

## Synthesis and antitumor activity of some novel thiophene, pyrimidine, coumarin, pyrazole and pyridine derivatives

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2-Cyano-*N*-(thiazol-2-yl) acetamide (**2a**) and 2-cyano-*N*-(oxazol-2-yl) acetamide (**2b**) were obtained *via* the reaction of ethyl cyanoacetate with either 2-aminothiazole (**1a**) or 2-aminoxazole (**1b**). The formed products were directed toward the reaction with cyclopentanone and elemental sulfur in the presence of triethylamine to give cyclopenta[*b*]thiophene derivatives (**3a,b**). The latter products were reacted with either ethyl cyanoacetate or malononitrile to form compounds **4a,b** and **5a,b**, respectively. Compounds **4a,b** were aimed at synthesizing some heterocyclic compounds; thus internal cyclization reactions were introduced to form compounds **6a,b**. Also, compounds **4a,b** reacted with salicylaldehyde, hydrazine derivatives and either urea or thiourea to produce coumarin derivatives (**7a,b**), pyrazole derivatives (**8a-d**) and pyrimidine derivatives (**9a-d**), respectively. Reaction of either benzaldehyde or benzene diazonium chloride (**11**) with compounds **4a,b** afforded compounds **10a,b** and **12a,b**, respectively. On the other hand, compounds **5a,b** underwent internal cyclization to form pyrimidine derivatives **13a,b**. Also, when compounds **5a,b** reacted with either ethyl cyanoacetate or malononitrile, they gave pyridine derivatives (**15a-d**) through the formation of intermediates (**14a-d**). Finally, formation of fused pyrimidine derivatives (**17a,b**) was achieved through the reaction of compounds **5a,b** and salicylaldehyde applying two different pathways. The first pathway used a catalytic amount of piperidine to form compounds **16a,b**; the latter products underwent cyclization to give compounds **17a,b**. The second pathway, using a catalytic amount of sodium ethoxide solution directly in one step, afforded compounds **17a,b**. Structures of the newly synthesized compounds were established using IR, <sup>1</sup>H NMR, <sup>13</sup>C NMR and mass spectrometry and their antitumor activity was investigated. Some of these compounds showed promising inhibitory effects on three different cell lines. However, fused pyrimidine acetonitrile derivatives **6a** and **6b** exerted the highest inhibitory effect, comparable to that of doxorubicin.

**Keywords:** thiophene, pyrimidine, coumarin, pyrazole, pyridine, antitumor activity

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Amino-thiophene derivatives were shown to be one of the most important groups of heterocyclic compounds with a wide spectrum of biological activities such as antitumor (1), antimetabolic (2) and antiviral (3), in addition to either thiazol-amide with fungicidal activity (4) or oxazol-amide with antimicrobial activity (5). Furthermore, thieno[2,3-*d*]pyrimidine derivatives show antibacterial (6) and antiproliferative activity (7). Also, chromene-3-carboxamide acts as an anti-*Helicobacter pylori* agent (8) and pyrazole derivatives have a specific effect with favorable antitumor activity (9). In addition, pyridine derivatives show anticonvulsant and anti-inflammatory (10), potential antitubercular (11) and anticonvulsant activity (12), among others.

In this article, we have described the synthesis of new heterocyclic compounds, thiophene, pyrimidine, coumarin, pyrazole and pyridine derivatives, in an attempt to improve the antitumor activity against three different cell lines: MCF-7 (breast adenocarcinoma), NCI-H460 (non-small cell lung cancer), and SF-268 (CNS cancer).

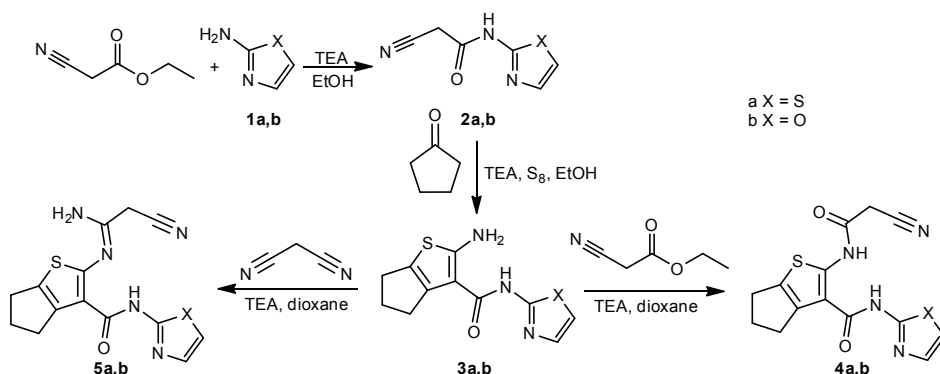
## EXPERIMENTAL

Melting points were determined in open capillaries and are not corrected. A Yanaco CHN CORDER MT-6 elemental analyzer (Japan) was used. IR spectra were recorded in KBr pellets using a PA-9721 IR spectrophotometer (Shimadzu, Japan). A Jeol 300 MHz (Japan) instrument was used to record the  $^1\text{H}$  NMR and  $^{13}\text{C}$  NMR spectra.  $\text{CD}_3\text{SOCD}_3$  was used as a solvent and TMS as internal standard. Chemical shifts were expressed as  $\delta$  (ppm). Kratos (75 eV) MS equipment (Germany) was used for mass spectra recording.

The synthetic pathways used are represented in Schemes 1–4 while the physicochemical and spectral data of the newly synthesized compounds are given in Tables I and II.

### Syntheses

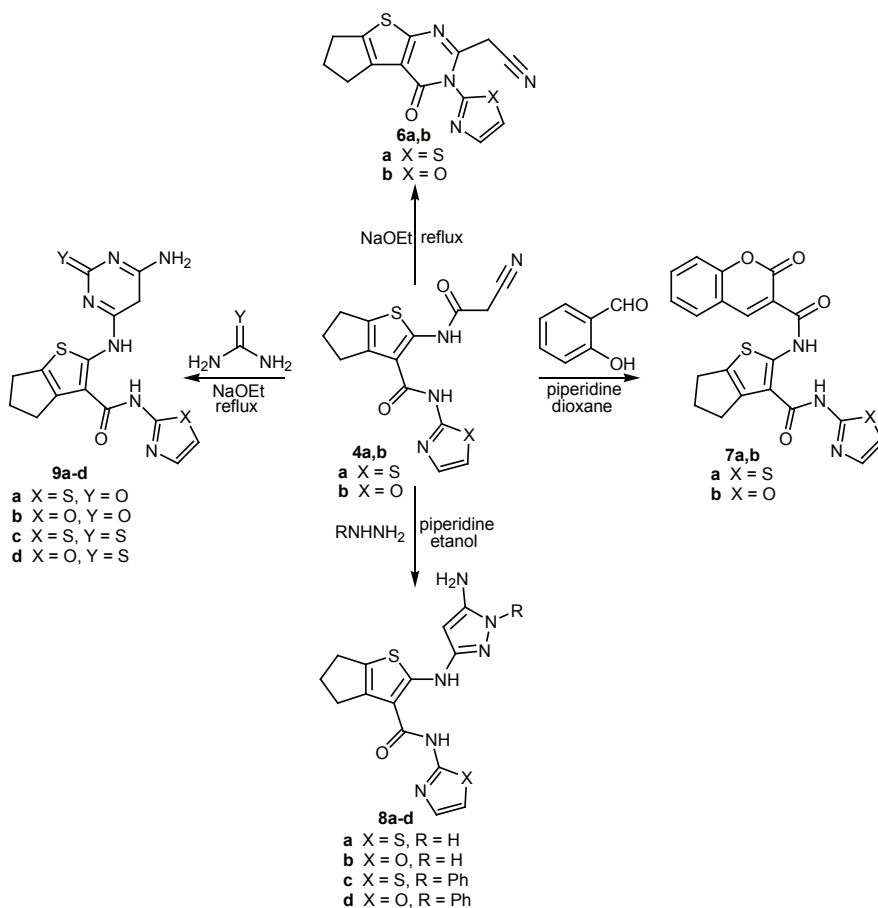
2-Cyano-N-(thiazol-2-yl)acetamide (**2a**) and 2-cyano-N-(oxazol-2-yl)acetamide (**2b**). – To a solution of either 2-aminothiazole (**1a**) (5.007 g, 0.05 mol) or 2-aminoxazole (**1b**) (4.204 g, 0.05 mol) in 50 mL of absolute ethanol containing triethylamine (0.5 mL) as a catalyst,



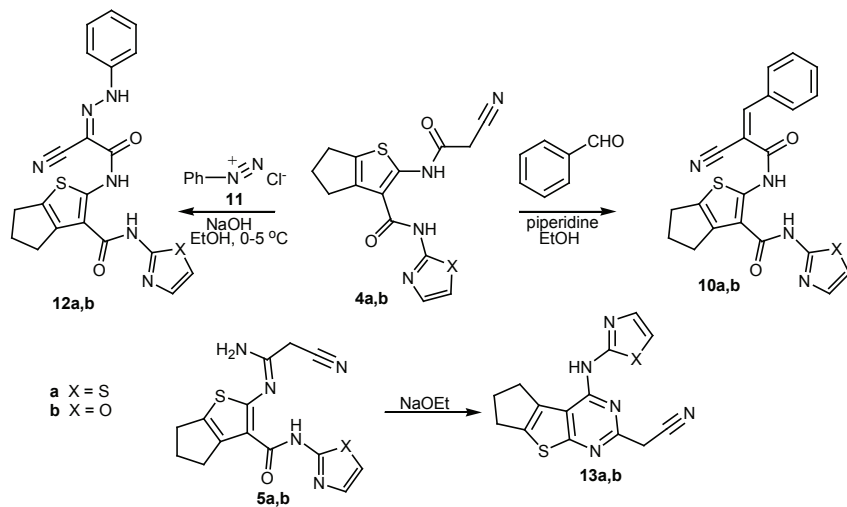
Scheme 1

ethyl cyanoacetate (5.656 g, 0.05 mol) was added and the reaction mixture was heated under reflux for 3 h, cooled and poured onto ice. Conc. HCl (a few drops) was added. The formed precipitate was filtered out and recrystallized from 1,4 dioxane to afford the desired product.

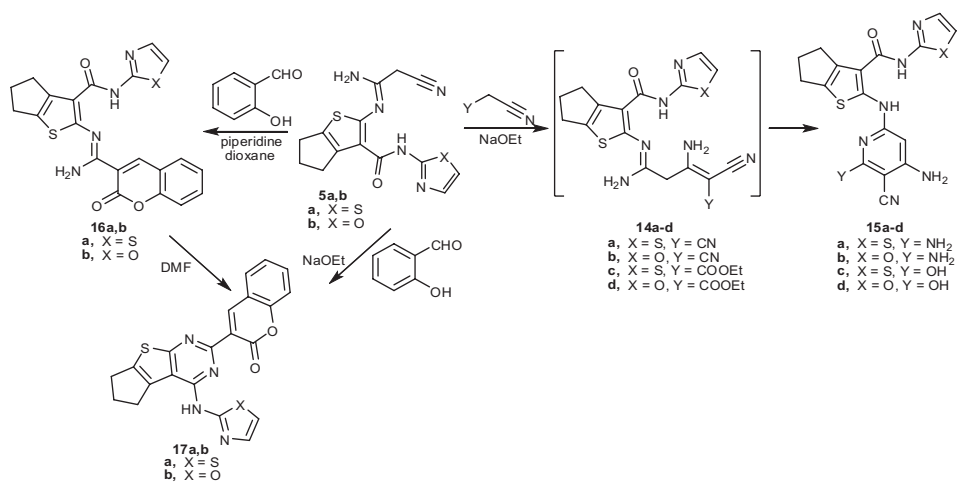
2-Amino-N-(thiazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**3a**) and 2-amino-N-(oxazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**3b**). – To a solution of either compound **2a** (5.016 g, 0.03 mol) or **2b** (4.534 g, 0.03 mol) in 50 mL of absolute EtOH containing triethylamine (1.0 mL), cyclopentanone (2.524g, 0.03 mol) and elemental sulfur (0.96 g, 0.03 mol) were added. The whole reaction mixture was heated under reflux for 2 h, then poured into an ice/water mixture containing a few drops of concentrated hydrochloric acid and the formed solid product, in each case, was collected by filtration and recrystallized from absolute ethanol.



Scheme 2



Scheme 3



Scheme 4

2-(2-Cyanoacetamido)-N-(thiazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**4a**) and 2-(2-cyanoacetamido)-N-(oxazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**4b**). – To a solution of either compound **3a** (5.307 g, 0.02 mol) or **3b** (4.986 g, 0.02 mol) in 1,4-dioxane (50 mL) containing triethylamine (0.5 mL), ethyl cyanoacetate (2.262 g, 0.02 mol) was added. The resulting reaction mixture was heated under reflux for 3 h, cooled and poured into an ice/water mixture with a few drops of conc. HCl added. The precipitate that formed was filtered and recrystallized from absolute ethanol.

Table I. Physicochemical data for synthesized compounds

Compd.	Mol. formula ( $M_r$ )	M. p. (°C)	Yield (%)	Calcd./found (%)			
				C	H	N	S
<b>2a</b>	C <sub>6</sub> H <sub>5</sub> N <sub>3</sub> OS	229–231	74	43.10	3.01	25.13	19.18
	167.19			43.32	3.23	24.98	18.96
<b>2b</b>	C <sub>6</sub> H <sub>5</sub> N <sub>3</sub> O <sub>2</sub>	201–202	70.5	47.69	3.33	27.81	–
	151.12			47.85	3.11	27.56	–
<b>3a</b>	C <sub>11</sub> H <sub>11</sub> N <sub>3</sub> OS <sub>2</sub>	169–171	81	49.79	4.18	15.84	24.17
	265.35			49.55	4.02	15.99	24.39
<b>3b</b>	C <sub>11</sub> H <sub>11</sub> N <sub>3</sub> O <sub>2</sub> S	209–211	77	53.00	4.45	16.86	12.86
	249.29			53.25	4.22	16.57	12.70
<b>4a</b>	C <sub>14</sub> H <sub>12</sub> N <sub>4</sub> O <sub>2</sub> S <sub>2</sub>	242–244	66	50.59	3.64	16.86	19.29
	332.4			50.40	3.44	16.63	19.13
<b>4b</b>	C <sub>14</sub> H <sub>12</sub> N <sub>4</sub> O <sub>3</sub> S	186–188	71	53.16	3.82	17.71	10.14
	316.34			53.41	3.59	17.50	9.88
<b>5a</b>	C <sub>14</sub> H <sub>13</sub> N <sub>5</sub> OS <sub>2</sub>	195–196	73	50.74	3.95	21.13	19.35
	331.42			50.96	3.66	21.35	19.17
<b>5b</b>	C <sub>14</sub> H <sub>13</sub> N <sub>5</sub> O <sub>2</sub> S	158–160	61	53.32	4.16	22.21	10.17
	315.35			53.15	3.97	22.49	10.47
<b>6a</b>	C <sub>14</sub> H <sub>10</sub> N <sub>4</sub> OS <sub>2</sub>	211–213	76	53.49	3.21	17.82	20.40
	314.39			53.77	3.45	17.55	20.16
<b>6b</b>	C <sub>14</sub> H <sub>10</sub> N <sub>4</sub> O <sub>2</sub> S	227–229	71	56.37	3.38	18.78	10.75
	298.32			56.58	3.52	18.99	10.46
<b>7a</b>	C <sub>21</sub> H <sub>15</sub> N <sub>3</sub> O <sub>4</sub> S <sub>2</sub>	171–173	65	57.65	3.46	9.60	14.66
	437.49			57.95	3.19	9.83	14.91
<b>7b</b>	C <sub>21</sub> H <sub>15</sub> N <sub>3</sub> O <sub>5</sub> S	150–151	61	59.85	3.59	9.97	7.61
	421.43			60.12	3.35	9.73	7.91
<b>8a</b>	C <sub>14</sub> H <sub>14</sub> N <sub>6</sub> OS <sub>2</sub>	123–125	62	48.54	4.07	24.26	18.51
	346.43			48.75	3.85	24.02	18.23
<b>8b</b>	C <sub>14</sub> H <sub>14</sub> N <sub>6</sub> O <sub>2</sub> S	155–157	58	50.90	4.27	25.44	9.71
	330.36			51.13	4.49	25.67	9.45
<b>8c</b>	C <sub>20</sub> H <sub>18</sub> N <sub>6</sub> OS <sub>2</sub>	178–180	55	56.58	4.29	19.89	15.18
	422.53			56.29	4.04	19.65	15.34
<b>8d</b>	C <sub>20</sub> H <sub>18</sub> N <sub>6</sub> O <sub>2</sub> S	218–220	51	59.10	4.46	20.68	7.89
	406.46			59.38	4.21	20.95	7.61

<b>9a</b>	C <sub>15</sub> H <sub>14</sub> N <sub>6</sub> O <sub>2</sub> S <sub>2</sub>	190–191	52	48.11	3.77	22.44	17.13
	374.44			48.36	3.53	22.33	17.36
<b>9b</b>	C <sub>15</sub> H <sub>14</sub> N <sub>6</sub> O <sub>3</sub> S	163–165	48	50.27	3.94	23.45	8.95
	358.38			50.54	3.68	23.16	8.66
<b>9c</b>	C <sub>15</sub> H <sub>14</sub> N <sub>6</sub> OS <sub>3</sub>	140–142	57	46.14	3.61	21.52	24.63
	390.51			46.44	3.90	21.23	24.40
<b>9d</b>	C <sub>15</sub> H <sub>14</sub> N <sub>6</sub> O <sub>2</sub> S <sub>2</sub>	221–223	55	48.11	3.77	22.44	17.13
	374.44			48.34	3.99	22.16	17.42
<b>10a</b>	C <sub>21</sub> H <sub>16</sub> N <sub>4</sub> O <sub>2</sub> S <sub>2</sub>	148–150	59	59.98	3.84	13.32	15.25
	420.51			60.24	3.56	13.12	15.42
<b>10b</b>	C <sub>21</sub> H <sub>16</sub> N <sub>4</sub> O <sub>3</sub> S	112–114	53	62.36	3.99	13.85	7.93
	404.44			62.18	3.68	13.61	8.15
<b>12a</b>	C <sub>20</sub> H <sub>16</sub> N <sub>6</sub> O <sub>2</sub> S <sub>2</sub>	131–133	64	55.03	3.69	19.25	14.69
	436.51			55.29	3.39	19.02	14.41
<b>12b</b>	C <sub>20</sub> H <sub>16</sub> N <sub>6</sub> O <sub>3</sub> S	110–111	55	57.13	3.84	19.99	7.63
	420.44			57.41	3.99	19.71	7.42
<b>13a</b>	C <sub>14</sub> H <sub>11</sub> N <sub>5</sub> S <sub>2</sub>	237–239	66	53.65	3.54	22.35	20.46
	313.40			53.35	3.32	22.62	20.19
<b>13b</b>	C <sub>14</sub> H <sub>11</sub> N <sub>5</sub> OS	217–219	60	56.55	3.73	23.55	10.78
	297.34			56.83	3.96	23.31	10.53
<b>15a</b>	C <sub>17</sub> H <sub>15</sub> N <sub>7</sub> OS <sub>2</sub>	215–217	56	51.37	3.80	24.67	16.13
	397.48			51.58	3.61	24.49	16.34
<b>15b</b>	C <sub>17</sub> H <sub>15</sub> N <sub>7</sub> O <sub>2</sub> S	222–224	51	53.53	3.96	25.71	8.41
	381.41			53.28	3.69	25.51	8.23
<b>15c</b>	C <sub>17</sub> H <sub>14</sub> N <sub>6</sub> O <sub>2</sub> S <sub>2</sub>	230–232	53	51.24	3.54	21.09	16.09
	398.46			51.03	3.28	20.87	16.32
<b>15d</b>	C <sub>17</sub> H <sub>14</sub> N <sub>6</sub> O <sub>3</sub> S	300 <	48	53.40	3.69	21.98	8.39
	382.40			53.63	3.91	21.69	8.21
<b>16a</b>	C <sub>21</sub> H <sub>16</sub> N <sub>4</sub> O <sub>3</sub> S <sub>2</sub>	105–107	63	57.78	3.69	12.84	14.69
	436.51			57.52	3.44	13.03	14.45
<b>16b</b>	C <sub>21</sub> H <sub>16</sub> N <sub>4</sub> O <sub>4</sub> S	89–91	66	59.99	3.84	13.33	7.63
	420.44			59.78	3.99	13.55	7.48
<b>17a</b>	C <sub>21</sub> H <sub>14</sub> N <sub>4</sub> O <sub>2</sub> S <sub>2</sub>	144–145	55	60.27	3.37	13.39	15.32
	418.89			59.98	3.59	13.14	15.12
<b>17b</b>	C <sub>21</sub> H <sub>14</sub> N <sub>4</sub> O <sub>3</sub> S	181–183	62	62.68	3.51	13.92	7.97
	402.43			62.95	3.28	13.74	7.67

Table II. Spectral data for synthesized compounds

Compd.	MS ( <i>m/z</i> )	<sup>13</sup> C NMR (δ, ppm)	<sup>1</sup> H NMR (δ, ppm)	IR (ν, cm <sup>-1</sup> )
<b>2a</b>	167	34.9 (CH <sub>2</sub> ), 119.4 (CN), 130.4, 131.1, 133.7 (thiazole C), 171.2 (C=O)	3.12-3.18 (s, 2H, CH <sub>2</sub> ) 6.53-6.64 (m, 2H, thiazole ring), 8.76 (s, 1H, NH, D <sub>2</sub> O-exchangeable)	3455-3380 (NH), 2891 (CH <sub>2</sub> ), 2225 (CN), 1666 (CO), 1653 (C=N)
<b>2b</b>	151	37.6 (CH <sub>2</sub> ), 115.2 (CN), 129.1, 132.7, 136.2 (oxazole C), 168.3 (C=O)	3.24-3.31 (s, 2H, CH <sub>2</sub> ), 6.44-6.51 (m, 2H, oxazole ring), 8.57 (s, 1H, NH)	3445-3395 (NH), 2884 (CH <sub>2</sub> ), 2223 (CN), 1661 (CO), 1652 (C=N)
<b>3a</b>	265	28.8, 29.9, 32.1 (3CH <sub>2</sub> ), 128.5, 129.4, 132.9, 133.5, 136.4 (thiophene C and thiazole C), 168.2 (C=O)	2.06-2.12 (m, 6H, 3CH <sub>2</sub> ), 4.23 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.38-6.48 (m, 2H, thiazole ring), 8.73 (s, 1H, D <sub>2</sub> O-exchangeable, NH)	3443-3325 (NH <sub>2</sub> , NH), 2874 (CH <sub>2</sub> ), 1662 (CO), 1656 (C=N)
<b>3b</b>	249	27.7, 29.8, 31.3 (3CH <sub>2</sub> ), 130.4, 131.1, 133.7, 135.6, 137.2 (thiophene C and oxazole C), 166.5 (C=O)	1.98-2.05 (m, 6H, 3CH <sub>2</sub> ), 4.39 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.44-6.51 (m, 2H, oxazole ring), 8.54 (s, 1H, D <sub>2</sub> O-exchangeable, NH)	3466-3232 (NH <sub>2</sub> , NH), 2878 (CH <sub>2</sub> ), 1660 (CO), 1652 (C=N)
<b>4a</b>	332	22.2, 26.7, 31.7, 40.8 (4 CH <sub>2</sub> ), 118.8 (CN), 124.3, 125.6, 126.9, 129.2, 134.5, 140.9, 143.2 (thiophene C, thiazole C), 172.3, 175.6 (2C=O)	2.11-2.19 (m, 6H, 3CH <sub>2</sub> ), 3.19-3.27 (s, 2H, CH <sub>2</sub> ), 6.55-6.61 (m, 2H, thiazole ring), 8.36, 8.68 (2s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3444-3431 (2NH), 2877 (CH <sub>2</sub> ), 2224 (CN), 1669, 1664 (2C=O), 1660 (C=N), 1647 (C=C)
<b>4b</b>	316	21.4, 26.5, 29.7, 38.8 (4 CH <sub>2</sub> ), 121.5 (CN), 122.4, 125.9, 127.7, 129.7, 133.8, 141.3, 145.1 (thiophene C, oxazole C), 168.3, 171.4 (2C=O)	2.07-2.12 (m, 6H, 3CH <sub>2</sub> ), 3.13-3.22 (s, 2H, CH <sub>2</sub> ), 6.53-6.67 (m, 2H, oxazole ring), 8.49, 8.83 (2s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3462-3449 (2NH), 2881 (CH <sub>2</sub> ), 2221 (CN), 1666, 1662 (2C=O), 1658 (C=N), 1649 (C=C)
<b>5a</b>	331	21.9, 24.5, 28.4, 36.3 (4CH <sub>2</sub> ), 120.9 (CN), 122.1, 124.8, 126.3, 129.9, 132.9, 142.7, 144.7 (thiophene C, thiazole C), 165.9 (C=N), 176.2 (C=O)	1.87-1.98 (m, 6H, 3CH <sub>2</sub> ), 3.04-3.11 (s, 2H, CH <sub>2</sub> ), 4.49 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.61-6.69 (m, 2H, thiazole ring), 8.52 (s, 1H, D <sub>2</sub> O-exchangeable, NH)	3463-3428 (NH <sub>2</sub> , NH), 2880 (CH <sub>2</sub> ), 2225 (CN), 1667 (C=O), 1660, 1655-1650 (2C=N), 1642 (C=C)
<b>5b</b>	315	24.6, 26.1, 28.3, 34.4 (4CH <sub>2</sub> ), 121.2 (CN), 122.7, 124.4, 127.8, 129.7, 132.2, 141.1, 142.8 (thiophene C, oxazole C), 167.6 (C=N), 172.7 (C=O)	1.96-2.05 (m, 6H, 3CH <sub>2</sub> ), 3.02-3.08 (s, 2H, CH <sub>2</sub> ), 4.58 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.73-6.84 (m, 2H, oxazole ring), 8.83 (s, 1H, D <sub>2</sub> O-exchangeable, NH)	3427-3386 (NH <sub>2</sub> , NH), 2865 (CH <sub>2</sub> ), 2227 (CN), 1669 (C=O), 1660, 1653 (2C=N), 1645 (C=C)

Compd.	MS ( <i>m/z</i> )	<sup>13</sup> CNMR (δ, ppm)	<sup>1</sup> H NMR (δ, ppm)	IR (ν, cm <sup>-1</sup> )
<b>6a</b>	314	20.7, 22.9, 27.1, 40.2 (4CH <sub>2</sub> ), 123.3 (CN), 124.3, 126.2, 128.1, 130.6, 133.8, 141.5, 145.9, 147.1 (thiophene C, thiazole C, pyrimidine C), 179.2 (C=O)	1.96–2.08 (m, 6H, 3CH <sub>2</sub> ), 3.08–3.12 (s, 2H, CH <sub>2</sub> ), 6.62–6.73 (m, 2H, thiazole ring)	2883 (CH <sub>2</sub> ), 2226 (CN), 1664 (C=O), 1662, 1657 (2C=N), 1651 (C=C)
<b>6b</b>	298	23.1, 25.2, 27.8, 37.2 (4CH <sub>2</sub> ), 127.1 (CN), 123.9, 125.2, 129.0, 131.5, 133.7, 142.1, 144.8, 147.4 (thiophene C, oxazole C, pyrimidine C), 178.1 (C=O)	1.93–2.11 (m, 6H, 3CH <sub>2</sub> ), 3.18–3.24 (s, 2H, CH <sub>2</sub> ), 6.54–6.66 (m, 2H, oxazole ring)	2891 (CH <sub>2</sub> ), 2223 (CN), 1669 (C=O), 1660, 1655 (2C=N), 1647 (C=C)
<b>7a</b>	437	22.7, 25.8, 29.3, (3CH <sub>2</sub> ), 122.7, 125.5, 128.7, 130.2, 132.4, 135.4, 138.1, 141.8, 144.2, 147.3, 149.5 (thiophene C, thiazole C, coumarin C), 175.6, 178.1, 182.9 (3C=O)	1.79–1.86 (m, 6H, 3CH <sub>2</sub> ), 6.65–6.75 (m, 2H, thiazole ring), 6.88 (s, 1H, coumarin H-4), 7.34–7.49 (m, 4H, C <sub>6</sub> H <sub>4</sub> ), 8.39, 8.56 (2s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3455–3334 (2NH), 3056 (CH-aromatic), 2882 (CH <sub>2</sub> ), 1802, 1669, 1663 (β CO), 1640 (C=C)
<b>7b</b>	421	20.5, 23.7, 28.2, (3CH <sub>2</sub> ), 123.7, 126.5, 127.4, 131.4, 132.8, 134.3, 139.1, 142.9, 145.7, 147.9, 149.9 (thiophene C, oxazole C, coumarin C), 177.2, 178.9, 182.6 (3C=O)	1.88–1.98 (m, 6H, 3CH <sub>2</sub> ), 6.64–6.78 (m, 2H, thiazole ring), 6.91 (s, 1H, coumarin H-4), 7.21–7.54 (m, 4H, C <sub>6</sub> H <sub>4</sub> ), 8.34, 8.69 (2s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3448–3326 (2NH), 3053 (CH-aromatic), 2887 (CH <sub>2</sub> ), 1796, 1666, 1661 (β CO), 1645 (C=C)
<b>8a</b>	346	22.7, 25.9, 31.4 (3CH <sub>2</sub> ), 128.1, 130.7, 132.6, 134.1, 137.3, 141.2, 144.5, 147.3 (thiophene C, thiazole C, pyrazole C), 169.8 (C=O)	1.95–2.05 (m, 6H, 3CH <sub>2</sub> ), 4.39 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.44–6.49 (m, 2H, thiazole ring, 1H, pyrazole ring), 8.67, 8.79, 8.95 (s, 3H, D <sub>2</sub> O-exchangeable, 3NH)	3468–3355 (NH <sub>2</sub> , 3NH), 2881 (CH <sub>2</sub> ), 1669 (CO), 1658 (C=N), 1647 (C=C)
<b>8b</b>	330	23.9, 25.4, 32.7 (3CH <sub>2</sub> ), 129.3, 130.9, 133.5, 134.8, 136.5, 142.1, 143.3, 146.7 (thiophene C, oxazole C, pyrazole C), 166.5 (C=O)	2.09–2.17 (m, 6H, 3CH <sub>2</sub> ), 4.47 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.53–6.67 (m, 2H, oxazole ring, 1H, pyrazole ring), 8.38, 8.59, 8.77 (s, 3H, D <sub>2</sub> O-exchangeable, 3NH)	3423–3363 (NH <sub>2</sub> , 3NH), 2888 (CH <sub>2</sub> ), 1663 (CO), 1657 (C=N), 1644 (C=C)
<b>8c</b>	422	20.6, 23.2, 27.3 (3CH <sub>2</sub> ), 126.3, 128.3, 129.1, 130.6, 132.3, 134.7, 136.7, 138.6, 140.5, 143.1, 147.5, 148.7, 149.8 (thiophene C, thiazole C, pyrazole C, C <sub>6</sub> H <sub>5</sub> ), 165.3 (C=O)	1.91–2.11 (m, 6H, 3CH <sub>2</sub> ), 4.51 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.49–6.63 (m, 2H, thiazole ring, 1H, pyrazole ring), 7.32–7.45 (m, 5H, C <sub>6</sub> H <sub>5</sub> ), 8.55, 8.73 (s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3477–3349 (NH <sub>2</sub> , 2NH), 3048 (CH aromatic), 2872 (CH <sub>2</sub> ), 1664 (CO), 1653 (C=N), 1649 (C=C)



Compd.	MS ( <i>m/z</i> )	<sup>13</sup> CNMR (δ, ppm)	<sup>1</sup> H NMR (δ, ppm)	IR (ν, cm <sup>-1</sup> )
<b>8d</b>	406	214.4, 22.9, 25.4 (3CH <sub>2</sub> ), 125.1, 127.5, 128.3, 130.9, 132.7, 135.1, 136.9, 138.3, 141.3, 143.6, 146.7, 148.9, 150.3 (thiophene C, oxazole C, pyrazole C, C <sub>6</sub> H <sub>5</sub> ), 166.2 (C=O)	2.07-2.16 (m, 6H, 3CH <sub>2</sub> ), 4.61 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.42-6.61 (m, 2H, oxazole ring, 1H, pyrazole ring), 7.28-7.43 (m, 5H, C <sub>6</sub> H <sub>5</sub> ), 8.44, 8.68 (s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3470-3351 (NH <sub>2</sub> , 2NH), 3052 (CH aromatic), 2881 (CH <sub>2</sub> ), 1669 (CO), 1655 (C=N), 1650 (C=C)
<b>9a</b>	374	23.4, 25.2, 29.5, 65.3 (3CH <sub>2</sub> , CH <sub>2</sub> pyrimidine ring), 125.5, 130.2, 132.9, 133.4, 136.7, 140.4, 143.7 (thiophene C, thiazole C, pyrimidine C), 166.6, 168.8 (2C=O)	1.83-1.94 (m, 6H, 3CH <sub>2</sub> ), 3.96 (s, 2H, pyrimidine ring), 4.51 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.61-6.68 (m, 2H, thiazole ring), 8.53, 8.71 (s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3433-3369 (NH <sub>2</sub> , 2NH), 2877 (CH <sub>2</sub> ), 1669, 1663 (2CO), 1655 (C=N), 1643 (C=C)
<b>9b</b>	358	22.5, 25.7, 27.6, 57.7 (3CH <sub>2</sub> , CH <sub>2</sub> pyrimidine ring), 124.6, 128.4, 131.7, 133.5, 136.8, 141.2, 143.9 (thiophene C, oxazole C, pyrimidine C), 164.5, 167.3 (2C=O)	1.93-2.07 (m, 6H, 3CH <sub>2</sub> ), 3.86 (s, 2H, pyrimidine ring), 4.42 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.69-6.76 (m, 2H, oxazole ring), 8.34, 8.58 (s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3422-3345 (NH <sub>2</sub> , 2NH), 2872 (CH <sub>2</sub> ), 1665, 1660 (2CO), 1651 (C=N), 1642 (C=C)
<b>9c</b>	390	24.1, 25.9, 27.5, 55.2 (3CH <sub>2</sub> , CH <sub>2</sub> pyrimidine ring), 123.5, 125.3, 128.5, 132.4, 135.7, 141.7, 143.5 (thiophene C, thiazole C, pyrimidine C), 165.3 (C=S), 175.6 (C=O)	1.77-1.86 (m, 6H, 3CH <sub>2</sub> ), 3.92 (s, 2H, pyrimidine ring), 4.54 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.61-6.73 (m, 2H, thiazole ring), 8.66, 8.92 (2s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3438-3355 (NH <sub>2</sub> , 2NH), 2883 (CH <sub>2</sub> ), 1667 (CO), 1653 (C=N), 1647 (C=C)
<b>9d</b>	374	25.2, 26.8, 27.9, 54.1 (3CH <sub>2</sub> , CH <sub>2</sub> pyrimidine ring), 119.9, 122.4, 125.7, 132.9, 134.2, 141.9, 143.6 (thiophene C, oxazole C, pyrimidine C), 157.7 (C=S), 169.8 (C=O)	1.87-1.99 (m, 6H, 3CH <sub>2</sub> ), 3.83 (s, 2H, pyrimidine ring), 4.47 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.52-6.62 (m, 2H, oxazole ring), 8.52, 8.86 (2s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3425-3341 (NH <sub>2</sub> , 2NH), 2880 (CH <sub>2</sub> ), 1666 (CO), 1652 (C=N), 1644 (C=C)
<b>10a</b>	420	24.1, 26.7, 29.2 (3CH <sub>2</sub> ), 109.8 (=CH), 111.3 (=C), 114.4 (CN), 120.3, 122.7, 124.6, 128.9, 131.4, 133.2, 135.4, 136.9, 138.7 (thiophene C, thiazole C, C <sub>6</sub> H <sub>5</sub> ), 166.8, 168.5 (2C=O)	1.83-1.96 (m, 6H, 3CH <sub>2</sub> ), 5.18 (s, 1H, =CH), 6.33-6.58 (m, 2H, thiazole ring), 7.35-7.48 (m, 5H, C <sub>6</sub> H <sub>5</sub> ), 8.37, 8.52 (s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3412-3388 (2NH), 3057 (CH aromatic), 2885 (CH <sub>2</sub> ), 2226 (CN), 1666, 1663 (2CO), 1654 (C=N), 1647 (C=C)

Compd.	MS ( <i>m/z</i> )	<sup>13</sup> CNMR (δ, ppm)	<sup>1</sup> H NMR (δ, ppm)	IR (ν, cm <sup>-1</sup> )
<b>10b</b>	404	20.7, 24.7, 28.1 (3CH <sub>2</sub> ), 108.9 (=CH), 114.3 (=C), 116.2 (CN), 121.2, 122.9, 125.4, 128.3, 132.6, 134.5, 135.9, 136.8, 139.5 (thiophene C, oxazole C, C <sub>6</sub> H <sub>5</sub> ), 164.3, 167.6 (2C=O)	1.78–1.87 (m, 6H, 3CH <sub>2</sub> ), 5.11 (s, 1H, =CH), 6.19–6.25 (m, 2H, oxazole ring), 7.39–7.57 (m, 5H, C <sub>6</sub> H <sub>5</sub> ), 8.31, 8.63 (s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3421–3376 (2NH), 3055 (CH aromatic), 2883 (CH <sub>2</sub> ), 2224 (CN), 1669, 1665 (2CO), 1653 (C=N), 1648 (C=C)
<b>12a</b>	436	27.1, 29.4, 30.8 (3CH <sub>2</sub> ), 114.5 (C=N), 118.1 (CN), 120.9, 123.8, 125.7, 128.3, 130.6, 132.2, 134.8, 136.7 (thiophene C, thiazole C, C <sub>6</sub> H <sub>5</sub> ), 163.9, 166.8 (2C=O)	1.97–2.12 (m, 6H, 3CH <sub>2</sub> ), 6.30–6.44 (m, 2H, thiazole ring), 7.22–7.37 (m, 5H, C <sub>6</sub> H <sub>5</sub> ), 8.37, 8.52, 8.77 (3s, 3H, D <sub>2</sub> O-exchangeable, 3NH)	3428–3391 (3NH), 3052 (CH aromatic), 2881 (CH <sub>2</sub> ), 2224 (CN), 1667, 1664 (2CO), 1659 (C=N), 1645 (C=C)
<b>12b</b>	420	24.7, 27.2, 31.2 (3CH <sub>2</sub> ), 112.6 (C=N), 119.2 (CN), 121.5, 124.7, 125.9, 127.4, 129.5, 131.4, 134.5, 136.2 (thiophene C, oxazole C, C <sub>6</sub> H <sub>5</sub> ), 160.6, 164.1 (2C=O)	2.07–2.18 (m, 6H, 3CH <sub>2</sub> ), 6.17–6.28 (m, 2H, oxazole ring), 7.33–7.45 (m, 5H, C <sub>6</sub> H <sub>5</sub> ), 8.28, 8.43, 8.69 (3s, 3H, D <sub>2</sub> O-exchangeable, 3NH)	3398–3349 (3NH), 3050 (CH aromatic), 2883 (CH <sub>2</sub> ), 2225 (CN), 1669, 1662 (2CO), 1657 (C=N), 1641 (C=C)
<b>13a</b>	313	21.2, 22.8, 25.4, 38.2 (4CH <sub>2</sub> ), 121.4 (CN), 123.1, 125.4, 127.3, 130.4, 132.9, 138.6, 143.3, 146.4 (thiophene C, thiazole C, pyrimidine C).	1.76–1.85 (m, 6H, 3CH <sub>2</sub> ), 3.12–3.19 (s, 2H, CH <sub>2</sub> ), 6.51–6.63 (m, 2H, thiazole ring), 8.11 (s, 1H, D <sub>2</sub> O-exchangeable, NH)	3343–3312 (NH), 2886 (CH <sub>2</sub> ), 2223 (CN), 1660, 1655 (2C=N), 1651 (C=C)
<b>13b</b>	297	22.3, 24.5, 25.8, 36.7 (4CH <sub>2</sub> ), 123.4 (CN), 125.1, 126.7, 127.9, 131.7, 133.8, 136.5, 140.3, 144.2 (thiophene C, oxazole C, pyrimidine C)	1.88–1.97 (m, 6H, 3CH <sub>2</sub> ), 3.14–3.21 (s, 2H, CH <sub>2</sub> ), 6.37–6.45 (m, 2H, oxazole ring), 8.06 (s, 1H, D <sub>2</sub> O-exchangeable, NH)	3340–3308 (NH), 2883 (CH <sub>2</sub> ), 2226 (CN), 1662, 1654 (2C=N), 1648 (C=C)
<b>15a</b>	397	19.7, 22.2, 26.4 (3CH <sub>2</sub> ), 122.4, 126.9, 131.2, 134.5, 137.9, 142.3, 144.3, 146.4, 148.2 (thiophene C, thiazole C, pyridine C), 166.7 (C=O)	1.81–1.93 (m, 6H, 3CH <sub>2</sub> ), 4.33, 4.52 (2s, 4H, D <sub>2</sub> O-exchangeable, 2NH <sub>2</sub> ), 6.61–6.78 (m, 2H, thiazole ring, 1H, pyridine ring), 8.53, 8.91 (2s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3437–3211 (2NH <sub>2</sub> , 2NH), 2889 (CH <sub>2</sub> ), 2227 (CN), 1666 (CO), 1652 (C=N), 1646 (C=C)
<b>15b</b>	381	20.6, 22.9, 25.1 (3CH <sub>2</sub> ), 118.2, 122.4, 126.3, 131.7, 135.4, 140.8, 143.8, 146.1, 147.9 (thiophene C, oxazole C, pyridine C), 169.3 (C=O)	1.94–2.11 (m, 6H, 3CH <sub>2</sub> ), 4.45, 4.67 (2s, 4H, D <sub>2</sub> O-exchangeable, 2NH <sub>2</sub> ), 6.56–6.69 (m, 2H, oxazole ring, 1H, pyridine ring), 8.43, 8.78 (2s, 2H, D <sub>2</sub> O-exchangeable, 2NH)	3416–3227 (2NH <sub>2</sub> , 2NH), 2883 (CH <sub>2</sub> ), 2224 (CN), 1665 (CO), 1655 (C=N), 1645 (C=C)

Compd.	MS ( <i>m/z</i> )	<sup>13</sup> CNMR (δ, ppm)	<sup>1</sup> H NMR (δ, ppm)	IR (ν, cm <sup>-1</sup> )
<b>15c</b>	398	20.2, 22.9, 24.8 (3CH <sub>2</sub> ), 120.3, 123.7, 130.3, 133.7, 136.4, 140.5, 142.5, 144.8, 146.9 (thiophene C, thiazole C, pyridine C), 169.5 (C=O)	1.90-1.98 (m, 6H, 3CH <sub>2</sub> ), 4.38 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.73-6.81 (m, 2H, thiazole ring, 1H, pyridine ring), 8.37, 8.79 (2s, 2H, D <sub>2</sub> O-exchangeable, 2NH), 8.97 (s, 1H, D <sub>2</sub> O-exchangeable, OH)	3498-3386 (OH, NH <sub>2</sub> , 2NH), 2882 (CH <sub>2</sub> ), 2225 (CN), 1665 (CO), 1648 (C=N), 1644 (C=C)
<b>15d</b>	382	21.3, 23.8, 26.9 (3CH <sub>2</sub> ), 117.3, 121.6, 130.8, 132.5, 135.9, 137.8, 140.4, 143.6, 145.7 (thiophene C, oxazole C, pyridine C), 163.7 (C=O)	1.76-1.88 (m, 6H, 3CH <sub>2</sub> ), 4.41 (s, 2H, D <sub>2</sub> O-exchangeable, NH <sub>2</sub> ), 6.65-6.76 (m, 2H, oxazole ring, 1H, pyridine ring), 8.42, 8.86 (2s, 2H, D <sub>2</sub> O-exchangeable, 2NH), 9.12 (s, 1H, D <sub>2</sub> O-exchangeable, OH)	3454-3372 (OH, NH <sub>2</sub> , 2NH), 2887 (CH <sub>2</sub> ), 2226 (CN), 1672 (CO), 1654 (C=N), 1647 (C=C)
<b>16a</b>	436	20.2, 22.8, 24.5 (3CH <sub>2</sub> ), 121.2, 124.7, 127.1, 130.2, 133.4, 135.5, 139.3, 141.4, 143.6, 145.9, 148.4 (thiophene C, thiazole C, chromene C), 168.5, 171.2 (2C=O)	1.95-2.07 (m, 6H, 3CH <sub>2</sub> ), 4.48 (s, 2H, NH <sub>2</sub> ), 6.69-6.95 (m, 2H, thiazole ring, s, 1H, coumarin H-4), 7.28-7.41 (m, 4H, C <sub>6</sub> H <sub>4</sub> ), 8.22 (s, 1H, D <sub>2</sub> O-exchangeable, NH)	3423-3364 (NH <sub>2</sub> , NH), 3050 (CH-aromatic), 2883 (CH <sub>2</sub> ), 1796, 1683 (2CO), 1644 (C=C)
<b>16b</b>	420	22.0, 23.9, 26.1 (3CH <sub>2</sub> ), 120.8, 123.5, 126.2, 129.6, 132.7, 137.4, 139.8, 141.7, 144.7, 146.5, 149.1 (thiophene C, oxazole C, chromene C), 165.7, 170.5 (2C=O)	2.03-2.12 (m, 6H, 3CH <sub>2</sub> ), 4.27 (s, 2H, NH <sub>2</sub> ), 6.52-6.84 (m, 2H, oxazole ring, s, 1H, coumarin H-4), 7.40-7.54 (m, 4H, C <sub>6</sub> H <sub>4</sub> ), 8.31 (s, 1H, D <sub>2</sub> O-exchangeable, NH)	3438-3326 (NH <sub>2</sub> , NH), 3051 (CH-aromatic), 2889 (CH <sub>2</sub> ), 1802, 1688 (2CO), 1647 (C=C)
<b>17a</b>	418	22.1, 23.7, 25.8 (3CH <sub>2</sub> ), 118.4, 122.4, 126.3, 130.1, 132.5, 134.7, 136.3, 139.9, 141.8, 143.7, 145.3, 147.7 (thiophene C, thiazole C, pyrimidine C, chromene C), 174.8 (C=O)	2.07-2.18 (m, 6H, 3CH <sub>2</sub> ), 6.54-6.77 (m, 2H, thiazole ring, s, 1H, coumarin H-4), 7.45-7.63 (m, 4H, C <sub>6</sub> H <sub>4</sub> ), 8.43 (s, 1H, D <sub>2</sub> O-exchangeable, NH)	3392-3355 (NH), 3052 (CH-aromatic), 2887 (CH <sub>2</sub> ), 1854, (CO), 1646 (C=C)
<b>17b</b>	402	20.3, 22.5, 25.6 (3CH <sub>2</sub> ), 119.9, 122.7, 125.1, 129.4, 131.8, 133.6, 135.4, 138.7, 142.1, 143.9, 145.7, 147.6 (thiophene C, oxazole C, pyrimidine C, chromene C), 175.7 (C=O)	2.11-2.24 (m, 6H, 3CH <sub>2</sub> ), 6.63-6.73 (m, 2H, oxazole ring, s, 1H, coumarin H-4), 7.33-7.51 (m, 4H, C <sub>6</sub> H <sub>4</sub> ), 8.51 (s, 1H, D <sub>2</sub> O-exchangeable, NH)	3412-3363 (NH), 3055 (CH-aromatic), 2883 (CH <sub>2</sub> ), 1851, (CO), 1643 (C=C)

2-(1-Amino-2-cyanoethylideneamino)-N-(thiazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**5a**) and 2-(1-amino-2-cyanoethylideneamino)-N-(oxazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**5b**). – To a solution of either compound **3a** (2.654 g, 0.01 mol) or **3b** (2.493 g, 0.01 mol) in 1,4-dioxane (40 mL) containing triethylamine (0.5 mL), malononitrile (0.66 g, 0.01 mol) was added. The reaction mixture was heated under reflux for 2 h, cooled and poured into an ice/water mixture containing a few drops of conc. HCl. The formed precipitate was filtered out and recrystallized from absolute ethanol.

2-(4-Oxo-3-(thiazol-2-yl)-3,4,6,7-tetrahydro-5H-cyclopenta[4,5]thieno[2,3-d]pyrimidin-2-yl)acetonitrile (**6a**) and 2-(4-oxo-3-(oxazol-2-yl)-3,4,6,7-tetrahydro-5H-cyclopenta[4,5]thieno[2,3-d]pyrimidin-2-yl)acetonitrile (**6b**). – A suspension of either **4a** (0.665 g, 0.002 mol) or **4b** (0.633 g, 0.002 mol) in sodium ethoxide (0.002 mol) [prepared by dissolving elemental sodium (0.046 g, 0.002 mol) in abs. EtOH (40 mL)] was heated over a boiling water bath for 6 h and then left to cool. The solid product formed upon pouring onto ice/water containing a few drops of hydrochloric acid (10 %) (until pH = 6). It was collected by filtration and then recrystallized from absolute ethanol.

N-(3-(thiazol-2-yl-carbamoyl)-5,6-dihydro-4H-cyclopenta[b]thiophen-2-yl)-2-oxo-2H-chromene-3-carboxamide (**7a**) and N-(3-(oxazol-2-yl-carbamoyl)-5,6-dihydro-4H-cyclopenta[b]thiophen-2-yl)-2-oxo-2H-chromene-3-carboxamide (**7b**). – To a solution of either compound **4a** (0.665 g, 0.002 mol) or **4b** (0.633 g, 0.002 mol) in 1,4-dioxane (40 mL) containing piperidine (0.50 mL), salicylaldehyde (0.244 g, 0.002 mol) was added. The reaction mixture was heated under reflux for 6 h and then evaporated under vacuum. The remaining product was triturated with absolute ethanol and the formed solid product was collected by filtration and recrystallized from 1,4-dioxane.

2-(5-Amino-1H-pyrazol-3-ylamino)-N-(thiazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**8a**), 2-(5-amino-1H-pyrazol-3-ylamino)-N-(oxazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**8b**), 2-(5-amino-1-phenyl-1H-pyrazol-3-ylamino)-N-(thiazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**8c**) and 2-(5-amino-1-phenyl-1H-pyrazol-3-ylamino)-N-(oxazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**8d**). – Either compound **4a** (0.665 g, 0.002 mol) or **4b** (0.633 g, 0.002 mol) was dissolved in absolute ethanol (50 mL) containing piperidine (0.50 mL), hydrazine hydrate (0.1 g, 0.002 mol) or phenylhydrazine (0.216 g, 0.002 mol) and the whole mixture was refluxed for 3 h and then poured into water containing ice and a few drops of concentrated HCl. Suction filtration was then used to collect the coagulated precipitate; the formed solid product was recrystallized from ethanol.

2-(6-Amino-2-oxo-2,5-dihydropyrimidin-4-ylamino)-N-(thiazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**9a**), 2-(6-amino-2-oxo-2,5-dihydropyrimidin-4-yl amino)-N-(oxazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**9b**), 2-(6-amino-2-thioxo-2,5-dihydropyrimidin-4-ylamino)-N-(thiazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**9c**) and 2-(6-amino-2-thioxo-2,5-dihydropyrimidin-4-ylamino)-N-(oxazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**9d**). – To a suspension of either compound **4a** (0.665 g, 0.002 mol) or **4b** (0.633 g, 0.002 mol) in sodium ethoxide (0.002 mol, 50 mL), either urea (0.12 g, 0.002 mol) or thiourea (0.152 g, 0.002 mol) was added. The whole reaction mixture was heated over a boiling water bath for 3 h, then poured onto ice/water containing a few drops

of concentrated hydrochloric acid (until pH 6) and the solid product was collected by filtration and recrystallized from absolute ethanol.

2-(2-Cyano-3-phenylacrylamido)-N-(thiazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**10a**) and 2-(2-cyano-3-phenylacrylamido)-N-(oxazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**10b**). – Either compound **4a** (0.665 g, 0.002 mol) or **4b** (0.633 g, 0.002 mol) was dissolved in 50 mL abs. EtOH with added piperidine (0.50 mL). Benzaldehyde (0.21 g, 0.002 mol) was added and the reaction mixture was heated under reflux for 3 h and then poured into ice-containing water with a few drops of conc. HCl added. The coagulated precipitate was collected by suction filtration and recrystallized from 1,4-dioxane.

2-(3-(Thiazol-2-yl-carbamoyl)-5,6-dihydro-4H-cyclopenta[b]thiophen-2-ylamino)-2-oxo-N'-phenylacetohydrazonoyl cyanide (**12a**) and 2-(3-(oxazol-2-ylcarbamoyl)-5,6-dihydro-4H-cyclopenta[b]thiophen-2-ylamino)-2-oxo-N'-phenylacetohydrazonoyl cyanide (**12b**). – To a cold solution (0–5 °C) of either compound **4a** (0.665 g, 0.002 mol) or **4b** (0.633 g, 0.002 mol) in abs. ethanol (50 mL) containing sodium hydroxide (0.08 g, 0.002 mol), benzenediazonium chloride (**11**) (0.002 mol) [prepared by adding an aqueous sodium nitrite solution (0.138 g, 0.002 mol) to a cold solution of aniline (0.002 mol) with an appropriate amount of concentrated HCl, at 0–5 °C, with continuous stirring] was added under continuous stirring. The reaction mixture was stirred at room temperature for an additional 4 h and the solid product so formed was collected by filtration and recrystallized from 1,4-dioxane.

2-(4-(Aminothiazol-2-yl)-6,7-dihydro-5H-cyclopenta[4,5]thieno[2,3-d]pyrimidin-2-yl) acetonitrile (**13a**) and 2-(4-(amino-oxazol-2-yl)-6,7-dihydro-5H-cyclopenta[4,5]thieno[2,3-d]pyrimidin-2-yl)acetonitrile (**13b**). – A suspension of either **5a** (0.663 g, 0.002 mol) or **5b** (0.631 g, 0.002 mol) in sodium ethoxide (50 mL, 0.002 mol) was heated over a boiling water bath for 6 h and then left to cool. The solid product was formed upon pouring it into ice/water containing a few drops of concentrated hydrochloric acid (until pH = 6); it was collected by filtration and recrystallized from absolute ethanol.

2-(4,6-Diamino-5-cyanopyridin-2-ylamino)-N-(thiazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**15a**), 2-(4,6-diamino-5-cyanopyridin-2-yl-amino)N-(oxazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**15b**), 2-(4-amino-5-cyano-6-hydroxypyridin-2-ylamino)-N-(thiazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**15c**) and 2-(4-amino-5-cyano-6-hydroxypyridine-2-ylamino)-N-(oxazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**15d**). – To a suspension of either **5a** (0.663 g, 0.002 mol) or **5b** (0.631 g, 0.002 mol) in sodium ethoxide (0.002 mol, 50 mL), either ethyl cyanoacetate (0.226 g, 0.002 mol) or malononitrile (0.132 g, 0.002 mol) was added. The reaction mixture was heated under reflux for 3 h. It was then poured onto an ice/water mixture containing a few drops of concentrated hydrochloric acid. The formed solid product was collected by filtration and recrystallized from absolute ethanol.

2-(Amino(2-oxo-2H-chromen-3-yl)methyleneamino)-N-(thiazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**16a**) and 2-(amino(2-oxo-2H-chromen-3-yl)-methyleneamino)-N-(oxazol-2-yl)-5,6-dihydro-4H-cyclopenta[b]thiophene-3-carboxamide (**16b**). – To a solution of either compound **5a** (0.663 g, 0.002 mol) or **5b** (0.631 g, 0.002 mol) in 1,4-dioxane (40 mL) containing piperidine (0.50 mL), salicylaldehyde (0.244 g, 0.002 mol) was added. The reaction mixture was heated under reflux for 4 h and then evaporated under vacuum. The

remaining product was triturated with absolute ethanol and the crude solid product was collected by filtration and recrystallized from 1,4-dioxane.

4-(2-Amino-thiazol-2-yl)-2-(2-oxo-2H-chromene-3-yl)-6,7-dihydro-5H-cyclopenta[4,5]thieno[2,3-d]pyrimidine (**17a**) and 4-(2-aminooxazol-2-yl)-2-(2-oxo-2H-chromene-3-yl)-6,7-dihydro-5H-cyclopenta[4,5]thieno[2,3-d]pyrimidine (**17b**). – *Method A.* A solution of either compound **16a** (0.872 g, 0.002 mol) or **16b** (0.84 g, 0.002 mol) in dimethylformamide (40 mL) containing a catalytic amount of triethylamine (0.5 mL) was heated under reflux for 3 h. It was then poured into an ice/water mixture containing a few drops of conc. HCl. The formed solid product was collected by filtration and recrystallized from 1,4-dioxane.

*Method B.* To a solution of either compound **5a** (0.663 g, 0.002 mol) or **5b** (0.631 g, 0.002 mol) in sodium ethoxide (0.002 mol, 50 mL), salicylaldehyde (0.244 g, 0.002 mol) was added. The reaction mixture was heated under reflux for 4 h and then evaporated under vacuum. The remaining product was triturated with absolute ethanol and the solid product so formed was collected by filtration and recrystallized from 1,4-dioxane.

#### *Antitumor activity tests: materials and methods*

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

Three different human tumor cell lines were used: MCF-7 (breast adenocarcinoma), NCI-H460 (non-small cell lung cancer) and SF-268 (CNS cancer). MCF-7 was obtained from the European Collection of Cell Cultures (ECACC, Salisbury, UK) while NCI-H460, SF-268 and normal fibroblast cells (WI-38) were kindly donated 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<sup>-1</sup> glutamine and antibiotics (penicillin 100 U mL<sup>-1</sup> and streptomycin 100 mg mL<sup>-1</sup>), at 37 °C in a humidified atmosphere containing 5 % CO<sub>2</sub>. Exponentially growing cells were obtained by plating 1.5 × 10<sup>5</sup> cell mL<sup>-1</sup> for MCF-7 and SF-268 and 0.75 × 10<sup>4</sup> cell mL<sup>-1</sup> for NCI-H460. This was followed by 24-h incubation. The influence of DMSO as a solvent on the growth of cell lines was evaluated in all experiments. It was performed by exposing untreated control cells to the maximum concentration of DMSO used in each assay (0.5 %).

The *in vitro* tumor cell growth assay was performed according to the procedure described by the National Cancer Institute (USA) (13). This method uses the protein-binding dye sulforhodamine B to indicate cell growth. Afterwards, exponentially growing cells in 96-well plates were exposed to five serial dilutions of each compound, starting from a maximum concentration of 150 mmol L<sup>-1</sup> for 48 h. Following this exposure period, adherent cells were fixed, washed and stained. The bound stain was then dissolved in DMSO and absorbance was measured at 492 nm in a plate reader (Power wave XS, Bio-Tek Instruments, USA). For each test compound and cell line, a dose-dependent curve was established. *GI*<sub>50</sub> (concentration of the compound that inhibits 50 % of net cell growth) was calculated (14). Doxorubicin was used as a reference drug (positive control).

## RESULTS AND DISCUSSION

### Chemistry

Syntheses of the new heterocyclic compounds, thiophene, pyrimidine, coumarin, pyrazole and pyridine derivatives, are depicted in Schemes 1–4. Elucidation of their structures was based on analytical and spectral data.

Synthesis of compounds **2a,b-4a,b** and **5a,b** is displayed in Scheme 1. The reaction of ethyl cyanoacetate with either 2-aminothiazole (**1a**) or 2-aminoxazole (**1b**) yielded the acetamido derivative of each thiazole and oxazole ring (**2a,b**), respectively. The latter, upon reaction with cyclopentanone in the presence of elemental sulfur and a catalytic amount of triethylamine, gave thiophene derivatives **3a,b**, which were directed toward the reaction with either ethyl cyanoacetate or malononitrile, and yielded two pairs of carboxamide derivatives **4a,b** and **5a,b**, respectively.

Reactivity of the first pair of carboxamide derivatives **4a,b** was studied through some different types of reactions. Thus, internal cyclization was introduced to give fused pyrimidine derivatives **6a,b**. The structure of compounds **6a,b** was verified by elemental analysis and spectral data. In compound **6a**, the <sup>1</sup>H NMR spectrum indicated the presence of a multiplet at  $\delta$  1.96–2.08 ppm, which could be assigned to 3 CH<sub>2</sub> groups. A singlet at  $\delta$  3.08–3.12 ppm indicated the presence of the CH<sub>2</sub> side group and a multiplet at  $\delta$  6.62–6.73 ppm corresponded to 2 H of the thiazole ring. The IR spectrum of compound **6a** showed disappearance of 2 NH stretching signals. Compounds **4a,b** also reacted with salicylaldehyde in the presence of piperidine to form coumarin derivatives **7a,b**. For compound **7a**, the <sup>1</sup>H NMR spectrum showed a multiplet at  $\delta$  1.79–1.86 ppm from the presence of 3 CH<sub>2</sub> groups, a multiplet at  $\delta$  6.65–6.75 ppm corresponding to 2 H of the thiazole ring, a singlet at  $\delta$  6.88 ppm corresponding to 1H of the coumarin ring, a multiplet at  $\delta$  7.34–7.49 ppm corresponding to 4 H of the benzene ring and two singlets, D<sub>2</sub>O-exchangeable at  $\delta$  8.39 and 8.56 ppm corresponding to 2 H of 2 NH groups. Furthermore, the reaction of compounds **4a,b** with either hydrazine hydrate or phenylhydrazine afforded compounds **8a-d**. Pyrimidine derivatives **9a-d** were obtained through the reaction of compounds **4a,b** with either urea or thiourea in sodium ethoxide. <sup>1</sup>H NMR spectrum of compound **9a** showed a multiplet at  $\delta$  1.83–1.94 ppm that indicated 3 CH<sub>2</sub> groups, a singlet at  $\delta$  3.96 ppm corresponding to 2 H of the pyrimidine ring, a singlet, D<sub>2</sub>O-exchangeable at  $\delta$  4.51 ppm corresponding to 2 H of the amino group, a multiplet at  $\delta$  6.61–6.68 ppm corresponding to 2 H of the thiazole ring and two singlets, D<sub>2</sub>O-exchangeable at  $\delta$  8.53 and 8.71 ppm corresponding to 2 H of 2 NH groups. In the reaction of compounds **4a,b** with either benzaldehyde or benzenediazonium chloride (**11**), compounds **10a,b** and **12a,b** were formed, respectively. <sup>1</sup>H NMR spectrum of compound **10a** indicated a multiplet at  $\delta$  1.83–1.96 ppm which represented 3 CH<sub>2</sub> groups, a singlet at  $\delta$  5.18 ppm corresponding to 1 H of =CH group, a multiplet at  $\delta$  6.33–6.58 ppm corresponding to 2 H of the thiazole ring, a multiplet at  $\delta$  7.35–7.48 ppm corresponding to 5 H of the benzene ring and two singlets, D<sub>2</sub>O-exchangeable at  $\delta$  8.37 and 8.52 ppm corresponding to 2 H of 2 NH groups.

Also, reactivity of the second pair of carboxamide derivatives **5a,b** was used. They underwent ready cyclization when heated in a sodium ethoxide solution, leading to the corresponding pyrimidine derivatives **13a,b**. Thus, the <sup>1</sup>H NMR spectrum of compound **13a** showed a multiplet at  $\delta$  1.76–1.85 ppm from the 3 CH<sub>2</sub> groups, a singlet at  $\delta$  3.12–3.19

ppm that indicated the presence of the CH<sub>2</sub> side group, a multiplet at  $\delta$  6.51–6.63 ppm corresponding to 2 H of the thiazole ring and a singlet D<sub>2</sub>O-exchangeable at  $\delta$  8.11 ppm corresponding to 1 H of the NH group. Moreover, <sup>13</sup>C NMR of the same compound showed signals at 21.2, 22.8, 25.4, 38.2 (cyclopentyl C, CH<sub>2</sub> side group), 121.4 (CN), 123.1, 125.4, 127.3, 130.4, 132.9, 138.6, 143.3, 146.4 ppm (thiophene C, thiazole C, pyrimidine C). Furthermore, when compounds **5a,b** reacted with either ethyl cyanoacetate or malononitrile in sodium ethoxide, they formed pyridine derivatives **15a-d**, through the formation of intermediate compounds **14a-d**. <sup>1</sup>H NMR spectrum of **15a** showed a multiplet at  $\delta$  1.81–1.93 ppm assignable to 3 CH<sub>2</sub> groups, two singlets D<sub>2</sub>O-exchangeable at  $\delta$  4.33 and 4.52 ppm that indicated the presence of 4 H of 2 NH<sub>2</sub> groups, a multiplet at  $\delta$  6.61–6.78 ppm corresponding to 2 H of the thiazole ring and 1 H of the pyridine ring and two singlets D<sub>2</sub>O-exchangeable at  $\delta$  8.53 and 8.91 ppm indicating the presence of 2 H for 2 NH groups. Finally, the reaction of **5a,b** with salicylaldehyde took two specific pathways. The first was carried out in piperidine affording compounds **16a,b**. The other products underwent cyclization to form fused pyrimidine derivatives **17a,b** in dimethylformamide. The second pathway took place in a sodium ethoxide solution to form compounds **17a,b** directly. The analytical and spectral data of products **16a,b** and **17a,b** are consistent with the assigned structures.

#### *Effects on the growth of human tumor cell lines and QSAR*

Compounds **2a,b-17a,b** were examined for their inhibitory effect on the *in vitro* growth of human tumor cell lines of different tumor types, namely, breast adenocarcinoma (MCF-7), non-small cell lung cancer (NCI-H460) and CNS cancer (SF-268). All tested compounds inhibited the growth of the tested tumor cell lines in a dose-dependent manner (data not shown). The results from Table III show that fused pyrimidine acetonitrile derivatives **6a,b** exerted the highest inhibitory effect against all three tumor cell lines, comparable with the reference standard. Among the fused pyrimidine derivatives containing coumarin moiety, compounds **17a,b** showed the highest inhibitory effects against all three tumor cell lines. Compounds **2a,b**, **13a,b** and **15a,b,c,d** showed moderate inhibitory effects against all three cancer cell lines, while the rest of the compounds (**3a,b**, **4a,b**, **5a,b**, **7a,b**, **8a-d**, **9a-d**, **10a,b**, **12a,b** and **16a,b**) showed low effects.

When comparing fused pyrimidine acetonitrile derivatives, it was found that compound **6b** was slightly more effective than **6a**, possibly due to the presence of the oxazole ring in **6b** instead of the thiazole ring in **6a**. On the other hand, for the fused pyrimidine derivatives containing coumarin moiety, it was found that compound **17a** with the thiazole ring showed stronger effect than compound **17b** containing the oxazole ring. Cyanoacetamide derivatives **2a** and **2b** showed comparable effects in the presence of the thiazole ring in compound **2a** instead of oxazole ring in compound **2b**. The same was seen when comparing fused pyrimidine derivatives, **13a** with the thiazole ring and **13b** with the oxazole ring. Among pyridine derivatives **15a-d**, compound **15a** was most effective against all three cell lines, possibly due to the presence of the NH<sub>2</sub> group instead of OH group (in **15c** and **15d**), in addition to the presence of the thiazole ring instead of the oxazole ring (in **15b**).

When comparing the amino-thiophene derivatives **3a,b**, acetamido-thiophene derivatives **4a,b** and acetimido-thiophene derivatives **5a,b**, it was found that these compounds showed almost the same effects despite structural differences (namely, the presence of NH<sub>2</sub> in **3a,b**, CNCH<sub>2</sub>CONH in **4a,b** and CNCH<sub>2</sub>C=N(NH<sub>2</sub>) in **5a,b**). On the other hand,



Table III. Effect of compounds **2a,b-17a,b** on the growth of three human tumor cell lines

Compd.	$GI_{50}$ (mmol L <sup>-1</sup> ) <sup>a</sup>			
	MCF-7	NCI-H460	SF-268	WI-38
<b>2a</b>	23.7 ± 7.4	24.1 ± 5.3	28.5 ± 5.9	NA
<b>2b</b>	27.1 ± 8.9	25.7 ± 6.8	26.2 ± 4.9	NA
<b>3a</b>	44.6 ± 7.8	45.1 ± 7.3	44.3 ± 7.7	65.5 ± 11.7
<b>3b</b>	41.9 ± 7.9	48.7 ± 6.3	42 ± 8.3	NA
<b>4a</b>	51.3 ± 11.8	50.4 ± 10.1	49.5 ± 11.5	> 100
<b>4b</b>	44.2 ± 9.3	46.1 ± 9.3	47.5 ± 9.5	> 100
<b>5a</b>	48.0 ± 9.9	48.2 ± 10.7	44.7 ± 9.5	> 100
<b>5b</b>	41.8 ± 7.6	43.5 ± 6.5	43.5 ± 8.1	38.1 ± 12.5
<b>6a</b>	0.1 ± 0.08	0.25 ± 0.4	1.6 ± 0.3	15.1 ± 8.3
<b>6b</b>	0.09 ± 0.06	0.07 ± 0.03	1.3 ± 0.2	14.4 ± 7.6
<b>7a</b>	37.9 ± 9.5	36.4 ± 8.7	32.1 ± 6.8	22.7 ± 11.3
<b>7b</b>	39.3 ± 10.2	34.6 ± 8.8	37.9 ± 9.7	49.1 ± 14.4
<b>8a</b>	40.3 ± 5.4	41.1 ± 7.6	36.6 ± 8.6	28.3 ± 12.3
<b>8b</b>	33.1 ± 7.2	34.5 ± 5.9	35.3 ± 6.2	21.1 ± 13.5
<b>8c</b>	29.0 ± 6.9	30.0 ± 7.4	27.5 ± 10.8	22.6 ± 11.5
<b>8d</b>	35.9 ± 11.3	39.2 ± 12.2	36.0 ± 14.1	23.1 ± 14.8
<b>9a</b>	44.2 ± 7.3	46.6 ± 6.4	34.9 ± 19.1	12.7 ± 11.8
<b>9b</b>	34.9 ± 11.2	41.0 ± 8.8	44.5 ± 11.6	29.3 ± 10.1
<b>9c</b>	36.7 ± 6.9	40.9 ± 6.6	42.4 ± 9.3	NA
<b>9d</b>	41.2 ± 8.6	34.0 ± 6.9	39.7 ± 8.7	NA
<b>10a</b>	42.7 ± 13.9	39.9 ± 14.3	32.3 ± 5.3	NA
<b>10b</b>	39.2 ± 6.8	37.3 ± 6.4	35.9 ± 6.9	NA
<b>12a</b>	45.2 ± 12.8	47.1 ± 16.4	38.5 ± 21.1	NA
<b>12b</b>	38.4 ± 7.7	35.2 ± 8.3	37.6 ± 8.9	NA
<b>13a</b>	21.7 ± 6.9	17.9 ± 4.7	21.5 ± 5.3	> 100
<b>13b</b>	21.2 ± 5.6	18.0 ± 3.9	19.7 ± 4.9	> 100
<b>15a</b>	16.2 ± 6.9	17.3 ± 7.8	15.1 ± 9.9	> 100
<b>15b</b>	25.2 ± 6.8	25.3 ± 9.4	26.9 ± 11.9	> 100
<b>15c</b>	19.9 ± 12.8	23.1 ± 11.4	30.2 ± 15.3	> 100
<b>15d</b>	20.2 ± 6.3	27.1 ± 8.3	25.5 ± 9.5	> 100
<b>16a</b>	41.1 ± 14.8	43.3 ± 16.4	39.8 ± 6.9	NA
<b>16b</b>	39.7 ± 8.5	37.4 ± 9.9	32.6 ± 10.5	NA
<b>17a</b>	3.1 ± 0.09	2.3 ± 1.4	2.9 ± 1.1	> 100
<b>17b</b>	6.2 ± 1.4	3.4 ± 0.7	4.8 ± 0.9	> 100
Doxorubicin	0.04 ± 0.008	0.09 ± 0.008	0.09 ± 0.007	> 100

NA – not applicable

Results are given in concentrations that were able to cause 50 % of cell growth inhibition ( $GI_{50}$ ) after a continuous exposure for 48 h.

<sup>a</sup> Mean ± SEM of three-independent experiments performed in duplicate.

when comparing coumarin derivatives **7a,b**, pyrazole derivatives **8a-d** and pyrimidine derivatives **9a-d**, it was found that compound **8c** was highly effective. This may be due to the presence of the pyrazole ring instead of either coumarin ring in compounds **7a,b** or pyrimidine ring in compounds **9a-d**, in addition to the phenyl group instead of H (in **8a,b**) and thiazole ring instead of oxazole ring (in **8d**).

Finally, when comparing benzylidene derivatives **10a,b**, phenylhydrazone derivatives **12a,b** and coumarin derivatives **16a,b**, it was found that these compounds may be of almost the same efficacy despite the presence of the benzylidene side chain in compounds **10a,b**, phenylhydrazone side chain in compounds **12a,b** and coumarin moiety in compounds **16a,b**.

## CONCLUSIONS

Among the newly synthesized products, fused pyrimidine acetonitrile derivatives **6a** and **6b** showed high inhibitory activity against all the three tumor cell lines, MCF-7, NCI-H460 and SF-268, comparable to that of doxorubicine. They were followed by fused pyrimidine derivatives containing the coumarin moiety, **17a** and **17b**.

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## REFERENCES

1. R. Romagnoli, P. G. Baraldi, C. Lopez-Cara, M. K. Salvador, D. Preti, M. A. Tabrizi, J. Balzarini, P. Nussbaumer, M. Bassetto, A. Brancale, X. H. Fu, Y. Gao, J. Li, S. Z. Zhang, E. Hamel, R. Bortolozzi, G. Basso and G. Viola, Design, synthesis and biological evaluation of 3,5-disubstituted 2- amino-thiophene derivatives as a novel class of antitumor agents, *Bioorg. Med. Chem.* **22** (2014) 5097–5109; DOI: 10.1016/j.bmc. 2013.12.030.
2. R. Romagnoli, P. G. Baraldi, M. D. Carrion, C. L. Cara, D. Perti, F. Fruttarolo, M. G. Pavani, M. A. Tabrizi, M. Tolomio, S. Grimaudo, A. Di Cristina, J. Balzarini, J. A. Hadfield, A. Bracale and E. Hamel, Synthesis and biological evaluation of 2- and 3-aminobenzo[b]thiophene derivatives as antimitotic agents and inhibitors of tubulin polymerization, *J. Med. Chem.* **50** (2007) 2273–2277; DOI: 10.1021/jm070050f.
3. C. E. Stephens, T. M. Felder, J. W. Sowell, G. Andrei, J. Balzarini, R. Snoeck and E. De Clercq, Synthesis and antiviral/antitumor evaluation of 2-amino- 2-carboxamido-3-aryl- sulfonylthiophenes and related compounds as a new class of diarylsulfones, *Bioorg. Med. Chem.* **9** (2001) 1123–1132; DOI: 10.1016/S0968-0896(00)00333-3.
4. S. Sulzer-Mosse, F. Cederbaum, C. Lamberth, G. Berthon, J. Umarye, V. Grasso, A. Schlereth, M. Blum and R. Waldmeier, Synthesis and fungicidal activity of N-thiazol-4-yl-salicylamides, a new family of anti-oomycete compounds, *Bioorg. Med. Chem.* **23** (2015) 2129–2138; DOI: 10.1016/j.bmc.2015.03.007.
5. V. Padmavathi, C. Prema Kumari, B. C. Venkatesh and A. Padmaja, Synthesis and antimicrobial activity of amido linked pyrrolyl and pyrazolyl-oxazoles, thiazoles and imidazoles, *Eur. J. Med. Chem.* **46** (2011) 5317–5326; DOI: 10.1016/j.ejmech.2011.08. 032.

6. M. B. Dewal, A. S. Wani, C. Vidaillac, D. Oupický, M. J. Rybak and S. M. Firestine, Thieno[2,3-*d*]pyrimidinedione derivatives as antibacterial agents, *Eur. J. Med. Chem.* **51** (2012) 145–153; DOI: 10.1016/j.ejmech.2012.02.035.
7. A. T. Mavrova, D. Wesselinova, J. A. Tsenov and L. A. Lubenov, Synthesis and antiproliferative activity of some new thieno[2,3-*d*]pyrimidine-4-(3*H*)-ones containing 1,2,4-triazole and 1,3,4-thiadiazole moiety, *Eur. J. Med. Chem.* **86** (2014) 676–683; DOI: 10.1016/j.ejmech.2014.09.032.
8. F. Chimenti, B. Bizzarri, A. Bolasco, D. Secci, P. Chimenti, S. Carradori, A. Granese, D. Rivanera, D. Lilli, A. Zicari, M. M. Scaltrito and F. Sisto, A novel class of selective anti-*Helicobacter pylori* agents 2-oxo-2*H*-chromene-3-carboxamide derivatives, *Bioorg. Med. Chem. Lett.* **17** (2007) 3065–3071; DOI: 10.1016/j.bmcl.2007.03.050.
9. S. Bindi, D. Fancelli, C. Alli, D. Berta, J. A. Bertrand, A. D. Cameron, P. Cappella, P. Carpinelli, G. Cervi, V. Croci, M. D'Anello, B. Forte, M. L. Giorgini, A. Marsiglio, J. Moll, E. Pesenti, V. Pittalà, M. Pulici, F. Riccardi-Sirtori, F. Roletto, C. Soncini, P. Storici, M. Varasi, D. Volpi, P. Zugnoni and P. Vianello, Thieno[3,2-*c*]pyrazoles: a novel class of Aurora inhibitors with favorable antitumor activity, *Bioorg. Med. Chem.* **18** (2010) 7113–7120; DOI: 10.1016/j.bmc.2010.07.048.
10. S. Ulloora, R. Shabaraya, R. Ranganathan and A. V. Adhikari, Synthesis, anticonvulsant and anti-inflammatory studies of new 1,4-dihydropyridin-4-yl-phenoxyacetohydrazones, *Eur. J. Med. Chem.* **70** (2013) 341–349; DOI: 10.1016/j.ejmech.2013.10.010.
11. A. A. Napoleon, F. R. N. Khan, E. D. Jeong and E. H. Chung, Potential antitubercular agents: Hexahydro-3-phenyl indazol-2-yl-(pyridine-4-yl) methanones from antitubercular drug isoniazid and bis(substituted benzyldene) cycloalkanones, *Chin. Chem. Lett.* **26** (2015) 567–571; DOI: 10.1016/j.ccllet.2015.01.008.
12. S. Malik, P. Ahuja, K. Sahu and S. A. Khan, Design and synthesis of new 3-(benzo [*d*] isoxazol-3yl)-1-substituted pyrrolidine-2,5-dione derivatives as anti-convulsants, *Eur. J. Med. Chem.* **84** (2014) 42–50; DOI: 10.1016/j.ejmech.2014.07.016
13. P. Skehan, R. Storeng, D. Scudiero, A. Monks, J. McMahon, D. Vistica, J. T. Warren, H. Bokesch, S. Kenney and M. R. Boyd, New colorimetric cytotoxicity assay for anticancer-drug screening, *J. Natl. Cancer Inst.* **82** (1990) 1107–1112; DOI: 10.1093/jnci/82.13.1107.
14. A. Monks, D. Scudiero, P. Skehan, R. Shoemaker, K. Paull, D. Vistica, C. Hose, J. Langley, P. Cronise, A. Vaigro-Wolff, M. Gray-Goodrich, H. Campbell, J. Mayo and M. J. Boyd, Feasibility of a high-flux anticancer drug screen using a diverse panel of cultured human tumor cell lines, *J. Natl. Cancer Inst.* **83** (1991) 757–766; DOI: 10.1093/jnci/83.11.757.