SYNTHESIS AND BIOLOGICAL EVALUATION OF SOME 1, 3-BENZTHIAZOLES DERIVATIVES

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INTRODUCTION
The enhancement of bacterial resistance of pathogens to currently available antibiotics constitutes a serious public health threat. So, there is constant need of new antibacterial agent having novel mechanism to act against the harmful pathogens. The present study deals with antimicrobial evaluation of some substituted benzothiazole were synthesized by reacting substituted aniline with potassium thiocyanate it forms N-substituted benzothiazol-2-yl) amines this upon reacting with DMF, CS₂ it forms N-Substituted dimethyl benzo[d]thiazol-2-yl) carbonodithioimidate finally on reacting with o-phenylenediamine it forms N-Substituted (benzo[d]thiazol-2-yl)-1H-benzo[d]imidazol-2-amine. All the synthesized compounds were screened for antibacterial activity against the representative panel of Gram-positive and Gram-negative bacteria strains. The biological screening identified compounds 3c and 3h as the most active ones showing an interesting antibacterial activity. It was also found that compounds 3c, 3e, 3f, 3h showed very good antibacterial activity whereas the other entire compound showed mild to moderate antibacterial activity as compared to standard drug.

Keywords: Antibacterial, Gram negative micro-organism, Gram positive micro-organism.

ABSTRACT
The enhancement of bacterial resistance of pathogens to currently available antibiotics constitutes a serious public health threat. So, there is constant need of new antibacterial agent having novel mechanism to act against the harmful pathogens. The present study deals with antimicrobial evaluation of some substituted benzothiazole were synthesized by reacting substituted aniline with potassium thiocyanate it forms N-substituted benzothiazol-2-yl) amines this upon reacting with DMF, CS₂ it forms N-Substituted dimethyl benzo[d]thiazol-2-yl) carbonodithioimidate finally on reacting with o-phenylenediamine it forms N-Substituted (benzo[d]thiazol-2-yl)-1H-benzo[d]imidazol-2-amine. All the synthesized compounds were screened for antibacterial activity against the representative panel of Gram-positive and Gram-negative bacteria strains. The biological screening identified compounds 3c and 3h as the most active ones showing an interesting antibacterial activity. It was also found that compounds 3c, 3e, 3f, 3h showed very good antibacterial activity whereas the other entire compound showed mild to moderate antibacterial activity as compared to standard drug.

Keywords: Antibacterial, Gram negative micro-organism, Gram positive micro-organism.
activity. Benzothiazoles are also found to be antimicrobial, antifungal, anticancer, and anti-inflammatory activity. These biological data prompted us to synthesize some new benzothiazole derivatives containing imidazol ring.

In the present study we reported here the synthesis and antimicrobial activity of several N-Substituted (benzol[d]thiazol-2-yl)-1H-benzo[d]imidazol-2-amine. Incorporation of imidazole moiety into the second position of the benzothiazole ring may result in compounds having better antimicrobial activity.

**Experimental Studies**

**Method and Materials**
The melting points were determined in open capillary tubes in a Hicon melting point apparatus. The elemental analyses (C, H, N) of all compounds were performed on the CHNS Elimentar (Anaylsen systime GmbH) Germany Vario EL III. All the Fourier transform infra red (FTIR) spectra were recorded in KBr pellets on a Jasco FT/IR 410 spectrometer. The 1H NMR spectra were taken on a Bruker 400 Ultra shieldTM (400 MHz) NMR spectrometer. Chemical shifts (d) are expressed in parts per million relative to tetramethylsilane (TMS) as an internal standard. The homogeneity of the compounds was checked by thin layer chromatography (TLC) on silica gel G (Merck) coated plates by using toluene: ethylacetate: formic acid (5:4:1) as solvent system. Iodine chamber and UV lamp were used for the visualization of TLC spots.

**Synthesis**

**General Procedure for Synthesis of N-Substituted-benzothiazol-2-yl)amines (I a-h)**
A mixture of substituted aniline (0.01 mol) and potassium thiocyanate (0.01 mol) in glacial acetic acid was cooled and stirred. To this solution, bromine (0.01 mol) was added drop wise at such a rate to keep the reaction temperature below 10 °C throughout the addition. Stirring was continued for additional 3 h and the separated hydrochloride salt was filtered, washed with acetic acid and dried. It was dissolved in hot water and neutralized with aqueous ammonia solution (25 %). The precipitate obtained was filtered, washed with water, dried and recrystallized to afford the N-substituted-1,3-benzothiazol-2-amine.

**General Procedure for Synthesis of N-Substituted dimethyl benzo[d]thiazol-2-yl)carbonodithioimidate (II a-h)**
To stirred cold solution of I (0.05 mol) in DMF (25cm3), 20 M-NaOH (5 cm3), carbon disulfide (8 cm3), and methyl iodide (0.05 mol) were added and the stirring was continued for additional 4 h. The mixture was poured into cold water and the formed solid was crystallized from benzene.

**General Procedure for Synthesis of N-Substituted (benzol[d]thiazol-2-yl)-1H-benzo[d]imidazol-2-amine (III a-h)**
A mixture of II (0.04 mol) and o-phenylenediamine (0.04 mol) in DMF (30 cm3) was refluxed for 8h. After cooling, the formed solid was crystallized from ethanol.

**Spectral Analysis**

**N-(benzo[d]thiazol-2-yl)-1H-benzo[d]imidazol-2-amine (3a)**
IR(CDCL3): NH (3339), Ar=C-H (3020), C=N (1610), CN (1453), CS (1259), 1H NMR (DMSO): 6.29-6.54 (8H of Ar-H), 3.5, 4.0 (2H, NH, exchangeable).

**N-(7-chlorobenzo[d]thiazol-2-yl)-1H-benzo[d]imidazol-2-amine (3b)**
IR NH (3325), 3034(Ar=C-H) 1597(C=N), 1455(C-N), 1255(C-S), 916 (C-Cl), 1H NMR (DMSO): 5.93-6.17 (8H of Ar-H), 4.0, 5.0 (2H, NH, exchangeable).

**N-(6-chlorobenzo[d]thiazol-2-yl)-1H-benzo[d]imidazol-2-amine (3c)**
IR NH (3330), 3036(Ar=C-H) 1602(C=N), 1470(C-N), 920 (C-Cl), 1H NMR (DMSO): 6.83-7.20 (8H of Ar-H), 4.5, 5.0 (2H, NH, exchangeable).

**2-(1H-benzo[d]imidazol-2-ylamino)benzo[d]thiazol-6-ol (3d)**
IR NH (3339), Ar=C-H (3020), C=N (1610), CN (1453), CS (1259), O-H Stretching(3466), 1H NMR (DMSO): 7.10-7.42 (8H of Ar-H), 2.0, 4.0 (2H, NH, exchangeable).

**N-(6-nitrobenzo[d]thiazol-2-yl)-1H-benzo[d]imidazol-2-amine (3e)**
IR NH (3338), 3033(Ar=C-H) 1617(C=N), 1480(C-N), 1266(C-S), 1359(C-N=O), 1H NMR (DMSO): 7.22-7.90 (8H of Ar-H), 3.0, 4.0 (2H, NH, exchangeable).
N2-(1H-benzo[d]imidazol-2-yl)benzo[d]
thiazole-2,6-diamine (3f)
IR- NH (3339), Ar-C-H (3020), C=N (1610),
CN (1453), CS (1259), N-H Stretching of
amine (3375), 1H NMR (DMSO): 6.84-6.52 (8H
of Ar-H), 3.0, 4.2 (2H, N-H, exchangeable).

N-(6-methylbenzo[d]thiazol-2-yl)-1H-benzo
d[d]imidazol-2-amine (3g)
IR- NH (3339), Ar-C-H (3020), C=N (1610),
CN (1453), CS (1259), (2839.22) C-H
Stretching in CH3, 1H NMR (DMSO): 6.28-6.95
(8H of Ar-H), 3.3, 4.6 (2H, N-H, exchangeable).

N-(6-methoxybenzo[d]thiazol-2-yl)-1H-benzo
d[d]imidazol-2-amine (3h)
IR- NH (3320), 3034 (Ar=C-H) 2951 (C-CH3),
1625 (C=N), 1470 (C-N), 1260 (C-S), 1291 (C-O),
1H NMR (DMSO): 6.39-6.72 (8H of Ar-H), 3.0,
5.0 (2H, N-H, exchangeable).

Antibacterial Activity
All the newly synthesized compounds (3a-h)
were evaluated for in vitro antibacterial
activity against gram positive and gram
negative bacterial strains such as Bacillus
subtilis, Bacillus pumilus, Escherichia coli and
Pseudomonas aeruginosa at concentration 100
µg/mL by disc diffusion method29 by using
DMSO as solvent control and nutrient agar
was employed as culture media. After 24 h
of incubation at 37°C, the zone of inhibition
was measured in mm. The activity was compared
with known antibiotic ciprofloxacin and
the data was represented in Table 2.

RESULTS AND DISCUSSION
The results of antimicrobial evaluation suggest
that all the compounds have very good
potential to act as antibacterial agents.
Compound 3c, 3e, 3f, 3h showed very good
activities against the entire test
microorganism. All synthesized compounds
are more active against gram positive
microorganism as compared to gram negative
one. Compound having substitution in the p-
position (3c) are more active than m-
substitution (3b). Compound 3c and 3h has
highest antibacterial activity against all the test
microorganisms. The zone inhibition is
described in Table 2.

Table 1: Characterization of Compounds

<table>
<thead>
<tr>
<th>Compound</th>
<th>R</th>
<th>Mol. Formula</th>
<th>Mol. Weight</th>
<th>Melting Point °C</th>
<th>Yield %</th>
<th>C</th>
<th>H</th>
<th>N</th>
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<tr>
<td>3a</td>
<td>H</td>
<td>C14H12N6S</td>
<td>266.32</td>
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<td>55</td>
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<td>m-Cl</td>
<td>C14H11ClN6S</td>
<td>300.77</td>
<td>206</td>
<td>61</td>
<td>55.91</td>
<td>3.02</td>
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<tr>
<td>3c</td>
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<td>C14H11ClN6S</td>
<td>300.77</td>
<td>202</td>
<td>57</td>
<td>55.92</td>
<td>3.00</td>
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<td>3d</td>
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<td>C14H11N6OS</td>
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<td>61</td>
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<td>3e</td>
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<td>C14H11N6O3</td>
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<td>196</td>
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<td>54.01</td>
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<tr>
<td>3f</td>
<td>p-NH2</td>
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<td>281.34</td>
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<td>P-CH3</td>
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<td>3h</td>
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<td>64</td>
<td>60.79</td>
<td>4.08</td>
<td>19.91</td>
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Table No 2: Antibacterial Activity of Compounds 3a-h.

<table>
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<tr>
<th>Compound Code</th>
<th>B. subtilis</th>
<th>B. pumilus</th>
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<th>P. aeruginosa</th>
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<tr>
<td></td>
<td>100 µg</td>
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<td>3a</td>
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</table>

*Each value is an average of three independent determination ± Standard deviation. Note: + denotes
no activity, 8-12 mm poor activity, 13-17 mm moderate activity, 18-20 mm and above good activity.
CONCLUSION
All the 08 newly synthesized compounds were screened for antibacterial activity studies at a concentration of 100 µg/ mL using DMSO as a control and ciprofloxacin used as standard against gram positive and gram negative bacteria. The data in the Table 2 indicates that among the synthesized compounds 3c, 3e, 3f and 3h compounds was found to possess a broad spectrum activity. However, the activities of the tested compounds are much less than those of standard antibacterial agents used.

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REFERENCES