table III shows the *in vitro* cytotoxic activity of the newly synthesized compounds. Most of the tested compounds exhibited significant activity compared to doxorubicin. It was found that thiophenes containing biologically active sulfathiazole **6**, sulfaphenazole **7**, sulfadiazine **9**, or benzothiazole **13** moieties with respective IC_{50} values of 10.25, 9.70, 9.55, and 9.39 μmol L^{-1} exhibited 3.1- to 3.4-fold higher anticancer activity than the reference drug with the IC_{50} value of 32.00 μmol L^{-1}. Further, thiophenes bearing the biologically active sulfamethoxazole **5**, sulfapyridine **8** and sulfamethazine **10** with respective IC_{50} values of 28.85, 23.48, and 27.51 μmol L^{-1} were nearly as active as doxorubicin. On the other hand, compounds **2**, **3**, **11**, **12**, **14** and **15** revealed slightly lower activity than that of doxorubicin. Thiophene derivatives showed that the cell killing potency against the breast cancer cell line as follows: **13** > **9** > **7** > **6** > **8** > **10** > **5** doxorubicin.



Sheme 2

CONCLUSIONS

The objective of the present study was to synthesize and investigate the anticancer activity of some novel thiophene derivatives carrying the biologically active sulfonamide, isoxazole, benzothiazole, quinoline and anthracene moieties. Compounds (*Z*)-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)-*N*-(thiazol-2-yl)benzenesulfonamide (**6**), (*Z*)-4-(3-oxo-3-(thio-phen-2-yl)prop-1-enylamino)-*N*-(1-phenyl-1*H*-pyrazol-5-yl)benzenesulfonamide (**7**), (*Z*)-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)-*N*-(pyrimidin-2-yl)benzenesulfonamide (**9**) and (*Z*)-3-(4-methoxybenzo[d]thiazol-2-ylamino)-1-(thiophen-2-yl)prop-2-en-1-one (**13**) showed a promising anti-breast cancer activity, even higher than that of doxorubicin, while compounds (*Z*)-*N*-(3,4-dimethyl-isoxazol-5-yl)-4-(3-oxo-3-(thiophen-2-yl)prop-1-

enylamino)benzene-sulfonamide (**5**), (*Z*)-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)-*N*-(pyridin-2-yl)benzenesulfonamide (**8**) and (*Z*)-*N*-(4-methylpyrimidin-2-yl)-4-(3-oxo-3-(thiophen-2-yl)prop-1-enylamino)benzenesulfonamide (**10**) were nearly as active as doxorubicin.

Biological screening of the test compounds could offer an encouraging framework in this field, which may lead to the discovery of even more potent anticancer agents.

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