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Original Article

SYNTHESIS, ANTIMICROBIAL, ANTIOXIDANT AND DOCKING STUDY OF (3-ALKOXY-5-NITROBENZOFURAN-2-YL) (PHENYL) METHANONE DERIVATIVES

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ABSTARCT

Objective: Synthesis, antimicrobial, antioxidant and docking studies of (3-methoxy-5-nitrobenzofuran-2-yl)(phenyl)methanone derivatives (**2a-j)**.

Methods: The products 2a-j, were obtained from different (3-hydroxy-5-nitrobenzofuran-2-yl)(phenyl)methanones[ref]and alkyl bromides in the presence of K_2CO_3 in DMF at room temperature. Afterwards, characterised through ^{1}H-NMR, [13]C-NMR, ESI-LCMS and elemental analyses. Finally, subjected them for antimicrobial and antioxidant studies through agar well diffusion technique and docking interaction studies through automated docking technique.

Results: From the current procedure, the compound 2a has showed the highest antibacterial activity and 2j showed lowest antibacterial activity. None of these (2a-j) have showed antifungal activity. The compound 2d has shown highest free radical scavenging activity and 2e has shown least activity. The biological properties of the synthesised compounds (2a-j) were further supported by docking studies.

Conclusion: Successfully synthesised and characterized ten individually novel (3-alkoxy-5-nitrobenzofuran-2-yl)(phenyl)methanone derivatives (**2a-j**) in one pot procedure and studied antimicrobial, antioxidant and docking properties. Perhaps the present work provides an evidence for the preparation, antimicrobial and antioxidant properties of the novel benzofuran derivatives.

Keywords: (3-Alkoxy-5-nitrobenzofuran-2-yl)(phenyl) methanone, (3-hydroxy-5-nitrobenzofuran-2-yl)(phenyl) methanones, Antimicrobial activity, Antioxidant activity, Molecular docking studies.

INTRODUCTION

Benzo[*b*]furans are present in the wide range of biologically active natural products. They are used in the manufacture of pharmacological materials, polymers and electronic devices.[1-11] Benzofurans have exhibited antitumor, antiviral activities.[12] They have possessed reversible-non-selective aromatase inhibitory effect[13] and selective toxicity against tumorigenic cell lines.[14] in particular, 2-aroylbenzofuran-3-ols are reported to display promising antitumor activity.[15-17] 2-Aroylbenzofuran-3-ols occupy a significant position among benzofurans because of their use as intermediates in the synthesis of several pharmaceutical compounds.[18-22a] In light of the above mentioned facts and in continuation of our research on 2-aroylbenzofuran-3-ols,[22b] we in this, reported a convenient method for the synthesis of various (3-alkoxy-5-nitrobenzofuran-2-yl)(phenyl)methanones (**2a-j)** and their preliminary antioxidant, antibacterial, antifungal and docking studies.

MATERIALS AND METHODS

^{1}H and [13]C NMR spectra were recorded on 400 MHz Brucker FT-NMR using TMS as the internal standard and $CDCl_3$ as solvent. The chemical shifts δ are expressed in ppm. Mass spectra were recorded on LC-MS (Shimatzu QP 5000 spectrometer). Purity was checked by TLC. All compounds were purified through column chromatographic technique on 60-120 silica gels by using 9.5:0.5 ratio of petroleum ether and ethyl acetate as eluent. Samples were crystallized from DCM or ethyl acetate. Chemicals and solvents used for the chemical synthesis were acquired from commercial sources of analytical grade. The agar well diffusion method was followed for the screening of antibacterial and antifungal activity. General Procedure for synthesis of (3-alkoxybenzofuran-2-yl)(phenyl) methanones (2a-j)

Synthesis of (3-(allyloxy)-5-nitrobenzofuran-2-yl)(4-chlorophenyl) methanone as an example (2a).

Activated (heated at 100 °C in a hot air oven) K_2CO_3 (47.87 mg, 0.34 mmol) was added into a 100 mL round bottomed flask containing a stirred mixture of 4-chlorophenyl)(3-hydroxy-5-nitrobenzofuran-2-yl)methanone (**1a**) (100.00 mg, 0.32 mmol) and allyl bromide (40.36 mg 0.32 mmol) in 5 mL of dry DMF at room temperature under nitrogen. The stirring was continued and the progress of the reaction was monitored by TLC. Most of the reactants were disappeared in about 30 min. However, the reaction mixture was stirred for a total period of 1 h to ensure the completion of the reaction. Then, evaporated the solvent completely under reduced pressure and diluted with ice cold water (20 mL). The aqueous layer was extracted with ethyl acetate (2x15 mL). The organic layer was washed with water and then by brine. The solvent mixture obtained was dried over anhydrous sodium sulphate, concentrated under reduced pressure and purified through column chromatography on 60-120 silica gel by using 0.5:9.5 mixture of ethyl acetate and petroleum ether to get 95 mg of 3-(allyloxy)-5-nitrobenzofuran-2-yl)(4-chlorophenyl)methanone (**2a**) (84%).

Adopting the same procedure, the other compounds (2b-j) were prepared.

Analytical data of the compounds 2a-j

(3-(Allyloxy)-5-nitrobenzofuran-2-yl)(4-chlorophenyl) methanone (2a)

^{1}H NMR (400 MHz, $CDCl_3$): δ 8.75 (d, *J* = 2.4 Hz, 1H), 8.43-8.40 (dd, *J* = 9.2, 2.4 Hz, 1H), 7.98-7.96 (dd, *J* = 6.8, 2.0 Hz, 2H), 7.61 (d, *J* = 9.2 Hz, 1H), 7.53-7.51 (dd, *J* = 6.8, 2.0 Hz, 2H), 6.02-5.95 (m, 1H), 5.38-5.28 (m, 2H), 4.95-4.93 (m, 2H); [13]C NMR (400 MHz, $CDCl_3$): δ 182.4, 155.2, 148.3, 144.3, 140.5, 139.6, 135.3, 131.9, 131.0, 128.7, 124.1, 123.0, 119.6, 118.5, 113.4, 75.2; MS *(m/z):* 358.06 $[M+H]^+$; Anal. calcd. for $C_{18}H_{12}ClNO_5$: C, 60.43; H, 3.38; N, 3.92; found: C, 60.03; H, 3.18; N, 3.88.

(3-(Allyloxy)-5-nitrobenzofuran-2-yl)(4-fluorophenyl) methanone (2b)

^{1}H NMR (400 MHz, $CDCl_3$): δ 8.75 (d, *J* = 2.4 Hz, 1H), 8.43-8.40 (dd, *J* = 8.8, 2.0 Hz, 1H), 8.09-8.05 (m, 2H), 7.62 (d, *J* = 9.0 Hz, 1H), 7.27-7.20 (m, 2H), 6.02-5.95 (m, 1H), 5.38-5.27 (m, 2H), 4.93-4.91 (m, 2H); [13]C NMR (400 MHz, $CDCl_3$): δ 182.2, 164.5, 155.2, 148.1, 144.3, 140.6, 133.2, 132.4, 132.3, 131.9, 124.0, 123.0, 119.6, 118.4, 115.7, 115.5, 113.4, 75.2; MS *(m/z):* 342.02 $[M+H]^+$; Anal. calcd. for $C_{18}H_{12}FNO_5$: C, 63.35; H, 3.54; N, 4.10; found: C, 63.02; H, 3.64; N, 4.01.

(4-Fluorophenyl)(3-methoxy-5-nitrobenzofuran-2-yl) methanone (2c)

^{1}H NMR (500 MHz, $CDCl_3$): δ 8.73 (d, *J* = 2.0 Hz, 1H), 8.37-8.34 (dd, *J* = 9.0, 2.0 Hz, 1H), 8.03-8.00 (m, 2H), 7.57 (d, *J* = 9.5 Hz, 1H), 7.18-7.14 (m, 2H), 4.20 (s, 3H); [13]C NMR (500 MHz, $CDCl_3$): δ 207.0, 182.1, 166.8, 164.7, 155.2, 149.8, 144.2, 139.8, 133.4, 132.4, 124.1, 122.1, 118.6, 115.7, 115.5, 113.5, 62.0, 29.7, 14.1; MS *(m/z):* 316.02 $[M+H]^+$; Anal. calcd. for $C_{16}H_{10}FNO_5$: C, 60.96; H, 3.20; N, 4.44; found: C, 60.26; H, 3.08; N, 4.24.

Ethyl 2-(2-(4-fluorobenzoyl) -5-nitrobenzo furan-3-yloxy) acetate (2d)

^{1}H NMR (400 MHz, $CDCl_3$): δ 8.82 (s, 1H), 8.43-8.40 (dd, *J* = 8.8, 2.5 Hz, 1H), 8.12-8.08 (m, 2H), 7.58 (d, *J* = 9.2 Hz, 1H), 7.26-7.19 (m, 2H), 5.10 (s, 2H), 4.21 (q, *J* = 7.2Hz, 2H), 1.25 (t, *J* = 7.2 Hz, 3H); [13]C NMR (400 MHz, $CDCl_3$): 182.1, 168.3, 166.3, 154.8, 148.8, 144.5, 140.4, 133.1, 132.5, 132.4, 124.2, 123.3, 118.7, 115.8, 115.6, 113.2, 70.6, 61.5, 14.0; MS *(m/z):* 388.02 $[M+H]^+$; Anal. calcd. for $C_{19}H_{14}FNO_7$: C, 58.92; H, 3.64; N, 3.62; found: C, 58.29; H, 3.24; N, 3.34.

(4-Fluorophenyl)(5-nitro-3-(prop-2-ynyloxy)benzofuran-2-yl)methanone (2e)

^{1}H NMR (500 MHz, $CDCl_3$): δ 8.79 (d, *J* = 2.5 Hz, 1H), 8.42-8.39 (dd, *J* = 8.8, 2.0 Hz, 1H), 8.10-8.07 (m, 2H), 7.61 (d, *J* = 8.8 Hz, 1H), 7.23-7.19 (m, 2H), 5.12 (s, 2H), 2.50 (s, 1H); [13]C NMR (500 MHz, $CDCl_3$): 182.3, 164.9, 160.9, 155.0, 147.1, 144.5, 141.3, 133.1, 132.6, 132.5, 124.2, 123.5, 118.6, 115.9, 115.7, 113.3 77.8, 61.6; MS *(m/z):* 340.02 $[M+H]^+$; Anal. calcd. for $C_{18}H_{10}FNO_5$: C, 63.72; H, 2.97; N, 4.13; found: C, 63.27; H, 2.79; N, 4.02.

4-(5-Nitro-3-propoxybenzofuran-2-carbonyl)benzonitrile (2f): ^{1}H NMR (400 MHz, $CDCl_3$)

δ 8.78 (d, *J* = 2.0 Hz, 1H), 8.44-8.42 (dd, *J* = 8.8, 2.4 Hz, 1H), 8.07-8.05 (dd, *J* = 7.6, 2.0 Hz, 2H), 7.84-7.82 (dd, *J* = 7.6, 2.0 Hz, 2H), 7.63 (d, *J* = 7.8 Hz, 1H), 4.45 (t, *J* = 6.4 Hz, 2H), 1.81-1.76 (m, 2H), 0.98 (t, *J* = 6.4 Hz, 3H); [13]C NMR (400 MHz, $CDCl_3$): 181.9, 155.7, 149.9, 144.2, 140.7, 139.6, 132.0, 129.8, 124.5, 122.3, 118.8, 117.7, 115.9, 113.7, 23.7, 10.1; MS *(m/z):* 351.20 $[M+H]^+$; Anal. calcd. for $C_{19}H_{14}N_2O_5$: C, 65.14; H, 4.03; N, 8.00; found: C, 64.93; H, 3.87; N, 7.84.

(4-Bromophenyl)(3-methoxy-5-nitrobenzofuran-2-yl)methanone (2g)

^{1}H NMR (400 MHz, $CDCl_3$): δ 8.79 (s, 1H), 8.43-8.40 (dd, *J* = 8.8, 2.4 Hz, 1H), 7.91-7.87 (m, 2H), 7.70-7.67 (m, 2H), 7.61 (d, *J* = 8.8 Hz, 1H), 4.26 (s, 3H); [13]C NMR (400 MHz, $CDCl_3$): 182.4, 155.2, 149.9, 144.2, 139.6, 135.8, 131.7, 131.0, 128.2, 124.1, 122.4, 118.6, 113.5, 62.0; MS *(m/z):* 375.20 $[M+H]^+$; Anal. calcd. for $C_{16}H_{10}BrNO_5$: C, 51.09; H, 2.68; N, 3.72; found: C, 50.63; H, 2.58; N, 3.62.

Ethyl 2-(2-(4-bromobenzoyl)-5-nitrobenzofuran-3-yloxy) acetate (2h)

^{1}H NMR (400 MHz, $CDCl_3$): δ 8.82 (d, *J* = 2.4 Hz, 1H), 8.43-8.40 (dd, *J* = 8.8, 2.4 Hz, 1H), 7.94-7.90 (m, 2H), 7.70-7.67 (m, 2H), 7.59 (d, *J* = 8.8 Hz, 1H), 5.12 (s, 2H), 4.21 (q, *J* = 7.2 Hz, 2H), 1.25 (t, *J* = 7.2 Hz, 3H); [13]C NMR (400 MHz, $CDCl_3$); 182.6, 168.2, 154.8, 149.0, 144.4, 140.2, 135.6, 131.8, 131.2, 128.4, 124.4, 123.2, 118.8, 113.2, 70.6, 61.2, 14.0; MS *(m/z):* 448.20 $[M+H]^+$; Anal. calcd. for $C_{19}H_{14}BrNO_7$: C, 50.91; H, 3.15; N, 3.12; found: C, 50.59; H, 3.06; N, 3.05.

(4-Fluorophenyl)(5-nitro-3-propoxybenzofuran-2-yl)methanone (2i)

^{1}H NMR (400 MHz, $CDCl_3$): δ 8.75 (d, *J* = 2.4 Hz, 1H), 8.42-8.39 (dd, *J* = 7.8, 2.4 Hz, 1H), 8.07-8.02 (m, 2H), 7.63-7.61 (dd, *J* = 7.8, 2.4 Hz, 1H), 7.23-7.19 (m, 2H), 4.37 (t, *J* = 7.2 Hz, 2H), 1.83-1.74 (m, 2H), 0.99 (t, *J* = 7.2 Hz, 3H); [13]C NMR (400 MHz, $CDCl_3$): δ 182.2, 166.9, 164.4, 155.4, 148.8, 144.2, 140.2, 133.4, 133.3, 132.3, 132.2, 124.0, 122.8, 118.5, 115.5, 115.4, 113.5, 23.1, 10.1; MS *(m/z):* 344.20 $[M+H]^+$; Anal. calcd. for $C_{18}H_{14}FNO_5$: C, 62.97; H, 4.11; N, 4.08; found: C, 62.67; H, 4.01; N, 3.98.

4-(5-Nitro-3-(prop-2-ynyloxy)benzofuran-2-carbonyl)benzonitrile (2j)

^{1}H NMR (400 MHz, $CDCl_3$): δ; 8.10-8.08 (2H, d, J=8.4 Hz), 7.83-7.79 (3H, m), 7.54-7.53 (1H, d, J=2 Hz), 7.36-7.33 (1H,dd, J=8.8 Hz, J=2 Hz), 5.12-5.11 (2H, d, J=2.4 Hz), 2.52-2.51 (1H, t, J=2.4 Hz); [13]C NMR (400 MHz, $CDCl_3$): δ 182.2, 151.5, 148.4, 146.9, 141.9, 140.0, 132.2, 130.1,127.8, 122.3, 118.9, 117.8, 116.4, 109.2, 77.8, 61.5; MS *(m/z):* 347.20 $[M+H]^+$; Anal. Calcd. for $C_{19}H_{10}N_2O_5$: C, 65.90; H, 2.91; N, 8.09; found: C, 65.70; H, 2.81; N, 7.99.

***In vitro* antimicrobial procedure**

Antibacterial susceptibility testing

The antibacterial activity of synthesized compounds was studied against Gram-negative and Gram-positive bacteria by the agar well diffusion method [23]. Nutrient agar (Hi Media, India) was used as the bacteriological medium. The compounds were dissolved in 10% aqueous dimethylsulfoxide (DMSO) to a final concentration of 100µg/100 µL. Pure DMSO was taken as the negative control and 100µg/100 µL Ciprofloxacin as the positive control. 100 µL of inoculum was aseptically introduced on to the surface of sterile agar plates and sterilized cotton swabs were used for even distribution of the inoculum. Wells was prepared in the agar plates using a sterile cork borer of 6.0 mm diameter. 100 µL of test and control compound was introduced in the well. The same procedure was used for all the strains. The plates were incubated aerobically at 35 °C and examined after 24 h [24, 25]. The diameter of the zone of inhibition produced by each agent was measured with a ruler and compared with those produced by the commercial antibiotic Ciprofloxacin.

Antifungal susceptibility testing

Antifungal activity of synthesized compounds was tested using agar well diffusion method. The potato dextrose agar plates were inoculated with each fungal culture (10 days old) by point inoculation. A well of about 6.0 mm diameter with sterile cock borer was aseptically punched on each agar plate. The synthesized test compound (100µg/100 µL) was introduced in to the well; a negative control well was too made with 100µl of the solvent DMSO and 100µg/100 µL Fluconazole as the positive control. Plates were kept in laminar flow for 30 minutes for pre diffusion of the compound to occur and then incubated at 28 °C for 48 h. Resulting zone of inhibition (in mm) was measured using a Hi media zone scale [26]

***In vitro* antioxidant activity procedure**

DPPH Assay: The DPPH free radical scavenging activity was assessed according to Okada and Okada [1998] with slight modification [27]. 300 mL of ethanolic solution of DPPH (0.05 mM) was added to 100 µg/mL of synthesised compound. The DPPH solution was freshly prepared and kept in the dark at 4 °C. Ethanol 96 % (2.7 mL) was added and the mixture was shaken vigorously. The mixture was left to stand for 5 min and the absorbance was measured using a spectrophotometer at 517 nm. Ethanol was used to zero the spectrophotometer. A blank sample containing the same amount of ethanol and DPPH was also prepared. All determinations were performed in duplicate. The radical scavenging activities of the tested samples, expressed as percentage of inhibition were calculated according to the following equation [28]

$$\text{Percentage inhibition} = [(AB-AT)/AB] \times 100$$

Where AT and AB are the absorbance values of the test and of the blank sample, respectively.

In silico molecular docking studies procedure

With *In vitro* antimicrobial and antioxidant results in hand it is thought worthwhile to do *in silico* studies to support the *In vitro* activity. Automated docking was used to determine the orientation

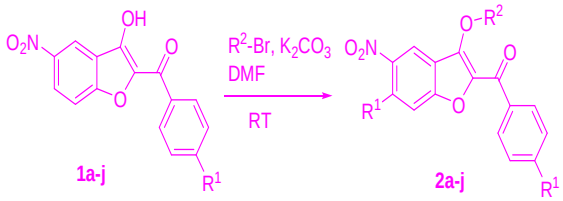
of inhibitors bound in the active site of GlcN-6-P synthase and Superoxide dismutase (SOD) as target for antimicrobial and antioxidant activity. A Lamarckian genetic algorithm method, implemented in the program AutoDock 3.0, was employed. The ligand molecules 1 to 10 were designed and the structure was analyzed by using ChemDraw Ultra 6.0. 3D coordinates were prepared using PRODRG server [29]. The protein structure file (PDB ID: 1XFF and 1CB4) was taken from PDB (www. rcsb. org/pdb) was edited by removing the heteroatoms, adding C terminal oxygen [30]. For docking calculations, Gasteigere – Marsili partial charges [31] were assigned to the ligands and non-polar hydrogen atoms were merged. All torsions were allowed to rotate during docking. The grid map was centered at particular residues of the proteins which was predicted from the ligplot and were generated with AutoGrid. The Lamarckian genetic algorithm and the pseudo-Solis and Wets methods were applied for minimization, using default parameters [32].

RESULTS AND DISCUSSION

Chemistry

Synthesis of (3-alkoxy-5-nitrobenzofuran-2-yl)(phenyl)methanone (**2a-j**) was achieved through straightforward alkylation of (3-hydroxy-5-nitrobenzofuran-2-yl)(phenyl)methanone (**1a-j**)[22b] with different alkyl bromides in the presence of K_2CO_3 as base and DMF as solvent at room temperature (Scheme 1). Most of the reactions were completed within 1h with 88-95% yield. The scope of this protocol was investigated with various substrates (**11a-j**) and alkyl halides as shown in table 1.

The structure of these individually new products was confirmed through their ^{1}H NMR, ^{13}C NMR spectroscopy, LCMS and elemental analyses.



Scheme 1: Synthesis of (3-alkoxy-5-nitrobenzofuran-2-yl)(phenyl)methanone derivatives (2a-j)

Table 1: Synthesis of (3-alkoxybenzofuran-2-yl)(phenyl)methanones (2a-j)

S. No.	Entry	R^1	R^2	% Yield
1	2a	Cl	Allyl	84
2	2b	F	Allyl	90
3	2c	F	Me	91
4	2d	F	CH_2COOEt	95
5	2e	F	Propergyl	93
6	2f	CN	Propyl	92
7	2g	Br	Me	88
8	2h	Br	CH_2COOEt	89
9	2i	F	Propyl	95
10	2j	CN	Propergyl	94

Antimicrobial activity

Synthesised compounds were effective in controlling the growth of bacterial strains tested (Table 2). Study revealed that, the antibacterial activity of compound **2a** was highest with the zone of inhibition of 19.00 ± 0.39 to 16.60 ± 0.31 respectively. Lowest inhibition values were recorded in compound **2j** in the range of 7.67 ± 0.25 to 4.87 ± 0.22. Rest of the compounds i. e., **2b-i** was showed moderate activity. *Bacillus flexus* was the most susceptible and *Pseudomonas aeruginosa* was the least amongst the bacterial strains investigated.

Ciprofloxacin was used as a positive experimental control against strains assayed and produced a zone of inhibition of 22.80 ± 0.18 to 15.00 ± 0.36. No inhibitory effect was observed for DMSO and used as negative control. None of these compounds (2a-j) were showed antifungal activity in *in-vitro* studies against the tested fungal strains (Table 2). But fluconazole was used as a positive experimental control and showed 7.33 ± 0.16 to 8.60 ± 0.31 mm zone of inhibition.

It is observed that, the synthesized compounds were found to be effective towards antibacterial substances for *Bacillus flexus* and *Pseudomonas aeruginosa*. Their activity is probably due to their ability to react with extracellular and soluble proteins and to complex with bacterial cell walls (Cowan 2002). From the results, it can be concluded that the compound **2a** is more effective than compound **2j** and proved to be a better antibacterial agent. In future, it can be used as leading molecule in drug design i. e., in inhibiting these bacterial strains. Thus, such screening of various compounds towards identifying the active agents is an essential factor for the successful prediction of lead molecules in drug designing.

Table 2: Antimicrobial activity of synthesized compounds (2a-j)

Compound	Antimicrobial activity			
	Bacteria		Fungus	
	Bacillus flexus	Pseudomonas aeruginosa	Scopulariopsis brevicaulis	Aspergillus tereus
2a	19.00 ± 0.39	16.60 ± 0.31	-	-
2b	12.53 ± 0.29	5.53 ± 0.24	-	-
2c	12.33 ± 0.20	6.60 ± 0.31	-	-
2d	14.27 ± 0.15	10.37 ± 0.20	-	-
2e	14.93 ± 0.18	10.77 ± 0.39	-	-
2f	15.43 ± 0.30	11.60 ± 0.23	-	-
2g	15.97 ± 0.15	11.83 ± 0.27	-	-
2h	14.60 ± 0.31	9.90 ± 0.38	-	-
2i	12.87 ± 0.47	7.93 ± 0.18	-	-
2j	7.67 ± 0.25	4.87 ± 0.22	-	-
Ciprofloxacin	22.80 ± 0.18	15.00 ± 0.36	-	-
Fluconazole	-	-	7.33 ± 0.16	8.60 ± 0.31

The values are the mean of three experiments ± S. E.

***In vitro* antioxidant activity**

Percentage inhibition of DPPH was the parameter widely used to measure antioxidant/free radical scavenging power. The synthesised compound showed potent free radical scavenging activity with the highest percentage inhibition of 85.78 at 100 µg/mL (fig. 1). DPPH is a reactively stable free radical. The assay is based on the scavenging ability of antioxidants towards the stable radical DPPH. DPPH gives a strong absorption band at 517 nm in the visible region.

When the odd electron becomes paired off in the presence of a free radical scavenger, the absorption reduces and the DPPH solution is decolourised as the colour changes from deep violet to light yellow. The degree of reduction in absorbance measurement is indicative of the radical scavenging power of the synthesised molecules. The compound **2d** appeared to be potent with a maximum inhibition of 85.78 % at 100 µg/mL concentrations and comparatively compound **2e** showed lesser activities with 30.47 % inhibition at the same concentration. From the study, it may be postulated that the compound **2d** reduces the radical into corresponding hydrazine upon reaction with the hydrogen donors in the antioxidant principles.

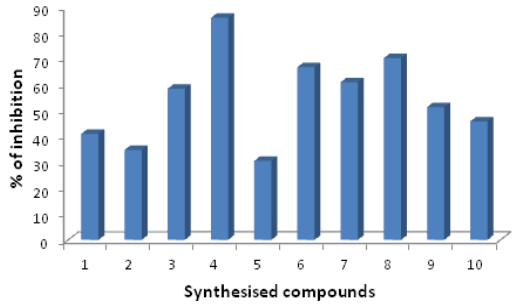


Fig. 1: Free radical scavenging using DPPH radical

In silico molecular docking studies

To identify potential antimicrobial and antioxidant lead among the compounds **2a-j**, docking calculations were performed using Autodock v3. The docking of molecules **2a-j** with Glutamine amido transferase domain reveals that, our synthesized molecule which are having inhibitory capability are exhibiting the interactions with one or the other amino acids in the active pockets which is showed in fig. 2. The docking results for inhibitor compounds are documented in table 2. All the ten molecules showed good binding energy and docking energy ranging from -6.25 kJmol^{-1} to -8.03 kJmol^{-1} and -7.04 kJmol^{-1} to -10.11 kJmol^{-1} respectively. Among the ten molecules, docking of GlcN-6-P synthase with compound **2a** revealed four hydrogen bonds and its binding energy and docking energy were -6.36 kJmol^{-1} and -8.01 kJmol^{-1} respectively, and it may be considered as good inhibitor of GlcN-6-P synthase. All the synthesized molecules were completely enfolded in the entire active pocket of GlcN-6-P synthase (fig. 2). The topology of the active site of GlcN-6-P synthase was similar in all synthesized molecules, which is lined by interacting amino acids as predicted from the ligplot (fig. 3). In *In vitro* studies also compound **2a** has emerged as a active molecule against all the screened microorganisms, so it can be predicted as the activity may be due to inhibition of enzyme GlcN-6-P synthase, which catalyses a complex reaction involving ammonia transfer from L-glutamine to Fru-6-P, followed by isomerisation of the formed fructosamine-6-phosphate to glucosamine-6-phosphate. Hence, this study has been proved that the molecule **2a** to be one of the potent antibacterial agents.

After docking the synthesised molecule with Superoxide dismutase, the compounds bound exactly at the active site of Superoxide dismutase, which was shown in fig. 4. A careful inspection of the binding pocket indicated that the active site of SOD was similar in all synthesised compounds at the Cu-Zn domain of SOD (fig. 5). Target information and docking details for the compounds were tabulated in table 3. Among the ten molecules, docking of SOD with compound **2d** revealed four hydrogen bonds and its binding energy and docking energy were -8.76 kJmol^{-1} and -11.27 kJmol^{-1} respectively, and it may be considered as good inhibitor of Superoxide dismutase.

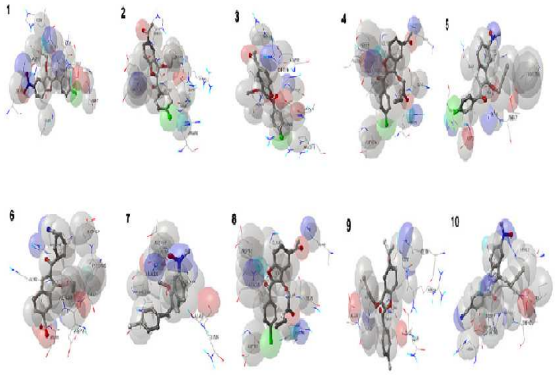


Fig. 2: Enfolding of synthesized molecules in the active pocket

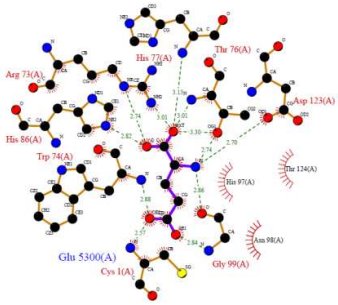


Fig. 3: Interacting amino acids as predicted from the ligplot

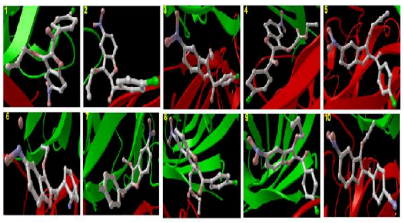


Fig. 4: Ligands docked in best of its conformation

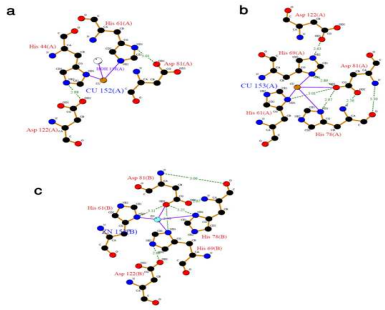


Fig. 5: Ligplot results for SOD. a. Showing the binding of ligand CU on A-chain amino acids present in an active pocket of SOD with two hydrogen bonds. b. Binding of ligand CU on A-chain amino acids present in another active pocket of SOD with six hydrogen bonds. c. Showing the binding of ligand ZN on B-chain amino acids present in an active pocket of SOD with six hydrogen bonds

CONCLUSION

In conclusion, synthesis of ten individually novel (3-alkoxy-5-nitrobenzofuran-2-yl)(phenyl)methanone (**2a-j**) derivatives have

been successfully made through industrially simple method. Studied preliminary antimicrobial, antioxidant and docking properties. Perhaps the present work provides an evidence for the preparation, consistent antioxidant activity as well as significant protection against bacterial infections of novel benzofuran derivatives.

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CONFLICT OF INTERESTS

Declared None

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