

SYNTHESIS, CHARACTERIZATION AND BIOLOGICAL ACTIVITIES OF SOME BENZOFUROPYRIMIDINE DERIVATIVES

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ABSTRACT

A series of new 8-bromo-3-[[phenylmethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one derivatives 4a-r have been synthesized by using ethyl 3-amino-5-bromo-¹-benzofuran-2-carboxylate 1 as a starting material. The amino ester 1 was converted into its hydrazide 2 by reacting with hydrazine hydrate, which upon treatment with various aldehydes and ketones yielded corresponding Schiff bases 3a-r. The desired compounds 4a-r were obtained by reacting Schiff bases 3a-r with triethyl orthoformate. The structures of all the synthesized compounds were established by analytical and spectral studies. The compounds 4a-r were evaluated for their antimicrobial by cup plate method and antioxidant activity by DPPH radical scavenging method. Some of the compounds exhibited considerable antimicrobial activities and compounds 4o and 4r were found to be excellent in scavenging DPPH radical at 50 µg and also retained almost same activity in 100 µg.

Keywords: Benzofuran, Fused pyrimidine, Antimicrobial, Antioxidant activity.

INTRODUCTION

The synthesis of benzofuran derivatives has received considerable attention due to their occurrence in large number of natural products [1] and synthetic pharmaceuticals [2]. Various benzofurans also find application as fluorescent sensors [3], oxidants [4], antioxidants, a variety of drugs and in other field of chemistry and agriculture [5]. The compounds encompassing pyrimidine moiety have been shown to be useful as bactericidals, antimalarials [6], analgesics [7], antihypertensive [8] and anti-tumor agents [9]. Pyrimidines and their ring-fused derivatives have a broad spectrum of biological activity; best known as the heterocyclic core of the nucleic acid bases. Fused heterocyclic structures containing pyrimidine and furan ring systems are well known to possess diverse biological activities such as antimicrobial [10, 11], anti-inflammatory [12], antiviral [13], anti-HIV [14] and antitumor [15]. Moreover the derivatives of benzofuroprymidines are of great importance because they are associated with analgesic, anti-inflammatory and antimicrobial activities [16, 17]. Some of these derivatives are also reported to exhibited good anticoccidial and blood sugar-lowering activities [18, 19]. Considering the importance of benzofuroprymidines, some of the synthetic methodologies for these fused heterocycles have been reported [20, 21].

Inspired by these reports, it was planned to synthesize a new series of benzofuroprymidines, i.e., various 8-bromo-3-[[1-phenylethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-ones (4a-r) and evaluate their antibacterial, antifungal and in-vitro antioxidant activities. The structures of the synthesized compounds were confirmed on the basis of IR, ¹H NMR, mass spectral data and supported by analytical data.

MATERIALS AND METHODS

Melting points were determined in an open capillary melting point apparatus and are uncorrected. Purity of the compounds was checked by TLC on silica gel. The IR spectra were recorded on a Nicolet Impact 410 FT-IR Spectrophotometer, using KBr pellets. ¹H NMR spectra were recorded on a Bruker-400 MHz spectrometer in CDCl₃ or DMSO using TMS as an internal standard. High-resolution mass spectra (HRMS-ESI) were obtained on a MicroTM Q-TOF Mass Spectrometer. All reagents were AR grade or chemically pure.

Synthesis of 3-amino-5-bromo-¹-benzofuran-2-carbohydrazide (2)

A mixture of ethyl 3-amino-5-bromobenzofuran-2-carboxylate (1) (2.9 g, 0.01 mole) and hydrazine hydrate (4 ml, 99%) was heated under reflux in ethanol (5 ml) on a steam bath for 3 hours and the reaction was monitored by TLC. The reaction product was cooled

thoroughly and colorless solid that separated and was collected by filtration. It was recrystallized from ethanol as colorless thin needles. Yield 74%; m.p. 238-240°C.

General procedure for synthesis of 3-amino-5-bromo-N'-[phenylmethylidene]-¹-benzofuran-2-carbohydrazides (3a-k) and 3-amino-5-bromo-N'-[1-phenylethylidene]-¹-benzofuran-2-carbohydrazides (3l-r)

The mixture of compound 2 (0.27 g, 1 mmol) and benzaldehyde (0.10 ml, 1 mmol) in DMF (4 ml) was refluxed at 110-120°C for about 3 hours. Progress of the reaction was monitored by TLC. After the completion of reaction, the reaction mixture was cooled to room temperature and added to ice cold water with constant stirring. The solid obtained was filtered, dried and recrystallised from ethanol.

The similar procedure was followed for the preparation of compounds 3b-r using substituted aldehydes and acetophenones.

3-Amino-5-bromo-N'-[phenylmethylidene]-¹-benzofuran-2-carbohydrazide (3a): IR ν_{max} (KBr) ν cm⁻¹: 3422, 3298, 3065, 2975, 2919, 1968, 1840, 1691. ¹H NMR (CDCl₃) δ ppm (TMS): 5.1 (s, -NH₂), 9.2 (s, -NH), 8.1 (s, 1H), 7.2-7.7 (m, 8H); MS: m/z = 359(M+H)⁺, 361(M+H+2)⁺.

8-Bromo-3-[[1-phenylethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one(3l): IR ν_{max} (KBr) ν cm⁻¹: 3440, 3359, 1952, 1635, 1568. ¹H NMR (CDCl₃) δ ppm (TMS): 2.2 (s, 3H), 5.0 (s, -NH₂), 11 (s, NH), 7.4-8.2 (m, 8H); MS: m/z = 373 (M+H)⁺, 375 (M+H+2)⁺.

General procedure for the synthesis of 8-bromo-3-[[phenylmethylidene] amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-ones (4a-k) and 8-bromo-3-[[1-phenylethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-ones (4l-r)

The compound 3a (0.5 g, 1.4 mmol) was refluxed with triethyl orthoformate (5 ml) for about 5 hours. Excess of triethyl orthoformate was removed by distillation under reduced pressure. The solid obtained was dried and recrystallized from ethanol.

The similar procedure was followed for the synthesis of compounds 3b-r.

8-Bromo-3-[[phenylmethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4a):

IR ν_{max} (KBr) ν cm⁻¹: 3058, 1697, 1611, 1566, 1525. ¹H NMR (CDCl₃) δ ppm (TMS): 9.5 (s, 1H), 8.4 (s, 1H), 7.4-8.2 (m, 8H), MS: m/z = 369 (M+H)⁺, 371 (M+H+2)⁺.

8-Bromo-3-[[2-chlorophenyl)methylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4b): IR ν_{\max} (KBr) ν cm^{-1} : 3059, 1680, 1600, 1550, 1480. ^1H NMR (CDCl_3) δ ppm (TMS): 9.6 (s, 1H), 8.8 (s, 1H), 7.5-8.2 (m, 7H). MS: m/z = 403 (M+H)⁺, 405(M+H+2)⁺.

8-Bromo-3-[[4-chlorophenyl)methylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4c): IR ν_{\max} (KBr) ν cm^{-1} : 3059, 2961, 1910, 1669, 1607, 1567, 1529. ^1H NMR (CDCl_3) δ ppm (TMS): 9.2 (s, 1H), 8.7 (s, 1H), 7.5-8.2 (m, 7H). MS: m/z 403(M+H)⁺, 405(M+H+2)⁺.

8-Bromo-3-[[3-nitrophenyl)methylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4d): IR ν_{\max} (KBr) ν cm^{-1} : 3085, 2963, 1698, 1613, 1571, 1528. ^1H NMR (CDCl_3) δ ppm (TMS): 9.7 (s, 1H), 8.9 (s, 1H), 7.5-8.3 (m, 7H), MS: m/z = 414 (M+H)⁺, 416 (M+H+2)⁺.

8-Bromo-3-[[4-methoxyphenyl)methylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4e): IR ν_{\max} (KBr) ν cm^{-1} : 3061, 3026, 2963, 2836, 1705, 1603, 1513. ^1H NMR (CDCl_3) δ ppm (TMS): 9.0 (s, 1H), 8.7 (s, 1H), 8.2(d, 1H) 7.8-7.9 (m, 4H), 7.1 (d, 2H), 3.8 (s, 3H). MS: m/z = 399 (M+H)⁺, 401 (M+H+2)⁺.

8-Bromo-3-[[4-hydroxyphenyl)methylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4f): IR ν_{\max} (KBr) ν cm^{-1} : 3278, 3088, 1902, 1676, 1598, 1517. ^1H NMR (CDCl_3) δ ppm (TMS): 11.3 (s, -OH), 9.6 (s, 1H), 8.6 (s, 1H), 7.5-8.3 (m, 7H). MS: m/z = 385 (M+H)⁺, 387 (M+H+2)⁺.

8-Bromo-3-[[2-hydroxyphenyl)methylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4g): IR ν_{\max} (KBr) ν cm^{-1} : 3300, 3100, 1902, 1679, 1598, 1517. ^1H NMR (CDCl_3) δ ppm (TMS): 11.1 (s, -OH), 9.4 (s, 1H), 8.5 (s, 1H), 7.4-8.3 (m, 7H). MS: m/z = 385 (M+H)⁺, 387 (M+H+2)⁺.

8-Bromo-3-[[5-bromo-2-hydroxyphenyl)methylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4h): IR ν_{\max} (KBr) ν cm^{-1} : 3300, 2988, 1900, 1670, 1598, 1517, 1440. ^1H NMR (CDCl_3) δ ppm (TMS): 11.5 (s, -OH), 9.4 (s, 1H), 7.5-8.3 (m, 7H). MS: m/z = 464 (M+H)⁺, 466 (M+H+2)⁺.

8-Bromo-3-[[4-hydroxy-3-methoxyphenyl)methylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4i): IR ν_{\max} (KBr) ν cm^{-1} : 3300, 3179, 2936, 2840, 1897, 1668, 1584, 1517. ^1H NMR (CDCl_3) δ ppm (TMS): 11.4 (s, -OH), 9.5 (s, 1H), 8.7 (s, 1H), 7.3-8.3 (m, 6H), 4.0 (s, 3H). MS: m/z = 415 (M+H)⁺, 417 (M+H+2)⁺.

8-Bromo-3-[[furan-2-ylmethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4j):

IR ν_{\max} (KBr) ν cm^{-1} : 3088, 2982, 2933, 1685, 1605. ^1H NMR (CDCl_3) δ ppm (TMS): 9.6 (s, 1H), 8.3 (s, 1H), 7.4-8.2 (m, 6H). MS: m/z = 359 (M+H)⁺, 361 (M+H+2)⁺.

8-Bromo-3-[[1E,2Z]-3-phenylprop-2-en-1-ylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4k): IR ν_{\max} (KBr) ν cm^{-1} : 3059, 2969, 1958, 1895, 1598, 1704, 1623. ^1H NMR (CDCl_3) δ ppm (TMS): 9.6 (s, 1H), 8.6 (s, 1H), 7.5-8.4(m, 8H), 6.0(s, 1H), 4.1 (s, 1H). MS: m/z = 395 (M+H)⁺, 397 (M+H+2)⁺.

8-Bromo-3-[[1-phenylethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4l): IR ν_{\max} (KBr) ν cm^{-1} : 3080, 2973, 2924, 1920, 1680, 1598. ^1H NMR (CDCl_3) δ ppm (TMS): 8.6 (s, 1H), 7.2-8.2 (m, 8H), 2.1 (s, 3H). MS: m/z = 383 (M+H)⁺, 385 (M+H+2)⁺.

8-Bromo-3-[[1-(4-chlorophenyl)ethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4m): IR ν_{\max} (KBr) ν cm^{-1} : 3050, 2955, 2899, 1900, 1669, 1600, 1567, 1529. ^1H NMR (CDCl_3) δ ppm (TMS): 8.7 (s, 1H), 7-8.2 (m, 7H), 2.3 (s, 3H). MS: m/z = 383 (M+H)⁺, 385 (M+H+2)⁺.

8-Bromo-3-[[1-(4-nitrophenyl)ethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4n): IR ν_{\max} (KBr) ν cm^{-1} : 3185, 2960, 2890, 1698, 1600, 1570, 1518. ^1H NMR (CDCl_3) δ ppm (TMS): 8.9 (s, 1H), 7.3-8.4 (m, 7H), 2.4 (s, 3H). MS: m/z = 428 (M+H)⁺, 430 (M+H+2)⁺.

8-Bromo-3-[[1-(4-methoxyphenyl)ethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4o): IR ν_{\max} (KBr) ν cm^{-1} : 3100, 3020, 2960, 2846, 1700, 1613, 1500. ^1H NMR (CDCl_3) δ ppm (TMS): 8.7 (s, 1H), 8.2-7.3 (m, 7H), 3.7 (s, 3H), 2.3(s, 3H). MS: m/z = 413 (M+H)⁺, 415 (M+H+2)⁺.

8-Bromo-3-[[1-(4-methylphenyl)ethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4p): IR ν_{\max} (KBr) ν cm^{-1} : 3026, 2950, 2845, 1700, 1618, 1520. ^1H NMR (CDCl_3) δ ppm (TMS): 8.5 (s, 1H), 7.2-8 (m, 7H), 2.3 (s, 3H), 1.7 (s, 3H). MS: m/z = 397 (M+H)⁺, 399 (M+H+2)⁺.

8-Bromo-3-[[1-(2-hydroxyphenyl)ethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4q): IR ν_{\max} (KBr) ν cm^{-1} : 3300, 3100, 1902, 1679, 1598, 1517. ^1H NMR (CDCl_3) δ ppm (TMS): 11.2 (s, -OH), 8.5 (s, 1H), 7.4-8.3 (m, 7H), 2.1 (s, 3H). MS: m/z = 399 (M+H)⁺, 401 (M+H+2)⁺.

8-Bromo-3-[[1-(4-hydroxyphenyl)ethylidene]amino][1]benzofuro[3,2-d]pyrimidin-4(3H)-one (4r): IR ν_{\max} (KBr) ν cm^{-1} : 3402, 3054, 2992, 2929, 1883, 1700, 1594, 1563, 1529. ^1H NMR (CDCl_3) δ ppm (TMS): 11.5 (s, -OH), 8.6 (s, 1H), 8.2 (d, 1H), 6.9-7.9 (m, 6H), 2.3 (s, 3H) MS: m/z = 399 (M+H)⁺, 401 (M+H+2)⁺.

Biological activity

Antimicrobial activity

Cultures of two gram-positive bacteria: *Bacillus subtilis* (AL009126) and *Staphylococcus aureus* (BX571856) and two gram-negative bacteria: *Escherichia coli* (AE014075) and *Pseudomonas aeruginosa* (AE004091) were used to investigate the antibacterial activity and *Aspergillus fumigatus* (CM000170) and *Candida albicans* (EAL02784) for the antifungal activity of the compounds (4a-r). The antimicrobial activity was assayed biologically using agar well diffusion method. In this method, wells of standard diameter were made in the nutrient agar medium and potato dextrose agar for the antibacterial and antifungal activity respectively containing standard microbial inoculums. The test compounds were introduced into the wells and diameter of the zone of inhibition was measured by antibiotic zone reader. The test was carried out in triplicates and average value of these as a zone of inhibition. The final values of the compounds were tested at a concentration of 50 $\mu\text{g/ml}$ and 100 $\mu\text{g/ml}$ in dimethyl sulphoxide as control against all the organisms. Ampicillin/Ciprofloxacin and Fluconazole were used as standards for comparison of antibacterial and antifungal activities, respectively at the same concentration as samples taken. The zone of inhibition was compared with the standard drugs after 24 h of incubation at 37 °C for antibacterial activity and 72 h at 25° C for antifungal activity.

Evaluation of Anti-oxidant activity-DPPH radical scavenging method

The free radical scavenging capacities of the compounds 4a-r were determined by using DPPH (1, 1-diphenyl-2-picrylhydrazyl radical). The DPPH radical scavenging assay is the simplest method to measure the ability of antioxidants to intercept free radicals. Antioxidants react with DPPH \cdot , which is a stable free radical, and then scavenge this radical by converting it to 1, 1-diphenyl-2-picrylhydrazine due to their H-donating ability. The degree of DPPH free radical scavenging potential of the compounds 4a-r was determined. Freshly prepared DPPH (3 ml of 0.1 mmole) was mixed with serial dilutions (50 μg and 100 μg) of compounds to a final volume of 4 ml and after 30 min of incubation in dark at room temperature, the absorbance was read at 517 nm using a spectrophotometer (Shimadzu UV-visible spectrophotometer, Japan). The DPPH control (containing no sample) was prepared using the same procedure. The inhibition concentration calculated using values obtained [22].

The capability to scavenge the DPPH \cdot radical was calculated using the following equation:

$$\text{DPPH}^\cdot \text{ Scavenging effect (\%)} = \left[\frac{A_{\text{control}} - A_{\text{sample}}}{A_{\text{control}}} \right] \times 100$$

Where A Control = The initial concentration of the stable DPPH radical without the test compound.

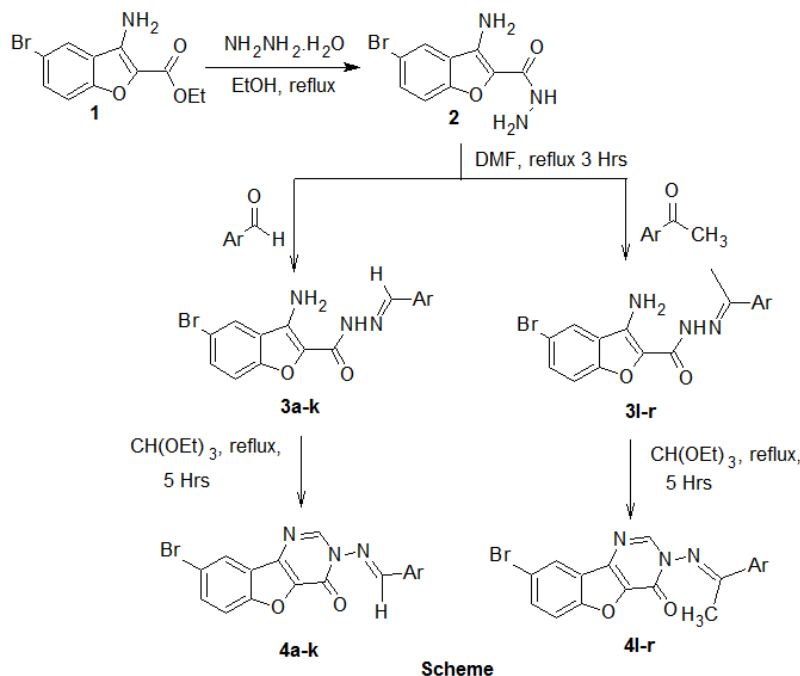
A Sample = Absorbance of the remaining concentration of DPPH \cdot in the presence of compound (4a-r).

Ascorbic acid, BHT (Butylated hydroxytoluene) and BHA (Butylated hydroxyanisole) have been used as standards.

RESULTS AND DISCUSSION

In the present work, the synthesis of fused bromo substituted benzofuroprymidines associated with imine group was accomplished by series of the reactions as shown in the Scheme. In order to achieve synthesis of the desired compounds, ethyl 3-amino-5-bromo benzofuran-2-carboxylate (1) was used as starting material which was synthesized by a reported method [23]. It was converted into 3-amino-5-bromo-1-benzofuran-2-carbohydrazide (2) by refluxing with excess of hydrazine

hydrate in ethanol. The condensation of hydrazide 2 with different aldehydes and acetophenones in DMF furnished corresponding Schiff bases 3a-r. The series of 3a-r compounds were treated with triethylorthoformate to yield 8-bromo-3-[[phenylmethylidene]amino] [1]benzofuro[3,2-d] pyrimidin-4(3H)-ones (4a-k) and 8-bromo-3-[[1-phenylethylidene]amino][1] benzofuro [3,2-d]pyrimidin-4(3H)-ones (4l-r). All the compounds synthesized were obtained in good yield, their corresponding melting points and analytical data shown in Table1.



Whereas Ar,

4a=C ₆ H ₅	4g=2-OH C ₆ H ₄ -	4m=4-Cl C ₆ H ₄ -
4b =2-Cl C ₆ H ₄ -	4h=5-Br-2-OH C ₆ H ₃ -	4n=4-NO ₂ C ₆ H ₄ -
4c =4-Cl C ₆ H ₄ -	4i=4-OH-3-OCH ₃ C ₆ H ₃ -	4o=4-OCH ₃ C ₆ H ₄ -
4d = 3-NO ₂ C ₆ H ₄ -	4j=C ₄ H ₄ O-	4p=4-CH ₃ C ₆ H ₄
4e =4-OCH ₃ C ₆ H ₄ -	4k=C ₆ H ₅ CH=CH-	4q=2-OH C ₆ H ₄ -
4f = 4-OH C ₆ H ₅ -	4l =C ₆ H ₅ -	4r=4-OH C ₆ H ₄ -

The characterizations of the synthesized compounds were carried out by FT-IR, ¹HNMR and mass spectral studies. The IR spectrum of hydrazide 2 exhibited absorption bands at 3402 cm⁻¹, 3307 cm⁻¹, 3201 cm⁻¹ and 1856 cm⁻¹ due to -NH, -NH₂, =C-H (aromatic) and -C=O groups respectively. The ¹HNMR spectrum of compound 2 showed three D₂O exchangeable peaks as three singlets. One singlet appeared at δ 4.3 due to two protons of -NH-NH₂ group, second singlet at δ 5.9 due to two protons at -NH₂ group at C-3 and third singlet at δ 9.2 due to one proton at -NH-NH₂ group respectively. Three aromatic protons appeared as one singlet at δ 8.1 and two doublets at δ 7.5 and δ 7.3. The mass spectrum of compound 2 exhibited molecular ion peak at m/z 270 (M+H)⁺ and at m/z 272 (M+H+2)⁺ of equal intensity as expected for the compounds containing bromine atom.

The conversion of hydrazide 2 into Schiff base 3a was evidenced by ¹HNMR and mass spectral data. The ¹HNMR spectrum exhibited a D₂O exchangeable singlet at δ 5.1 assignable to protons of -NH₂ group at C-3 and another D₂O exchangeable singlet at δ 9.2 due to -

NH proton. Disappearance of singlet at δ 4.3 due to -NH₂ protons of hydrazide group and appearance of peak integrating for one proton at δ 8.1 due to -CH=N- group along with the multiplet integrating for eight protons at δ 7.2-7.7 confirmed the formation of Schiff base. Its mass spectrum containing two peaks at m/z 359 (M+H)⁺ and m/z 361 (M+H+2)⁺ supported the assigned structure.

The formation of 8-bromo-3-[[phenylmethylidene]amino][1] benzofuro[3,2-d]pyrimidin-4(3H)-one 4a was established by conspicuous absence of singlets at δ 5.1 and δ 9.2 in ¹HNMR spectrum indicating the involvement of -NH₂ and -NH groups in cyclization. A proton of -CH of pyrimidine resulted in a peak as singlet at δ 9.5 whereas -CH=N- proton gave a singlet at δ 8.4 and aromatic protons appeared as multiplet between δ 7.4 and δ 8.4. As expected two molecular ion peaks at m/z 369 (M+H)⁺ and m/z 371 (M+H+2)⁺ were observed in its mass spectrum. Similarly, structures of all other compounds were confirmed and are given in experimental section.

Table 1: Physical and Analytical data of synthesized compounds

Compound	Molecular formula	Yield % ^a	Melting point °C	Analytical data % Found (Calculated)		
				C	H	N
4a	C17H10BrN3O2	90	246-248	55.30 (55.46)	2.62 (2.74)	11.30 (11.41)
4b	C17H9BrClN3O2	85	250-252	50.65 (50.71)	2.20 (2.25)	10.35 (10.44)
4c	C17H9BrClN3O2	85	252-254	50.60 (50.71)	2.10 (2.25)	10.20 (10.44)
4d	C17H9BrN4O4	90	168-170	49.20 (49.42)	2.10 (2.20)	13.40 (13.56)
4e	C18H12BrN3O3	86	220-222	54.20 (54.29)	2.95 (3.04)	12.00 (12.05)
4f	C17H10BrN3O3	91	264-266	53.05 (53.15)	2.10 (2.26)	10.80 (10.94)
4g	C17H10BrN3O3	87	266-268	53.00 (53.15)	2.20 (2.26)	10.80 (10.94)
4h	C17H9Br2N3O3	95	270-272	44.00 (44.09)	1.80 (1.96)	9.00 (9.07)
4i	C18H12BrN3O4	75	252-254	52.05 (52.19)	2.80 (2.92)	10.10 (10.14)
4j	C15H8BrN3O3	80	260-264	50.20 (50.30)	2.25 (2.10)	11.60 (11.73)
4k	C19H12BrN3O2	70	220-222	57.50 (57.89)	3.00 (3.07)	10.50 (10.66)
4l	C18H12BrN3O2	90	218-220	57.70 (57.89)	3.00 (3.07)	10.50 (10.66)
4m	C18H11BrClN3O2	70	110-112	51.75 (51.89)	2.50 (2.66)	10.00 (10.09)
4n	C18H11BrN4O4	75	128-130	51.50 (51.89)	2.40 (2.66)	10.02 (10.09)
4o	C19H14BrN3O3	70	162-164	55.20 (55.36)	3.35 (3.42)	10.05 (10.19)
4p	C19H14BrN3O2	70	110-112	57.20 (57.59)	3.40 (3.56)	10.40 (10.60)
4q	C18H12BrN3O3	72	250-252	54.10 (54.29)	2.95 (3.04)	10.40 (10.56)
4r	C18H12BrN3O3	80	118-120	54.10 (54.29)	3.00 (3.04)	10.40 (10.56)

^a=Isolated yield

Investigation of antimicrobial activity revealed that 4c, 4d, 4e, 4g, 4j, 4p and 4q exhibit considerable activity against all the four bacteria whereas 4a, 4f and 4m were moderately active against *Bacillus subtilis*, *Escherichia coli* and *Pseudomonas aeruginosa* but inactive against *Staphylococcus aureus* when compared with standard drugs at the concentration of 50 µg and 100 µg. Among all the tested

compounds 4c, 4d, 4f, 4i, 4l, 4m, 4n, 4o and 4r showed moderate antifungal activity and 4a, 4b, 4g, 4h and 4p were significant antifungal activity against only *Candida albicans* when compared with the standard flucanazole at the concentration of 50 µg and 100 µg. The remaining compounds exhibited poor antimicrobial activity. The results are reported in Table 2.

Table 2: Antimicrobials activity of synthesized compounds (4a-r)

Tested samples	Zone of inhibition in mm											
	Antibacterial activity						Antifungal activity					
	B. s		S. a		E. c		P. a		A. f		C. a	
	A	B	A	B	A	B	A	B	A	B	A	B
4a	-	-	5	12	-	-	6	10	6	8	13	15
4b	6	10	-	-	8	10	4	8	3	4	12	15
4c	8	12	6	10	10	14	10	12	8	10	10	15
4d	6	10	8	10	7	10	9	13	8	15	10	10
4e	7	10	8	10	9	11	10	10	8	15	10	10
4f	-	-	10	12	5	10	5	8	10	10	10	10
4g	5	12	8	10	6	10	8	10	8	8	13	15
4h	7	11	8	12	-	5	-	-	7	9	12	10
4i	-	-	5	8	-	-	7	10	8	10	10	17
4j	8	12	6	10	7	10	5	12	7	8	12	20
4k	3	5	-	-	5	12	-	4	5	10	8	12
4l	5	8	8	14	6	10	2	5	8	10	10	12
4m	-	-	5	10	7	14	-	-	8	10	8	14
4n	5	7	3	8	8	12	5	8	6	10	6	12
4o	3	5	-	4	4	8	-	-	6	12	8	12
4p	6	10	8	10	8	14	8	12	-	-	12	14
4q	10	14	8	10	5	8	6	10	6	10	8	10
4r	6	12	4	9	-	-	7	14	10	12	12	14
Control	0	0	0	0	0	0	0	0	0	0	0	0
Ciprofloxacin	22	24	20	22	20	20	20	20	N	N	N	N
Ampicillin	15	15	10	10	15	14	11	10	N	N	N	N
Fluconazole	N	N	N	N	N	N	N	N	25	25	22	24

Where control=DMSO, N=not applicable, -=inactive, B.s=*Bacillus subtilis*, S.a=*Staphylococcus aureus*, E.c=*Escherichia coli*, P.a=*Pseudomonas aeruginosa*, A.f=*Aspergillus fumigatus*, C.a=*Candida albicans*, A=50 µg and B=100 µg.

In order to evaluate the antioxidant activity and free radical scavenging potentials of compounds 4a-r were tested at 50 µg and 100 µg by the DPPH radical scavenging method. Antioxidant reacts with DPPH, which is a stable free radical, and converts it to 1, 1-diphenyl-2-picrylhydrazine. The degree of discoloration indicates the scavenging potentials of the antioxidant compound. The experiment was done in triplicate and average value has taken as

% of scavenging. On comparison of screened samples with standards ascorbic acid (A A), BHT and BHA, compounds 4o and 4r were shown to be excellent in scavenging DPPH radical at 50 µg and also retained almost same activity in 100 µg. But compound 4m showed appreciable scavenging activity compared to all derivatives. The results are results were graphically presented in Figure 1.

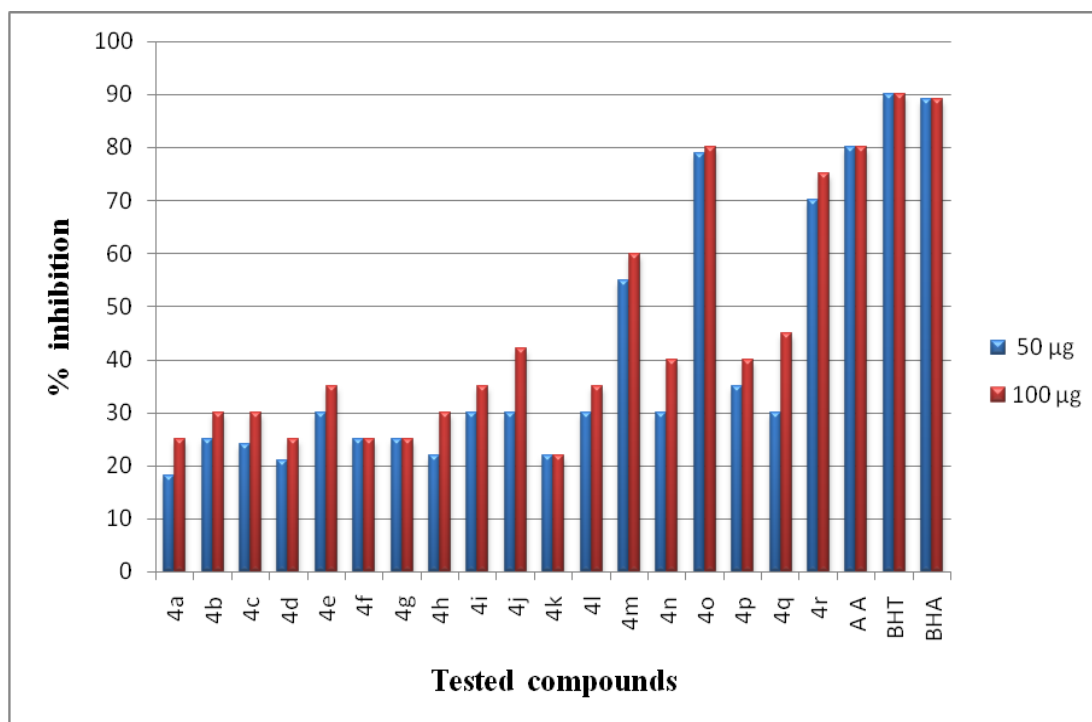


Fig 1: DPPH radical scavenging activity of synthesized compounds 4a-r

CONCLUSION

In conclusion, the synthesis of fused benzofuroprymidines was achieved by an efficient and simple approach. The results of present investigation clearly indicate that the antibacterial and antifungal activity vary with the species of organisms used and molecular structure. In most of the tested samples, they showed comparable fungicidal activity against *Candida albicans* than *Aspergillus fumigatus* and in case antibacterial activity most of the molecules screened were showed appreciable inhibition at 100 µg compared with lower concentrations. The compounds 4o and 4r showed promising free radical scavenging activity as compared with standards used.

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REFERENCES

- Zeni G, Larock RC, Synthesis of heterocycles via palladium p-olefin and p-alkyne chemistry. *Chem Rev*, 2004;104:2285-2310.
- Nagahara T, Yokoyama T, Inamura K, Katakura S, Komoriya S, Yamaguchi H, et al., Dibasic (amidinoaryl)propanoic acid derivatives as novel blood coagulation factor xa inhibitors. *J Med Chem*, 1994;37:1200-1207.
- Oter O, Ertekin K, Kirilmis C, Koca M, Ahmedzade M, Characterization of a newly synthesized fluorescent benzofuran derivative and usage as a selective fiber optic sensor for Fe(III). *Sensor Actuat B-Chem*, 2007;122:450-456.
- Karatas F, Koca M, Kara H, Servi S, Synthesis and oxidant properties of novel (5-bromobenzofuran-2-yl)(3-methyl-3-mesitylcyclobutyl)ketonethiosemicarbazone. *Eur J Med Chem*, 2006;41:664-669.
- Habermann J, Ley SV, Smits R, Three-step synthesis of an array of substituted benzofurans using polymer-supported reagents. *J Chem Soc Perkin Trans 1*, 1999;1:2421-2423.
- Campaigne, E, Ellis RL, Bradford M, Ho J, Synthesis of Some Lreidodihydrofurans and Related Pyrimidones as Potential Antimalarials. *J Med Chem*, 1969;12:339-342.
- Regnier G, Canevar L, Le RJ, Douarec JC, Halstop S, Daussy J, Triphenylpropylpiperazine Derivatives as New Potent Analgetic Substances. *J Med Chem*, 1972;15:295-298.
- Winter CA, Fisley EAR, Nuss GW, Carrageenin-induced edema in hind paw of the rat as an assay for antiinflammatory drugs. *Proc Soc Exp Biol Med*, 1962;111:544-547.
- Sugaira K, Schmid AF, Schmid MM, Brown FG, Effect of compounds on a spectrum of rat tumors. *Cancer Chemother Rep 1*, 1973;23 (1): 231-233.
- Manna F, Chimenti F, Fioravanti R, Bolasco A, Secci D, Chimenti P, Ferlini C, Scambia G, Synthesis of some pyrazoles derivatives and preliminary investigation of their affinity binding to p-glycoprotein. *Bioorg Med Chem Lett*, 2005;15:4632-4635.
- Jantova S, Stankovsky S, Spirkova K, In vitro antibacterial activity of ten series of substituted quinazolines. *Biologia Bratislava*, 2004;59:741-752.
- Harinadhababu V, Manna SK, Srinivasan KK, Bhat VG Synthesis and biological evaluation of 1,3,5-trisubstituted pyrazolines bearing benzofurans. *Indian J Heterocycl Chem*, 2004;13:253-256.
- Nagaraja GK, Prakash GK, Satyanarayan ND, Vaidya VP, Mahadevan KM, Synthesis of novel 2-aryl-2,3-dihydronaphtho[2,1-b]furo[3,2-b]pyridin-4(1H)-ones of biological importance. *Arkivoc*, 2006;15:142-152.
- Maruyama T, Kozai S, Demizu Y, Witvrouw M, Pannecouque C, Balzarini J, Snoecks R, Andrei G, De Clercq E, Synthesis and anti-HIV¹ and anti-HCMV activity of 1-substituted 3-(3,5-dimethyl-benzyl)uracil derivatives. *Chem Pharm Bull*, 2006;5:325-333.
- Naito H, Ohsuki S, Sugimori M, Atsumi R, Minami M, Nakamura Y, et al., Synthesis and antitumor activity of novel pyrimidinyl pyrazole derivatives. II. Optimization of the phenylpiperazine moiety of 1-[5-methyl¹-(2-pyrimidinyl)-4-pyrazolyl]-3-phenylpiperazinyl¹-trans-propenes. *Chem Pharm Bull*, 2002;50:453-462.
- Kikuchi H, Yamamoto K, Horoiwa S, Hirai S, Kasahara R, Hariguchi N, Matsumoto M, Oshima Y, Exploration of a new type of antimalarial compounds based on febrifugine. *J Med Chem*, 2006;49:4698-4706.
- Duval E, Case A, Stein RL, Cuny GD, Structure-activity relationship study of novel tissue transglutaminase inhibitors. *Bioorg Med Chem Lett*, 2005;15:1885-1889.
- Teimouria MB, Bazhrang R, Shaken not stirred: A facile synthesis of 1,4-bis(furo[2,3-d]pyrimidine-2,4(1H,3H)-dione-5-

- yl)benzenes by one-pot reaction of isocyanides, N,N-dimethylbarbituric acid, and terephthalaldehyde. *Bioorg Med Chem Lett*, 2006;16:3697-3701.
19. Zanatta N, Schneider JMFM, Schneider PH, Wouters AD, Bonacorso HG, Martins MAP, Wessjohann LA, Regiospecific Synthesis of 4-Alkoxy and 4-Amino Substituted 2-Trifluoromethyl Pyrroles, *J Org Chem*, 2006;71:6996-6998.
 20. Bodke Y, Sangapure SS, Synthesis and biological activity of some benzofuro[3,2-d]pyrimidines. *J Indian Chem Soc*, 2003;80:187-189.
 21. Parameshwarappa G, Sangapure SS, Synthesis and antimicrobial activities of 2-aryl-3,4-dihydro-4-oxo-5-bromobenzofuro[3,2-d]pyrimidines. *Organic Chemistry An Indian Journal*, 2009;5(1):250-253.
 22. Oyaizu M, Studies on product of browning reaction prepared from glucose amine. *Jpn J Nutri*, 1986;44:307-315.
 23. G. Parameshwarappa, B. Raga and S.S. Sangapure. Synthesis of new benzofuran analogues from 5-Bromosalicylaldehyde and their biological activities. *Organic Chemistry: An Indian Journal*, 2008;4(5):375-379.