Research Article

PHARMACOLOGICAL STUDIES ON DERIVATIVES OF CYTOSINE

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Abstract

The Schiff bases of cytosine (I) 4-((2-hydroxy-3,5-diiodobenzylidene)amino)pyrimidin-2(1H)-one, (II) 4-((4-hydroxy-3-methoxybenzylidene)amino)pyrimidin-2(1H)-one, (III) 4-((3,4,5-trimethoxybenzylidene)amino)pyrimidin-2(1H)-one, (IV) 4-((furan-2-ylmethylene)amino)pyrimidin-2(1H)-one, (V) 4-((pyridin-4-ylmethylene)amino)pyrimidin-2(1H)-one, (VI) 4-((2-hydroxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one, (VII) 4-((4-hydroxy-3-methoxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one and (VIII) 4-((4-diethylamino)-2-hydroxybenzylidene)amino)pyrimidin-2(1H)-one were prepared and characterized by physical and analytical data, FTIR, ¹H NMR, ¹³C NMR spectra and were screened against gram positive bacteria Staphylococcus aureus, Bacillus subtilis and gram negative bacteria Escherichia coli, Klebsiella aerogenes for antibacterial activity and were screened against Aspergillus niger and Candida albicans for antifungal activity by disc diffusion method. Ciprofloxacin and Nystatin were used as standard for bacteria and fungi.

Keywords: Cytosine, Staphylococcus aureus, Bacillus subtilis, Escherichia coli, Klebsiella aerogenes, antifungal activity, Aspergillus niger, Candida albicans, Ciprofloxacin and Nystatin.

Introduction

The findings of the Lancet study are shocking that superbugs kill babies as antibiotic resistance rises. Nearly 25 percent of babies with sepsis died despite timely medical intervention and because of unresponsiveness to treatment, is worrisome. If 80 per cent of infection caused by acinetobacter and in more than 50 per cent of klebsiella infections, the first line treatment fails, it clearly indicates the rampant misuse of antibiotics. There is an indiscriminate use of antibiotics and analgesic drugs. Unaffordable and inaccessible qualitative health-care services easy availability of drugs over the counter without any prescription and inadequate dosage recommendations by unqualified medical practitioners are the main reasons for the superbug menace. It would be major public health disaster if the rampant use of drugs were left unchecked. Recently, derivatives of cytosine were found to have potential non antibiotic resistance antibacterial, antiviral and anticancer properties. Our study clearly indicates on the antibacterial antifungal activities of derivatives of cytosine derived from condensation of aldehyde and ketone with cytosine.
Materials and Methods

Materials

All the reagents used were of AR grade. Commercially available rectified spirit was dried over anhydrous quicklime for 24 hours, filtered and distilled before use (BP 78°C). Dimethylsulphoxide (sigma) and N,N-dimethylformamide (sigma) were used as such cytosine, 3,5-diiodosalicylaldehyde, vaniline, 34,5-trimethoxybenzaldehyde, furfural, pyridine-4-carboxaldehyde, 5-nitrosalicylaldehyde, 5-nitrovaniline and 4-diethylaminosalicylaldehyde were purchased from Alfa Aesar.

Instruments

Melting points were determined using Elico melting point apparatus. Elemental analyses were performed using Elementar Vario EL III. IR spectra of the compounds were recorded with KB pellets with carry 630 FTIR Spectrometer in the 4000-400 cm\(^{-1}\) range. The \(^1\)HNMR and \(^{13}\)CNMR spectra were recorded on a Bruker 400 MHz FT-PMR Spectrometer.

General preparation of derivatives of cytosine

All the azomethine compounds of derivatives of cytosine were prepared as reported in the literature\(^{[3-11]}\) by the followings scheme – 1.

![Scheme 1](attachment:image.png)

Where, Ar-CHO =

![Substituents](attachment:image2.png)
Preparation of (I)4-((2-hydroxy-3,5-diiodobenzylidene)amino)pyrimidin-2(1H)-one

To the hot solution of cytosine 1.11 g (0.01mol) in minimum amount of dimethyl formamide and the DMF solution of 3,5-diiodosalicylaldehyde 3.73 g (0.01mol) were mixed. The reaction mixture was refluxed for 3 hrs. The yellowish-brown coloured solid mass 4-((2-hydroxy-3,5-diiodobenzylidene)amino)pyrimidin-2(1H)-one was formed during refluxing. The crude product was cooled, filtered, washed with ethanol, ether and recrystallized from DMF and then dried over vacuum desiccator.

Preparation of (II)4-((4-hydroxy-3-methoxybenzylidene)amino)pyrimidin-2(1H)-one

Cytosine (1.11 g, 0.01mol) was dissolved in 5ml of hot glacial acetic acid, 1.52 g (0.01mol) of vaniline was dissolved in 5ml of glacial acetic acid and were mixed. The reaction mixture was refluxed with stirring for 5 hours. The mixture was allowed to cool, and poured into ice. The crude solid 4-((4-hydroxy-3-methoxybenzylidene)amino)pyrimidin-2(1H)-one was filtered off and washed with distilled water, then re-crystallized from acetic acid and then dried over vacuum desiccator.

Preparation of (III)4-((3,4,5-trimethoxy benzylidene)amino)pyrimidin-2(1H)-one

25ml of ethanolic solution of cytosine(1.11 g, 0.01mol) was added to 25ml of ethanolic solution of 3,4,5-trimethoxybenzaldehyde(1.96g, 0.01mol). Then three drops of glacial acetic acid was added to the reaction mixture. The reaction mixture was heated and refluxed for about 5 hours at 90°C. The resulting solution was further concentrated by water bath. The product 4-((3,4,5-trimethoxybenzylidene)amino)pyrimidin-2(1H)-one obtained was precipitated, cooled and collected after filtration. The precipitate was purified by washing with distilled water and then with ethanol. The 4-((3,4,5-trimethoxybenzylidene)amino)pyrimidin-2(1H)-one was again recrystallized in ethanol and then dried over vacuum desiccator.

Preparation of (IV)4-((furan-2-ylmethylene)amino)pyrimidin-2(1H)-one

The 4-((furan-2-ylmethylene)amino)pyrimidin-2(1H)-one was prepared by stirring a methanolic solution of cytosine (1.11 g, 0.01mol) with furfural (96 g, 0.01mol) in 1:1 stoichiometric ratio at room temperature over 24 hours. The precipitate obtained were filtered and washed with methanol and recrystallized from methanol and then dried over vacuum desiccator.

Preparation of (V)4-((pyridin-4-ylmethylene)amino)pyrimidin-2(1H)-one

A mixture of Pyridine-4-carboxaldehyde 1.07 g (0.01mol) and cytosine 1.11 g (0.01mol) were grained with a pestle in an open mortar at room temperature for 3 minutes. To this reaction mixture sulphuric acid 2 drops and 20ml DMF were added and grained for 5 minutes. On completion of reaction as monitored by TLC, the light greenish-colored solid 4-((pyridin-4-ylmethylene)amino)pyrimidin-2(1H)-one was separated out. The obtained solid was isolated by simple Buchner filtration and was recrystallized from DMF and then dried over vacuum desiccator.

Preparation of (VI)4-((2-hydroxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one

2.2 grams of cytosine (0.02mol) was mixed with 3.3g of 5-nitosalicylaldehyde (0.02mol) and was grained well in acidic acid medium at room temperature. The mixture was transferred into hundred milliliter Round Bottom flask and was refluxed for six hours in oil bath. The solid product 4-((2-hydroxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one was filtered and washed with ethanol and recrystallized in DMSO and then dried over vacuum desiccator.
Preparation of (VII) 4-((4-hydroxy-3-methoxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one

A mixture of 5-nitrovaniline (1.97 g, 0.01 mol) and cytosine (1.11 g, 0.01 mol) was grained in a mortar with a pestle made of porcelain for 10 minutes. The mixture turned pasty after few minutes of grinding. It was grained till yellow colour product appears. The mixture was left overnight. The resultant product 4-((4-hydroxy-3-methoxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one was recrystallized using ethanol and then dried over vacuum desiccator.

Preparation of (VIII) 4-((4-(diethylamino)-2-hydroxybenzylidene)amino)pyrimidin-2(1H)-one

4-((4- (diethylamino)-2-hydroxybenzylidene)amino)pyrimidin-2(1H)-one was prepared from equimolar quantity of cytosine (1.11 g, 0.01 mol) and 4-diethylaminosalicylaldehyde (1.93 g, 0.01 mol) in 30 ml of methanol were heated at 70°C on water bath for 4-hrs in presence of few drops of glacial acetic acid. The crude product were obtained after removal of methanol under reduced pressure. The products were recrystallized from methanol and then dried over vacuum desiccator.

Antimicrobial susceptibility

Principle

Disc impregnated with known concentration of antibacterial drug are placed on an agar plate that has been inoculated uniformly over the entire plate with a culture of the bacterium to be tested. The plate is incubated for 18 to 24 hours at 37°C. During this period, the antibacterial agent diffuses through the agar and may prevent the growth of the organism. Effectiveness of susceptibility is proportional to the diameter of the inhibition zone around the disc. Organisms which grow up to the edge of the disc are resistant.

Procedure

The plate was labeled with the name of the culture, sample and standard at the bottom of the plate. Then sterile cotton swab on a wooden applicator stick was dipped into the bacterial suspension. Excess fluid was removed by rotating the swab and rubbed gently over the plate to obtain uniform distribution of the inoculums. The sterile disc was held on the inoculated plate with the help of micropipette. The sample was leveled in the sterile disc and incubated at 37°C in an incubator. After incubation, the diameter of the zone of inhibition of growth was measured.

Results and Discussion

The physical and analytical data of (I) 4-((2-hydroxy-3,5-diodobenzylidene)amino)pyrimidin-2(1H)-one, (II) 4-((4-hydroxy-3-methoxybenzylidene)amino)pyrimidin-2(1H)-one, (III) 4-((3,4,5-trimethoxybenzylidene)amino)pyrimidin-2(1H)-one, (IV) 4-((furan-2-ylmethylene)amino)pyrimidin-2(1H)-one, (V) 4-((pyridin-4-ylmethylen)amino)pyrimidin-2(1H)-one, (VI) 4-((2-hydroxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one, (VII) 4-((4-hydroxy-3-methoxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one and (VIII) 4-((4-diethylamino)-2-hydroxybenzylidene)amino)pyrimidin-2(1H)-one are given in Table 2.
[I] 4-((2-hydroxy-3,5-diiodobenzylidene) amino)pyrimidin-2(1H)-one

FTIR (cm⁻¹): 3587 & 679 cm⁻¹ (\(\text{\(-\text{O-H}\)}\)), 3285 & 650 cm⁻¹ (\(\text{\(-\text{N-H}\)}\)), 1687 cm⁻¹ (\(\geqslant\text{C=O}\)), 1633 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)), 1624 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)), 1210 cm⁻¹ (\(\text{Ar-OH}\)) & 589 cm⁻¹ (\(\text{Ar-I}\))

¹HNMR δ (ppm): 8.48 (s, 1H), 8.43 (d, 1H), 8.07 (s, 1H), 7.85 (s, 1H), 7.88 (s, 1H), 5.40 (d, 1H) & 5.35 (s, 1H)

¹³CNMR δ (ppm): 163.7 (s), 160.1 (s), 156.3 (s), 153.2 (s), 141.5 (s), 128.0 (s), 127.4 (s), 104.8 (s), 104.0 (d), 60.8 (s) & 56.1 (s)

[II] 4-((4-hydroxy-3-methoxybenzylidene) amino)pyrimidin-2(1H)-one

FTIR (cm⁻¹): 3606 & 690 cm⁻¹ (\(\text{\(-\text{O-H}\)}\)), 3410 & 750 cm⁻¹ (\(\text{\(-\text{N-H}\)}\)), 1710 cm⁻¹ (\(\geqslant\text{C=O}\)), 1640 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)), 1610 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)), 1240 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)), 1190 cm⁻¹ (\(\text{Ar-CR}\)) & 1140 cm⁻¹ (\(\text{Ar-O-R}\))

¹HNMR δ (ppm): 8.48 (s, 1H), 8.43 (d, 1H), 8.00 (s, 1H), 7.52 (s, 1H), 7.34 (d, 1H), 6.91 (d, 1H), 5.40 (d, 1H), 5.35 (s, 1H) & 3.83 (s, 3H)

¹³CNMR δ (ppm): 163.7 (s), 160.1 (s), 156.3 (s), 151.0 (s), 149.3 (s), 127.4 (s), 127.3 (s), 122.9 (s), 117.0 (s), 112.1 (s), 104.8 (s) & 56.1 (s)

[III] 4-((3,4,5-trimethoxybenzylidene)amino)pyrimidin-2(1H)-one

FTIR (cm⁻¹): 3289 & 805 cm⁻¹ (\(\text{\(-\text{N-H}\)}\)), 1732 cm⁻¹ (\(\geqslant\text{C=O}\)), 1633 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)), 1606 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)), 1237 cm⁻¹ (\(\text{Ar-CR}\)) & 1120 cm⁻¹ (\(\text{Ar-O-R}\))

¹HNMR δ (ppm): 8.48 (s, 1H), 8.43 (d, 1H), 8.00 (s, 1H), 7.14 (s, 2H), 5.40 (d, 1H) & 3.83 (s, 9H)

¹³CNMR δ (ppm): 163.7 (s), 160.1 (s), 156.3 (s), 153.2 (s), 141.5 (s), 128.0 (s), 127.4 (s), 104.8 (s), 104.0 (d), 60.8 (s) & 56.1 (s)

[IV] 4-((furan-2-ylmethylene)amino)pyrimidin-2(1H)-one

FTIR (cm⁻¹): 3270 & 750 cm⁻¹ (\(\text{\(-\text{N-H}\)}\)), 1700 cm⁻¹ (\(\geqslant\text{C=O}\)), 1640 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)), 1620 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)), 1230 cm⁻¹ (\(\text{Ar-O-R}\)) & 1040 cm⁻¹ (\(\text{\(-\text{N-CH}\)}\))

¹HNMR δ (ppm): 8.43 (d, 1H), 8.00 (s, 1H), 7.75 (d, 1H), 7.50 (s, 1H), 6.93 (d, 1H), 6.52 (t, 1H) & 5.40 (d, 1H)

¹³CNMR δ (ppm): 163.7 (s), 160.1 (s), 156.3 (s), 149.1 (s), 144.4 (s), 127.4 (s), 118.9 (s), 112.6 (s) & 104.8 (s)

[V] 4-((pyridin-4-ylmethylene)amino) pyrimidin-2(1H)-one

FTIR (cm⁻¹): 3300 & 770 cm⁻¹ (\(\text{\(-\text{N-H}\)}\)), 1640 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)), 1620 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)) & 1110 cm⁻¹ (\(\text{\(-\text{N-CH}\)}\))

¹HNMR δ (ppm): 8.66 (d, 2H), 8.43 (s, 1H), 8.00 (s, 1H), 7.98 (d, 2H), 7.50 (s, 1H) & 5.40 (d, 1H)

¹³CNMR δ (ppm): 163.7 (s), 160.1 (s), 156.3 (s), 149.4 (d), 144.3 (s), 127.4 (s), 120.4 (s) & 104.8 (s)

[VI] 4-((2-hydroxy-5-nitrobenzylidene)amino) pyrimidin-2(1H)-one

FTIR (cm⁻¹): 3622 & 778 cm⁻¹ (\(\text{\(-\text{O-H}\)}\)), 3271 & 814 cm⁻¹ (\(\text{\(-\text{N-H}\)}\)), 1741 cm⁻¹ (\(\geqslant\text{C=O}\)), 1651 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)), 1633 cm⁻¹ (\(\text{\(-\text{N=CH}\)}\)) & 1480 cm⁻¹ (\(\text{Ar-NO}_{2}\))

\[ ^1\text{HNMR} \delta \text{ (ppm): } 8.48 \text{ (s, 1H)}, 8.43 \text{ (d, 1H)}, 8.35 \text{ (s, 1H)}, 8.05 \text{ (d, 1H)}, 8.00 \text{ (s, 1H)}, 7.28 \text{ (d, 1H)}, 5.40 \text{ (d, 1H)} \text{ and } 5.35 \text{ (s, 1H)} \]

\[ ^{13}\text{CNMR} \delta \text{ (ppm): } 167.2 \text{ (s)}, 163.7 \text{ (s)}, 160.1 \text{ (s)}, 156.3 \text{ (s)}, 140.6 \text{ (s)}, 128.6 \text{ (s)}, 127.4 \text{ (s)}, 125.5 \text{ (s)}, 119.4 \text{ (s)}, 116.9 \text{ (s) and } 104.8 \text{ (s)} \]

[\text{VII}] 4-(4-hydroxy-3-methoxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one

\[ \text{FTIR (cm}^{-1}\text{): } 3280 \text{ and } 796 \text{ cm}^{-1} (\text{N=H}), 3262 \text{ and } 751 \text{ cm}^{-1} (\text{O=H}), 1741 \text{ cm}^{-1} (\text{C=O}), 1642 \text{ cm}^{-1} (\text{N=C}), 1633 \text{ cm}^{-1} (\text{C=C}) \text{ and } 1570 \text{ cm}^{-1} (\text{C=O}) \]

\[ ^1\text{HNMR} \delta \text{ (ppm): } 8.48 \text{ (s, 1H)}, 8.43 \text{ (d, 1H)}, 7.91 \text{ (s, 2H)}, 5.40 \text{ (s, 1H)}, 5.35 \text{ (s, 1H)} \text{ and } 3.83 \text{ (s, 3H)} \]

\[ ^{13}\text{CNMR} \delta \text{ (ppm): } 163.7 \text{ (s), 160.1 \text{ (s), 156.3 \text{ (s), 140.3 \text{ (s), 138.1 \text{ (s), 128.2 \text{ (s), 127.4 \text{ (s), 118.2 \text{ (s), 116.5 \text{ (s), 104.8 \text{ (s) and 56.1 (s)} \text{.} }} \]

[\text{VIII}] 4-((4-(diethylamino)-2-hydroxy benzylidene)amino)pyrimidin-2(1H)-one

\[ \text{FTIR (cm}^{-1}\text{): } 3253 \text{ and } 841 \text{ cm}^{-1} (\text{N=H}), 3145 \text{ and } 688 \text{ cm}^{-1} (\text{O=H}), 3064 \text{ cm}^{-1} (\text{C=O}), 1678 \text{ cm}^{-1} (\text{C=O}), 1624 \text{ cm}^{-1} (\text{N=O}) \text{ and 1237 cm}^{-1} (\text{O=O}) \]

\[ ^1\text{HNMR} \delta \text{ (ppm): } 8.48 \text{ (s, 1H)}, 8.43 \text{ (d, 1H)}, 8.00 \text{ (s, 1H)}, 7.48 \text{ (d, 1H)}, 6.37 \text{ (d, 1H)}, 6.30 \text{ (s, 1H)}, 5.40 \text{ (d, 1H)}, 3.41 \text{ (q, 4H)} \text{ and 1.15 (t, 6H)} \]

\[ ^{13}\text{CNMR} \delta \text{ (ppm): } 163.7 \text{ (s), 162.0 \text{ (s), 160.1 \text{ (s), 156.3 \text{ (s), 153.3 \text{ (s), 132.8 \text{ (s), 127.4 \text{ (s), 108.4 \text{ (s), 104.8 \text{ (s), 104.5 \text{ (s), 47.1 (d) and 12.9 (s)}} \text{.} }} \]

Table 2. The physical and analytical data of derivatives of cytosine

<table>
<thead>
<tr>
<th>Derivatives of Cytosine</th>
<th>Molecular Weight</th>
<th>Nature of Appearance</th>
<th>% of yield</th>
<th>Elemental Analysis (in %)</th>
</tr>
</thead>
<tbody>
<tr>
<td>[I] C₁₁H₇I₂N₅O₂</td>
<td>467.00</td>
<td>Yellow Crystalline Solid</td>
<td>85</td>
<td>C 28.29, H 1.51, O 6.85, N 9.00, I 54.35</td>
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<tr>
<td>[II] C₁₂H₁₁N₃O₃</td>
<td>245.23</td>
<td>Yellow Crystalline Solid</td>
<td>87</td>
<td>C 58.77, H 4.52, O 19.57, N 17.13, I -</td>
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<tr>
<td>[III] C₁₄H₁₄N₃O₄</td>
<td>289.28</td>
<td>Yellow Crystalline Solid</td>
<td>68</td>
<td>C 58.13, H 5.23, O 22.12, N 14.53, I -</td>
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<tr>
<td>[IV] C₉H₇N₃O₂</td>
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<td>79</td>
<td>C 57.14, H 3.73, O 16.92, N 22.21, I -</td>
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<td>[V] C₁₀H₈N₄O</td>
<td>200.19</td>
<td>Yellow Crystalline Solid</td>
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<td>[VI] C₁₁H₈N₄O₄</td>
<td>260.19</td>
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<td>84</td>
<td>C 50.77, H 3.10, O 24.60, N 21.53, I -</td>
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<tr>
<td>[VII] C₁₂H₁₀N₄O₅</td>
<td>290.23</td>
<td>Yellow Crystalline Solid</td>
<td>86</td>
<td>C 49.66, H 3.47, O 27.56, N 19.30, I -</td>
</tr>
<tr>
<td>[VIII] C₁₅H₁₈N₄O₂</td>
<td>286.329</td>
<td>Yellow Crystalline Solid</td>
<td>77</td>
<td>C 62.92, H 6.34, O 11.18, N 19.57, I -</td>
</tr>
</tbody>
</table>
Antibacterial bioassay

Antibacterial activities\cite{12,13} of derivatives of cytosine were screened against bacterial gram positive bacteria *Staphylococcus aureus*, and gram negative bacteria *Escherichia coli*, *Klebsiella aerogenes* and *Bacillus subtilis* by disc diffusion method and the results obtained were formulated in Table.3 and Fig. 1-4 The experiments were carried out in DMSO solution at a concentration of 100ppm using Muller Hinton agar media. Ciprofloxacin was used as a standard.

Antifungal bioassay

Antifungal\cite{14,15} screening of derivatives of cytosine were carried out against *Aspergillus niger* and *Candida albicans* by disc diffusion method and the results obtained were formulated in Table.3 and Fig. 5 and 6. The test was carried out in DMSO solution at a concentration of 100 ppm. Results were compared with Nystatin at the same concentration.

Table 3. Antibacterial and antifungal activity of derivatives of Cytosine

<table>
<thead>
<tr>
<th>Compounds</th>
<th>Gram negative Bacterias</th>
<th>Gram positive Bacterias</th>
<th>Fungi</th>
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<tbody>
<tr>
<td></td>
<td><em>E. coli</em></td>
<td><em>K. aerogenes</em></td>
<td><em>B. subtilis</em></td>
</tr>
<tr>
<td>I</td>
<td>33</td>
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<td>II</td>
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<tr>
<td>VIII</td>
<td>18</td>
<td>15</td>
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<tr>
<td>Standard</td>
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<td>30</td>
<td>40</td>
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</table>
The antibacterial and antifungal activity of azomethine compounds I- VIII (table. 3 and figure 1-6) clearly indicate that they inhibit the growth of tested bacteria and fungi in the decreasing order I>II>III>IV>V>VI>VII>VIII. Azomethine compounds I- VIII prevent bacterial reproduction by acting as an antimetabolite to paraamino benzoic acid (PABA), where PABA is a vital component in the biosynthesis of tetrahydrofolic acid. Competitive inhibition of PABA processing enzymes by I-VIII ultimately blocks the action of dihydrofolic acid synthetase, and therefore prevents dihydrofolic acid formation. As bacteria are unable to take up tetrahydrofolic acid from their surroundings, inhibition of dihydrofolic acid synthetase will starve the bacteria of thymidine and uridine. These two nucleosides are required for DNA replication and transcription, therefore cell growth and division is disrupted, and thus provides enough time for the body’s own immune system to eliminate the bacterial threat[17].

The nature of bonding and structure of azomethine organic compounds were elucidated[3- 5] by the elemental analysis, Melting Point, FTIR, 1H NMR, 13C NMR, Chromatography and Molar ratio methods Gomathiet.al were prepared 4-(3-ethoxy-2-hydroxybenzoxazolylideneamino-N-(thiazole-2-yl)-benzenesulfonamide, Mohamed et.al and were prepared 4-(phenyl-propylideneamino)-benzenesulfonamide. In accordance with the data obtained from antibacterial activities of 4-((2-hydroxy-3,5-diiodobenzylidene)amino)benzenesulfonamide and 4-((2-hydroxy-3,5-diiodobenzylidene)amino)-N-(thiazole-2-yl)-benzenesulfonamide, were moderately inhibited the growth of tested bacteria but our derivatives of cytosine cytokine (I) 4-((2-hydroxy-3,5-diiodobenzylidene)amino)pyrimidin-2(1H)-one, (II) 4-((4-hydroxy-3-methoxybenzylidene)amino)pyrimidin-2(1H)-one, (III) 4-((3,4,5-trimethoxybenzylidene)amino)pyrimidin-2(1H)-one, (IV) 4-((furan-2-ylmethylene)amino)pyrimidin-2(1H)-one, (V) 4-((pyridin-4-ylmethylene)amino)pyrimidin-2(1H)-one, (VI) 4-((2-hydroxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one, (VII) 4-((4-hydroxy-3-methoxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one and (VIII) 4-((4-diethylamino)-2-hydroxybenzylidene)amino)pyrimidin-2(1H)-one were highly inhibited the growth of tested bacteria.

In accordance with the data obtained from antifungal activities of 4-(3-ethoxy-2-hydroxybenzoxazolylideneamino-N-(thiazole-2-yl)-benzenesulfonamide[16] (Gomathi et.al) were moderately inhibited the growth of tested fungi but our derivatives of cytosine (I) 4-((2-hydroxy-3,5-diiodobenzylidene) amino)pyrimidin-2(1H)-one, (II) 4-((4-hydroxy-3-methoxybenzylidene) amino)pyrimidin-2(1H)-one, (III) 4-((3,4,5-trimethoxybenzylidene) amino)pyrimidin-2(1H)-one, (IV) 4-((furan-2-ylmethylene)amino) pyrimidin-2(1H)-one, (V) 4-((pyridin-4-ylmethylene)amino)pyrimidin-2(1H)-one, (VI) 4-((2-hydroxy-5-nitrobenzylidene) amino)pyrimidin-2(1H)-one, (VII) 4-((4-hydroxy-3-methoxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one and (VIII) 4-((4-diethylamino)-2-hydroxybenzylidene)amino)pyrimidin-2(1H)-one were highly inhibited the growth of tested fungi. From the results and previous work, antibacterial and antifungal activity studies were indicated that iodine substituted derivatives of cytosine were more active against bacteria and fungi than other derivatives of cytosine.

**Conclusion**

The derivatives of cytosine (I) 4-((2-hydroxy-3,5-diiodobenzylidene) amino) pyrimidin-2(1H)-one, (II) 4-((4-hydroxy-3-methoxybenzylidene) amino) pyrimidin-2(1H)-one, (III) 4-((3,4,5-trimethoxybenzylidene)amino)pyrimidin-2(1H)-one, (IV) 4-((furan-2-ylmethylene)amino)pyrimidin-2(1H)-one, (V) 4-((pyridin-4-ylmethylene)amino) pyrimidin-2(1H)-one, (VI) 4-((2-hydroxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one, (VII) 4-((4-hydroxy-3-methoxy-5-nitrobenzylidene)amino)pyrimidin-2(1H)-one and (VIII) 4-((4-diethylamino)-2-hydroxybenzylidene)amino)pyrimidin-2(1H)-one were highly inhibited by the derivatives of cytosine.
References

7. Miller and Rhode,Ber 2 5, 2053 (1892).