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Green synthesis and antibacterial evaluation of isatin–oxime–triazole conjugates

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Abstract: A series of isatin–oxime–triazole conjugates (**4**) has been synthesized utilizing copper nanoparticle catalyzed cycloaddition between 3-(hydroxyimino)-1-(prop-2-ynyl)indolin-2-one (**2**) and organic azides in water. 3-(hydroxyimino)-1-(prop-2-ynyl)indolin-2-one was in turn prepared from *N*-propargylated isatin by reacting with hydroxylamine hydrochloride in presence of sodium sulfate. The structures of all the synthesized compounds were established on the basis of their ^1H NMR, ^{13}C NMR, FTIR and MS data. All the synthesized compounds were screened *in vitro* for antibacterial activity against two gram-positive (*Staphylococcus epidermidis*, *Bacillus subtilis*) and two gram-negative bacteria (*Escherichia coli*, *Pseudomonas aeruginosa*). The antibacterial data revealed that some of the compounds exhibited good to high activity and the incorporation of triazole ring with isatin-oxime has enhanced the activity against *S. epidermidis* and *E. Coli*.

Keywords: Isatin, 1,2,3-Triazoles, Oximes, Copper nanoparticles, Antimicrobial activity

Introduction

The emergence of multidrug-resistance has threatens the effective prevention and treatment of infections caused by variety of bacteria and fungi. Despite extensive progress in medicinal chemistry, there remains a challenge for the medicinal chemist to develop more effective and safer antimicrobial agents. Molecular hybridization is an approach that combines two or more pharmacophores into a single molecule with enhanced bioactivity [1,2]. Isatin (indoline-2,3-dione) contains indole nucleus with two distinct cyclic carbonyl groups, keto and

lactam. Isatin derivatives have been the subject of investigation owing to their widespread applications in synthetic and medicinal chemistry [3]. Isatin derivatives are also reported as versatile synthons for the synthesis of various biologically significant heterocyclic compounds [4]. Additionally, compounds containing isatin moiety are of great interest because of their wide spectrum of pharmacological activities such as antimicrobial [5], anticonvulsant [6], anticancer [7], antioxidant [8], anti-inflammatory activity [9], etc. Oximes are well recognized for the characterization of carbonyl compounds and as versatile precursors in synthetic organic

chemistry ^[10]. Hybrid compounds containing isatin and oxime moieties with interesting biological activities are reported in literature. For example, Cao *et al.* reported the synthesis of inhibitors of the mitogen-activated kinase, JNK3 (**I**) ^[11]. Similarly, some isatin oxime derivatives (**II**) were found to inhibit proliferation of apoptosis-resistant cancer cell lines (Fig. 1) ^[12].

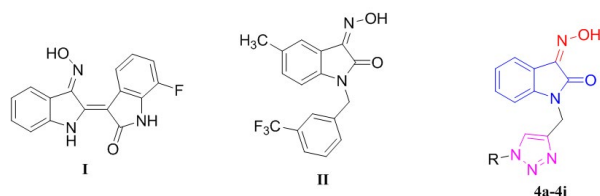


Fig. 1. Chemical structures of some biologically active isatin-oxime derivatives (**I**, **II**) and designed isatin-oxime-triazole conjugates (**4a-4j**).

1,2,3-Triazoles have gained paramount interest from organic and medicinal chemists due to their significant biological activities *viz.* antimicrobial ^[13,14], anticancer ^[15,16], antitubercular ^[17], antimalarial ^[18], etc. The Cu(I)-catalyzed azide-alkyne cycloaddition is the most important and powerful route for the regioselective synthesis of 1,4-disubstituted 1,2,3-triazoles ^[19]. A number of homogeneous copper catalysts have been working well for the synthesis of 1,2,3-triazoles, however, they suffer from the difficulty of separating the traces of catalyst from the products. Recently, supported copper nanoparticles as heterogeneous catalysts have been utilized for the preparation of 1,2,3-triazoles to circumvent the above mentioned problem ^[20]. The heterogenization of click catalysts constitutes an attractive alternative because of the benefits of easy removal, recovery and reusability of the catalyst. Therefore, in order to produce single molecular scaffold by combining two or more biologically active entities it was envisioned to synthesize a series of hybrids of isatin-oxime and 1,2,3-triazole moieties by molecular hybridization approach. In continuation of our

work on the biologically relevant 1,2,3-triazoles ^[21,22], herein, we describe the green synthesis, characterization and evaluation of some isatin-oxime-triazole conjugates (**4a-4j**) as promising antibacterial agents utilizing copper nanoparticles catalyzed alkyne-azide cycloaddition using water as solvent.

Experimental

General. Starting materials and solvents were procured from the local suppliers and were used as such. Melting points were recorded by using electro thermal melting apparatus and are uncorrected. The IR spectra were recorded with KBr on a SHIMAZDU IR AFFINITY-I FTIR spectrophotometer. The ¹H NMR and ¹³C NMR spectra were recorded on Bruker Avance III 400 nano bay in DMSO-*d*₆ at 400 and 100 MHz, respectively and TMS was used as an internal standard, chemical shift are reported in δ (ppm) and coupling constants (*J*) are measured in Hz. Mass spectra (MS) were recorded on waters 2618 EQD, ESI instrument. The progress of the reaction and the purity of compounds were verified by TLC on readymade silica gel plates (SIL G/UV254, ALUGRAM).

Synthesis of 3-(hydroxyimino)-1-(prop-2-ynyl)indolin-2-one (2**)** ^[23]: A solution of *N*-propargylated isatin **1** (1 mmol), hydroxylamine hydrochloride (2 mmol), and anhydrous sodium sulfate (1 mmol) in ethanol (10 mL) was refluxed while stirring. After completion of the reaction as monitored by TLC, the reaction mixture was filtered and the filtrate was concentrated in vacuum to afford the corresponding *N*-propargylated isatin oxime **2** in a good yield which was further recrystallized with ethanol.

3-(hydroxyimino)-1-(prop-2-ynyl)indolin-2-one (2**):** IR (KBr, ν_{max} /cm⁻¹): 3236 (N-OH), 3086, 2959, 2120, 1719, 1609 (C=N), 1450, 1360, 1196, 1113, 1086, 1055, 1012, 955, 928,

750, 708, 646. ^1H NMR (DMSO- d_6 , 400 MHz): δ 2.60 (t, 1H, J = 2.4 Hz, CH), 4.57 (t, 2H, NCH_2), 7.12 (d, 1H, H-5, J = 7.4 Hz), 7.16 (t, 1H, H-6, J = 7.96 Hz), 7.48 (t, 1H, H-4, J = 7.72 Hz), 8.02 (d, 1H, H-7, J = 7.4 Hz), 13.57 (bs, NOH, 1H). ^{13}C NMR (DMSO- d_6 , 100 MHz): δ 162.76 (C=N), 143.66, 142.28, 132.52, 127.39, 123.56, 115.75, 110.14, 78.34, 74.94, 29.09. MS: m/z (M^+) Calcd. for $\text{C}_{11}\text{H}_8\text{N}_2\text{O}_2$: 200.1; found: 201.2 ($\text{M}+\text{H}$) $^+$

General procedure for synthesis of isatin-oxime-triazole conjugates (4a-4g) [24]: A solution of propargyl isatin oxime (**2**) (1 mmol), substituted benzyl bromides (**3a-3g**) (1 mmol), and sodium azide (1.2 mmol) were placed in a round bottom flask. Water (5-7 mL) and cellulose-CuI (0.1 g) were added to it and the reaction mixture was heated at 70 °C. After completion as monitored by TLC, the reaction mixture was filtered and residue was washed with ethyl acetate three times (3x30 mL). The organic extract was washed with water, brine and dried over Na_2SO_4 . The filtrate was concentrated under reduced pressure to give the final product.

1-((1-benzyl-1H-1,2,3-triazol-4-yl)methyl)-3-(hydroxyimino)indolin-2-one (4a): IR (KBr, $\nu_{\text{max}}/\text{cm}^{-1}$): 3189 (N-OH), 3121 (C-H str, triazole), 3084, 3030, 2947, 2803, 1722 (C=O), 1609 (C=N), 1462, 1435, 1360, 1339, 1188, 1122, 1051, 1020, 947, 752, 725, 696. ^1H NMR (DMSO- d_6 , 400 MHz): δ 4.05 (s, 2H, NCH_2), 5.54 (s, 2H, NCH_2), 7.06 (d, 1H, H-5, J = 7.4 Hz), 7.10 (t, 1H, H-6, J = 7.96 Hz), 7.19-7.54 (m, 5H, benzyl), 7.60 (t, 1H, H-4, J = Hz), 7.99 (d, 1H, H-7, J = 7.2 Hz), 8.17 (s, 1H, triazolyl-H), 13.49 (bs, NOH, 1H). ^{13}C NMR (DMSO- d_6 , 100 MHz): δ 163.24 (C=O), 143.90 (C=N), 143.06 (C-4), 142.72, 136.43, 132.40, 129.20, 128.60, 128.36, 127.34, 124.06, 125.23, 123.23 (C-5), 115.78, 110.11, 53.27 (NCH_2), 35.13 (NCH_2). MS: m/z (M^+) Calcd. for $\text{C}_{18}\text{H}_{15}\text{N}_5\text{O}_2$: 333.1 found; 332.1 ($\text{M}+\text{H}$) $^+$.

1-((1-(2-nitrobenzyl)-1H-1,2,3-triazol-4-yl)methyl)-3-(hydroxyimino)indolin-2-one (4b): IR (KBr, $\nu_{\text{max}}/\text{cm}^{-1}$): 3198 (N-OH), 3138 (C-H str, triazole), 3028, 3007, 2927, 2801, 1728 (C=O), 1607 (C=N), 1528 (NO_2), 1462, 1362 (NO_2), 1337, 1310, 1190, 1126, 1016, 1005, 937, 731. ^1H NMR (DMSO- d_6 , 400 MHz): δ 4.05 (s, 2H, NCH_2), 5.84 (s, 2H, NCH_2), 6.97 (d, 1H, ArH, J = 8.0 Hz), 7.07 (d, 1H, H-5, J = 8.0 Hz), 7.11 (t, 1H, H-6, J = 8.0 Hz), 7.40 (t, 1H, H-4, J = 8.0 Hz), 7.61 (d, 1H, J = 8.0 Hz), 7.70 (d, 1H, J = 8.0 Hz), 7.98 (d, 1H, H-7, J = 8.0 Hz), 8.11 (d, 1H, J = 8.0 Hz), 8.17 (s, 1H, triazolyl-H), 13.49 (bs, NOH, 1H). ^{13}C NMR (DMSO- d_6 , 100 MHz): δ 163.25 (C=O), 148.02, 143.03 (C=N), 142.71 (C-4), 134.78, 132.41, 131.28, 130.40, 130.09, 125.52, 124.90, 123.25 (C-5), 115.78, 110.15, 50.38 (NCH_2), 35.10 (NCH_2). MS: m/z (M^+) Calcd. for $\text{C}_{18}\text{H}_{14}\text{N}_6\text{O}_4$: 378.1 found; 379.3 ($\text{M}+\text{H}$) $^+$.

1-((1-(3-nitrobenzyl)-1H-1,2,3-triazol-4-yl)methyl)-3-(hydroxyimino)indolin-2-one (4c): IR (KBr, $\nu_{\text{max}}/\text{cm}^{-1}$): 3190 (N-OH), 3142 (C-H str, triazole), 3118, 2957, 2841, 1726 (C=O), 1609 (C=N), 1522 (NO_2), 1466, 1429, 1346 (NO_2), 1308, 1231, 1184, 1130, 1086, 1051, 1011, 995, 937, 854, 795, 754, 731, 696. ^1H NMR (DMSO- d_6 , 400 MHz): δ 4.05 (s, 2H, NCH_2), 5.48 (s, 2H, NCH_2), 7.06 (d, 1H, H-5, J = 8.0 Hz), 7.12 (t, 1H, H-6, J = 8.0 Hz), 7.65 (t, 1H, J = 4.0 Hz), 7.72 (d, 1H, J = 8.0 Hz), 7.98 (t, 1H, H-4, J = 8.0 Hz), 7.99 (d, 1H, H-7, J = 8.0 Hz), 8.18 (d, 2H, J = 8.0 Hz), 8.33 (s, 1H, triazolyl-H), 13.49 (bs, NOH, 1H). ^{13}C NMR (DMSO- d_6 , 100 MHz): δ 163.25 (C=O), 148.01, 143.89 (C=N), 143.03 (C-4), 142.71, 134.78, 132.41, 131.28, 130.39, 130.09, 125.52, 124.90, 125.25 (C-5), 115.77, 110.15, 50.38 (NCH_2), 35.10 (NCH_2). MS: m/z (M^+) Calcd. for $\text{C}_{18}\text{H}_{14}\text{N}_6\text{O}_4$: 378.1 found; 379.2 ($\text{M}+\text{H}$) $^+$.

1-((1-(4-nitrobenzyl)-1H-1,2,3-triazol-4-yl)methyl)-3-(hydroxyimino)indolin-2-one (4d): IR (KBr, $\nu_{\text{max}}/\text{cm}^{-1}$): 3186 (N-OH), 3180 (N-

OH), 3119 (C–H str, triazole), 3080, 1726 (C=O), 1607 (C=N), 1520 (NO₂), 1466, 1431, 1346 (NO₂), 1182, 1049, 1016, 935, 854, 795, 729, 694. ¹H NMR (DMSO-*d*₆, 400 MHz): δ 4.05 (s, 2H, NCH₂), 5.55 (s, 2H, NCH₂), 7.07 (d, 1H, H-5, *J* = 8.0 Hz), 7.11 (t, 1H, H-6, *J* = 8.0 Hz), 7.40 (t, 1H, H-4, *J* = 8.0 Hz), 7.49 (d, 2H, ArH, *J* = 8.0 Hz), 7.99 (d, H-7, *J* = 8.0 Hz), 8.21 (d, 2H, ArH, *J* = 8.0 Hz), 8.24 (s, 1H, triazolyl-H), 13.49 (bs, NOH, 1H). ¹³C NMR (DMSO-*d*₆, 100 MHz): δ 163.39 (C=O), 147.19, 144.32 (C=N), 142.91 (C-4), 141.56, 132.47, 127.35, 125.96, 123.39 (C-5), 122.59, 121.08, 115.75, 110.07, 52.40 (NCH₂), 35.04 (NCH₂). MS: *m/z* (M⁺) Calcd. for C₁₈H₁₄N₆O₄ : 378.1 found; 379.3 (M+H)⁺.

1-((1-(2-methylbenzyl)-1H-1,2,3-triazol-4-yl)methyl)-3-(hydroxyimino)indolin-2-one (4e): IR (KBr, *v*_{max}/cm⁻¹): 3198 (N-OH), 3144 (C–H str, triazole), 3051, 2810, 1719 (C=O), 1609 (C=N), 1462, 1342, 1186, 1013, 785, 745. ¹H NMR (DMSO-*d*₆, 400 MHz): δ 2.50 (s, 3H, CH₃), 4.98 (s, 2H, NCH₂), 5.55 (s, 2H, NCH₂), 7.01 (d, 1H, ArH, *J* = 8.0 Hz), 7.07-7.11 (m, 2H, ArH), 7.13-7.24 (m, 3H, ArH, H-5, H-6), 7.39 (t, 1H, H-4, *J* = 8.0 Hz), 7.98 (d, 1H, H-7, *J* = 8.0 Hz), 8.06 (s, 1H, triazolyl-H), 13.47 (bs, NOH, 1H). ¹³C NMR (DMSO-*d*₆, 100 MHz): δ 163.24 (C=O), 143.91 (C=N), 142.55 (C-4), 136.73, 134.55, 132.36, 130.87, 129.03, 128.76, 127.33, 126.68, 124.11, 123.22 (C-5), 115.78, 110.11, 51.42 (NCH₂), 35.11 (NCH₂), 19.07 (CH₃). MS: *m/z* (M⁺) Calcd. for C₁₉H₁₇N₅O₂ : 347.1 found; 348.2 (M+H)⁺.

1-((1-(3-methylbenzyl)-1H-1,2,3-triazol-4-yl)methyl)-3-(hydroxyimino)indolin-2-one (4f): IR (KBr, *v*_{max}/cm⁻¹): 3190 (N-OH), 3121 (C–H str, triazole), 3022, 2803, 1724 (C=O), 1611 (C=N), 1464, 1435, 1348, 1188, 1047, 1018, 983, 748. ¹H NMR (DMSO-*d*₆, 400 MHz): δ 2.25 (s, 3H, CH₃), 4.98 (s, 2H, NCH₂), 5.49 (s, 2H, NCH₂), 7.08 (t, 1H, H-6, *J* = 8.0 Hz), 7.11-7.24 (m, 5H, H-5, Ar), 7.38 (t, 1H, H-4,

J = 8.0 Hz), 7.98 (d, 1H, H-7, *J* = 8.0 Hz), 8.15 (s, 1H, triazolyl-H), 13.47 (bs, NOH, 1H). ¹³C NMR (DMSO-*d*₆, 100 MHz): δ 163.20 (C=O), 143.66 (C=N), 142.12 (C-4), 135.83, 134.24, 132.78, 130.09, 128.93, 128.43, 127.12, 126.79, 124.01, 123.09 (C-5), 115.55, 110.09, 51.35 (NCH₂), 35.07 (NCH₂), 19.13 (CH₃). MS: *m/z* (M⁺) Calcd. for C₁₉H₁₇N₅O₂ : 347.1; found 348.4 (M+H)⁺.

1-((1-(4-methylbenzyl)-1H-1,2,3-triazol-4-yl)methyl)-3-(hydroxyimino)indolin-2-one (4g): IR (KBr, *v*_{max}/cm⁻¹): 3185 (N-OH), 3146 (C–H str, triazole), 3090, 2935, 2874, 1732 (C=O), 1609 (C=N), 1530, 1462, 1435, 1350, 1310, 1229, 1190, 1128, 1105, 1082, 1049, 1009, 935, 785, 758, 727, 696. ¹H NMR (DMSO-*d*₆, 400 MHz): δ 2.89 (s, 3H, CH₃), 4.96 (s, 2H, NCH₂), 5.48 (s, 2H, NCH₂), 6.97 (d, 1H, H-5, *J* = 8.6 Hz), 7.07 (t, 1H, H-6, *J* = 8.0 Hz), 7.11-7.16 (m, 4H, Ar), 7.39 (t, 1H, H-4, *J* = 7.64 Hz), 7.98 (d, 1H, H-7, *J* = 7.2 Hz), 8.12 (s, 1H, triazolyl-H), 13.48 (bs, NOH, 1H). ¹³C NMR (DMSO-*d*₆, 100 MHz): δ 163.24 (C=O), 143.91 (C=N), 143.06 (C-4), 142.55, 136.73, 134.56, 132.36, 130.87, 129.02, 128.76, 127.33, 126.69, 124.12, 123.22 (C-5), 115.78, 110.12, 51.41 (NCH₂), 35.10 (NCH₂), 19.07 (CH₃). MS: *m/z* (M⁺) Calcd. for C₁₉H₁₇N₅O₂ : 347.1; found 348.3 (M+H)⁺.

General procedure for synthesis of isatin-oxime-triazole conjugates (4h-4j): For the synthesis of compounds (4h-4j), first we synthesized phenyl azides (3h-3j) from different aniline derivatives by diazotization-azidation method as reported in the literature [25]. A mixture of *N*-propargylated isatin oxime **2** (1 mmol), phenyl azides (3h-3j) (1 mmol), cellulose-CuI (0.1 g) in water (5-7 mL) was heated at 70 °C. After completion, the reaction mixture was filtered and residue was washed with ethyl acetate three times (3x30 mL). The organic extract was washed with water, brine and dried over Na₂SO₄. The solvent was concentrated under reduced pressure to give the

desired conjugates (**4h-4j**).

3-(hydroxyimino)-1-((1-*p*-tolyl-1*H*-1,2,3-triazol-4-yl)methyl)indolin-2-one (4h**):**

IR (KBr, $\nu_{\max}/\text{cm}^{-1}$): 3187 (N-OH), 3163 (C-H str, triazole), 3080, 2914, 2851, 1713 (C=O), 1609 (C=N), 1518, 1464, 1381, 1346, 1233, 1188, 1061, 1015, 943, 826, 754. ^1H NMR (DMSO- d_6 , 400 MHz): δ 2.50 (s, 3H, CH_3), 5.07 (s, 2H, NCH_2), 6.97 (d, 1H, H-5, $J = 8.6$ Hz), 7.07 (t, 1H, H-6, $J = 8.0$ Hz), 7.11-7.29 (m, 4H, Ar), 7.39 (t, 1H, H-4, $J = 7.64$ Hz), 8.0 (d, 1H, H-7, $J = 8.0$ Hz), 8.77 (s, 1H, triazolyl-H), 13.48 (bs, NOH, 1H). ^{13}C NMR (DMSO- d_6 , 100 MHz): δ 163.29 (C=O), 143.52 (C=N), 143.06 (C-4), 138.78, 134.74, 132.44, 130.65, 127.35, 123.25 (C-5), 121.96, 120.33, 115.7, 110.11, 35.13 (NCH_2), 21.01 (CH_3). MS: m/z (M^+) Calcd. for $\text{C}_{18}\text{H}_{15}\text{N}_5\text{O}_2$: 333.1; found 334.0 ($\text{M}+\text{H}$) $^+$.

3-(hydroxyimino)-1-((1-bromo-1*H*-1,2,3-triazol-4-yl)methyl)indolin-2-one (4i**):**

IR (KBr, $\nu_{\max}/\text{cm}^{-1}$): 3185 (N-OH), 3169 (C-H str, triazole), 3075, 2970, 2897, 1713 (C=O), 1609 (C=N), 1499, 1464, 1381, 1344, 1233, 1186, 1059, 1015, 943, 826, 756. ^1H NMR (DMSO- d_6 , 400 MHz): δ 5.08 (s, 2H, NCH_2), 7.08 (t, 1H, H-5, $J = 8.0$ Hz), 7.15 (d, 1H, H-6, $J = 8.0$ Hz), 7.41 (t, 1H, H-4, $J = 8.0$ Hz), 7.76 (d, H-7, $J = 8.0$ Hz), 7.85 (d, 2H, ArH, $J = 8.0$ Hz), 8.00 (d, 2H, ArH, $J = 8.0$ Hz), 8.86 (s, 1H, triazolyl-H), 13.49 (bs, NOH, 1H). ^{13}C NMR (DMSO- d_6 , 100 MHz): δ 163.30 (C=O), 143.95 (C=N), 143.88, 143.01 (C-4), 136.16, 133.20, 132.45, 127.36, 123.27 (C-5), 122.38, 122.15, 121.79, 115.88, 110.09, 35.10 (NCH_2). MS: m/z (M^+ ; Br^{79}) Calcd. for $\text{C}_{17}\text{H}_{12}\text{BrN}_5\text{O}_2$: 397.0; found 398.1 ($\text{M}+\text{H}$; ^{79}Br) $^+$, Calcd. 398.0; found 400.1 ($\text{M}+\text{H}$; ^{81}Br) $^+$.

3-(hydroxyimino)-1-((1-(4-nitrophenyl)-1*H*-1,2,3-triazol-4-yl)methyl)indolin-2-one (4j**):**

IR (KBr, $\nu_{\max}/\text{cm}^{-1}$): 3195 (N-OH), 3167 (C-H str, triazole), 3088, 2954, 2657, 1724 (C=O),

1607 (C=N), 1526 (NO_2), 1460, 1433, 1344 (NO_2), 1246, 1186, 1113, 1084, 1045, 1018, 943, 856, 752, 691. ^1H NMR (DMSO- d_6 , 400 MHz): δ 5.12 (s, 2H, NCH_2), 7.10 (t, 1H, H-5, $J = 7.6$ Hz), 7.16 (d, 1H, H-6, $J = 8.0$ Hz), 7.41 (t, 1H, H-4, $J = 8.0$ Hz), 8.01 (d, H-7, $J = 8.0$ Hz), 8.20 (d, 2H, ArH, $J = 8.0$ Hz), 8.42 (d, 2H, ArH, $J = 8.0$ Hz), 9.05 (s, 1H, triazolyl-H), 13.51 (bs, NOH, 1H). ^{13}C NMR (DMSO- d_6 , 100 MHz): δ 163.29 (C=O), 147.71, 145.08, 143.89 (C=N), 143.73, 143.00 (C-4), 142.88, 129.50, 129.33, 124.46, 124.34, 124.18, 123.30 (C-5), 115.76, 110.09, 35.10 (NCH_2). MS: m/z (M^+) Calcd. for $\text{C}_{17}\text{H}_{12}\text{N}_6\text{O}_4$: 364.1; found: 365.3 ($\text{M}+\text{H}$) $^+$.

Results and discussion

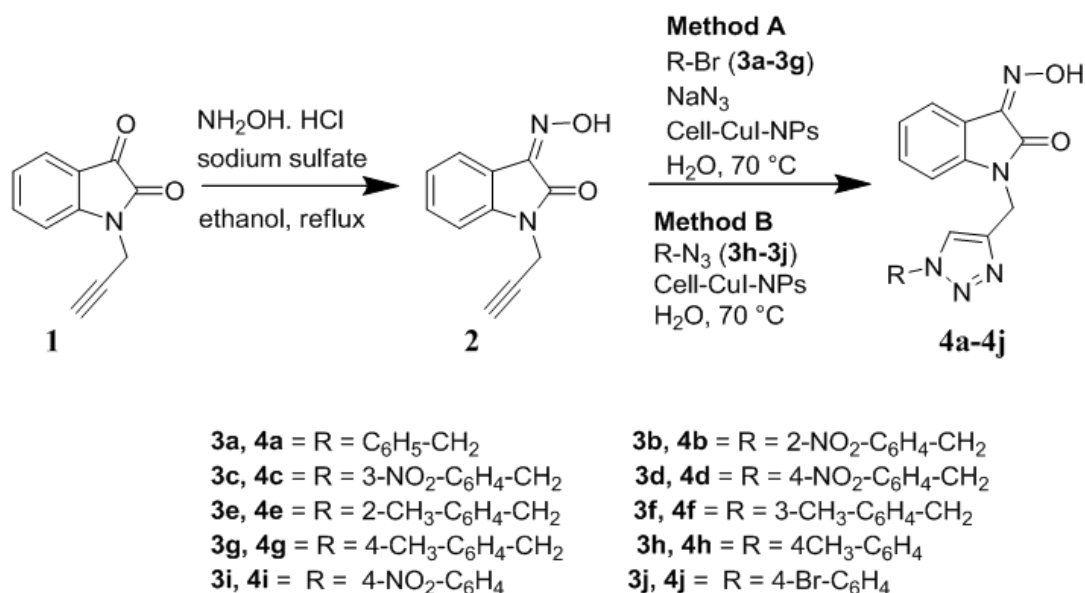
Chemistry

The outline for the synthesis of isatin-oxime-triazole conjugates is presented in Scheme 1. Initially, the *N*-alkylation of commercially available isatin was performed with propargyl bromide in presence of K_2CO_3 and KI to furnish *N*-propargylated isatin (**1**) [26]. The *N*-propargylated isatin was reacted with hydroxylamine hydrochloride in the presence of anhydrous sodium sulfate in ethanol under reflux to yield the expected 3-(hydroxyimino)-1-(prop-2-ynyl)indolin-2-one (**2**) as per literature procedure [23].

3-(hydroxyimino)-1-(prop-2-ynyl)indolin-2-one (**2**) was then reacted in one-pot multicomponent click reaction with variedly substituted benzyl bromides, and sodium azide in presence of catalytic amount of cellulose supported cuprous iodide nanoparticles in water yielding the 3-(hydroxyimino)-1-((1-substituted-1*H*-1,2,3-triazol-4-yl)methyl)indolin-2-one (**4a-4g**) in good to high yields. The cellulose supported cuprous iodide nanoparticles were synthesized by reacting cellulose and cuprous iodide *via* the reported procedure [24]. Further, 3-(hydroxyimino)-1-(prop-2-ynyl)indolin-

2-one (**2**) underwent cycloaddition with organic azides (**3h-3j**) in presence of catalytic amount of cellulose supported cuprous iodide nanoparticles in water furnished desired triazole conjugates (**4h-4j**). Organic azides (**3h-3j**) were synthesized from their corresponding commercially available aniline derivatives

using previously reported procedure ^[25]. It is also worthy to mention that no chromatographic purification was needed to obtain the desired triazole conjugates (**4h-4j**). To the best of our knowledge this is the first report on the isatin-oxime-triazole conjugates prepared in water. The structures of all the newly synthesized



SCHEME 1: Synthesis of isatin–oxime-triazole conjugates (**4a-4j**)

Table 1. Synthesis of isatin–oxime-triazole conjugates (**4a-4j**) using cuprous iodide nanoparticles in water

Entry	Compounds	R	Yield (%)	MP (°C)
1	1	---	94	194-196
2	2	---	89	198-200
3	4a	C ₆ H ₅ -CH ₂ -	83	208-210
4	4b	2-O ₂ N-C ₆ H ₄ -CH ₂ -	86	224-226
5	4c	3-O ₂ N-C ₆ H ₄ -CH ₂ -	85	224-226
6	4d	4-O ₂ N-C ₆ H ₄ -CH ₂ -	89	220-222
7	4e	2-CH ₃ -C ₆ H ₄ -CH ₂ -	88	226-228
8	4f	3-CH ₃ -C ₆ H ₄ -CH ₂ -	89	216-218
9	4g	4-CH ₃ -C ₆ H ₄ -CH ₂ -	89	218-220
10	4h	4-CH ₃ -C ₆ H ₄ -	81	232-234
11	4i	4-O ₂ N-C ₆ H ₄ -	88	238-240
12	4j	4-Br-C ₆ H ₄ -	87	236-238

compounds were analyzed on the basis of their FTIR, ^1H NMR, ^{13}C NMR and MS spectral data. In the IR spectrum of compound **4g**, the appearance of a broad peak due to $=\text{N}-\text{OH}$ stretching in the region 3185 cm^{-1} showed formation of oxime. The presence of triazole ring was confirmed by a peak at 3146 cm^{-1} due to $=\text{C}-\text{H}$ stretching of triazole ring and the peak at 1732 cm^{-1} was assigned to the $\text{C}=\text{O}$ stretching of lactam. The ^1H NMR spectrum of compound **4g** in $\text{DMSO}-d_6$ exhibited a broad singlet exchangeable with D_2O at δ 13.48, corresponding to the proton of hydroxyl group of oxime. A characteristic singlet observed at δ 8.12 was assigned to proton of triazole functionality. The ^{13}C NMR spectrum of compound **4g** displayed a peak at δ 143.91, which was assigned to $-\text{C}=\text{N}$ of oxime moiety. The peaks at δ 143.06 and δ 123.22 were assigned to the C-4 and C-5 carbon atoms of the triazole ring, respectively. The mass spectrum of compound **4g** showed a peak at m/z 348.2 $(\text{M}+\text{H})^+$, which is in good agreement with the molecular formula of the compound. Likewise, the remaining compounds showed the expected pattern as reported in case of **4g**.

Pharmacology

The *in silico* study for ADME (absorption, distribution, metabolism, and

elimination) prediction of all the synthesized compounds (**1**, **2**, **4a-4j**) to find out the drug-likeness was carried out and illustrated in Table 3. To predict physico-chemical parameters such as molecular weight (MW), logarithm of partition coefficient (milog P), number of hydrogen bond acceptors (n-ON), number of hydrogen bonds donors (n-OHNNH), topological polar surface area (TPSA), number of rotatable bonds (n-ROTB), molecular volume (MV) (Lipinski's parameters) Molinspiration online property calculation toolkit was used (<http://www.molinspiration.com/cgi-bin/properties>) [28].

Antibacterial activity

All the synthesized compounds (**1**, **2**, **4a-4j**) were screened *in vitro* for their antibacterial activity against two Gram-positive bacteria (*Staphylococcus epidermidis* MTCC 6880 and *Bacillus subtilis* MTCC 441) and two Gram-negative bacteria (*Escherichia coli* MTCC 16521 and *Pseudomonas aeruginosa* MTCC 424) following standard serial dilution method [27]. Ciprofloxacin was used as standard drug and the minimum inhibitory concentrations (MIC) in $\mu\text{M}/\text{mL}$ are listed in Table 2.

From table 2, it can be analyzed that most of the compounds exhibited good to high

Table 2. *In vitro* antibacterial screening of compounds **1-2**; **4a-4j** (MIC in $\mu\text{M}/\text{mL}$)

Entry	Compounds	R	<i>S. epidermidis</i>	<i>B. subtilis</i>	<i>E. coli</i>	<i>P. aeruginosa</i>
1	1	---	0.0337	0.0675	0.0675	0.0675
2	2	---	0.0312	0.0312	0.0624	0.0312
3	4a	$\text{C}_6\text{H}_5\text{CH}_2$	0.0187	0.0375	0.0375	0.0375
4	4b	$2-\text{O}_2\text{NC}_6\text{H}_4-\text{CH}_2$	0.0165	0.033	0.033	0.033
5	4c	$3-\text{O}_2\text{NC}_6\text{H}_4-\text{CH}_2$	0.0083	0.033	0.033	0.0165
6	4d	$4-\text{O}_2\text{NC}_6\text{H}_4-\text{CH}_2$	0.0165	0.033	0.033	0.033
7	4e	$2-\text{CH}_3\text{C}_6\text{H}_4-\text{CH}_2$	0.018	0.036	0.036	0.036
8	4f	$3-\text{CH}_3\text{C}_6\text{H}_4-\text{CH}_2$	0.009	0.036	0.036	0.036
9	4g	$4-\text{CH}_3\text{C}_6\text{H}_4-\text{CH}_2$	0.018	0.036	0.036	0.036
10	4h	$4-\text{CH}_3\text{C}_6\text{H}_4$	0.0187	0.0375	0.0375	0.0375
11	4i	$4-\text{O}_2\text{NC}_6\text{H}_4$	0.0086	0.0343	0.0343	0.0343
12	4j	$4-\text{BrC}_6\text{H}_4$	0.0157	0.0314	0.0157	0.0157
13	Ciprofloxacin	----	0.0047	0.0047	0.0047	0.0047

In silico ADME prediction

Table 3: Molinspiration bioactivity score of compounds **1**, **2**, **4a-4j**.

Entry	Compounds	% ABS	MW	ClogP	TPSA	nON	NoHNH	Nrotb	Volume
1	1	95.52	185.18	1.06	39.08	3	0	1	163.01
2	2	90.16	200.20	1.42	54.60	4	1	1	175.31
3	4a	79.57	333.35	2.64	85.31	7	1	4	289.72
4	4b	63.76	378.35	2.55	131.14	10	1	5	313.06
5	4c	63.76	378.35	2.57	131.14	10	1	5	313.06
6	4d	63.76	378.35	2.60	131.14	10	1	5	313.06
7	4e	79.57	347.38	3.04	85.31	7	1	4	306.28
8	4f	79.57	347.38	3.06	85.31	7	1	4	306.28
9	4g	79.57	347.38	3.08	85.31	7	1	4	306.28
10	4h	79.57	333.35	2.76	85.31	7	1	3	289.48
11	4i	63.76	364.32	2.27	131.14	10	1	4	296.25
12	4j	79.57	398.22	3.12	85.31	7	1	3	290.81

antibacterial activity. The activity of compound *N*-propargylated isatin **2** was found to be better than its precursor *N*-propargylated isatin **1**. The activity of most of the isatin–oxime-triazole conjugates was found better than the compound **2**, suggesting that the incorporation of triazole moiety has improved the activity against *S. epidermidis* and *E. coli*. In case of *S. epidermidis*, compounds **4c** (0.0083 $\mu\text{M}/\text{mL}$), **4f** (0.009 $\mu\text{M}/\text{mL}$), **4i** (0.0086 $\mu\text{M}/\text{mL}$) displayed very good and comparable activity to standard ciprofloxacin. Further, compound **4j** containing a 4-bromophenyl attached to triazole ring exhibited broad spectrum activity and was found to be active against all the tested strains.

From the [Molinspiration bioactivity score](#) it was observed that all the synthesized compounds (**1**, **2**, **4a-4j**) displayed good conformational flexibility because these are having sufficient number of rotatable bonds (3-5). Absorption was calculated by the formula as $\% \text{ABS} = 109 - (0.345 \times \text{TPSA})$ ^[29] and all the synthesized

compounds exhibits good absorption ($\% \text{ABS} = 63.76\text{-}95.52$). As per Lipinski rule-of-five a drug should not violate no more than one of the following Lipinski's rule: $\text{mlog } P$ (octanol-water partition coefficient) ≤ 5 , molecular weight ≤ 500 , number of hydrogen bond acceptors ≤ 10 and number of hydrogen bond donors ≤ 5 . It was also observed that all the synthesized compounds conformed to Lipinski's rule of five and having good bioavailability. Therefore, these predicted properties showed that these compounds are possessing good drug-like properties.

Conclusion

In summary, we reported the synthesis of some isatin–oxime-triazole conjugates *via* copper nanoparticles catalyzed alkyne-azide cycloaddition between 3-(hydroxyimino)-1-(prop-2-ynyl)indolin-2-one (**2**) and substituted organic azides using water as green solvent. All the synthesized compounds were characterized by ^1H NMR, ^{13}C NMR, FTIR, and MS data

and tested *in vitro* for antibacterial activity. The activity results revealed that compound **4j** exhibited broad spectrum activity and was found to be active against all the tested strains. Further, the incorporation of triazole moiety has improved the bioactivity of the synthesized compounds against *S. epidermidis* and *E. coli*, suggesting the utility of molecular hybridization approach. These results provide an insight for the construction of triazole based hybrid compounds for the generation of efficient leads.

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