INTRODUCTION

An antimicrobial is an agent that kills microorganisms or inhibits their growth. The search for new molecules with broad spectrum of antimicrobial properties is challenging task to researchers. In view of continuous enhancement of microbial infection in society, the triazole ring system possess appreciable biocompatibility and can play a pivotal role in designing new potent antimicrobials. Triazole moieties are found to show antimalarial [1,2], antioxidant [3], antitrypanosomal [4], anti-inflammatory [5,6], antibacterial [7-9], antifungal [10-12], antiproliferative [13], antiprotozoal [14], antituberculosis [15,16] and anticancer [17,18]. Further, 1,4-disubstituted 1,2,3-triazoles serve as effective bioactive molecule being stable to reaction conditions i.e., oxidation, reduction, hydrolysis and participate in hydrogen bonding with biological system.

The dipolar [3+2] cycloaddition of organic azides and terminal alkynes is the usual method for the synthesis of 1,2,3-triazoles. This reaction was comprehensively studied by Huisgen and Padwa [19] yielding 1,4 and 1,5-disubstituted 1,2,3-triazoles variant of substituted 1,2,3-triazole. Later, Sharpless et al. [20] and Meldal [21] developed Cu(I) catalyzed azide-alkyne cycloaddition (CuAAC) through “Click Chemistry” concept results into formation of 1,4-disubstituted 1,2,3-triazoles. The “Click Chemistry” projected cycloaddition reactions are prime important due to its several advantages in form of regioselectivity, wide substrate scope, mild reaction conditions and excellent yields.

Here, we sought to synthesize 1,4-disubstituted 1,2,3-triazoles having amide and hydroxy functionality from the reaction of terminal alkynes and 2-azido-N-substituted N-phenylpropanamides (generated in situ from reaction of 2-bromo-N-substituted-N-phenylpropanamides and sodium azide). The synthesized triazoles were characterized by various spectroscopic techniques viz., FT-IR, 1H NMR, 13C NMR, HRMS and evaluated for microbicidal activity against Escherichia coli, Enterobacter aerogenes, Klebsiella pneumoniae; one Gram-positive bacteria Staphylococcus aureus and two fungal strains Candida albicans and Aspergillus niger.

EXPERIMENTAL

All the chemicals and solvents were purchased from Sigma-Aldrich, Alfa Aesar and Hi-Media. The reactions were monitored by thin layer chromatography using n-hexane, ethyl acetate and chloroform in different ratios as mobile phase. The melting points of synthesized compounds were recorded in °C by applying an open capillary method and are uncorrected. 1H NMR and 13C NMR spectra were recorded on a Bruker Avance II 400 MHz at 400 MHz and 100 MHz, respectively, using DMSO as solvents. Chemical shift values are recorded on δ scale and the coupling constants (J) in Hertz. HRMS were obtained using a Waters Micromass Q-T of Micro (ESI) spectrophotometer and values were recorded in m/z. The IR spectra were recorded on Shimadzu IR affinity-I FTIR spectrophotometer using KBr powder.

General procedure for the synthesis of 1,4-disubstituted 1,2,3-triazoles: A solution of aromatic amines (1a-1e) (1.0...
mmol in dichloromethane was taken in round bottom flask with potassium carbonate (1.5 mmol) as base and drop-wise addition of 2-bromopropanoyl bromide (1.2 mmol) was carried out at 0-5 °C and stirred the contents for 15-20 min. After the completion of reaction, ice cold water was added to the reaction mixture, the resulting solid, 2-bromo-N-substituted N-phenylpropanamide (3a-3c) was filtered and dried.

For the synthesis of target compounds 5a-5i, the starting reactant 2-bromo-N-substituted propanamide (3a-3c) was stirred with aqueous sodium azide (3.0 mmol) in dimethyl formamide for 30 min at 35-50 °C in round bottom flask. In above mixture terminal hydroxyalkyne (4a-4c), aqueous CuSO4 5H2O (0.1 mmol) and sodium ascorbate (0.4 mmol) were added and stirring was continued for 8-10 h at same temperature. After the completion of reaction, ice cold water was added to the reaction mixture, then filtered the precipitated solid, washed with aqueous ammonia solution followed by water. The solid product was recrystallized with ethyl acetate to get pure product (5a-5i).

2-[4-(Hydroxymethyl)-1H-1,2,3-triazol-1-yl]-N-methyl-N-phenylpropanamide (5a): White solid; Yield: 86 %; m.p. 94-96 °C; IR (KBr, νmax, cm⁻¹): 3380 (O-H str.), 3148 (C-H str. triazole), 3064 (C-H str. aromatic ring), 2980 (C-H str., aliphatic), 1663 (C=O str. amide), 1542, 1463 (C=C str., aromatic ring); 1H NMR (400 MHz, DMSO-d6): δ 7.81 (s, 1H, C-H triazole), 7.53-7.38 (m, 5H, Ar-H), 5.47 (q, 1H, J = 7.2 Hz), 5.10 (s, 1H, OH), 2.95 (s, 3H), 1.54 (d, 3H, J = 7.2 Hz), 1.48 (s, 6H); 13C NMR (100 MHz, DMSO-d6): δ 168.4 (C=O), 155.9 (C3 triazole), 142.8, 130.8, 128.8, 127.8, 119.7 (C5 triazole), 67.8, 55.3, 38.0, 31.1, 18.5; HRMS (m/z) calculated for C16H16N6O2 [M+H]+: 289.1586. Found: 289.1661.

2-[4-(2-Hydroxypropan-2-yl)-1H-1,2,3-triazol-1-yl]-N-phenylpropanamide (5e): White solid; Yield: 85 %; m.p. 142-144 °C; IR (KBr, νmax, cm⁻¹): 3384 (O-H str.), 3140 (C-H str. triazole), 3070 (C-H str. aromatic ring), 2980 (C-H str., aliphatic), 1658 (C=O str. amide), 1543, 1463 (C=C str., aromatic ring); 1H NMR (400 MHz, DMSO-d6): δ 7.80 (s, 1H, C-H triazole), 7.53-7.38 (m, 5H, Ar-H), 5.27 (q, 1H, J = 8 Hz), 5.08 (s, 1H, O), 3.19 (q, 2H, J = 8 Hz), 1.54 (d, 3H, J = 8 Hz), 1.48 (s, 6H), 1.01 (t, 3H, J = 8 Hz); 13C NMR (100 MHz, DMSO-d6): δ 168.4 (C=O), 155.9 (C3 triazole), 142.8, 130.5, 128.8, 127.8, 119.9 (C5 triazole), 67.5, 55.3, 38.0, 31.1, 18.5, 12.9; HRMS (m/z) calculated for C16H16N6O2 [M+H]+: 303.1403. Found: 303.1461.

2-[4-(2-Hydroxypropan-2-yl)-1H-1,2,3-triazol-1-yl]-N,N-diphenylpropanamide (5f): White solid; Yield: 78 %; m.p. 116-118 °C; IR (KBr, νmax, cm⁻¹): 3390 (O-H str.), 3163 (C-H str. triazole), 3043 (C-H str. aromatic ring), 2978 (C-H str., aliphatic), 1676 (C=O str. amide), 1593, 1452 (C=C str., aromatic ring); 1H NMR (400 MHz, DMSO-d6): δ 7.80 (s, 1H, C-H triazole), 7.43 (brs, 10H, Ar-H), 5.47 (q, 1H, J = 7.2 Hz), 5.09 (s, 1H), 1.65 (d, 3H, J = 7.2 Hz), 1.47 (s, 6H); 13C NMR (100 MHz, DMSO-d6): δ 168.8 (C=O), 156.0 (C3 triazole), 128.8, 127.3, 120.1 (C5 triazole), 67.5, 56.2, 31.3, 18.5; HRMS (m/z) calculated for C20H22N6O2 [M+H]+: 351.1743. Found: 351.1813.

2-[4-(2-Hydroxybutan-2-yl)-1H-1,2,3-triazol-1-yl]-N-methyl-N-phenylpropanamide (5g): White solid; Yield: 90 %; m.p. 66-68 °C; IR (KBr, νmax, cm⁻¹): 3378 (O-H str.), 3140 (C-H str. triazole), 3057 (C-H str. aromatic ring), 2978 (C-H str., aliphatic), 1665 (C=O str. amide), 1593, 1454 (C=C str., aromatic ring); 1H NMR (400 MHz, DMSO-d6): δ 7.77 (s, 1H, C-H triazole), 7.52-7.34 (m, 5H, Ar-H), 5.47 (q, 1H, J = 6.8 Hz), 5.10 (s, 1H, OH), 2.95 (s, 3H), 1.71 (t, 2H, J = 6.7 Hz), 1.54-1.40 (m, 6H), 0.74 (t, 3H, J = 7.6 Hz); 13C NMR (100 MHz, DMSO-d6): δ 167.9 (C=O), 155.0 (C3 triazole), 141.0, 130.5, 129.0, 128.8, 122.3 (C5 triazole), 55.7, 55.5, 44.6, 18.3, 12.9; HRMS (m/z) calculated for C16H11N6O2 [M+H]+: 303.1743. Found: 303.1874.

N-Ethyl-2-[4-(hydroxymethyl)-1H-1,2,3-triazol-1-yl]-N-phenylpropanamide (5h): White solid; Yield: 90 %; m.p. 68-70 °C; IR (KBr, νmax, cm⁻¹): 3380 (O-H str.), 3140 (C-H str. triazole), 3075 (C-H str. aromatic ring), 2978 (C-H str., aliphatic), 1662 (C=O str. amide), 1593, 1454 (C=C str., aromatic ring); 1H NMR (400 MHz, DMSO-d6): δ 7.57 (s, 1H, C-H triazole), 7.51-7.34 (m, 5H, Ar-H), 5.19 (q, 1H, J = 6.8 Hz), 4.93 (s, 1H, OH), 3.67 (q, 2H, J = 6.4 Hz), 1.73-1.40 (m, 6H), 1.01 (t, 3H, J = 6.4 Hz), 0.74 (t, 3H, J = 8 Hz); 13C NMR (100 MHz, DMSO-d6): δ 167.9 (C=O), 155.0 (C3 triazole), 158.6, 148.2, 130.7, 129.6, 129.1, 127.2 (C5 triazole), 56.2, 55.5, 18.3; HRMS (m/z) calculated for C16H16N6O2 [M+H]+: 323.1403. Found: 323.1501.
triazole), 141.0, 130.5, 128.8, 120.6 (C5 triazole), 70.2, 55.6, 44.5, 35.9, 28.6, 18.5, 12.9, 8.8; HRMS (m/z) calculated for C17H24N4O2 [M+H]+: 317.1899. Found: 317.1970.

2-[4-(2-Hydroxybutan-2-yl)-1H-1,2,3-triazol-1-yl]-N,N-diphenylpropanamide (5i): White solid; Yield: 86 %; m.p. 76-80 °C; IR (KBr, ν max, cm⁻¹): 3370 (O-H str.), 3159 (C-H str. triazole), 3061 (C-H str. aromatic ring), 2972 (C-H str., aliphatic), 1678 (C=O str. amide), 1593, 1454 (C=C str., aromatic ring); 1H NMR (400 MHz, DMSO-d6): δ 7.79 (s, 1H, C-H triazole), 7.43 (brs, 10H, Ar-H), 5.46 (q, 1H, J = 4.8 Hz), 4.94 (s, 1H, OH), 1.75-1.64 (m, 5H), 1.42 (s, 3H), 0.75 (t, 3H, J = 8 Hz); 13C NMR (100 MHz, DMSO-d6): δ 168.8 (C=O), 155.0 (C4 triazole), 130.6, 129.6, 129.0, 127.3, 120.8 (C5 triazole), 70.2, 56.2, 36.0, 28.7, 18.5, 8.8; HRMS (m/z) calculated for C21H24N4O2[M+H]+: 365.1899. Found: 365.1968.

Antimicrobial activity: Compounds (5a-5i) were screened for in vitro antimicrobial activity against Escherichia coli, Enterobacter aerogenes, Klebsiella pneumoniae, Staphylococcus aureus, Candida albicans and Aspergillus niger by serial dilution method using a stock solution of 100 µg/mL concentration in five sets at different concentrations of 50, 25, 12.5, 6.25, 3.12 µg/mL. DMSO used as control solvent while, norfloxacin and fluconazole used as standard drugs, respectively for bacteria and fungus. All these dilutions were inoculated with respective bacterial and fungal strain in saline solution. Growth was assessed after incubation for 24 h at 37 °C in case of all bacteria, for 48 h at 25 °C in case of C. albicans and for 120 h at 25 °C for in case of A. niger and the minimum inhibitory concentration (MIC) value was noted. The MIC is defined as the lowest concentration of the antimicrobial agent that prevents visible growth of a microorganism under defined conditions.

**RESULTS AND DISCUSSION**

2-Bromopropanoyl bromide (2) was added in aromatic amines (1a-1c) in dichloromethane using potassium carbonate as a base to synthesize 2-bromo- N-substituted-N-phenylpropanamides (3a-3c). 1,4-Disubstituted 1,2,3-triazoles (5a-5i) were synthesized by reacting 2-azido-N-substituted-N-phenylpropanamides (generated in situ from 2-bromo-N-substituted N-phenylpropanamides and sodium azide) with hydroxyl terminal alkynes using dimethylformamide:water as solvent in the presence of copper sulphate pentahydrate and sodium ascorbate with stirring of 8-10 h at 35-50 °C (Scheme-I).

The characterization of synthesized compounds was carried out by the techniques FTIR, 1H NMR, 13C NMR spectroscopy and HRMS. The triazole formation was confirmed by the presence of copper sulphate pentahydrate and sodium ascorbate with stirring of 8-10 h at 35-50 °C (Scheme-I).

The characterization of synthesized compounds was carried out by the techniques FTIR, 1H NMR, 13C NMR spectroscopy and HRMS. The triazole formation was confirmed by the presence of absorption bands in the region 3384-3370 (O-H str.), 3172-3140 (C-H, str., triazole ring), 1678-1662 (C=O str., amide) cm⁻¹ in IR spectra. The 1H NMR spectra of the

![Scheme-I](image-url)
compounds 5a-5i displayed one characteristic singlet in the region at δ 7.97-7.76 due to triazolyl proton. In the 13C NMR spectra signal for C4 of the triazole moiety appeared in the region at δ 156.0-148.1, for C5 signal appeared in region δ 119.7-122.3 and signals due to carbonyl carbons of amide appeared in range of δ 168.9-167.9.

**Antibacterial activity:** All the synthesized compounds (5a-5i) were tested for in vitro antibacterial activity against Gram-negative bacteria - *Escherichia coli* (MTCC 443), *Enterobacter aerogenes* (NCDC 106), *Klebsiella pneumoniae* (NCDC 138) and Gram-positive bacteria - *Staphylococcus aureus* (MTTC 3160) by the standard serial dilution method [22]. The minimum inhibitory concentration values (MIC; corresponds to the lowest concentration that inhibits the microbial growth) of the synthesized triazoles are represented in µmol/mL and the results were compared to the control drug norfloxacin (Table-1). The screening results revealed that the compound 5f and 5i was found to be the broadly active against bacterial strains amongst tested compounds, while compounds 5c, 5f, 5h and 5i showed excellent activity comparable to standard against *S. aureus*.

**Conclusion**

In the present study, regioselective synthesis of a series of 2-(4-(hydroxyalkyl)-1H-1,2,3-triazol-1-yl)-N-substituted N-phenylpropanamide employing click reaction has been reported. Efficient 1,3-dipolar cycloaddition between terminal hydroxalkynes and 2-azido-N-substituted N-phenylpropanamides yielded 1,4-disubstituted 1,2,3-triazole having amide and hydroxy functionality in good yield. Some of the synthesized compound also displayed appreciable microbicidal activity.

**REFERENCES**


