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Original research paper

Synthesis and cytotoxicity evaluation of thiazole derivatives obtained from 2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophene-3-carbonitrile

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Reactivity of 2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophene-3-carbonitrile towards thioglycolic acid resulted in thiazole derivative **1**. The latter reacted with different chemical reagents to give thiazole, pyrano[2,3-*d*]thiazole and thiazolo[4,5-*d*]thiazole derivatives. Cytotoxicity effects of the newly synthesized products against six cancer cell lines, namely, human gastric cancer (NUGC), human colon cancer (DLD-1), human liver cancer (HA22T and HEPG-2), human breast cancer (MCF) and nasopharyngeal carcinoma (HONE-1) as well as against a normal fibroblast cell (WI-38) were evaluated. The study showed that the 4,5,6,7 tetrahydrobenzo[*b*] thiophene derivatives **6a**, **7**, **8a**,**b**, **9b** and **10b**,**c** were the most active compounds. Their potencies were attributed to the presence of the electron withdrawing groups.

Keywords: tetrahydrobenzo[*b*]thiophene, thiazole, pyrano[2,3-*d*]thiazole, thiazolo[4,5-*d*]thiazole, cytotoxicity, anticancer activity

A number of thiazole derivatives were synthesized according to the Hantzsch thiazole synthesis (1), along with other methods (2–7). Heterocyclic compounds containing thiazole moiety were found to exhibit a wide spectrum of biological activities such as antioxidant (8), antitubercular (9, 10), diuretic (11), antischizophrenia (12), antibacterial (13, 14), anti-inflammatory (15), anti-HIV (16), antihypertensive (17), antiallergic (18), hypnotic (19), analgesic (20), antitumor and cytotoxic (21, 22). Thiazole moiety is present in many drugs such as thiamine (vitamin B_1), penicillin (antibiotic), sulfathiazole (antibacterial drug), 2-(4-chlorophenyl)thiazole-4-ylacetic (anti-inflammatory agent), thiabendazole [2-(4-thiazolyl)benzimidazole] (anthelmintic and fungicide), and niridazole [1-(5-nitro-2-thiazolyl)-2-imidazolidinone] (schistosomicidal agent) (23, 24). Some thiazole derivatives have been recently proven to be anticancer agents (25). In the present study, we demonstrated the reaction of 4,5,6,7-tetrahydrobenzo[*b*]thiophene with thioglycolic acid to produce new thiazole derivatives incorporating thiophene moiety and studied their cytotoxicity against different cancer cell lines.

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EXPERIMENTAL

General

All melting points were uncorrected and determined on an electrothermal apparatus (Büchi 535, Switzerland) in an open capillary tube. IR spectra (KBr discs) were recorded on a FTIR plus 460 IR spectrophotometer (Shimadzu, Japan). ^{13}C NMR and ^{1}H NMR spectra were recorded on a Varian Gemini-200 (200 MHz) (USA) spectrometer in DMSO-d_6 as solvent, using TMS as internal reference and chemical shifts (δ, ppm). Mass spectra were recorded using a Hewlett Packard 5988 (USA) GC/MS system and GCMS-QP 1000 Ex Shimadzu (Japan) using EI (electron impact method). Elemental analyses were carried out on a Vario EL III Elemental CHNS analyzer (Elementar Analysensysteme GmbH, Germany).

Syntheses

*2-(2-Amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl)thiazol-4(5*H*)-one (**1**).* – To a solution of 2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophene-3-carbonitrile (1.78 g, 0.01 mol) in acetic acid (30 mL), thioglycolic acid (0.92 g, 0.01 mol) was added. The reaction mixture was heated under reflux for 3 h, then poured into ice/water and the formed solid product was collected by filtration and crystallized from ethanol.

*2-Cyano-*N*-(4,5,6,7-tetrahydro-3-(4,5-dihydro-4-oxothiazol-2-yl)benzo[*b*]thiophen-2-yl)acetamide (**2**).* – To compound **1** (2.52 g, 0.01 mol) in dimethylformamide (30 mL), ethyl cyanoacetate (1.13 g, 0.01 mol) was added, then heated in a reflux system for 4 h and poured into an ice/water mixture. The formed solid product was collected by filtration and crystallized from dimethylformamide.

*1-(4,5,6,7-Tetrahydro-3-(4,5-dihydro-4-oxothiazol-2-yl)benzo[*b*]thiophen-2-yl)-3-phenylthiourea (**3**).* – To the dry solid of compound **1** (2.52 g, 0.01 mol) in 1,4-dioxane (35 mL) containing a catalytic amount of triethylamine (0.50 mL), phenylisothiocyanate (1.35 g, 0.01 mol) was added. The whole reaction mixture was heated under reflux for 4 h, then poured into an acidified ice/water mixture. The formed solid product was collected by filtration and crystallized from 1,4-dioxane.

*1-(3-(5-(2-Phenylhydrazono)-4,5-dihydro-4-oxothiazol-2-yl)-4,5,6,7-tetrahydrobenzo[*b*] thiophen-2-yl)-3-phenylthiourea (**4**).* – To a cold solution (0–5 °C) of compound **3** (3.87 g, 0.01 mol) in ethanol (50 mL) containing sodium hydroxide (0.40 g, 0.01 mol), benzenediazonium chloride (0.01 mol) [prepared by adding a cold solution of sodium nitrite (0.69 g, 0.01 mol) in water (10 mL) to a cold solution (0–5 °C) of aniline (0.93 g, 0.01 mol) in concentrated hydrochloric acid (12 mL) under continuous stirring] was added under continuous stirring. The whole reaction mixture was left at room temperature for 1 h and the solid product formed was collected by filtration and crystallized from ethanol.

*2-(2-Amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl)-5-bromo-thiazol-4(5*H*)-one (**5**)* – To a solution of compound **1** (2.52 g, 0.01 mol) in acetic acid (40 mL) at 50 °C, bromine (1.80 g, 0.01 mol) was added dropwise. The reaction mixture was kept at room temperature for 1 h under continuous stirring. The solid product, when poured into an ice/water mixture, was collected by filtration and recrystallized from acetic acid.

*2-(2-Amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl)-7-imino-7*H*-pyrano[2,3-d]thiazol-5-amine (**6a**) and 2-(2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl)-7-imino-7*H*-pyrano[2,3-d] thiazol-5-ol (**6b**). General procedure.* – To a solution of compound **1** (2.52 g, 0.01 mol) in 1,4-dioxane (40 mL) containing triethylamine (0.50 mL), either malononitrile (0.66 g, 0.01 mol) or ethyl cyanoacetate (1.13 g, 0.01 mol) was added. The reaction mixture, in each case, was heated under reflux for 5 h, left to cool and then poured into an ice/water mixture containing a few drops of hydrochloric acid. The formed solid product, in each case, was collected by filtration and re-crystallized from 1,4-dioxane.

*5-(2-Amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl)-3-phenyl-thiazolo[4,5-d]thiazole-2(3*H*)-thione (**7**).* – To a mixture of compound **1** (2.52 g, 0.01 mol) in 1,4-dioxane (35 mL) containing triethylamine (0.50 mL), elemental sulfur (0.32 g, 0.01 mol) and phenylisothiocyanate (1.35 g, 0.01 mol) were added. The reaction mixture was heated under reflux for 5 h and then poured into a beaker containing an acidified ice/water mixture. The solid product was collected by filtration, dried and then recrystallized from 1,4-dioxane.

*5-(2-Phenylhydrazono)-2-(2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl)thiazol-4(5*H*)-one (**8a**), (5*E*)-5-(2-(4-chlorophenyl)hydrazono)-2-(2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl) thiazol-4(5*H*)-one (**8b**), 5-(2-(4-methoxyphenyl)-hydrazono)-2-(2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl)thiazol-4(5*H*)-one (**8c**) and 5-(2-*p*-tolylhydrazono)-2-(2-amino-4,5,6,7-tetrahydrobenzo[b]thiophen-3-yl)thiazol-4(5*H*)-one (**8d**). General procedure.* – To a cold solution (0–5 °C) of compound **1** (2.52 g, 0.01 mol) in ethanol (50 mL) containing sodium hydroxide (10 %, 10 mL), a solution of either benzenediazonium chloride (0.01 mol) or *p*-chlorobenzenediazonium chloride (0.01 mol) or *p*-methoxybenzenediazonium chloride (0.01 mol) or *p*-methylbenzenediazonium chloride (0.01 mol) [prepared by dissolving sodium nitrite (0.70 g, 0.01 mol) in water (2 mL) and added to a cold solution of aniline (0.93 g, 0.01 mol), *p*-chloroaniline (1.27 g, 0.01 mol), *p*-methoxyaniline (1.23 g, 0.01 mol) or *p*-toluidine (1.07 g, 0.01 mol), containing an appropriate amount of hydrochloric acid under continuous stirring] was added under continuous stirring. The solid product formed, in each case, was collected by filtration and dried, and then recrystallized from ethanol.

*Ethyl-2-cyano-2-(2-(3-(4-oxo-4,5-dihydrothiazol-2-yl)-4,5,6,7-tetrahydrobenzo[*b*]thiophen-2-yl)hydrazono)acetate (**9a**), (3-(4-oxo-4,5-dihydrothiazol-2-yl)-4,5,6,7-tetrahydrobenzo[*b*]thiophen--2-yl)carbonohydrazonoyl dicyanide (**9b**), dimethyl-2-(2-(3-(4-oxo-4,5-dihydrothiazol-2-yl)-4,5,6,7-tetrahydrobenzo[*b*]thiophen-2-yl)hydrazono)malonate (**9c**) and diethyl-2-(2-(3-(4-oxo-4,5-dihydrothiazol-2-yl)-4,5,6,7-tetrahydrobenzo[*b*]thiophen-2-yl)-hydrazono)malonate (**9d**). General procedure.* – To a cold solution (0–5 °C) of the diazotized compound **1** [prepared by adding a $NaNO_2$ (0.69 g, 0.01 mol) solution to a cold solution of **1** (2.52 g, 0.01 mol) in acetic acid (20 mL) and HCl (6 mL, 18 %)], either ethyl cyanoacetate (1.13 g, 0.01 mol) or malononitrile (0.66 g, 0.01 mol) or acetyl acetone (1.00 g, 0.01 mol) or malonic acid diethyl ester (1.60 g, 0.01 mol) in ethanol (20 mL) containing sodium hydroxide (1.00 g) was gradually added under stirring. Upon cooling in an ice-bath, a solid product formed in each case. It was collected by filtration, washed with water and crystallized from ethanol.

*2-(2-Amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl)-5-benzylidenethiazol-4(5*H*)-one (**10a**), 5-(4-chlorobenzylidene)-2-(2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl)thiazol-4(5*H*)-one (**10b**), 5-(4-methoxybenzylidene)-2-(2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl)* *thiazol-4(5*H*)-one (**10c**) and 5-(2-hydroxybenzylidene)-2-(2-amino-4,5,6,7-tetrahydrobenzo[*b*] thiophen-3-yl)thiazol-4(5*H*)-one (**10d**). General procedure.* – To a solution of compound **1** (2.52 g, 0.01 mol) in 1,4-dioxane and a catalytic amount of piperidine (0.50 mL), either benzaldehyde (1.06 g, 0.01 mol) or *p*-chlorobenzaldehyde (1.12 g, 0.01 mol) or *p*-methoxy-benzaldehyde (1.08 g, 0.01 mol) or salicylaldehyde (1.22 g, 0.01 mol) were added. The reaction mixture was heated under reflux for 5 h, then poured into an acidified ice/water mixture. The formed solid product, in each case, was collected by filtration and recrystallized from 1,4-dioxane.

In vitro *cytotoxic assay*

Fetal bovine serum (FBS) and *L*-glutamine were purchased from the Gibco Invitrogen Company (UK). RPMI-1640 medium was purchased from Cambrex (USA). Dimethyl sulfoxide (DMSO), CHS-828, penicillin, streptomycin and sulforhodamine B (SRB) were purchased from the Sigma Chemical Company (USA).

No experiments were done on humans. Cancer and normal human cell lines were purchased. Cell cultures were obtained from the European Collection of Cell Cultures (ECACC, Salisbury, UK) while human gastric cancer (NUGC and HR), human colon cancer (DLD-1), human liver cancer (HA22T and HEPG-2), human breast cancer (MCF-7), nasopharyngeal carcinoma (HONE-1) and normal fibroblast cells (WI-38) were kindly provided by the National Cancer Institute (NCI, Cairo, Egypt). Cell lines grew as monolayers and were routinely maintained in RPMI-1640 medium supplemented with 5 % heat inactivated FBS, 2 mmol L^{-1} glutamine and antibiotics (penicillin 100 U mL^{-1}, streptomycin 100 μg mL^{-1}), at 37 °C in a humidified atmosphere containing 5 % CO_2. Exponentially growing cells were obtained by plating 1.5×10^5 cells mL^{-1} for the six human cancer cell lines, followed by 24 h of incubation.

The prepared heterocyclic compounds were evaluated according to standard protocols for their *in vitro* cytotoxicity (26–28) against the six human cancer cell lines: human gastric cancer (NUGC), human colon cancer (DLD-1), human liver cancer (HA22T and HEPG-2), human breast cancer (MCF-7) and nasopharyngeal carcinoma (HONE-1), as well as normal fibroblast cells (WI-38).

The reference compound was (*Z*)-(6-(4-chlorophenoxy)hexyl)-3-cyano-2-(pyridin-4-yl)guanidine (CHS-828), which is an antitumor agent. The effect of vehicle solvent (DMSO) on the growth of these cell lines was evaluated in all experiments by exposing untreated control cells to the maximum concentration (0.5 %) of DMSO used in each assay.

RESULTS AND DISCUSSION

Chemistry

The reaction of 2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophene-3-carbonitrile with thioglycolic acid gave the thiazole derivative **1**. The structure of compound **1** was confirmed on the basis of analytical and spectral data. Thus, the ^1H NMR spectrum showed the presence of multiplets at δ 1.69–1.75 and δ 2.50–2.57 ppm for the four CH_2 groups of

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

Compd.	Molecular formula (M_r)	M. p. (°C)	Yield (%)	Crystal color	Analysis (calcd./found) (%)			
					C	H	N	S
1	$C_{11}H_{12}N_2OS_2$ (252.36)	187–190	71	Canary yellow crystals	52.35/52.75	4.79/4.57	11.10/11.42	25.41/25.73
2	$C_{14}H_{13}N_3O_2S_2$ (319.40)	202–205	60	Gray crystals	52.65/52.99	4.10/4.38	13.16/12.96	20.08/20.34
3	$C_{18}H_{17}N_3OS_3$ (387.54)	210–213	70	Brown crystals	55.79/56.10	4.42/4.66	10.84/11.02	24.82/25.11
4	$C_{24}H_{21}N_5OS_3$ (491.65)	117–120	80	Brown crystals	58.63/58.20	4.31/4.22	14.24/14.64	19.57/19.17
5	$C_{11}H_{11}N_2OS_2Br$ (331.25)	127–130	60	Yellowish white crystals	39.88/40.10	3.35/2.99	8.46/8.43	19.36/19.66
6a	$C_{14}H_{14}N_4OS_2$ (318.42)	147 –150	72	Brown crystals	52.81/53.10	4.43/4.83	17.60/17.29	20.14/19.80
6b	$C_{14}H_{13}N_3O_2S_2$ (319.40)	207–210	95	Green crystals	52.65/52.99	4.10/4.50	13.16/13.12	20.08/19.75
7	$C_{18}H_{15}N_3S_4$ (401.59)	132–135	70	Brown crystals	48.31/48.61	4.24/3.89	10.53/10.46	31.62/31.94
8a	$C_{17}H_{16}N_4OS_2$ (356.47)	197–200	70	Orange crystals	57.28/57.23	4.52/4.23	15.72/15.53	17.99/17.66
8b	$C_{17}H_{15}N_4OS_2Cl$ (390.91)	137–140	78	Orange crystals	52.23/52.53	3.87/3.50	14.33/14.33	16.41/16.71
8c	$C_{18}H_{18}N_4O_2S_2$ (386.49)	97–100	50	Brown crystals	55.94/56.23	4.69/4.35	14.50/14.73	16.59/16.90
8d	$C_{18}H_{18}N_4OS_2$ (370.49)	137–140	82	Faint brown crystals	58.35/58.65	4.90/5.23	15.12/14.80	17.31/16.95
9a	$C_{16}H_{16}N_4O_3S_2$ (376.45)	127–130	60	Faint brown crystals	51.05/51.15	4.28/4.00	14.88/14.68	17.04/16.93
9b	$C_{14}H_{11}N_5OS_2$ (329.40)	177–180	70	Brown crystals	51.05/51.08	3.37/3.48	21.26/20.90	19.47/19.10
9c	$C_{16}H_{17}N_3O_3S_2$ (363.45)	107–110	75	Brown crystals	52.87/53.01	4.71/4.75	11.56/11.70	17.64/17.82
9d	$C_{18}H_{21}N_3O_5S_2$ (423.51)	127–130	60	Faint brown crystals	51.05/51.02	5.00/4.99	9.92/10.20	15.14/15.44
10a	$C_{18}H_{16}N_2OS_2$ (340.46)	177–180	75	Yellow crystals	63.50/63.64	4.74/4.81	8.23/8.48	18.84/19.10
10b	$C_{18}H_{15}N_2OS_2Cl$ (374.91)	175–178	71	Yellow crystals	57.67/57.54	4.03/4.32	7.47/7.70	17.11/16.80
10c	$C_{19}H_{18}N_2O_2S_2$ (370.49)	182–185	72	Yellow crystals	61.60/61.81	4.90/5.29	7.56/7.86	17.31/17.00
10d	$C_{18}H_{16}N_2O_2S_2$ (356.46)	187–190	80	Canary yellow crystals	60.65/61.01	4.52/4.40	7.86/8.22	17.99/18.30

Table II. Spectral and mass data of the newly synthesized compounds

Compd.	^{1}H NMR (DMSO-d_6) (δ, ppm)	^{13}C NMR (DMSO-d_6) (δ, ppm)	IR (ν_{max}, cm^{-1})	MS: m/z (%) = $[M]^+$
1	1.69-1.75 (m, 4H, $2CH_2$), 2.50-2.57 (m, 4H, $2CH_2$), 3.90 (s, 2H, CH_2), 6.91 (s, 2H, NH_2)	21.68, 23.23, 23.43, 23.96 ($4CH_2$ cyclohexene), 38.93 (CH_2 thiazole), 126.89, 130.45, 131.05, 146.64 (thiophene 4C), 162.67 (thiazole C=N), 168.02 (C=O)	3426, 3333 (NH_2), 2932-2842 (CH_2), 1692 (C=O), 1623 (C=N), 1576, 1436 (C=C)	253 $[M+1]^+$ (1.90), 252 $[M]^+$ (4.30), 251 $[M-1]^+$ (4.10)
2	1.70-1.74 (m, 4H, $2CH_2$), 2.39-2.55 (m, 4H, $2CH_2$), 3.35 (s, 2H, CH_2), 4.09 (s, 2H, CH_2), 11.46 (s, 1H, NH, D_2O exchange-able)	22.92, 23.24, 23.44, 23.97 ($4CH_2$ cyclohex-ene), 25.00 (CH_2), 39.22 (CH_2 thiazole), 114.21 (CN), 103.00, 126.00, 126.90., 146.65 (thiophene 4C) 163.00 (thiazole C=N), 168.03, 175.00 (2C=O)	3427-3218 (NH), 2934-2841 (CH_2), 2216 (CN), 1710, 1694 (2C=O), 1623 (C=N), 1576, 1457 (C=C)	321 $[M+2]^+$ (0.29), 320 $[M+1]^+$ (0.09), 319 $[M^+]^+$ (0.18), 318 $[M-1]^+$ (0.21), 317 $[M-2]^+$ (0.80), 59 (100.00)
3	1.74-1.82 (m, 4H, $2CH_2$), 2.48-2.56 (m, 4H, $2CH_2$), 3.56 (s, 2H, CH_2), 6.90-7.51 (m, 5H, C_6H_5), 9.76 (s, 1H, NH, D_2O exchangeable), 11.48 (s, 1H, NH, D_2O exchangeable)	22.56, 22.91, 23.24, 23.44 ($4CH_2$ cyclohex-ene), 38.95 (CH_2 thiazole), 120.84, 123.57, 124.33, 125.85, 126.91, 128.35 (phenyl 6C), 130.48, 138.00, 139.41, 146.64 (thiophene 4C), 162.00 (thiazole C=N), 168.04 (C=O), 180.00 (C=S)	3434-3219 (2NH), 3080 (CH aromatic), 2933-2841 (CH_2), 1694 (C=O),1625 (C=N), 1575, 1438 (C=C), 1369, 1282 (C=S)	388 $[M+1]^+$ (1.02), 387 $[M]^+$ (8.29), 386 $[M-1]^+$ (2.80), 77 $[C_6H_5]^+$ (14.09), 156 (100.00)
4	1.74-1.83 (m, 4H, $2CH_2$), 2.50-2.57 (m, 4H, $2CH_2$), 6.93-7.88 (m, 10H, $2C_6H_5$), 8.70, 9.00, 11.49 (3s, 3H, 3NH, D_2O exchangeable)	22.30, 22.49, 23.17, 23.37 ($4CH_2$ cyclohexene), 114.09, 114.09, 122.01, 126.89, 126.89, 128.58, 128.58, 129.29, 129.29, 130.42, 138.02, 143.10, (phenyl 12C), 118.05, 127.20, 137.10, 150.40, (thiophene 4C), 146.56, 163.02 (thiazole 2C=N), 167.93 (C=O), 179.01 (C=S)	3430-3275 (3NH), 3076 (CH aromatic), 2929 (CH_2), 1692 (C=O), 1640 (C=N), 1620, 1440 (C=C), 1373, 1248 (C=S)	493 $[M+1]^+$ (0.19), 492 $[M]^+$ (0.26), 491 $[M-1]^+$ (0.18), 490 $[M-2]^+$ (0.19), 128 (100.00), 77 $[C_6H_5]^+$ (74.27)
5	1.73-1.91 (m, 4H, $2CH_2$), 2.37-2.60 (m, 4H, $2CH_2$), 6.6 (s, 1H, CH thiazole), 7.28 (s, 2H, NH_2, D_2O exchangeable)	22.03, 22.77, 23.31, 24.82 ($4CH_2$ cyclohexene), 59.01 (CH thiazole), 118.07, 125.88, 131.43, 157.98 (thiophene 4C), 159.53 (thiazole C=N), 167.61 (C=O)	3314, 3194 (NH_2), 2931-2855 (CH_2), 1664 (C=O), 1583, 1443 (C=C), 1527 (C=N)	333 $[M+2]^+$ (4.03), 332 $[M+1]^+$ (27.84), 331 $[M]^+$ (2.58), 330 $[M-1]^+$ (14.10), 329 $[M-2]^+$ (1.75), 192 (100.00)

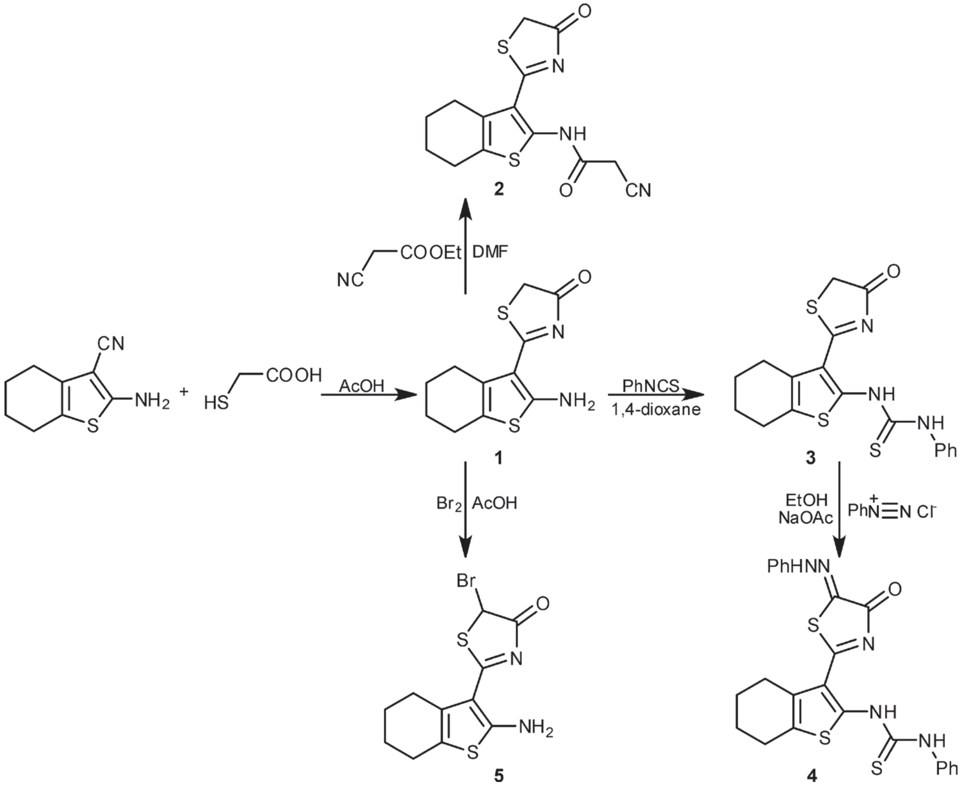
6a	1.68-1.95 (m, 4H, $2CH_2$), 2.33-2.55 (m, 4H, $2CH_2$), 6.92 (s, 1H, CH pyran), 7.09, 7.25 (2s, 4H, $2NH_2$, D_2O exchangeable), 11.46 (s, 1H, NH, D_2O exchangeable)	22.90, 23.23, 23.43, 23.96 ($4CH_2$ cyclohexene), 66.31 (pyran C), 126.90, 130.46, 131.05, 140.00 (thiophene 4C), 146.63 (thiazole C=N), 152.00, 162.67, 164.00, 168.03 (pyran 3C, C=NH)	3428, 3333 ($2NH_2$), 3271-3218 (NH), 2997-2842 (CH_2), 1690 (C=N), 1622, 1438 (C=C)	320 $[M+2]^+$ (0.07), 319 $[M+1]^+$ (0.13), 318 $[M]^+$ (0.12), 317 $[M-1]^+$ (0.46), 59 (100.00)
6b	1.69-1.75 (m, 4H, $2CH_2$), 2.31-2.56 (m, 4H, $2CH_2$), 6.90 (s, 1H, CH pyran), 7.15 (s, 2H, NH_2, D_2O exchangeable), 11.47 (s, 1H, NH, D_2O exchangeable), 15.10 (s, 1H, OH)	22.94, 23.26, 23.47, 23.99 ($4CH_2$ cyclohexene), 66.00 (pyran C), 126.95, 130.51, 131.08, 140.00 (thiophene 4C), 146.67 (thiazole C=N), 153.00, 162.70, 168.08, 178.00 (pyran 3C, C=NH)	3428, 3334 (NH_2), 3273-3222 (NH, OH), 2997-2842 (CH_2), 1694 (C=N), 1623, 1437 (C=C)	321 $[M+2]^+$ (0.69), 320 $[M+1]^+$ (1.72), 319 $[M]^+$ (0.36), 318 $[M-1]^+$ (0.41), 317 $[M-2]^+$ (0.23), 64 (100.00)
7	1.70-1.74 (m, 4H, $2CH_2$), 2.32-2.55 (m, 4H, $2CH_2$), 6.77 (s, 2H, NH_2, D_2O exchangeable), 6.91-7.54 (m, 5H, C_6H_5)	22.92, 23.25, 23.45, 23.98 ($4CH_2$ cyclohexene), 124.24, 124.24, 126.90, 128.30, 128.66, 128.66, 130.46, 131.50, 134.00, 139.47 (thiophene 4C, phenyl 6C), 146.64, 162.67, 168.02 (thiazole 2C, C=N), 179.49 (C=S)	3325, 3332 (NH_2), 3079-3000 (CH aromatic), 2929, 2842 (CH_2), 1691 (C=N) 1623, 1441 (C=C), 1368, 1282 (C=S)	404 $[M+2]^+$ (0.22), 402 $[M]^+$ (0.27), 401 $[M-1]^+$ (0.27), 400 $[M-2]^+$ (0.51), 77 $[C_6H_5]^+$ (100.00)
8a	1.74-1.75 (m, 4H, $2CH_2$), 2.49-2.62 (m, 4H, $2CH_2$), 6.40 (s, 2H, NH_2, D_2O exchangeable), 6.90-7.61 (m, 5H, C_6H_5), 11.48 (s, 1H, NH, D_2O exchangeable)	22.38, 22.56, 23.23, 23.43 ($4CH_2$ cyclohexene), 114.21, 114.21, 122.00, 126.88, 126.88, 129.02 (phenyl 6C), 130.45, 137.01, 143.03, 146.64 (thiophene 4C), 152.01, 162.03 (thiazole 2C=N), 168.02 (C=O)	3431, 3276 (NH_2), 3226 (NH), 3081 (CH aromatic), 2933, 2858 (CH_2), 1696 (C=O), (C=N) 1640, (C=C), 1576, 1440, 1555 (=N-NH)	357 $[M+1]^+$ (0.91), 356 $[M]^+$ (1.14), $^+$ 355 $[M-1]^+$ (0.23), 92 (100.00)
8b	1.71-1.75 (m, 4H, $2CH_2$), 2.50-2.65 (m, 4H, $2CH_2$), 6.91 (s, 2H, NH_2, D_2O exchangeable), 7.36-7.76 (m, 4H, C_6H_4), 11.49 (s, 1H, NH, D_2O exchangeable)	22.37, 22.55, 23.23, 23.43 ($4CH_2$ cyclohexene), 114.20, 114.20, 119.01, 126.87, 126.87, 127.01 (phenyl 6C), 128.29, 130.45, 142.01, 146.63 (thiophene 4C), 150.01, 162.01 (thiazole 2C=N), 168.01 (C=O)	3431, 3274 (NH_2), 3225 (NH), 3081 (CH aromatic), 2995-2858 (CH_2), 1696 (C=O), 1645 (C=N), 1600, 1440 (C=C), 1554 (=N-NH)	393 $[M+2]^+$ (0.25), 392 $[M]^+$ (0.34), 76 $[C_6H_4]^+$ (5.04), 150 (100.00)
8c	1.20 (s, 3H, CH_3), 1.69-1.91 (m, 4H, $2CH_2$), 2.32-2.64 (m, 4H, $2CH_2$), 6.90 (s, 2H, NH_2, D_2O exchangeable), 6.98-7.80 (m, 4H, C_6H_4), 11.45 (s, 1H, NH, D_2O exchangeable)	22.93, 23.25, 23.45, 23.98 ($4CH_2$ cyclohexene), 55.62 (OCH_3), 115.40, 115.40, 116.85, 116.85, 119.01, 127.74 (phenyl 6C), 131.06, 137.01, 144.00, 151.01 (thiophene 4C), 153.02, 162.69 (thiazole 2C=N), 168.01 (C=O)	3430, 3333 (NH_2), 3216 (NH), 3050 (CH aromatic), 2934, 2838 (CH_2, CH_3), 1690 (C=O), 1640 (C=N) 1605, 1440 (C=C), 1510 (=N-NH)	387 $[M+1]^+$ (0.60), 386 $[M]^+$ (0.68), 80 (100.00), 76 $[C_6H_4]^+$ (3.53)

8d	1.10 (s, 3H, CH_3), 1.69-1.95 (m, 4H, $2CH_2$), 2.37-2.57 (m, 4H, $2CH_2$), 6.91 (s, 2H, NH_2, D_2O exchangeable), 7.16-7.49 (m, 4H, C_6H_4), 11.48 (s, 1H, NH, D_2O exchangeable)	21.67, 22.38, 22.56, 23.23, 23.43, (4CH_2 cyclohexene, CH_3), 114.21, 114.21, 126.88, 126.88, 128.01, 129.02 (phenyl 6C), 130.45, 137.01, 143.03, 146.64 (thiophene 4C), 152.01, 162.03 (thiazole 2C=N), 168.02 (C=O)	3433, 3277 (NH_2), 3228 (NH), 3083 (CH aromatic), 2932, 2860 (CH_2, CH_3), 1696 (C=O), 1645 (C=N), 1610, 1437 (C=C), 1555 (=N-NH)	373 $[M+2]^+$ (0.54), 150 (100.00), 76 $[C_6H_4]^+$ (6.29)
9a	1.21-1.24 (t, 3H, CH_3), 1.75-1.91 (m, 4H, $2CH_2$), 2.50-2.83 (m, 4H, $2CH_2$), 3.80 (s, 2H, CH_2), 4.10-4.20 (q, 2H, CH_2), 11.37 (s, 1H, NH, D_2O exchangeable)	17.56, 23.09, 23.69, 24.11, 24.63 (4CH_2 cyclohexene, CH_3), 39.87 (CH_2 thiazole), 66.39 (CH_2), 113.15 (CN), 109.34, 116.96, 120.78, 135.62, 137.64, (thiophene 4C, C=N), 158.74 (thiazole C=N), 159.25, 172.10 (2C=O)	3432 (NH), 2935 (CH_2, CH_3), 2219 (CN), 1740, 1680 (2C=O), 1639 (C=N), 1600, 1437 (C=C), 1526 (=N-NH)	377 $[M+1]^+$ (0.67), 376 $[M]^+$ (0.73), 64 (100.00)
9b	1.76-1.91 (m, 4H, $2CH_2$), 2.50-2.82 (m, 4H, $2CH_2$), 3.86 (s, 2H, CH_2), 11.23 (s, 1H, NH, D_2O exchangeable)	23.40, 23.52, 23.80, 24.23 (4CH_2 cyclohex-ene), 38.90 (CH_2 thiazole), 113.01, 113.01 (2CN), 86.05, 119.01, 125.10, 137.60, 137.62 (thiophene 4C, C=N), 160.21 (thiazole C=N), 173.31 (C=O)	3434 (NH), 2933 (CH_2), 2260, 2199 (2CN), 1680 (C=O), 1638 (C=N), 1600, 1436 (C=C), 1555 (=N-NH)	327 $[M-2]^+$ (8.94), 178 (100.00)
9c	1.10, 1.23 (2s, 6H, $2CH_3$), 1.77-1.96 (m, 4H, $2CH_2$), 2.51-2.82 (m, 4H, $2CH_2$), 3.30 (s, 2H, CH_2), 11.49 (s, 1H, NH, D_2O exchangeable)	22.54, 22.68, 23.35, 23.56, 23.77, 24.23 (4CH_2 cyclohexene, 2CH_3), 38.87 (CH_2 thiazole), 117.07, 127.10, 130.61, 133.97, 137.72 (thio-phene 4C, C=N), 159.28 (thiazole C=N), 172.02, 181.10, 181.10 (3C=O)	3436 (NH), 2935 (CH_2, CH_3), 1690, 1685, 1670 (3C=O), 1637 (C=N), 1600, 1437 (C=C), 1546 (=N-NH)	364 $[M+1]^+$ (0.58), 363 $[M]^+$ (0.62), 362 $[M-1]^+$ (0.52), 64 (100.00)
9d	1.06-1.23 (t, 6H, $2CH_3$), 1.76-1.91 (m, 4H, $2CH_2$), 2.61-2.82 (m, 4H, $2CH_2$), 3.88 (s, 2H, CH_2), 4.18-4.44 (q, 4H, $2CH_2$), 11.69 (s, 1H, NH, D_2O exchangeable)	13.55, 13.55, 23.08, 23.27, 23.71, 24.14 (4CH_2 cyclohexene, 2CH_3), 39.98 (CH_2 thiazole), 65.66, 65.66 (2CH_2), 114.81, 117.11, 120.81, 135.61, 137.64 (thiophene 4C, C=N), 157.71 (thiazole C=N), 158.22, 158.74, 172.05 (3C=O)	3433 (NH), 2935 (CH_2, CH_3), 1750, 1745, 1675 (3C=O), 1636 (C=N), 1600, 1439 (C=C), 1544 (=N-NH)	422 $[M-1]^+$ (1.09), 421 $[M-2]^+$ (1.37), 64 (100.00)
10a	1.73-1.81 (m, 4H, $2CH_2$), 2.50-2.71 (m, 4H, $2CH_2$), 6.90 (s, 1H, CH), 7.52 (s, 2H, NH_2, D_2O exchange-able), 7.54-7.99 (m, 5H, C_6H_5)	23.22, 23.42, 23.69, 24.52 (4CH_2 cyclohex-ene), 120.64, 120.64, 128.98, 128.98, 129.20, 130.46 (phenyl 6C), 132.52, 132.88, 134.35, 134.72, 146.63, 159.49, 160.47 (thiophene 4C, CH, thiazole C, C=N), 168.03 (C=O)	3270, 3224 (NH_2), 3079 (CH aromatic), 2997-2840 (CH, CH_2), 1695 (C=O), 1640 (C=N), 1597, 1448 (C=C)	342 $[M+2]^+$ (3.97), 341 $[M+1]^+$ (2.83), 340 $[M]^+$ (8.15), 339 $[M-1]^+$ (1.37), 338 $[M-2]^+$ (1.10), 77 $[C_6H_5]^+$ (28.37), 59 (100.00)

10b	1.74-1.81 (m, 4H, $2CH_2$), 2.49-2.71 (m, 4H, $2CH_2$), 6.91 (s, 1H, CH), 7.36 (s, 2H, NH_2, D_2O exchange-able), 7.60-7.99 (m, 4H, C_6H_4)	23.21, 23.42, 23.66, 24.54 ($4CH_2$ cyclohexene), 114.19, 126.85, 126.85, 129.24, 129.24, 130.43 (phenyl 6C), 131.03, 133.27, 133.59, 137.08, 146.63, 158.96, 167.95 (thiophene 4C, CH, thiazole C, C=N), 167.99 (C=O)	3273, 3223 (NH_2), 3079 (CH aromatic), 2994-2850 (CH, CH_2), 1694 (C=O), 1630 (C=N), 1595, 1439 (C=C)	377 $[M+2]^+$ (0.30), 376 $[M+1]^+$ (0.74), 375 $[M]^+$ (0.47), 374 $[M-1]^+$ (0.94), 373 $[M-2]^+$ (0.24), 58 (100.00)
10c	1.73-1.76 (m, 4H, $2CH_2$), 2.46-2.65 (m, 4H, $2CH_2$), 3.85 (s, 3H, CH_3), 6.90 (s, 1H, CH), 7.06 (s, 2H, NH_2, D_2O exchange-able), 7.09-7.91 (m, 4H, C_6H_4)	23.24, 23.44, 23.72, 24.49 ($4CH_2$ cyclohexene), 55.50 (OCH_3), 114.21, 114.21, 114.55, 127.51, 127.51, 130.44 (phenyl 6C), 131.27, 131.71, 134.04, 146.64, 159.69, 160.30, 162.91, (thiophene 4C, CH, thiazole C, C=N), 168.01 (C=O)	3264, 3219 (NH_2), 3078 (CH aromatic), 2995-2842 (CH, CH_2, CH_3), 1691 (C=O), 1620 (C=N), 1600, 1429 (C=C)	371 $[M+1]^+$ (0.03), 370 $[M]^+$ (0.03), 368 $[M-2]^+$ (0.04), 76 $[C_6H_4]^+$ (0.65), 59 (100.00)
10d	1.73-1.79 (m, 4H, $2CH_2$), 2.47-2.69 (m, 4H, $2CH_2$), 6.96 (s, 1H, CH), 6.97 (s, 2H, NH_2, D_2O exchange-able), 6.98-7.80 (m, 4H, C_6H_4), 11.45 (s, 1H, OH)	23.22, 23.42, 23.65, 24.51 ($4CH_2$ cyclohexene), 116.74, 119.45, 119.69, 121.00, 126.88, 130.45 (phenyl 6C), 130.98, 132.88, 134.46, 146.63, 158.53, 159.72, 159.93 (thiophene 4C, CH, thiazole C, C=N), 168.02 (C=O)	3431 (OH), 3273, 3225 (NH_2), 3079 (CH aromatic), 2935-2842 (CH, CH_2), 1695 (C=O), 1630 (C=N), 1599, 1442 (C=C)	358 $[M+2]^+$ (0.12), 357 $[M+1]^+$ (0.22), 356 $[M]^+$ (0.77), 76 $[C_6H_4]^+$ (1.30), 59 (100.00)

the cyclohexene ring, a singlet at δ 3.90 ppm for CH_2 and a singlet at δ 6.91 ppm for NH_2. In addition, the ^{13}C NMR spectrum revealed four signals at δ 21.68, 23.23, 23.43, 23.96 ppm for four CH_2 groups in cyclohexene, a signal at δ 38.93 ppm for the CH_2 thiazole moiety. Another four signals at δ 126.89, 130.45, 131.05, 146.64 ppm were for the thiophene ring, a signal at δ 162.67 ppm for thiazole C=N and a signal at δ 168.02 ppm for the C=O group. Also, compound **1** reacted with ethyl cyanoacetate in dimethylformamide to give the *N*-cyanoacetamido derivative. In addition, compound **1** reacted with phenyl isothiocyanate to give the *N*-phenylthiourea derivative **3**. ^1H NMR spectrum of compound **3** showed two multiplets, at δ 1.74–1.82 and 2.48–2.56 ppm, for the four CH_2 groups of the cyclohexene moiety, a singlet at δ 3.56 ppm for the thiazole CH_2 group, a multiplet at δ 6.90–7.51 ppm for the phenyl ring and two singlets at δ 9.76 and 11.48 ppm for two NH groups. Moreover, the mass spectrum revealed *m/z* at 388 $[M+1]^+$, *m/z* at 387 $[M]^+$ and *m/z* at 77 $[C_6H_5]^+$ for the phenyl moiety. Compound **3** reacted with benzenediazonium chloride in a basic ethanolic solution at 0–5 °C to give the phenylhydrazo derivative **4**.

Compound **1** reacted with bromine in an acetic acid solution to afford the 5-bromothiazole derivative **5**. In addition, compound **1** reacted with either ethyl cyanoacetate or malononitrile in 1,4-dioxane and in the presence of a catalytic amount of triethylamine to give the pyrano[2,3-*d*]thiazole-2-yl)benzo[*b*]thiophene derivatives **6a** and **6b**, respectively. Analytical and spectral data of com-

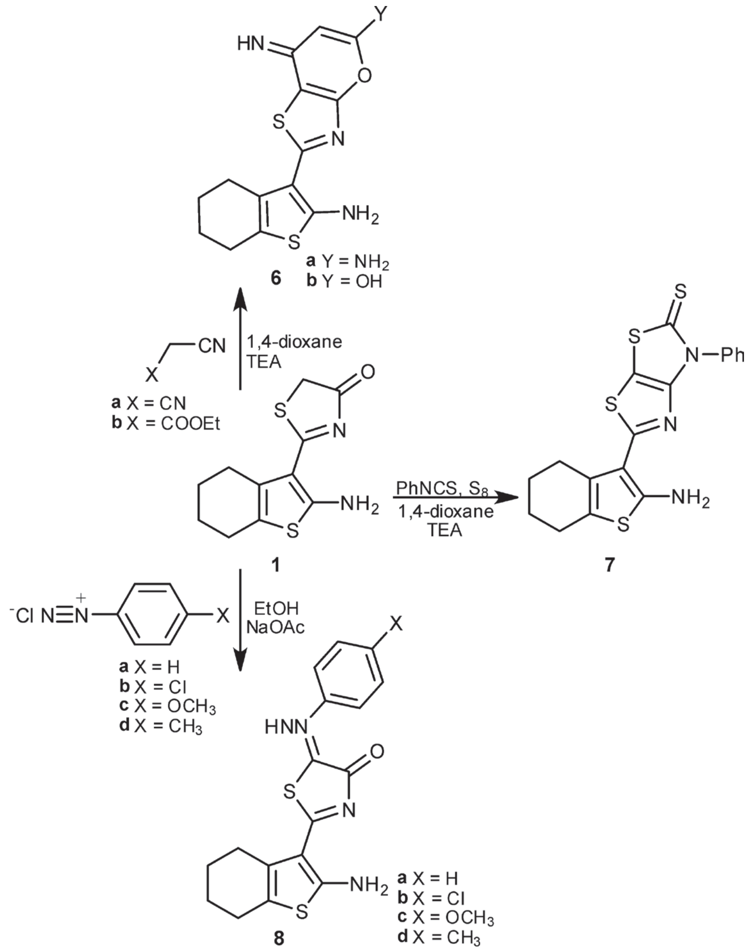


Scheme 1

pounds **6a**,**b** were consistent with their respective structures. Thus, the [1]H NMR spectrum of **6a** (as an example) showed two multiplets at *δ* 1.68-1.95 and 2.33-2.55 ppm for four CH_2 groups of the cyclohexene ring, a singlet at *δ* 6.92 ppm for pyran CH, two singlets at *δ* 7.09 and 7.25 ppm (D_2O exchangeable) for two NH_2 groups and a singlet at *δ* 11.46 ppm for the NH group. The [13]C NMR spectrum showed four signals at *δ* 22.90, 23.23, 23.43, 23.96 ppm for four CH_2 groups in the cyclohexene ring, a signal at *δ* 66.31 ppm for the pyran carbon moiety, four signals at *δ* 126.90, 130.46, 131.05, 140.00 ppm for the thiophene carbon ring, a signal at *δ* 146.63 ppm for the thiazole C=N and four signals at *δ* 152.00, 162.67, 164.00, 168.03 ppm for the pyran ring.

Further, compound **1** underwent the Hantzch reaction (1) through its reaction with elemental sulfur and phenylisothiocyanate to give the 3-phenylthiazolo[4,5-*d*]thiazole derivative **7**. The structure of the latter product was based on its analytical and spectral data (see experimental section and Tables I and II).

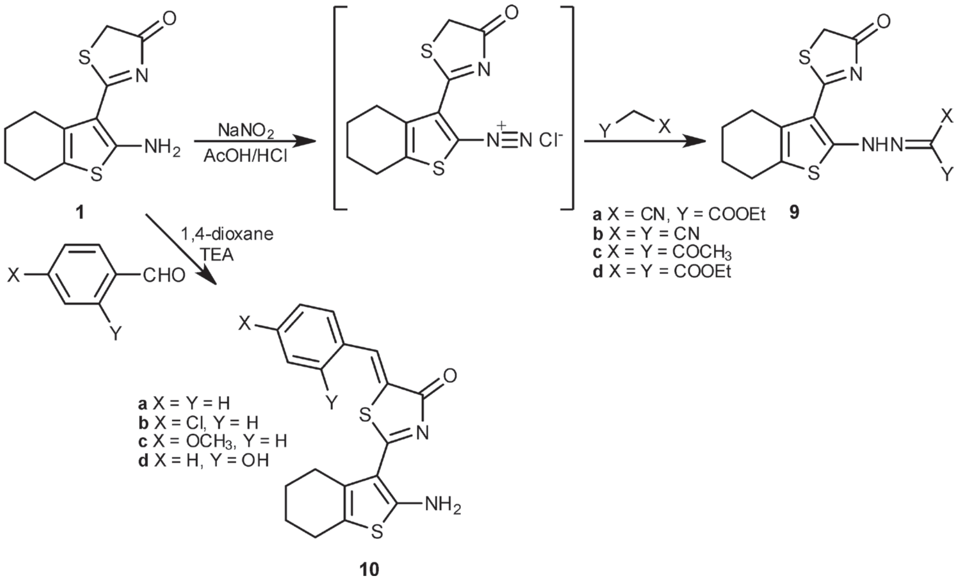
Compound **1** showed high reactivity towards diazonium salts; thus, it reacted with any of the diazonium salts, namely, benzenediazonium chloride, 4-chlorobenzenediazonium chloride, 4-methoxybenzenediazonium chloride or 4-methylbenzenediazonium



Scheme 2

chloride, at 0–5 °C to give the arylhydrazo derivatives **8a-d**, respectively. On the other hand, the 2-amino group present in the tetrahydrobenzo[*b*]thiophene underwent diazotization and coupling with active methylene reagents. A solution of compound **1** in acetic/hydrochloric acid, when treated with sodium nitrite, gave the intermediate diazonium salt. The latter was coupled with any of ethyl cyanoacetate, malononitrie, acetylacetone or ethyl acetoacetate to give the hydrazone derivatives **9a-d**, respectively. Finally, the reaction of compound **1** with any of benzaldehyde, 4-chlorobenzaldehyde, 4-methoxybenzaldehyde or salicylaldehyde afforded the corresponding arylidene derivatives **10a-d**, respectively.

Synthetic pathways are presented in Schemes 1-3 and physicochemical and spectral data of the synthesized compounds are given in Tables I and II. Cytotoxicity of the newly synthesized products is displayed in Table III.



Scheme 3

Evaluation of in vitro *cytotoxic activity*

The prepared heterocyclic compounds were tested against six human cancer cell lines, human gastric cancer (NUGC), human colon cancer (DLD-1), human liver cancer (HA22T and HEPG-2), human breast cancer (MCF-7) and nasopharyngeal carcinoma (HONE-1), as well as against normal fibroblast cells (WI-38).

The effects of the newly prepared compounds on six cancer cell lines are presented in Table III. Some heterocyclic compounds exerted marked cytotoxicity against most of the cancer cell lines tested (IC_{50} 20–100 nmol L^{-1}). Normal fibroblasts cells (WI-38) were affected to a much lesser extent (IC_{50} > 100 nmol L^{-1}). It was found that some compounds showed cytotoxicity even higher than the reference CHS-828. Thus, compounds **5**, **7**, **8a, 9b** and **10b** showed high cytotoxicity towards NUGC, **8a** being of comparable activity to the reference drug. Compounds **3**, **4**, **5**, **6b**, **8a**, **8b**, **8d**, **9a** and **9d** showed high cytotoxicity against DLD-1, with the most active **7**, **9b** and **10b** with IC_{50} markedly lower than that of CHS-828. The same applies to compounds **6a**, **9b** and **9c** against HA22T. Moreover, compounds **3**, **4**, **7**, **8a**, **8b**, **8d**, **9b**, **9d, 10a** and **10b** showed higher cytotoxicity against HEPG-2 than the reference drug. In the case of HONE-1, **8b** could be considered to be of comparable activity to CHS-828, while compounds **10c** and **8b** showed cytotoxicity comparable to CHS-828 against MCF-7.

Compound **8a** was the most active towards the NUGC cancer cell line, compound **10b** for DLD-1, compound **6a** for HA22T, compound **8b** for HEPG-2 and HONE-1 and finally compound **10c** for MCF-7, all compared to the standard reference CHS-828. It is important to mention that most of the newly synthesized products showed either no or low cytotoxicity towards the normal cell line WI-38.

Table III. Cytotoxicity of the newly synthesized products against six cancer cell lines and a normal fibroblast cell

Compd.	IC_{50} (nmol L^{-1})[a,b]						
	NUGC	DLD-1	HA22T	HEPG-2	HONE-1	MCF-7	WI-38
1	2101±86	2432±59	2358±80	1350±63	2180±58	1140±58	NA
2	3138±13	2366±14	2228±12	2130±69	1584±79	326±94	650±77
3	549±80	220±68	318±35	150±42	248±59	291±48	120±22
4	201±12	127±17	118±22	219±18	1170±22	1029±34	NA
5	38±18	163±38	120±68	3744±13	441±38	1264±64	860±59
6a	122±32	3210±96	59±22	1245±39	1140±60	1130±84	NA
6b	228±49	569±42	213±70	1112±59	2052±60	2011±84	632±55
7	48±16	55±12	128±80	128±42	248±59	128±77	838±48
8a	23±80	220±44	183±68	224±29	487±38	390±90	NA
8b	350±57	116±38	290±73	120±38	26±12	48±14	NA
8c	2116±21	2765±21	2838±17	3220±32	2440±24	2239±16	NA
8d	320±59	749±36	194±57	499±29	2871±17	840±68	NA
9a	537±75	440±38	1165±70	2766±12	6273±32	2533±21	419±78
9b	55±25	48±12	87±22	350±32	449±43	290±43	NA
9c	1135±76	2183±21	89±39	1220±49	2180±80	2120±69	NA
9d	302±67	143±94	173±48	392±66	80±55	284±44	NA
10a	1105±54	2460±17	2160±21	214±84	380±90	1086±29	NA
10b	80±22	24±18	160±53	284±79	130±68	73±42	872
10c	2265±60	2139±54	2257±73	2177±69	2250±12	18±80	262±52
10d	1232±69	1166±79	2225±94	2216±13	326±79	1286±87	NA
CHS-828	25±10	2315±13	2067±13	1245±69	15±60	18±70	NA

DLD-1 – colon cancer, HA22T – liver cancer, HEPG-2 – liver cancer, HONE-1 – nasopharyngeal carcinoma, MCF-7 – breast cancer, NA – not active, NUGC – gastric cancer, WI-38 – normal fibroblast cells

[a] Drug concentration required to inhibit tumor cell proliferation by 50 % after continuous exposure for 48 h; CHS-828 was used as positive control; DMSO 0.5 % negative control.

[b] Mean ± SEM, $n = 3$.

Structure activity relationship

It is clear from the results in Table III that the thiazole moiety was crucial for the cytotoxic effect of cyclic compounds **1-10a-d**. Compounds **3**, **4**, **5**, **6a**, **7**, **8a**, **8b, 8d**, **9b-d, 10b** and **10c** exhibited a marked cytotoxic effect against the different cancer cell lines with

IC_{50}'s in the nanomolar range. Compound **3** showed high cytotoxicity against the six cell lines and showed some activity against the normal cell line WI-38. Reactivity of compound **3** was attributed to the presence of thiazole together with the *N*-phenylthiourea moiety. On the other hand, compound **5** showed high cytotoxicity against NUGC, HA22T and DLD-1 cell lines with IC_{50} 38, 120 and 163 nmol L^{-1}. The presence of two cyano groups in compound **9b** was probably responsible for its higher activity compared to compounds **9a**, **9c** and **9d.** It is obvious that compound **10b** showed higher cytotoxicity than **10a**, **10c** and **10d** due to the presence of the Cl group. In conclusion, based on the presented data, the presence of an electron withdrawing group enhanced the potency of the compound.

CONCLUSIONS

Briefly, we have reported the synthetic strategies for the synthesis of new thiazole derivatives starting from 2-(2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl)thiazol-4(5*H*)-one (**1)**. The newly prepared compounds were studied for their anticancer activities on six human cancer cell lines and a normal human cell line. The data showed that the pyrano[2,3-*d*]thiazole derivative (**6a**), thiazolo[4,5-*d*]thiazole derivative (**7**), 2-(2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophen-3-yl)thiazole derivatives (**8a**,**b**, **10b**,**c**) and the 4,5,6,7-tetrahydrobenzo[*b*]thiophen-2-yl)carbonohydrazonoyl derivative (**9b**) were most active against all the tested cancer cell lines. Taking into account their non-toxicity towards the normal cell line, compounds **6a**, **8a**, **8b** and **9b** might be considered to be of potential therapeutic assistance.

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