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## Azomethine-(5-methyl-1*H*-tetrazol-1-yl) conjugates: Synthesis, antibacterial and antifungal evaluation

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**Abstract:** Reaction of 4-methyl-3-(5-methyl-1*H*-tetrazol-1-yl)benzenamine with nine different aromatic aldehydes gives the target compounds (**5a-i**) in good yields. A total of nine new entities were synthesized and were characterized by IR, <sup>1</sup>H & <sup>13</sup>C NMR and mass spectroscopic analysis and evaluated for their antibacterial and antifungal activities. Antibacterial activity data showed that compounds **5a**, **5b**, **5g**, **5h** and **5i** were most notable against the bacterial strains. Besides, all the compounds were screened for their antifungal activities.

**Keywords:** antibacterial, antifungal, *E. coli*, tetrazole, schiff base.

## 1. INTRODUCTION

Multidrug-resistant (MDR) bacteria, a group of frequent causative pathogens in healthcare, are typically associated with nosocomial infections. MDR bacteria often cause complex illnesses, mainly infections of urinary tract, respiratory, ocular, endocarditis, bone and joint, and skin, etc. Indeed, these infections are difficult to treat, kind of antibiotics used which lead to the emergence of new mutant strains having high level of resistance to many antibiotics.<sup>1</sup> Besides, the prevalent infections leads progressively more complex therapy in recent years as their mechanisms of resistance and sensitivity patterns differ widely across different regions.<sup>2-5</sup> This emphasizes the urgent need for the discovery and development of structurally-novel antimicrobial agents with good pharmacological profile and excellent activity towards resistant strains.<sup>6,7</sup>

At present, principally in the field of medicinal chemistry, tetrazoles have received much attention due to their prevalent biological activities.<sup>8,9</sup> The tetrazole ring is often used as carboxylic acid surrogates,<sup>10-14</sup> and lipophilic spacers which improves oral absorption.<sup>15</sup> The pharmaceutical entities like Losartan,<sup>16</sup> Valsartan,<sup>17</sup> Irbesartan,<sup>18</sup> Flomoxef<sup>19</sup> and Cefonicid<sup>20</sup> contains the tetrazole ring serve as highly effective drugs. Recently, the tetrazole conjugates have found to possess broad spectrum of antibacterial activity notably thieno[2,3-*d*] pyrimidines,<sup>21,22</sup> pyrrolo[3,2-*e*]pyrimidines,<sup>23</sup> quinoline,<sup>24</sup> quinoxalines,<sup>25</sup> thiazole,<sup>26</sup> etc. Consequently, tetrazole derivatives such as biphenyl tetrazoles,<sup>27</sup> triazolo[4,3-*d*]tetrazolo[5,1-*f*]-1,2,4-triazines<sup>28</sup> substituted aryl tetrazolo[1,5-*b*]1,2,5- oxadiazepin-9-ones<sup>29</sup> and 2-(5- substituted phenyl-1*H*-tetrazol-1-yl) pyridine<sup>30</sup> have been found to possess promising antibacterial activity.

Furthermore, tetrazole derivatives bearing hydrazone and thiazoline moieties<sup>31</sup> and triazine dendrimeric chalcones<sup>32</sup> exhibit a potent antifungal activity. Besides, tetrazole scaffolds like 3-aryl-1-(5-phenyl-1*H*-tetrazol-1-yl)prop-2-en-1-one,<sup>33</sup> 5-thiosubstituted tetrazole derivatives,<sup>34</sup> 2-(1-methyl-1*H*-1,2,3,4-tetrazol-5-yl)sulfanyl-*N*-(5-methylisoxazol-3-yl)acetamide,<sup>35</sup> and tetrazole derivatives containing quinoline,<sup>36,37</sup> thiophene,<sup>38</sup> pyridyl and pyrimidyl,<sup>39,40</sup> catecholthioethers,<sup>41</sup> purine,<sup>42</sup> piperazine and triazole<sup>43</sup> display comparable antifungal activity. Therefore, it was envisaged that a new series of tetrazoles containing various Schiff bases would possess good activity due to its structural resemblance with reported heterocycles.

In continuation of our work, we have designed and synthesized a new series of (Benzylidene)(4-methyl-3-(5-methyl-1*H*-tetrazol-1-yl)phenyl)amine derivatives (**5a-i**) from 4-methyl-3-(5-methyl-1*H*-tetrazol-1-yl)benzenamine by reacting it with various aromatic aldehydes in ethanol. All the target molecules were evaluated for their antimicrobial activity against selected bacterial and fungal strains.

## 2. EXPERIMENTAL

**2.1. Materials and Methods:** All the chemicals used were of AR grade and were purchased from Sd-Fine chemicals. The progress of reaction and purity of the product were monitored by thin layer chromatography using precoated Silica 60/UV<sub>254</sub> (SDFCL). IR spectra were recorded (KBr) on a Perkin Elmer 1650 spectrophotometer and reported in cm<sup>-1</sup>. <sup>1</sup>H NMR and <sup>13</sup>C NMR spectra were recorded on 400 MHz Varian Mercury plus 400 MHz FT NMR spectrometer using CDCl<sub>3</sub> as solvents and TMS as an internal standard. Chemical shifts are expressed as  $\delta$  ppm scale relative to TMS ( $\delta$  = 0.00 ppm). Melting points were obtained using melting points apparatus (Model MP-96) and are uncorrected.

## 2.2. General procedure

**Synthesis of (substituted-benzylidene)(4-(5-methyl-1H-tetrazol-1-yl)phenyl)amine (5a-i)** : To a solution of 4-methyl-3-(5-methyl-1H-tetrazol-1-yl)benzenamine (**3**, 0.5 mmol) in 20 ml ethanol were added substituted benzaldehydes (**4a-I**, 0.5 mmol). The mixture was stirred for 30 min. at room temperature, poured into cold water, filtered and dried. Column Chromatography (4:1 Pet. Ether:EtOAc) gave compound **5a**.

## 2.3 Physical and Spectral Data

**(4-Methoxybenzylidene)(4-(5-methyl-1H-tetrazol-1-yl)phenyl)amine (5a)**: Yield: 90%; mp 154-155 °C; IR:  $\nu/\text{cm}^{-1}$ : 3102, 3063 (Ar-H), 2978, 2863 (CH<sub>3</sub>), 1619 (C=N azomethine), 1587 (C=N tetrazole ring), 1490 (N=N tetrazole ring), 1441, 1403 and 1252 (N-N=N), 1096 and 1005 (tetrazole ring); <sup>1</sup>H-NMR:  $\delta$  ppm: 2.08 (s, 3H, CH<sub>3</sub>); 3.92 (s, 3H, OCH<sub>3</sub>); 6.98 (d, 2H, Ar-H); 7.21 (d, 2H, Ar-H); 7.36 (d, 2H, Ar-H); 7.83 (d, 2H, Ar-H); 8.40 (s, 1H, N=CH); <sup>13</sup>C-NMR:  $\delta$  ppm: 9.0 (CH<sub>3</sub>-Tetrazole), 55.4 (CH<sub>3</sub>-O), 119.4 (C, tetrazole), 123.7 (2C), 128.5 (2C), 130.2 (2C), 131.1 (2C), 132.9 (C-Tetrazole), 133.8 (C-N), 151.1 (C, Ar), 152.4 (C, Ar), 162.2 (N=CH); MS (ESI) 294 (M+1)<sup>+</sup>. Anal. calcd for C<sub>16</sub>H<sub>15</sub>N<sub>5</sub>O: C, 65.52; H, 5.15; N, 23.88; O, 5.45. Found: C, 65.43; H, 5.18; N, 23.91; O, 5.46.

**(2-Chlorobenzylidene)(4-(5-methyl-1H-tetrazol-1-yl)phenyl)amine (5b)** : Yield: 91%; mp 128-130 °C; IR:  $\nu/\text{cm}^{-1}$ : 3116, 3077 (Ar-H), 2981, 2876 (CH<sub>3</sub>), 1604 (C=N azomethine), 1591 (C=N tetrazole ring), 1492 (N=N tetrazole ring), 1406 and 1277 (N-N=N), 1090 and 998 (tetrazole ring), 758 (C-Cl); <sup>1</sup>H-NMR:  $\delta$  ppm: 2.12 (s, 3H, CH<sub>3</sub>); 7.13 (d, 2H, Ar-H); 7.39-7.46 (m, 3H, Ar-H); 7.55 (d, 2H, Ar-H); 7.73 (m, 1H, Ar-H); 8.95 (s, 1H, N=CH); <sup>13</sup>C-NMR:  $\delta$  ppm: 9.1 (CH<sub>3</sub>-Tetrazole), 120.1 (C, tetrazole), 121.08 (C, Ar), 123.3 (2C, Ar), 130.01 (C, Ar), 132.56 (2C, Ar), 133.32 (C-Tetrazole), 133.96 (C-N), 137.31 (C, Ar), 150.01 (C, Ar), 151.01 (C, Ar), 151.99 (C, Ar), 160.88 (N=CH); MS (ESI) 299 (M+1)<sup>+</sup>. Anal. calcd for C<sub>16</sub>H<sub>12</sub>ClN<sub>5</sub>: C, 60.51; H, 4.06; Cl, 11.91; N, 23.52 Found: C, 60.48; H, 4.10; Cl, 11.93; N, 23.56.

**(3-Chlorobenzylidene)(4-(5-methyl-1H-tetrazol-1-yl)phenyl)amine (5c)** : Yield: 95%; mp 136-138 °C; IR:  $\nu/\text{cm}^{-1}$ : 3123, 3089 (Ar-H), 2992, 2869 (CH<sub>3</sub>), 1633 (C=N azomethine), 1597 (C=N tetrazole ring), 1503 (N=N tetrazole ring), 1405 and 1273 (N-N=N), 1097, 1068 and 995 (tetrazole ring), 717 (C-Cl); <sup>1</sup>H-NMR:  $\delta$  ppm: 2.11 (s, 3H, CH<sub>3</sub>); 7.11 (d, 2H, Ar-H); 7.46 (d, 2H, Ar-H); 7.53-7.58 (m, 3H, Ar-H); 7.82 (m, 1H, Ar-H); 8.51 (s, 1H, N=CH); <sup>13</sup>C-NMR:  $\delta$  ppm: 9.08 (CH<sub>3</sub>-Tetrazole), 119.4 (C, tetrazole), 119.69 (C, Ar), 123.41 (2C, Ar), 128.47 (C, Ar), 130.0 (C, Ar), 132.89 (2C, Ar), 133.43 (C-Tetrazole), 133.97 (C-N), 137.37 (C, Ar), 150.09 (C, Ar), 151.89 (C, Ar), 160.8 (N=CH); MS (ESI) 299 (M+1)<sup>+</sup>. Anal. calcd for C<sub>16</sub>H<sub>12</sub>ClN<sub>5</sub>: C, 60.51; H, 4.06; Cl, 11.91; N, 23.52 Found: C, 60.49; H, 4.11; Cl, 11.92; N, 23.54.

**(4-Chlorobenzylidene)(4-(5-methyl-1H-tetrazol-1-yl)phenyl)amine (5d)**: Yield: 97%; mp 142-144 °C; IR:  $\nu/\text{cm}^{-1}$ : 3101, 3062 (Ar-H), 2962, 2850 (CH<sub>3</sub>), 1629 (C=N azomethine), 1598 (C=N tetrazole ring), 1500 (N=N tetrazole ring), 1407 and 1279 (N-N=N), 1130, 1099 and 1030 (tetrazole ring), 751 (C-Cl); <sup>1</sup>H-NMR:  $\delta$  ppm: 2.10 (s, 3H, CH<sub>3</sub>); 7.13 (d, 2H, Ar-H); 7.28 (d, 2H, Ar-H); 7.48 (d, 2H, Ar-H); 7.82 (d, 2H, Ar-H); 8.49 (s, 1H, N=CH); <sup>13</sup>C-NMR:  $\delta$  ppm: 9.07 (CH<sub>3</sub>-Tetrazole), 119.46 (C, tetrazole), 123.34 (2C, Ar), 129.35 (2C, Ar), 130.1 (2C, Ar), 132.38 (2C, Ar), 138.24 (C-Tetrazole), 142.41 (C-N), 150.82

(C, Ar), 152.48 (C, Ar), 160.17 (N=CH); MS (ESI) 299 (M+1)<sup>+</sup>. Anal. calcd for C<sub>16</sub>H<sub>12</sub>ClN<sub>5</sub>: C, 60.51; H, 4.06; Cl, 11.91; N, 23.52 Found: C, 60.48; H, 4.05; Cl, 11.96; N, 23.57.

**(4-(5-Methyl-1H-tetrazol-1-yl)phenyl)(2-nitrobenzylidene)amine (5e)** : Yield: 93%; mp 128-130 °C; IR:  $\nu/\text{cm}^{-1}$ : 3105, 3076 (Ar-H), 2989, 2887 (CH<sub>3</sub>), 1629 (C=N azomethine), 1607 (C=N tetrazole ring), 1494 (N=N tetrazole ring), 1403 and 1299 (N=N=N), 1105 and 1009 (tetrazole ring), 854 (C-NO<sub>2</sub>); <sup>1</sup>H-NMR:  $\delta$  ppm: 2.10 (s, 3H, CH<sub>3</sub>); 7.24 (d, 2H, Ar-H); 7.44 (d, 2H, Ar-H); 7.51-7.63 (m, 3H, Ar-H) 8.26 (m, 1H, Ar-H); 8.94 (s, 1H, N=CH); <sup>13</sup>C-NMR:  $\delta$  ppm: 9.11 (CH<sub>3</sub>-Tetrazole), 119.4 (C, tetrazole), 123.3 (C, Ar), 125.09 (2C, Ar), 129.04 (C, Ar), 130.66 (C, Ar), 131.06 (2C, Ar), 133.2 (C-Tetrazole), 133.65 (2C, Ar), 137.42 (C-N), 149.27 (C, Ar), 151.57 (C, Ar) 159.68 (N=CH); MS (ESI) 309 (M+1)<sup>+</sup>. Anal. calcd for C<sub>16</sub>H<sub>12</sub>N<sub>5</sub>O<sub>2</sub>: C, 58.44; H, 3.92; N, 27.26; O, 10.38 Found: C, 58.40; H, 3.93; N, 27.29; O, 10.41.

**(4-(5-Methyl-1H-tetrazol-1-yl)phenyl)(3-nitrobenzylidene)amine (5f)** : Yield: 96%; mp 136-138 °C; IR:  $\nu/\text{cm}^{-1}$ : 3130, 3106 (Ar-H), 2982, 2859 (CH<sub>3</sub>), 1624 (C=N azomethine), 1597 (C=N tetrazole ring), 1485 (N=N tetrazole ring), 1400 and 1293 (N=N=N), 1097 and 1003 (tetrazole ring), 845 (C-NO<sub>2</sub>); <sup>1</sup>H-NMR:  $\delta$  ppm: 2.10 (s, 3H, CH<sub>3</sub>); 7.28 (d, 2H, Ar-H); 7.38 (d, 2H, Ar-H); 7.48-7.51 (m, 3H, Ar-H); 8.51 (s, 1H, N=CH); 8.79 (m, 1H, Ar-H); <sup>13</sup>C-NMR:  $\delta$  ppm: 9.13 (CH<sub>3</sub>-Tetrazole), 119.5 (C, tetrazole), 123.5 (C, Ar), 124.22 (C, Ar), 128.01 (2C, Ar), 130.0 (C, Ar), 131.56 (2C, Ar), 133.25 (C-Tetrazole), 133.88 (C-N), 136.02 (C, Ar), 148.72 (C, Ar) 152.51 (C, Ar), 158.64 (N=CH); MS (ESI) 309 (M+1)<sup>+</sup>. Anal. calcd for C<sub>16</sub>H<sub>12</sub>N<sub>5</sub>O<sub>2</sub>: C, 58.44; H, 3.92; N, 27.26; O, 10.38 Found: C, 58.43; H, 3.94; N, 27.28; O, 10.44.

**(4-(5-Methyl-1H-tetrazol-1-yl)phenyl)(4-nitrobenzylidene)amine (5g)** : Yield: 98%; mp 190-192 °C; IR:  $\nu/\text{cm}^{-1}$ : 3100, 3052 (Ar-H), 2971, 2856 (CH<sub>3</sub>), 1631 (C=N azomethine), 1596 (C=N tetrazole ring), 1506 (N=N tetrazole ring), 1409 and 1279 (N=N=N), 1162 and 1096 (tetrazole ring), 841 (C-NO<sub>2</sub>); <sup>1</sup>H-NMR:  $\delta$  ppm: 2.11 (s, 3H, CH<sub>3</sub>); 7.44 (dd, 2H, Ar-H); 7.52 (dd, 2H, Ar-H); 8.11 (d, 2H, Ar-H); 8.32 (d, 2H, Ar-H); 8.63 (s, 1H, N=CH); <sup>13</sup>C-NMR:  $\delta$  ppm: 9.12 (CH<sub>3</sub>-Tetrazole), 119.67 (C, tetrazole), 123.1 (2C, Ar), 129.16 (2C, Ar), 131.36 (2C, Ar), 132.38 (2C, Ar), 133.49 (C, Ar), 140.89 (C-Tetrazole), 142.02 (C-N), 150.09 (C, Ar), 152.22 (C, Ar), 160.12 (N=CH); MS (ESI) 309 (M+1)<sup>+</sup>. Anal. calcd for C<sub>16</sub>H<sub>12</sub>N<sub>5</sub>O<sub>2</sub>: C, 58.44; H, 3.92; N, 27.26; O, 10.38 Found: C, 58.41; H, 3.93; N, 27.23; O, 10.42.

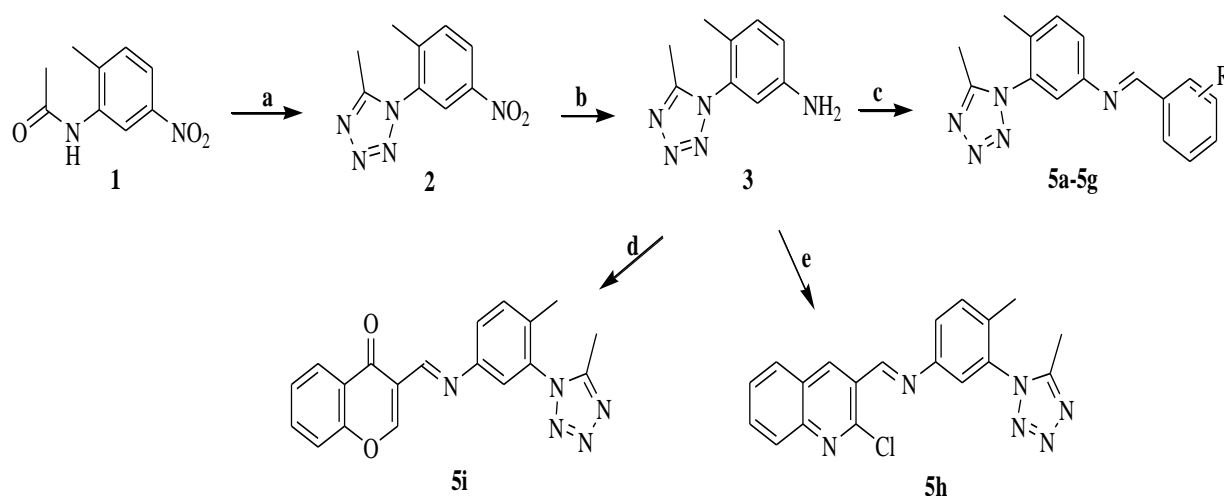
**N-((2-chloroquinolin-3-yl)methylene)-4-(5-methyl-1H-tetrazol-1-yl)benzenamine (5h)** : Yield: 78%; mp 201-202 °C; IR:  $\nu/\text{cm}^{-1}$ : 3114, 3062 (Ar-H), 2963, 2891 (CH<sub>3</sub>), 1623 (C=N azomethine), 1590 (C=N tetrazole ring), 1503 (N=N tetrazole ring), 1400 and 1287 (N=N=N), 1151 and 1088 (tetrazole ring); <sup>1</sup>H-NMR:  $\delta$  ppm: 2.52 (s, 3H, CH<sub>3</sub>); 7.47-7.52 (m, 5H, Ar-H); 7.56-7.57 (d, 2H, Ar-H); 8.61 (s, 1H, Ar-H); 8.92 (s, 1H, Ar-H); 9.02 (s, 1H, N=CH); <sup>13</sup>C-NMR:  $\delta$  ppm: 9.86 (CH<sub>3</sub>-Tetrazole), 122.46 (C, tetrazole), 125.61 (C, Ar), 126.21 (2C, Ar), 127.1 (C, Ar), 131.62 (C, Ar), 134.23 (2C, Ar), 136.51 (C, Ar), 137.21 (C, Ar), 139.05 (C-Tetrazole), 146.0 (C-N), 147.2 (C, Ar), 148.7 (C, Ar), 151.9 (C, Ar), 153.4 (C, Ar), 158.26 (N=CH); MS (ESI) 350 (M+1)<sup>+</sup>. Anal. calcd for C<sub>18</sub>H<sub>13</sub>ClN<sub>6</sub>: C, 61.98; H, 3.76; Cl, 10.16; N, 24.09 Found: C, 61.89; H, 3.81; Cl, 10.23; N, 24.41.

**3-((4-(5-methyl-1H-tetrazol-1-yl)phenylimino)methyl)-4H-chromen-4-one (5i)** : Yield: 87%; mp 162-164 °C; IR:  $\nu/\text{cm}^{-1}$ : 3101, 3064 (Ar-H), 2977, 2865 (CH<sub>3</sub>), 1618 (C=N azomethine), 1591 (C=N tetrazole ring), 1500 (N=N tetrazole ring), 1421 and 1267 (N=N=N), 1168 and 1110 (tetrazole ring); <sup>1</sup>H-NMR:  $\delta$  ppm: 2.62 (s, 3H, CH<sub>3</sub>); 5.72 (s, 1H, Chrom), 7.08 (m, 1H, Ar-H); 7.14 (m, 1H, Ar-H); 7.28-7.31 (m, 2H,

Ar-H); 7.45-7.57 (m, 4H, Ar-H); 7.99 (s, 1H, N=CH);  $^{13}\text{C}$ -NMR:  $\delta$  ppm: 9.78 (CH<sub>3</sub>-Tetrazole), 101.44 (C, Chrom), 105.24 (C, Chrom), 117.26 (C, Tetrazole), 118.05 (C, Chrom), 122.35 (C, Chrom), 122.58 (2C, Chrom), 126.18 (C, Chrom), 126.39 (C, Chrom), 129.18 (2C, Ar), 134.91 (C, Chrom), 141.46 (C-Tetrazole), 142.69 (C-N), 151.57 (C, Ar), 155.91 (N=CH), 182.06 (C=O), MS (ESI) 350 (M+1)<sup>+</sup>. Anal. calcd for C<sub>18</sub>H<sub>13</sub>N<sub>5</sub>O<sub>2</sub>: C, 65.25; H, 3.95; N, 21.14; O, 9.66 Found: C, 65.21; H, 3.97; N, 21.17; O, 9.65.

### 3. RESULTS AND DISCUSSION

**3.1. Chemistry:** The key intermediate 4-methyl-3-(5-methyl-1H-tetrazol-1-yl)benzenamine (**3**) was synthesized according to our previously reported method<sup>44</sup> (Scheme 1). Initially, the cyclization of *N*-(2-methyl-5-nitrophenyl)acetamide (**1**) into 5-methyl-1-(2-methyl-5-nitrophenyl)-1H-tetrazole (**2**) was carried using NaN<sub>3</sub> in the presence of TiCl<sub>4</sub> in acetonitrile under reflux condition. Subsequent reduction of compound (**2**) using NaBH<sub>4</sub> in the presence of Ni(OAc)<sub>2</sub> in water results 4-methyl-3-(5-methyl-1H-tetrazol-1-yl)benzenamine (**3**). Considering the importance of Schiff base templates as bioactive scaffolds, the compound (**3**) was condensed with various substituted aromatic aldehydes in alcohol over a period of 10 - 40 min. to yield the target compounds (**5a-i**) (Scheme 1).



**5; R. a, 4-OCH<sub>3</sub>, b, 2-Cl, c, 3-Cl, d, 4-Cl, e, 2-NO<sub>2</sub>, f, 3-NO<sub>2</sub>, g, 4-NO<sub>2</sub>**

**Scheme 1.** Reaction conditions: a) NaN<sub>3</sub>, TiCl<sub>4</sub>/Acetonitrile, b) NaBH<sub>4</sub>, Ni(OAc)<sub>2</sub>/Water, c) Substituted aldehyde **4**/EtOH, d) 3-formyl chromone/EtOH, e) 2-chloro, 3-formylquinoline/EtOH.

### 3.2. Biological Activities

**In vitro antibacterial activity:** The synthesized 1,5-disubstituted tetrazole linked Schiff base compounds were screened for their *in vitro* antibacterial activities using disk diffusion method by finding the zone of inhibition of the sample against the standard drug, tetracycline. The antibacterial activities of all the

synthesized compounds were tested against four gram-positive pathogens, i.e. i) *Bacillus subtilis*, ii) *Bacillus Megaterium*, iii) *Bacillus cereus* and iv) *Staphylococcus aureus* (ATCC 6538), and six gram-negative pathogens, i.e. i) *Salmonella typhi*, ii) *Salmonella abony*, iii) *Enterobacter aerogenes*, iv) *Escherichia coli* (ATCC 25922), v) *Pseudomonas aerogenosa* (ATCC 9027) and vi) *Shigella boydii*.<sup>28</sup> DMSO was used as solvent to get desired concentrations. The results were recorded by two fold serial dilution method to determine the minimum inhibitory concentration (MIC) as the average diameter of inhibition zone (IZ) of bacterial growth around the disk in mm. All the experiments were carried in triplicates and the mean values were determined.

Antibacterial activity result data (**Table 1**) showed that some of the synthesized compounds exhibited significant activity against all the strains. On the basis of results, it was clear that among the tested compounds, the compounds **5a**, **5b**, **5g** and **5h** showed promising broad spectrum antibacterial activity against *Salmonella typhi* with MIC values ranging between 6 to 13 mg/mL. Compounds **5a**, **5b**, **5g**, **5h** and **5i** exhibited notable antibacterial activity against *Enterobacter aerogenes* while the compound **5a** and **5g** exhibited comparable activity 16 and 17 mg/mL respectively against *Escherichia coli* as compared to standard drug tetracyclin (20 mg/mL). The activity data also showed the compound **5g** having nitro group substitution on aldehyde moiety possesses notable activity against *Pseudomonas aerogenosa*. The compounds **5c**, **5d**, **5e** and **5f** were found to possess poor activity against all the strains used. Based on the structure-activity relationship (SAR) study, it was observed that among all the synthesized tetrazole bearing Schiff bases, the compounds **5b**, **5g** and **5h** were found active towards all the strains used for analysis (**Table 1**).

**In vitro antifungal activity:** The *in vitro* evaluation of the synthesized compounds for antifungal activity was carried out using agar diffusion method by finding the zone of inhibition of the drug sample against the standard drug. The organisms employed in the *in vitro* testing of the compounds were *Candida albicans*, *Saccharomyces cerevisiae* and *Aspergillus niger*. The culture was maintained on Sabouraud's agar (Microbiology grade, Hi Media LABORATORY) medium by periodic sub culturing. DMSO was used as solvent to get desired concentrations. All the experiments were carried in triplicates and the mean values were determined. Apart from putting the controls of standard drug (Nystatin), controls with dimethyl sulphoxide (positive control) and without dimethyl sulphoxide (negative control) were also included in the test.

The antifungal properties of the compounds were tested against the fungal strains *Candida albicans*, *Saccharomyces cerevisiae* and *Aspergillus niger* and the diameters of the zone of inhibition (in mm) of the compounds are listed in Table 2. The compounds **5b**, **5g** and **5h** notably prevented the growth of all the fungal strains whereas rest of the other compounds were inactive or showed the minimum antifungal property.

**Table 1:** *In vitro* antibacterial activity of compounds (**5a–i**).

Compound	Gram-negative bacteria						Gram-positive bacteria			
	<i>Salmonella typhi</i>	<i>Enterobacter aerogenes</i>	<i>Escherichia coli</i>	<i>Pseudomonas aerogenosa</i>	<i>Salmonella typhi</i>	<i>Shigella boydii</i>	<i>Bacillus subtilis</i>	<i>Bacillus megaterium</i>	<i>Staphylococcus aureus</i>	<i>Bacillus cereus</i>
<b>5a</b>	12	11	16	-	-	-	-	-	-	12
<b>5b</b>	12	11	07	07	-	12	09	05	11	09
<b>5c</b>	-	-	-	-	-	11	-	-	-	-
<b>5d</b>	-	-	-	-	-	-	-	-	-	-
<b>5e</b>	-	08	06	-	10	08	-	-	-	-
<b>5f</b>	-	-	11	-	-	-	10	-	-	-
<b>5g</b>	10	07	17	16	08	15	15	09	13	13
<b>5h</b>	06	12	09	08	-	12	14	13	13	12
<b>5i</b>	-	10	-	09	-	-	-	-	-	11
<b>Tetracyclin</b>	22	20	20	33	21	26	25	20	30	25



. **Table 2.** *In vitro* antifungal activity of compounds (**5a–i**)

Compounds	<i>Candida albicans</i>	<i>Saccharomyces cerevisiae</i>	<i>Aspergillus niger</i>
<b>5a</b>	06	-	08
<b>5b</b>	08	08	10
<b>5c</b>	-	-	-
<b>5d</b>	-	-	-
<b>5e</b>	-	-	-
<b>5f</b>	-	-	-
<b>5g</b>	14	12	14
<b>5h</b>	-	10	12
<b>5i</b>	06	-	-
<b>Nystatin</b>	25	20	30

#### 4. CONCLUSIONS

In the present work we have described synthesis, characterization and antimicrobial screening of tetrazole bearing Schiff bases. A total of nine new entities were synthesized and were characterized by IR,  $^1\text{H}$  &  $^{13}\text{C}$  NMR and mass spectral data. The biological data revealed that the newly synthesized compounds showed moderate to significant activity profile. Antibacterial activity data recognized that compounds **5a**, **5b**, **5g**, **5h** and **5i** were most notable against the bacterial strains as compared to the standard drug tetracyclin, while compound **5g** was found the most potent against *Pseudomonas aeruginosa*. Besides, all the compounds were screened for their antifungal activities.

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#### REFERENCES

1. R.N. Jones, M.A., Bacterial resistance: a worldwide problem, *Diagn Microbiol Infect Dis* 31, 1998, 379-388.
2. A. Sandiumenge, T. Lisboa, F. Gomez, P. Hernandez, L. Canadell, J. Rello, Effect on antibiotic diversity on ventilator associated pneumonia caused by ESKAPE organisms. *Chest*. 2011.



3. J.L. Vincent, J. Rello, J. Marshall, E. Silva, A. Anzueto, C.D. Martin, R. Moreno, J. Lipman, C. Gomersall, Y. Sakr, K. Reinhart, EPIC II Group of Investigators, International study of the prevalence and outcomes of infection in intensive care units. *JAMA*. 2009, 302(21), 2323-2329.
4. G. Cornaglia, J. Garau, D.M. Livermore, Living with ESBLs. Introduction. *Clin Microbiol Infect*. 2008;14 Suppl1:1-2. Erratum in *Clin Microbiol Infect*. 2008;14 Suppl 5:21-4.
5. H.W. Boucher, G.H. Talbot, J.S. Bradley, J.E. Edwards, D. Gilbert, L.B. Rice, Bad bugs, no drugs: no ESKAPE! An update from the Infectious Diseases Society of America, *Clin. Infect. Dis*, 2009, 48(1), 1-12.
6. S.F. Cui, Y. Ren, S.L. Zhang, X.M. Peng, G.L.V. Damu, R.X. Geng, C.H. Zhou, synthesis and biological evaluation of a class of quinoline triazoles as potential antimicrobial agents and their interactions with calf thymus DNA, *Bioorg. Med. Chem. Lett*, 2013, 23(11), 3267-3272.
7. B.T. Yin, C.Y. Yan, X.M. Peng, S.L. Zhang, S. Rasheed, R.X. Geng, C. Zhou, Synthesis and biological evaluation of  $\alpha$ -triazolyl chalcones as a new type of potential antimicrobial agents and their interaction with calf thymus DNA and human serum albumin, *Eur. J. Med. Chem*, 2014, 71, 148-159.
8. R.M. Demarinis, J.R.E. Hoover, G.L. Dunn, P. Actor, J.V. Uri, J.A. Weisbach, A new parenteral cephalosporin. SK&F 59962: 7-trifluoromethylthioacetamido-3-(1-tetrazol-5-ylthiomethyl)-3-cephem-4-carboxylic acid. Chemistry and structure activity relationships, *J. Antibiotics*, 1975, 28(6), 463-470.
9. R.S. Upadhyaya, S. Jain, N. Sinha, N. Kishore, R. Chandra, S.K. Arora, Synthesis of novel substituted tetrazoles having antifungal activity, *Eur. J. Med. Chem*, 2004, 39(7), 579-592.
10. K. Pegklidou, C. Koukoulitsa, I. Nicolaou, V.J. Demopoulos, Design and synthesis of novel series of pyrrole based chemotypes and their evaluation as selective aldose reductase inhibitors. A case of bioisosterism between a carboxylic acid moiety and that of a tetrazole, *Bioorg. Med. Chem*, 2010, 18(6), 2107-2114.
11. Matta, C. F.; Arabi, A. A.; Weaver, D. F. The bioisosteric similarity of the tetrazole and carboxylate anions: Clues from the topologies of the electrostatic potential and of the electron density, *Eur. J. Med. Chem*, 2010, 45(5), 1868-1872.
12. B.J. Al-Hourani, S.K. Sharma, J.Y. Mane, J. Tuszyński, V. Baracos, T. Kniess, M. Suresh, J. Pietzsch, F. Wuest, Synthesis and evaluation of 1,5-diaryl-substituted tetrazoles as novel selective cyclooxygenase-2 (COX-2) inhibitors, *Bioorg. Med. Chem. Lett*, 2011, 21(6), 1823.
13. W.A. Carroll, D.M. Kalvin, A.P. Medrano, A.S. Florjancic, Y. Wang, D.L. Donnelly-Roberts, M.T. Namovic, G. Grayson, P. Honoré, M.F. Jarvis, Structure-activity relationships of bioisosteres of a carboxylic acid in a novel class of bacterial translation inhibitors, *Bioorg. Med. Chem. Lett*, 2007, 17(14), 4044-4043.
14. F.F. Zhang, L.L. Gan, C.H. Zhou, Synthesis, antibacterial and antifungal activities of some carbazole derivatives, *Bioorg. Med. Chem. Lett*, 2010, 20(6), 1881.
15. H. Singh, A.S. Chawla, V.K. Kapoor, D. Paul, R.K. Malhotra, 4-Medicinal chemistry of tetrazoles, *Prog. Med. Chem*, 1980, 17, 151-183.

16. R.J. MacFadyen, M. Tree, A.F. Lever, J.L. Reid, Effects of the angiotensin II receptor antagonist Losartan (DuP 753/MK 954) on arterial blood pressor response to infused angiotensin II in the salt-deplete dog, *Clin. Sci*, 1992, 83(5), 549-556.
17. M. Karaman, S. Balta, S.A. Ay, M. Cakar, I. Naharci, S. Demirkol, T. Celik, Z. Arslan, O. Kurt, N. Kocak, H. Sarlak, S. Demirbas, F. Bulucu, E. Bozoglu, The comparative effects of valsartan and amlodipine on vWf levels and N/L ratio in patients with newly diagnosed hypertension. *Clin. Exp. Hypertens*, 2013, 35(7), 516-522.
18. M. Bauer, R.K. Harris, R.C. Rao, D.C. Apperley, C.A. Rodger, NMR study of desmotropy in Irbesartan, a tetrazole-containing pharmaceutical compound, *J. Chem. Soc., Perkin Trans. 2*, 1998, 475-482.
19. C.H. Lee, J.W. Liu, C.C. Li, Y.F. Tang, L.H. Su, Spread of ISCR1 elements containing bla<sub>DHA-1</sub> and multiple antimicrobial resistance genes leading to increase of Flomoxef resistance in extended-spectrum- $\beta$ -lactamase producing *Klebsiella pneumoniae* *Antimicrob. Agents Chemother*, 2011, 55(9), 4058-4063.
20. L. Pochini, M. Galluccio, D. Scumaci, N. Giangregorio, A. Tonazzi, F. Palmieri, C. Indiveri, Interaction of  $\beta$ -lactam antibiotics with the mitochondrial carnitine/acylcarnitine transporter, *Chem. Biol. Interact*, 2008, 173, 187-194.
21. M. Salahuddin, S. Singh, S.M. Shantakumar, Synthesis and antimicrobial activity of some novel benzo thieno pyrimidines, *Rasayan J. Chem*, 2009, 2(1), 167-73.
22. P.B. Mohite, R.B. Pandhare, S.G. Khanage, Synthesis, characterization and antimicrobial activity of some new pyrimidines containing tetrazole, *Bioint. Res. Appl. Chem*, 2012, 2(1), 258-263.
23. C.G. Dave, R.D. Shah, Annellation of triazole and tetrazole systems onto pyrrolo[2,3-d]pyrimidines: Synthesis of tetrazolo[1,5-c]-pyrrolo[3,2-e]-pyrimidines and triazolo[1,5-c]pyrrolo-[3,2-e]pyrimidines as potential antibacterial agents, *Molecules*, 2002, 7(7), 554-565
24. A.A. Bekhit, O.A. El-Sayed, E. Aboulmag , J.Y. Park, Tetrazolo[1,5- a]quinoline as a potential promising new scaffold for the synthesis of novel antiinflammatory and antibacterial agents, *Eur. J. Med. Chem*, 2004, 39, 249–255.
25. U. Natrajan , I. Kaliappan, N.K. Singh, A facile design and efficient synthesis of schiff's bases of tetrazolo [1,5-a] quinoxalines as potential anti-inflammatory and anti-microbial agents, *Der Pharma Chemica*, 2010, 2(1), 159-167.
26. H.N. Patil, D. Varadaraji, S.S. Suban, V.R. Ramasamy, K. Kubendiran, J.S.K.G. Raguraman, S.K. Nalilu, H.N. Pati, Synthesis and evaluation of a series of 1-substituted tetrazole derivatives as antimicrobial agents, *Org. Commun*, 2010; 3(3), 45-56.
27. S.N. Rao, T. Ravisankar, J. Latha, K.S. Babu, Synthesis, characterization and antimicrobial activity of novel biphenyl tetrazoles, *Der Pharma Chemica*, 2012, 4(3), 1093-1103.
28. M.A.M. Taha, S.M. El-Badry, Antimicrobial assessment of some heterocyclic compounds utilizing ethyl 1-aminotetrazole-5-carboxylate, *J. Korean Chem. Soc*, 2010, 54(4), 414-418.

29. M.A.M. Taha, S.M. El-Badry, Novel and efficient synthesis of tetrazolo[1,5-b]-1,2,5-oxadiazepines as antibacterial activities from 1-aminotetrazole-5-carboxylate, *J. Korean Chem. Soc.*, 2011, 55(6), 974-977.
30. S. George, P. Shanmugapandiyan, Synthesis and antimicrobial evaluation of 2-(5-(substituted phenyl)-1H-tetrazol-1-yl)pyridines, *Int. J. Pharm. Pharm. Sci.*, 2011, 4 (3) 104-106.
31. M.D. Altıntop, Z.A. Kaplancikli, G.A. Ciftci, R. Demirel, R. Synthesis and biological evaluation of thiazoline derivatives as new antimicrobial and anticancer agents, *Eur. J. Med. Chem.* 2014, 74(3), 264.
32. S. Vembu, S. Pazhamalai, M. Gopalakrishnan, Synthesis, spectral characterization, and effective antifungal evaluation of 1H-tetrazole containing 1,3,5-triazine dendrimers, *Med. Chem. Res.*, 2016, 25(9), 1916-1924.
33. M. Shakir, S. Khanam, F. Firdaus, A. Latif, M. Aatif, S.I. Al-Resayes, Synthesis, spectroscopic characterization, DNA interaction and antibacterial study of metal complexes of tetraazamacrocyclic Schiff base, *Spectrochim. Acta, A* 93 (2012) 354-362.
34. V. Dhayanithi, S.S. Syed, K. Kumaran, K.R.J. Sankar, R.V. Ragavan, P.S.K. Goud, N.S. Kumari, H.N. Pati, Synthesis of selected 5-thio-substituted tetrazole derivatives and evaluation of their antibacterial and antifungal activities, *J. Serb. Chem. Soc.*, 2011, 76, 165-175.
35. Y. Ozkay, Z. Incesu, N.I. Onder, Y. Tunalı, H. Karaca, I. Isikdag, U. Ucucu, Antimicrobial and anticancer effects of some 2-(substitutedsulfanyl)-N-(5-methyl-isoxazol-3-yl)acetamide derivatives, *Med. Chem. Res.* 2013, 22(1), 211.
36. A.H. Kategaonkar, R.U. Pokalwar, S.S. Sonar, V.U. Gawali, B.B. Shingate, M.S. Shingare, Synthesis, in vitro antibacterial and antifungal evaluations of new  $\alpha$ -hydroxyphosphonate and new  $\alpha$ -acetoxyposphonate derivatives of tetrazolo [1,5-a] quinoline, *Eur. J. Med. Chem.* 2010, 45(3), 1128-1132.
37. N.D. Shashikumar, G. Krishnamurthy, H.S. Bhojyanaik, M.R. Lokesh, K.S. Jithendrakumara, Synthesis of new biphenyl-substituted quinoline derivatives, preliminary screening and docking studies, *J. Chem. Sci.*, 2014, 126, 205-212.
38. A.A. Abu-Hashem, K.M. Abu-Zied, M.F. El-Shehry, Synthetic utility of bifunctional thiophene derivatives and antimicrobial evaluation of the newly synthesized agents, *Monatsh. Chem.*, 2011, 142(5), 539-545.
39. S. George, P. Shanmugapandiyan, Synthesis and antimicrobial evaluation of 2-(5-(substituted phenyl)-1H-tetrazol-1-yl)pyridines, *Int. J. Pharm. Pharm. Sci.* 2012, 4(3), 104-106.
40. S. Vembu, P. Parasuraman, M. Gopalakrishnan, Synthesis, in vitro antifungal and antitubercular evaluation of novel amino pyrimidines based tetrazole derivatives, *J. Pharm. Res.* 2014, 8(10), 1552-1558.
41. H. Adibi, A. Rashidi, M.M. Khodaei, A. Alizadeh, M.B. Majnooni, N. Pakravan, R. Abiri, D. Nematollahi, Catecholthioether derivatives: Preliminary study of *in-vitro* antimicrobial and antioxidant activities, *Chem. Pharm. Bull.*, 2011, 59(9), 1149-1152.

42. Kinali-Demirci, S.; Idil, O.; Disli, A. Synthesis of some novel purine derivatives incorporating tetrazole ring and investigation of their antimicrobial activity and DNA interaction, *Med. Chem. Res.*, 2015, 24, 1218-1225.
43. R.S. Upadhayaya, N. Sinha, S. Jain, N. Kishore, R. Chandra, S.K. Arora, Optically active antifungal azoles: synthesis and antifungal activity of (2R,3S)-2-(2,4-difluorophenyl)-3-(5-{2-[4-aryl-piperazin-1-yl]-ethyl}-tetrazol-2-yl/1-yl)-1-[1,2,4]-triazol-1-yl-butan-2-ol, *Bioorg. Med. Chem.*, 2004, 12(9), 2225-2238.
44. S.G. Vedpathak, R.G. Momle, G.K. Kakade, V.S. Ingle, An improved and convenient route for the synthesis of 5-methyl-1*H*-tetrazol-1-yl substituted benzenamines, *World J. Pharm. Res.*, 2016, 5(12), 1049.

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