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**SYNTHESIS, REACTIONS, AND ANTI-BACTERIAL ACTIVITY OF SOME NEW N-BENZYL-4-OXOTHIAZOLIDIN-2-YLIDENE)ACETAMIDE DERIVATIVES**

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**Abstract**

Treatment of N-benzyl-2-cyanoacetamide (**1**) with ethyl isothiocyanate (**2**) and p-phenylenediisothiocyanate (**11**) gave the non-isolable intermediates **3** and **12**, respectively. Subsequent treatment of **3** and **12**, respectively with  $\alpha$ -halo esters and/or chloroacetone gave the corresponding 4-oxothiazolidin-2-ylidene **5a-c**, bis(4-oxothiazolidin-2-ylidene) (**14**), thiazol-2-ylidene (**6**) and bis(5-acetyl-4-amino-3-N-benzylthiophenecarboxamido)-1-,4-phenylenediamine (**13**) derivatives, respectively. Reaction of **5a** with electrophilic carbon was studied where derivatives (**8a,b**), **10** were obtained. Cyclocondensation of **1** with thioglycolic acid afforded thiazolidin-4-one derivative (**15**). Condensation of **15** with 1-naphthaldehyde, arylidenemalononitriles and ethyl  $\alpha$ -cyanocinnamate gave 4,5-dihydrothiazol-2-ylacrylamide (**17**), thiazolo[3,2-a]pyridines (**16a,b**) and (**18**), respectively. The structures of these new compounds were confirmed by IR, ( $^1\text{H}$ - and  $^{13}\text{C}$ -NMR) and mass spectral analyses. Some of the synthesized compounds were tested in vitro for their antimicrobial activity, where compounds **5a**, **6**, **8b**, **16a**, **16b**, and **17** exhibited the best antibacterial activity against *Salmonella typhi* NCIM130331.

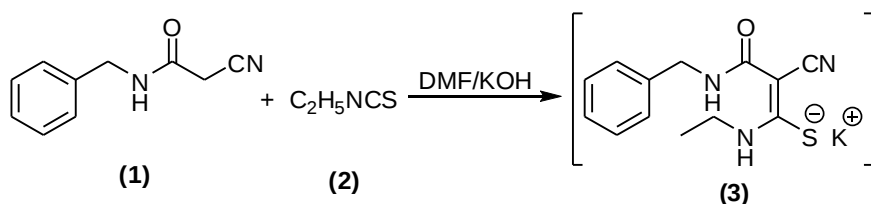
**Keywords** : N-benzyl-2-cyanoacetamide, thiazolidinone and bithiazolidinone derivatives.

**Introduction**

The literature survey revealed that a large number of thiazolidinone derivatives are known in medicinal chemistry for their therapeutic value<sup>1</sup>. Many derivatives of these compounds showed interesting of anti-bacterial<sup>2</sup>, antifungal<sup>3</sup>, anticonvulsant<sup>4</sup>, anticancer<sup>5</sup> and anti-tuberculosis<sup>6</sup> activities. It was of interest to synthesize some new thiazolidinone derivatives to investigate their biological properties.

**Results and Discussion**

The N-benzyl-2-cyanoacetamide (**1**) was used as a key intermediate to synthesize hitherto unknown thiazolidinone and bithiazolidinone derivatives. The reaction of compound (**1**) with ethyl isothiocyanate (**2**) in the presence of potassium hydroxide at room temperature gave the non-isolable potassium sulfide salt (**3**), Eq. 1.



Eq. 1

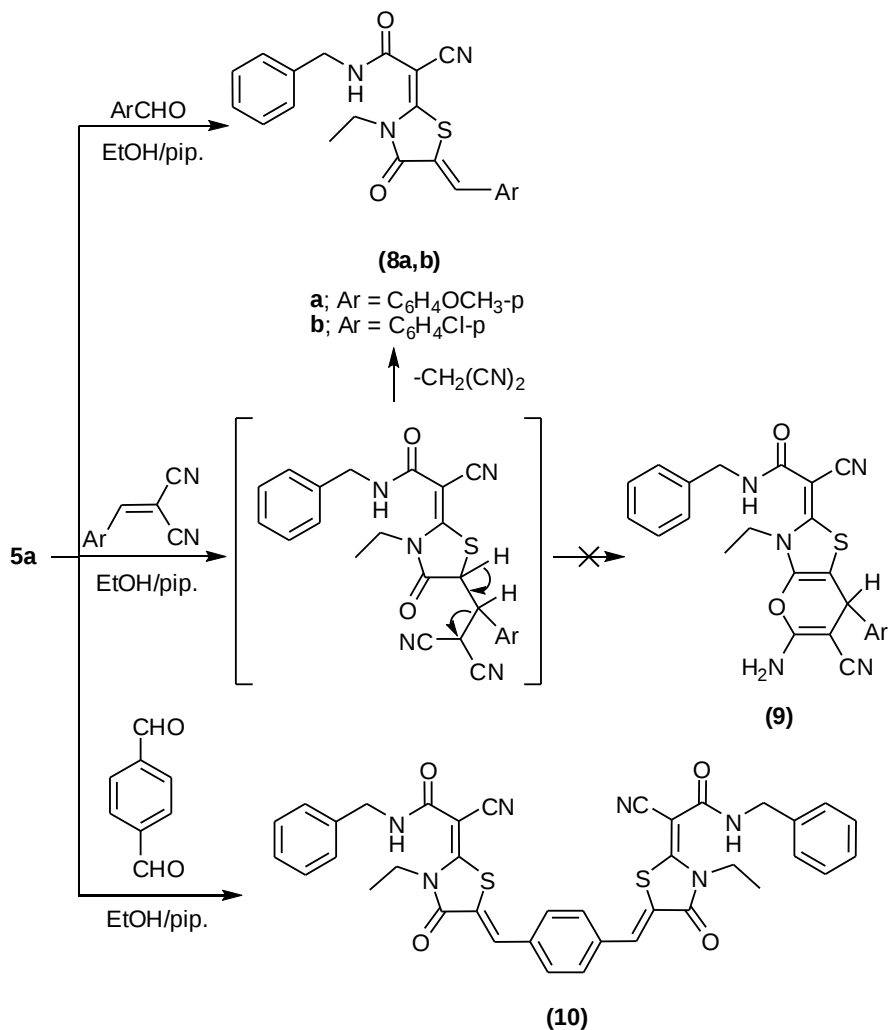
Treatment of the non-isolable potassium salt **(3)** with  $\alpha$ -halo esters **(4a-c)** at room temperature gave thiazolidin-4-one derivatives **(5a-c)**, Scheme 1. Structure of compounds **(5a-c)** was confirmed on the basis of elemental analyses and spectral data (*cf.* table 2).

The  $^{13}\text{C}$  NMR of compound **5a** showed signals at  $\delta$  13.81 ( $\text{CH}_3$ ), 31.10 ( $\text{SCH}_2$ ), 38.34 ( $\text{CH}_2\text{CH}_3$ ), 42.92 ( $\text{CH}_2\text{NH}$ ), 76.04 ( $\text{C-CN}$ ), 116.46 (CN), 126.73, 127.27, 128.21, 139.52 (aromatic), 163.87 (CO), 168.90 (C2, thiazolidinone), 173.78 (CO). Mass spectrum of **5a** revealed a molecular ion peak at  $m/z$  301 (10.6%) and the base peak at  $m/z$  91 (stable tropylium cation). The mass spectrum of **5b** showed a molecular ion peak at  $m/z$  315 (11.8%) and the base peak was found in the spectrum at  $m/z$  91. Mass spectrum of **5c** showed a molecular ion peak at  $m/z$  329 (16.6%) and the base peak was observed at  $m/z$  91 (stable tropylium cation).

The formation of structures **5a-c** was assumed to proceed via the initial alkylation followed by intramolecular cyclization with elimination of ethanol<sup>7</sup>. Cyclocondensation of the intermediate **3** with chloroacetone at room temperature yielded the corresponding 4-methylthiazole derivative **6**, Scheme 1. The structure **7** was excluded on the basis of elemental analyses and spectral data (*cf.* table 2). Mass spectrum of compound **6** exhibited a molecular ion peak at  $m/z$  299 (25%) and the base peak at  $m/z$  91 (stable tropylium cation). The formation of **6** was obtained via initial alkylation followed by intramolecular cyclization through dehydration.

**Scheme 1**

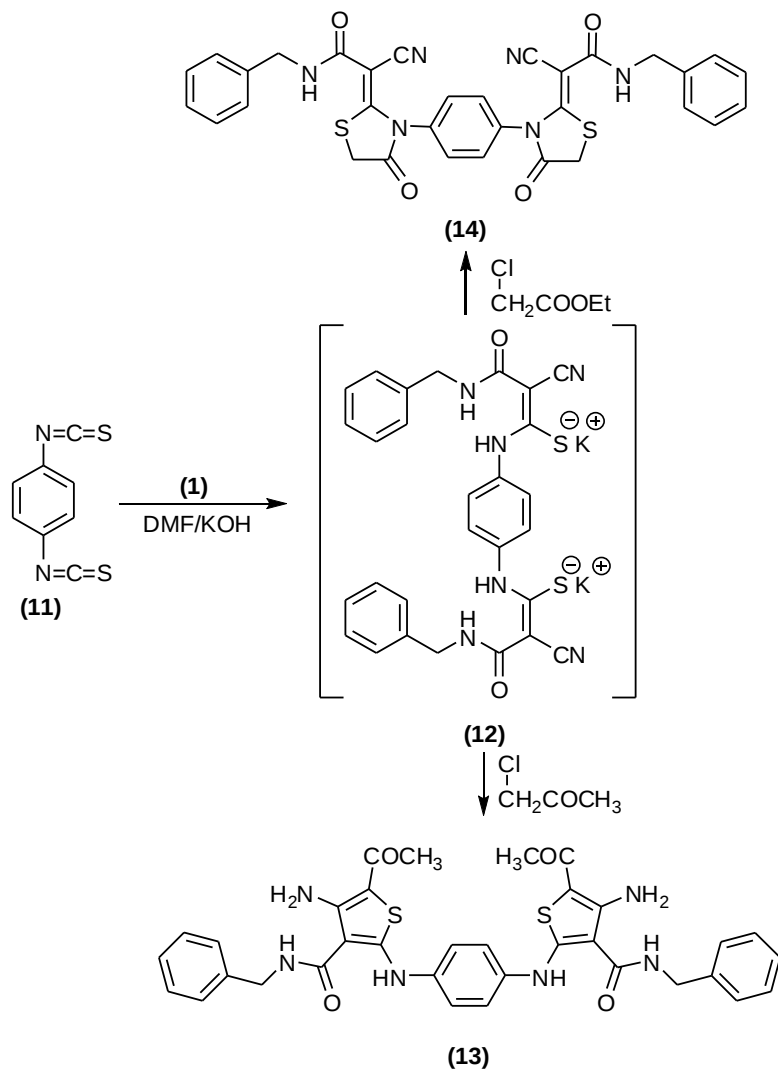
Condensation of compound **5a** with aromatic aldehydes in refluxing ethanol in the presence of piperidine afforded the corresponding benzylidene derivatives **8a,b**, Scheme 2. The structure of compounds **8a,b** was established by analytical and spectral data (*cf.* table 2). The mass spectrum of **8a** showed a molecular ion peak at  $m/z$  419 (19.6%) together with a base peak at  $m/z$  164 ( $\text{CH}_3\text{OC}_6\text{H}_4\text{CHCS}$ ). When compound **5a** was reacted with respective arylidenemalononitrile in refluxing ethanol containing a catalytic amount of piperidine gave the same molecular structure of **8** and the other possible structure of pyranothiazole derivative **9** was ruled out on the basis of analytical and spectral data. Bisthiazolidinone derivative (**10**) was achieved by refluxing compound **5a** with terephthalaldehyde (2 : 1 molar ratio) in ethanolic piperidine, Scheme 2. The structure of compound **10** was confirmed by analytical and spectral data (*cf.* table 2).



Scheme 2

The present contribution was extended to synthesize hitherto unknown bithiophene and bithiazolidinone derivatives. Treatment of *N*-benzyl-2-cyanoacetamide (**1**) with *p*-phenylenediisothiocyanate (**11**) in the presence of potassium hydroxide at room temperature gave the non-isolable sulfide potassium salt (**12**). The latter was converted into bithiophene derivative (**13**) by treatment with chloroacetone at room temperature, through Thorpe cyclization<sup>7</sup>. Also, cyclization of the adduct **12** with ethyl chloroacetate afforded the corresponding bithiazolidinone derivative **14**, via initial alkylation and elimination of ethanol, Scheme 3. Structures

of compounds **13** and **14** were established by analytical and spectral data (cf. table 2).



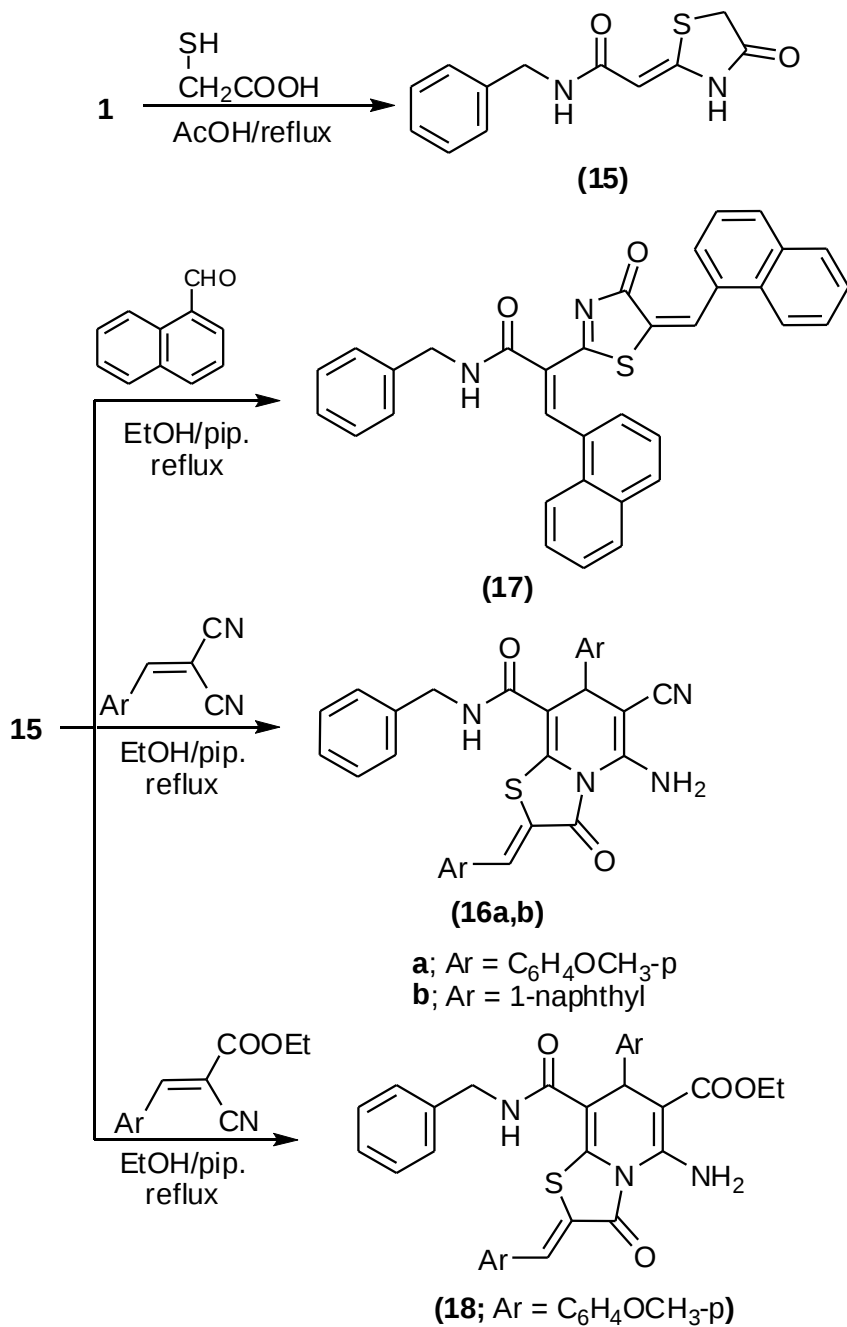
**Scheme 3**

Our investigation was extended to study the reaction of *N*-benzyl-2-cyanoacetamide (**1**) with thioglycolic acid. Thus, cyclocondensation of compound **1** with thioglycolic acid at reflux temperature afforded thiazolidin-4-one derivative **15** on the basis of analytical and spectral data (cf. table 2). Mass spectrum of **15**

afforded the molecular ion peak at  $m/z$  248 (52.8%) with base peak at  $m/z$  91 (stable tropylium cation).  $^{13}\text{C}$ NMR spectrum of compound **15** revealed signals at  $\delta$  31.94 ( $\text{SCH}_2$ ), 40.40 ( $\text{CH}_2\text{CO}$ ), 41.89 ( $\text{CH}_2\text{benzyl}$ ), 126.66, 127.24, 128.25, 140.01 (aromatic), 152.18 ( $\text{C}=\text{N}$ ), 166.54 ( $\text{CO}$ ), 174.00 ( $\text{CO}$ ).

The formation of compound **15** resulting from initial nucleophilic addition of mercapto group to nitrile center followed by intramolecular cyclization by elimination of water<sup>8</sup>.

Transformation of **15** to the corresponding thiazolo[3,2-*a*]pyridine derivatives **16a,b** was achieved upon refluxing of **15** with arylidenemalononitriles in ethanol in the presence of piperidine, Scheme 4. The structure of **16a,b** was established by analytical and spectral data (cf. table 2). Mass spectra of compound **16a** and **16b** were revealed the molecular ion peak at  $m/z$  550 (5.1%) and 590 (2.9%) together with base peaks at  $m/z$  91 for both, respectively. Also, condensation of **15** with 1-naphthaldehyde in ethanol in the presence of piperidine yielded the novel *N*-benzyl-3-(naphthalen-1-yl)-2-(5-(naphthalen-1-ylmethylene)-4-oxo-4,5-dihydro-thiazol-2-yl)acrylamide (**17**). The molecular structure of **17** was confirmed by analytical and spectral data (cf. table 2). Mass spectrum of compound **17** exhibited the molecular ion peak at  $m/z$  524 (2.2%) and the base peak at  $m/z$  91 (stable tropylium cation). In the same manner, the novel thiazolopyridine derivative (**18**) was obtained from the reaction of **15** with ethyl  $\alpha$ -cyano(4-methoxy)cinnamate in ethanol in the presence of piperidine, Scheme 4. The structure of compound **18** was established by analytical and spectral data (cf. table 2).



Scheme 4

## Experimental

All melting points are uncorrected. IR spectra (KBr) were measured on Shimadzu 440 spectrometer, <sup>1</sup>H NMR spectra were obtained in DMSO on a Varian Gemini 200 MHz spectrometer using TMS as internal standard; chemical shifts are reported as (ppm). Mass spectra were obtained on GCMS\QP 1000 Ex mass spectrometer at 70 eV. Elemental analyses were carried out at the Microanalytical Center, Faculty of Science (Cairo University, Egypt). The characteristics data for prepared compounds are given in table 1. *N*-benzyl-2-cyanoacetamide (**1**) was prepared as previously reported<sup>9</sup>.

**Preparation of compounds 5a-c, 6, 13, and 14 : General procedure:** *N*-benzyl-2-cyano-2-(3-ethyl-4-oxothiazolidin-2-ylidene)acetamide (**5a**), *N*-benzyl-2-cyano-2-(3-ethyl-5-methyl-4-oxo-thiazolidin-2-ylidene)-acetamide (**5b**), *N*-benzyl-2-cyano-2-(3,5-diethyl-4-oxo-thiazolidin-2-ylidene)-acetamide (**5c**), *N*-benzyl-2-cyano-2-(3-ethyl-4-methylthiazol-2(3h)-ylidene)acetamide (**6**), 2,2'-(1,4-phenylenebis(azanediyl))bis(5-acetyl-4-amino-*N*-benzyl-thiophene-3-carboxamide) (**13**), 2,2'-(3,3'-(1,4-phenylene))bis(4-oxothiazolidin-3-yl-2-ylidene))bis(*N*-benzyl-2-cyanoacetamide) (**14**).

To a suspension of finely powdered potassium hydroxide (0.01 mole) in dry dimethylformamide (10 ml) at 0°C, the cyanoacetamide derivative (**1**, 0.01 mole or 0.02 mole) and then the requisite isothiocyanate (0.01 mole) were added in portions. The reaction mixture was stirred at room temperature for 3h and then treated with  $\alpha$ -halogenated compound (0.01 mole) and left at room temperature for 24h, then it was poured into ice/water and acidified with 0.1 N HCl at pH 3-4. The resulting precipitate was filtered off, dried and recrystallized from the proper solvent, table 1.

**Formation of compounds *N*-benzyl-2-cyano-2-(3-ethyl-5-(4-arylmethylidene)-4-oxothiazolidin-2-ylidene)acetamide (8a,b), and 5,5'-(1,4-phenylenebis(methan-1-yl-1-ylidene))bis(3-ethyl-4-oxothiazolidine-5,2-diylidene))bis(*N*-benzyl-2-cyanoacetamide) (10): General procedure:**

A mixture of compound **5a** (0.01 mole or 0.02 mole), aromatic aldehyde or terephthalaldehyde (0.01 mole) and few drops of piperidine in absolute ethanol (40 ml) was refluxed for 3h. The solid product formed on hot was filtered off, washed



with ethanol and dried. The crude product was crystallized to give **8a,b**, and **10**, respectively, table 1.

**Synthesis of *N*-benzyl-2-(4-oxo-thiazolidine-2-ylidene) acetamide (15):**

A solution of **1** (0.01 mole) in glacial acetic acid (10 ml) was treated with thioglycolic acid (0.01 mole). The reaction mixture was refluxed for 3h and then evaporated in vacuo. The remaining product was triturated with water and the resulting solid product was collected by filtration and recrystallized from dioxane to give **15**, table 1.

***N*-Benzyl-3-(naphthalen-1-yl)-2-(5-(naphthalen-1-ylmethylene)-4-oxo-4,5-dihydrothiazol-2-yl)acrylamide (17).**

A mixture of compound **15** (0.01 mole) and 1-naphthaldehyde (0.01 mole) in ethanol (40 ml) containing a few drops of piperidine heated under reflux for 3h. The resulting solid product was collected and recrystallized from dioxane to give **17**, table 1.

**Formation of compounds 5-amino-*N*-benzyl-6-cyano-2-(arylmethylidene)-7-(aryl)-3-oxo-3,8-dihydro-2*H*-thiazolo[3,2-*a*]pyridine-8-carboxamide (16a,b), and ethyl 5-amino-8-(benzylcarbamoyl)-2-(4-methoxybenzylidene)-7-(4-methoxy-phenyl)-3-oxo-3,8a-dihydro-2*H*-thiazolo[3,2-*a*]-pyridine-6-carboxylate (18): General procedure:**

A mixture of compound **15** (0.01 mole), respective cinnamionitrile (0.01 mole) and piperidine (0.01 mole) in ethanol (40 ml) was heated under reflux for 3h, the solid product was collected and recrystallized from DMF to give **16a,b** and **18**, respectively, table 1.

Table 1. Characteristics Data for the Synthesized Compounds.

Compd. No.	Yield (%)	Solvent Cryst.	M.p. (°C)	Mol. Formula (Mol. wt.)	Elemental analyses Calcd./Found %		
					C%	H%	N%
<b>5a</b>	78	Dioxane	198-200	C <sub>15</sub> H <sub>15</sub> N <sub>3</sub> O <sub>2</sub> S	59.78	5.02	13.94
				(301.35)	59.70	4.80	13.80
<b>5b</b>	58	Dioxane	152-154	C <sub>16</sub> H <sub>17</sub> N <sub>3</sub> O <sub>2</sub> S	60.93	5.43	13.32
				(315.38)	60.90	5.30	13.20
<b>5c</b>	65	Dioxane	165-167	C <sub>17</sub> H <sub>19</sub> N <sub>3</sub> O <sub>2</sub> S	61.98	5.81	12.76
				(329.40)	61.90	5.70	12.70
<b>6</b>	72	Dioxane	160-162	C <sub>16</sub> H <sub>17</sub> N <sub>3</sub> OS	64.19	5.72	14.04
				(299.38)	64.10	5.60	13.90
<b>8a</b>	75	Dioxane	215-217	C <sub>23</sub> H <sub>21</sub> N <sub>3</sub> O <sub>3</sub> S	65.85	5.05	10.02
				(419.5)	65.80	4.90	9.85
<b>8b</b>	77	Dioxane	236-238	C <sub>22</sub> H <sub>18</sub> ClN <sub>3</sub> O <sub>2</sub> S	62.33	4.28	9.91
				(423.94)	62.20	4.20	9.80
<b>10</b>	72	Dioxane/ DMF	230-232	C <sub>38</sub> H <sub>32</sub> N <sub>6</sub> O <sub>4</sub> S <sub>2</sub>	65.12	4.60	11.99
				(700.81)	65.00	4.50	11.90
<b>13</b>	61	Dioxane	202-204	C <sub>34</sub> H <sub>32</sub> N <sub>6</sub> O <sub>4</sub> S <sub>2</sub>	62.56	4.94	12.87
				(652.77)	62.50	4.90	12.80
<b>14</b>	65	AcOH	299-301	C <sub>32</sub> H <sub>24</sub> N <sub>6</sub> O <sub>4</sub> S <sub>2</sub>	61.92	3.90	13.54
				(620.68)	61.80	3.80	13.50
<b>15</b>	76	Dioxane	207-208	C <sub>12</sub> H <sub>12</sub> N <sub>2</sub> O <sub>2</sub> S	58.05	4.87	11.28
				(248.30)	57.90	4.80	11.20
<b>16a</b>	57	DMF	240-242	C <sub>31</sub> H <sub>26</sub> N <sub>4</sub> O <sub>4</sub> S	67.62	4.76	10.18
				(550.61)	67.50	4.70	10.10
<b>16b</b>	66	DMF	265-267	C <sub>37</sub> H <sub>26</sub> N <sub>4</sub> O <sub>2</sub> S	75.23	4.44	9.48
				(590.67)	75.10	4.40	9.50
<b>17</b>	71	Dioxane	153-154	C <sub>34</sub> H <sub>24</sub> N <sub>2</sub> O <sub>2</sub> S	77.84	4.61	5.34
				(524.61)	77.80	4.50	5.30
<b>18</b>	65	DMF	253-255	C <sub>33</sub> H <sub>31</sub> N <sub>3</sub> O <sub>6</sub> S	66.32	5.23	7.03
				(597.66)	66.20	5.10	6.90

Table 2. Spectral data of the synthesized compounds .

Compd. No.	IR (KBr, cm <sup>-1</sup> )	<sup>1</sup> HNMR (DMSO- <i>d</i> <sub>6</sub> ) (δ, ppm).
5a	3364 (NH), 3032 (CH-arom.), 2936 (CH-aliph.), 2194 (C≡N), 1732, 1652 (C=O; thiazolidinone & amide).	1.23 (t, 3H, CH <sub>3</sub> ), 3.86 (s, 2H, CH <sub>2</sub> -thiazolidinone), 4.13 (q, 2H, CH <sub>2</sub> CH <sub>3</sub> ), 4.38 (s, 2H, CH <sub>2</sub> NH), 7.35-7.42 (m, 5H, Ar-H), 8.46 (s, 1H, NH; exchangeable with D <sub>2</sub> O).
5b	3360 (NH), 2928 (CH-aliph.), 2198 (C≡N), 1728, 1642 (C=O; thiazolidinone & amide).	1.27 (t, 3H, CH <sub>3</sub> CH <sub>2</sub> ), 1.51 (d, 3H, CH <sub>3</sub> ), 4.04-4.17 (m, 3H, CH <sub>2</sub> CH <sub>3</sub> + CH-thiazole), 4.40 (s, 2H, CH <sub>2</sub> ), 7.35 (m, 5H, Ar-H), 8.50 (s, 1H, NH; exchangeable with D <sub>2</sub> O).
5c	3346 (NH), 3032 (CH-arom.), 2970, 2924 (CH-aliph.), 2200 (C≡N), 1726, 1644 (C=O; thiazolidinone & amide).	0.95, 1.26 (2t, 6H, 2CH <sub>3</sub> ), 1.30 (p, 2H, CH <sub>2</sub> ), 4.1-4.39 (m, 3H, CH <sub>2</sub> + thiazole-H), 7.35-7.61 (m, 5H, Ar-H), 8.51 (s, 1H, NH; exchangeable with D <sub>2</sub> O).
6	3366 (NH), 3106 (CH-arom.), 2982, 2916 (CH-aliph.), 2170 (C≡N), 1658 (C=O; amide).	1.37 (t, 3H, CH <sub>3</sub> ), 2.32 (s, 3H, CH <sub>3</sub> ), 4.37 (q, 2H, CH <sub>2</sub> CH <sub>3</sub> ), 4.40 (s, 2H, CH <sub>2</sub> ), 6.77 (s, 1H, CH-thiazole), 7.33-7.60 (m, 5H, Ar-H), 7.68 (s, 1H, NH; exchangeable with D <sub>2</sub> O).
8a	3352 (NH), 2930 (CH-aliph.), 2194 (C≡N), 1700, 1646 (C=O; thiazolidinone & amide).	1.35 (t, 3H, CH <sub>3</sub> CH <sub>2</sub> ), 3.89 (s, 3H, OCH <sub>3</sub> ), 4.31 (q, CH <sub>2</sub> CH <sub>3</sub> ), 4.43 (s, 2H, CH <sub>2</sub> NH), 7.17-7.78 (m, 10H, Ar-H + benzylidene-H), 8.69 (s, 1H, NH; exchangeable with D <sub>2</sub> O).
8b	3334 (NH), 2986, 2932 (CH-aliph.), 2194 (C≡N), 1700, 1646 (C=O; thiazolidinone & amide).	
10	3332 (NH), 2930 (CH-aliph.), 2200 (C≡N), 1702, 1646 (C=O; thiazolidinone & amide).	1.31 (t, 6H, 2 CH <sub>3</sub> CH <sub>2</sub> ), 4.27 (q, 4H, 2 CH <sub>2</sub> CH <sub>3</sub> ), 4.42 (s, 4H, 2 CH <sub>2</sub> NH), 7.32-8.08 (m, 14H, Ar-H), 8.69 (s, 2H, benzylidene-H), 10.05 (s, 2H, 2NH; exchangeable with D <sub>2</sub> O).
13	3392, 3146 (NH <sub>2</sub> /NH), 3050 (CH-arom.), 2970 (CH-aliph.), 1766, 1640 (C=O acetyl and amide).	2.50 (s, 6H, 2COCH <sub>3</sub> ), 4.3, 4.83 (2s, 4H, 2CH <sub>2</sub> NH), 7.30-7.5 (m, 14H, Ar-H), 8.02 (s, 4H, 2NH <sub>2</sub> ), 9.63, 9.82 (4H, 4NH; exchangeable with D <sub>2</sub> O).
14	3346 (NH), 2982 (CH-aliph.), 2200 (C≡N), 1710, 1646 (C=O; thiazolidinone & amide).	4.43, 4.90 (2s, 8H, 4CH <sub>2</sub> ), 7.2-7.51 (m, 14H, Ar-H), 9.64, 9.84 (2s, 2H, 2NH; exchangeable with D <sub>2</sub> O).
15	3318 (NH), 3050 (CH-arom.), 2988, 2896 (CH-aliph.), 1702, 1640 (C=O; thiazolidinone & amide).	3.69, 4.34 (2s, 4H, 2CH <sub>2</sub> ), 5.70 (s, 1H, methyldene-H), 7.20-7.62 (m, 5H, Ar-H), 8.29, 11.34 (2s, 2H, 2NH; exchangeable with D <sub>2</sub> O).
16a	3446, 3330 (NH <sub>2</sub> /NH), 3026 (CH-arom.), 2930 (CH-aliph.), 2184 (C≡N), 1696, 1654 (C=O; thiazolidinone & amide).	3.81, 3.89 (2s, 6H, 2 OCH <sub>3</sub> ), 4.4 (S, 2H, CH <sub>2</sub> ), 4.89 (s, 1H, pyridine-H), 6.87-7.72 (m, 16H, Ar-H + benzylidene-H + NH <sub>2</sub> ), 8.3 (br.s, 1H, NH; exchangeable with D <sub>2</sub> O).
16b	3418, 3336 (NH <sub>2</sub> /NH), 3044 (CH-arom.), 2192 (C≡N), 1698, 1658 (C=O; thiazolidinone & amide).	4.5 (s, 2H, CH <sub>2</sub> ), 5.9 (s, 1H, pyridine-H), 6.5 (s, 2H, NH <sub>2</sub> ; exchangeable with D <sub>2</sub> O), 6.90-8.3 (m, 20H, Ar-H + benzylidene-H), 8.5 (s, 1H, NH; exchangeable with D <sub>2</sub> O).
17	3250 (NH), 3052 (CH-arom.), 1694, 1638 (C=O; thiazolidinone & amide).	4.50 (s, 2H, CH <sub>2</sub> ), 7.40-8.22 (m, 21H, Ar-H + benzylidene-H), 10.40 (s, 1H, NH; exchangeable with D <sub>2</sub> O).
18	3402, 3266 (NH <sub>2</sub> /NH), 3046 (CH-arom.), 1700, 1662 (C=O; ester, thiazolidinone & amide).	0.83 (t, 3H, CH <sub>3</sub> ), 3.83 (s, 6H, 2 OCH <sub>3</sub> ), 4.22 (s, 2H, CH <sub>2</sub> NH), 6.01 (s, 1H, pyridine-H), 6.63-8.3 (m, 16H, Ar-H) + benzylidene-H + NH <sub>2</sub> ), 8.85 (s, 1H, NH; exchange-able with D <sub>2</sub> O).

**Antibiogram susceptibility and resistance to twelve complex compounds:****a) Bacterial inoculums:**

Only one single 24 h. old colony was picked up from the surface of nutrient agar plates and then inoculated into nutrient broth and incubated in a shaking incubator for 24 h. The assay medium was inoculated by 1.0 ml bacterial broth per each 100ml

**b) Preparation of the assay medium:**

Mueller-Hinton agar medium was used for this assay. It contains (g/l): Infusion from meat, 2.0; casein hydrolysate, 17.5; starch, 1.5; agar, 13.0, distilled water up to 1000 ml. Dissolve the ingredients in distilled water by heating in boiling water, adjusted pH at  $7.4 \pm 0.2$ , distribute in small flask, autoclave for 15.0 minutes at  $115^{\circ}\text{C}$  and allow to cool to about  $45^{\circ}\text{C}$  (Mueller and Hinton<sup>10</sup>, 1941; Dewees *et al*<sup>11</sup>, 1970).

**c) Inoculation of assay plates:**

Pour plate method technique (15 cm in diameter) was applied by inoculating the Mueller-Hinton agar medium (25 ml/plate) while at  $45^{\circ}\text{C}$  by 1.0 ml of each bacterial isolate broth culture, then pouring the homogenized seeded medium and left for solidification. Antibiotic discs are placed on the surface of solid medium and kept in refrigerator for diffusion just before incubation. Detection of inhibition zones around antibiotic discs on inoculated plates is an indication of antibacterial activities of antibiotics.

**d) Antibiotic discs:**

The disc diffusion method was applied using commercial paper discs impregnated with antibiotics and loaded on the inoculated plates. Twelve discs were applied on seven microbes viz. *Escherichia coli* NCTC-10418, *Pseudomonas syringae* ATCC-19310, *Salmonella typhi* NCIM 130331, *Staphylococcus aureus* NCTC-7447, *Bacillus subtilis* NCTC-70400, *Candida albicans* CBS-652 and *Aspergillus niger* LTV-1.

**Results****Antibiogram susceptibility and resistance of twelve complexes compounds:**

Eleven compounds were applied during this study against the growth of the five bacterial, two unicellular and multicellular fungal strains viz. *Escherichia coli*

NCTC-10418, *Pseudomonas syringae* ATCC-19310, *Salmonella typhi* NCIM 130331, *Staphylococcus aureus* NCTC-7447, *Bacillus subtilis* NCTC-70400, *Candida albicans* CBS-652 and *Aspergillus niger* LTV-1 by paper disc diffusion method according to Dewees *et al*<sup>11</sup>, (1970), Iakshkina *et al*<sup>12</sup>, (1988), Fuchs *et al*<sup>13</sup>. (1990), National Committee for Clinical Laboratory Standards (1988 & 1990) and Ronald and Meridith<sup>14</sup> (1991).

It was obvious from the results recorded in table (3) showed that six compounds only (**5a**, **6**, **8b**, **16a**, **16b** and **17**) exhibited antibacterial activity against only one bacterial strain viz. *Salmonella typhi* NCIM 130331 with (9, 9, 10, 10, 10 and 8 mm) respectively represented Gram-negative bacilli. All other tested compounds give negative results as antimicrobial agents against all tested microbes.

**Table 3:** A summary of resistance and susceptibility of the eleven compounds against seven tested microbial strains.

Compd. No.	Mean diameter of inhibition zone (mm)						
	<i>Escherichia coli</i> NCTC-10418	<i>Pseudomonas syringae</i> ATCC-19310	<i>Salmonella typhi</i> NCIM 130331	<i>Bacillus subtilis</i> NCTC-70400	<i>Staphylococcus aureus</i> NCTC-7447	<i>Candida albicans</i> CBS-652	<i>Aspergillus niger</i> LTV-1
<b>5a</b>	-	-	9	-	-	-	-
<b>5b</b>	-	-	-	-	-	-	-
<b>6</b>	-	-	9	-	-	-	-
<b>8a</b>	-	-	-	-	-	-	-
<b>8b</b>	-	-	10	-	-	-	-
<b>10</b>	-	-	-	-	-	-	-
<b>15</b>	-	-	-	-	-	-	-
<b>16a</b>	-	-	10	-	-	-	-
<b>16b</b>	-	-	10	-	-	-	-
<b>18</b>	-	-	-	-	-	-	-
<b>17</b>	-	-	8	-	-	-	-

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## References

1. S. G. KÜCÜKGÜZEL, E. E. ORUC, S. ROLLAS, F. SAHIN AND A. OZBEK, *Eur. J. Med. Chem.*, **37**, 197 (2002).
2. G. CAPAN, N. ULUSOY, N. ERGENC AND M. KIRAZ, *Monatshefte für Chemie*, **130**, 1399 (1999).
3. N. ERGENC AND G. CAPAN, *IL Farmaco*, **49**, 133 (1994).
4. J. J. BHAT, B. R. SHAH, H. P. SHAH, P. B. TRIVEDI, N. K. UNDAVIA AND N.C. DESAI, *Indian J. Chem.*, **33B**, 189 (1994).
5. L. BUKOWSKI, M. JANOWIEC, Z. ZWOLSKA AND Z. ANDREJCZYK, *Pharmazie*, **53**, 373 (1998).
6. Y. A. AMMAR, A. M. SH. EL-SHARIEF, A. G. AL-SEHEMI, Y. A. MOHAMED, G. A. M. EL-HAG ALI, M. A. SENUSSI AND M. S. A. EL-GABY, *Phosphorus, Sulfur and Silicon*, **180 (11)**, 2503 (2005).
7. Y. A. AMMAR, M. M. ALY, A. G. AL-SEHEMI, Y. A. MOHAMED, M. A. SALEM AND M. S. A. EL-GABY, *Phosphorus, Sulfur and Silicon*, **183 (7)**, 1710 (2008).
8. M. S. A. EL-GABY, M. M. KHAFAGY, G. A. M. EL-HAG ALI, A. A. EL-MAGHRABY AND M. H. M. HELAL, *Phosphorus, Sulfur and Silicon*, **178**, 1681 (2003).
9. K. M. AL-ZAYDI, R. M. BORIK, M. H. EL-NAGDI, *Ultrasonics Sonochemistry*, **169 (5)**, 660 (2009).
10. H. J. MUELLER, J. Hinton, *Proc. Soc. Expt. Biol. Med.*, **48**, 330 (1941).
11. L. B. DEWEES, J. A. POUPARD, H. E. MORTON, *J. Appl. Microbiol.*, **20**, 293 (1970).
12. I. V. IAKSUHKINA, I. V. MALKOVA, I. N. PROZOROVA, *Antibiot. Khimioter.*, **33(5)**, 342 (1988).
13. P. C. FUCHS, R. N. JONES, A. L. BARRY, *Antimicrob. Agents Chemother.*, **34**, 414 (1990).
14. N. J. RONALD, E. E. MERIDITH, *J. Clin. Microbiol.*, **29 (12)**, 2890 (1991).