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Microwave-Assisted, Solvent Free and Parallel Synthesis of Some Newer 2, 4-Disubstituted 1, 5- Benzodiazepines of Biological Interest

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ABSTRACT

Benzodiazepines and their derivatives were reported to have wide biological activities and were synthesized by the reaction between substituted benzaldehydes and substituted ketones in presence of sodium hydroxide to afford chalcones and further reaction between 1, 2-diamine under smooth condensation with chalcones in presence of glacial acetic acid afforded a new class of 1, 5-benzodiazepines in good yield. 2, 4-disubstituted 1, 5-benzodiazepine derivatives (**1B-19B**) were synthesized by microwave and conventional methods. The synthesized compounds were evaluated for their antibacterial, antifungal, anthelmintic and cytotoxic activity. The chemical structures of the newly synthesized compounds have been confirmed by IR, ^{1}H-NMR, MASS spectral data and elemental analysis. All the synthesized substituted benzodiazepines have shown good antimicrobial activity, moderate to good anthelmintic activity and possessed significant cytotoxic activity.

Keywords: 1, 5- Benzodiazepines, chalcones, antibacterial, antifungal, anthelmintic and cytotoxic activity.

INTRODUCTION

1,5-benzodiazepine derivatives have received significant attention and the core is indeed a "privileged scaffold" found in compounds active against a variety of target types including peptide hormones (such as CCK), interleukin converting enzymes (ICE) and potassium blockers (I_k). More recently, the area of biological interest of 1, 5-benzodiazepines has been extended to various diseases such as cancer, viral infection (non-nucleoside inhibitors of HIV-1 reverse transcriptase), and cardiovascular disorders. In addition, 1, 5-benzodiazepines show antidepressive [1], analgesic [2], antianxiety [3], antifungal [4], antibacterial [5], anthelmintic [6-7], anti-inflammatory [8] and anticancer [9-11] activities. Besides these derivatives are also used as dyes for acrylic fibre in photography. Moreover, 1, 5-benzodiazepines are valuable synthons used for the preparation of other fused ring compounds such as triazolo, oxadiazolo, oxazino, or furano benzodiazepines. Due to their wide range of biological, industrial and synthetic applications, the development of mild and efficient protocols continues to be a challenging endeavour in synthetic organic chemistry.

Green synthesis is defined as environmentally benign chemical synthesis. Various approaches of green synthesis are reported in literature including the use of Green catalyst [12-13], PTC and Microwave method. All these approaches help in reducing the atmospheric pollution. In mid 1980's microwave irradiation was first applied towards synthetic chemistry. In 1986 first microwave assisted synthesis paper was published by Gedye *et al*. [14] The Microwave assisted organic synthesis (MAOS) is one of the non conventional techniques used now a day in the laboratory and is superior in many ways to traditional heating for the synthesis of novel compounds because the reactions are cost effective, increased yield and ecofriendly. Now, Microwave has become a new era in the field of synthetic chemistry.

We have made an attempt to synthesize 1, 5 benzodiazepine derivatives by using both microwave assisted as well as conventional synthetic method. All the newly synthesized compounds have been characterized by spectroscopic data, elemental analysis and screened for antibacterial, antifungal, anthelmintic and cytotoxic activity.

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MATERIALS AND METHODS

The melting points were determined in open capillary tubes and are uncorrected. The homogeneity of all the newly synthesized compounds were checked by TLC on silica gel-protected aluminum sheets (Type 60 F_{254}, Merck) and the spots were detected by exposure to UV-lamp at 254 nm for few seconds. The infrared (IR) spectra were recorded on 470-Shimadzu Infrared Spectrophotometer using KBr disc technique and expressed in cm^{-1}. ^{1}H NMR spectra were recorded on Bruker DRX-300 in DMSO-d_6 as a solvent. The

chemical shift was given in δ (ppm) downfield from tetramethylsilane (TMS) as an internal standard. Splitting patterns were designated as follows: s: singlet; d: doublet; t: triplet; q: quartet; m: multiplet. Elemental analysis was carried on Elemental Vario EL III Carlo Erba 1108 and the values were within ±0.4% of the theoretical values.

Scheme-1

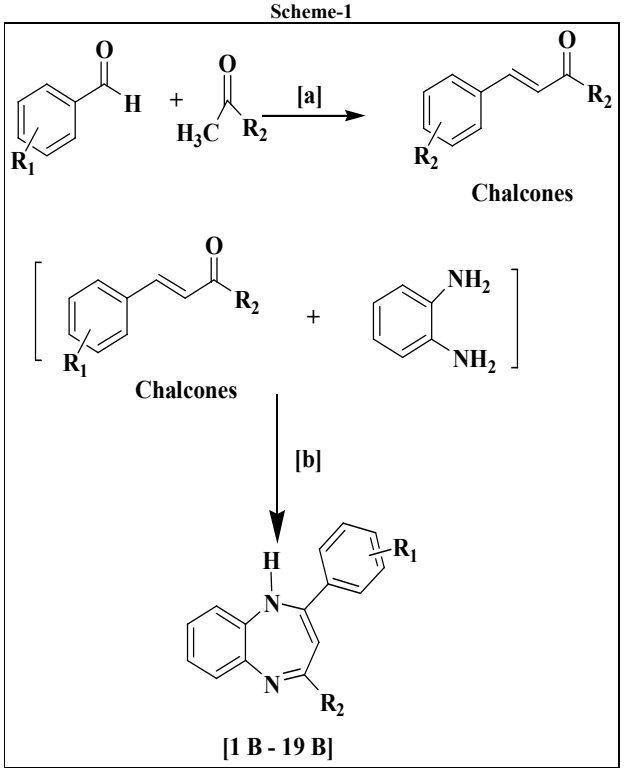


Fig. 1: Synthesis of 2, 4 disubstituted 1, 5- benzodiazepines (1B-19B) from substituted chalcones ([a] NaOH, Stirring 1-11 h. [b] Glacial acetic acid)

General procedure for the synthesis of Chalcones (1A-19A)

A solution of 2.2 g of sodium hydroxide in 20 ml of water and 20 ml of ethanol was placed in a beaker. Beaker was immersed in a bath of crushed ice, 0.01 mol of ketone derivatives was added in the beaker and 0.01 mol of benzaldehyde derivatives was added to it with constant stirring. The temperature of the mixture was kept at about 25ºC and the mixture was vigorously stirred for 1-11 hours. The stirrer was removed and reaction mixture was filtered and washed with cold water until the washing was neutral to litmus and recrystalized from ethanol.

General procedure for the preparation of 2, 4-Disubstituted 1, 5-benzodiazepines (1B-19B)

Conventional method

A mixture of 2 g chalcone (0.01mol), 1 g of *o*-phenylenediamine (0.01mol) and 5 ml glacial acetic acid in 15 ml DMF was taken in a round bottom flask and refluxed for 8-15 hours. The reaction mixture was allowed to attain room temperature and it was treated with cold water, the solid was separated with the help of filtration and washed with water and recrystalized from methanol.

Microwave method

A mixture of 2 g chalcone (0.01mol), 1 g of *o*-phenylenediamine (0.01mol) and 5 ml glacial acetic acid in 15 ml DMF was taken in a beaker and exposed to microwave irradiation at 750 Watt for 5-10 minutes. The reaction mixture was allowed to attain room temperature and it was treated with cold water, the solid was separated with the help of filtration and washed with water and recrystalized from methanol.

Spectral analysis

2-(3'-Nitrophenyl)-4-phenyl-1*H*-benzo[*b*][1,5]diazepine (1B)

Dark brown crystals. **UV (λ_{max}) (DMSO):** 253.0 nm; **FTIR (KBr):** 3360.82 (N-H, str.), 3050.62 (C-H, str.), 1591.16 (C=N, str.), 1563.23 (C=C, str.), 1272.64 (C-N, str.), 804.10 cm^{-1} (C-N, str. for NO_2); **^{1}H-NMR (DMSO-d_6, ppm) δ:** 3.36 (1H, NH, s,D_2O exchangeable), 7.25-8.61 (14H, m, Ar-H); **^{13}C-NMR δ:** 81.0, 116.6, 119.1, 119.2, 120.3, 124.1, 127.1, 127.8 (2C), 128.1 (2C), 128.2, 130.0, 130.5, 132.2, 134.2, 136.2, 140.6, 147.8, 140.0, 163.6; **MS (m/z):** 341.09.342 $[M+1]^+$, 296.12, 265.09, 220.09, 144.06; **Elemental analysis:** Calcd. for $C_{21}H_{15}N_3O_2$: C, 73.89; H, 4.43; N, 12.31. Found: C, 73.82; H, 4.40; N, 12.29%.

2-(4'-Chlorophenyl)-4-phenyl-1*H*-benzo[*b*][1,5]diazepine (2B).

Creamy Crystals; **UV (λ_{max}) (DMSO):** 309.0 nm; **FTIR (KBr)**: 3406.94 (N-H, str.), 3067.82 (C-H, str.), 1587.30 (C=N, str.), 1556.74 (C=C, str.), 1272.64 (C-N, str.), 682.75 cm^{-1} (C-Cl, str.). **^{1}H-NMR ($CDCl_3$-d_6, ppm) δ:** 3.82 (1H, s, NH, D_2O exchangeable), 7.22-8.04 ppm (14H, m, Ar- H); **^{13}C-NMR δ:** 81.3, 116.4, 119.0, 121.8, 122.0, 125.8, 126.4, 127.1 (3C), 127.6, 127.9, 128.1, 130.1, 131.2, 131.3, 131.8, 136.1, 140.2, 147.6, 163.2; **MS (m/z)**: 330.09, 331 $[M+1]^+$, 296.13, 254.08, 220.09, 144.06; **Elemental analysis:** Calcd. for $C_{21}H_{15}ClN_2$: C; 76.24, H; 4.53, Cl; 10.68, N; 8.47%. Found: C; 76.21, H; 4.48, Cl; 10.50, N; 8.41 %.

2-(3'-Methoxyphenyl)-4-phenyl-1*H*-benzo[*b*][1,5]diazepine (3B)

Yellow amorphous solid.; **UV (λ_{max}) (DMSO)**: 307.0 nm. **FTIR (KBr)**: 3403.41 (N-H, str.), 3060.12 (C-H, str.), 2925.81 (C-H, str. in CH_3), 1586.43 (C=N, str.), 1559.02 (C=C, str.), 1255.28 (C-N, str.), 1097.42 cm^{-1} (C-O-C, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ:** 3.31 (3H, s, OCH_3), 4.31 (1H, s, NH, D_2O exchangeable), 6.96-8.09 (14H, m, Ar- H); **^{13}C-NMR δ:** 53.8, 81.8, 109.5, 111.2, 116.1, 116.6, 119.0, 122.1, 127.3, 127.5, 127.8 (2C), 128.1, 128.2, 130.1, 131.2, 134.1, 137.2, 140.6, 148.3, 159.1, 163.4\; **MS (m/z):** 326.13, 327 $[M+1]^+$, 296.13, 250.10, 220.09, 144.06; **Elemental analysis:** Calcd. for $C_{22}H_{18}N_2O$: C; 80.96, H; 5.56, N; 8.58. Found: C; 80.91, H; 5.52, N; 8.56%.

2-(4'-Fluorophenyl)-4-phenyl-1*H*-benzo[*b*][1,5]diazepine (4B)

Brown crystals; **UV (λ_{max}) (DMSO):** 302.0 nm; **FTIR (KBr)**: 3389.23 (N-H, str.), 3060.82 (C-H, str.), 1568.67 (C=N, str.), 1550.29 (C=C, str.), 1330.99 (C-F, str.), 1272.64 cm^{-1} (C-N, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ:** 3.47 (1H, s, N-H, D_2O exchangeable), 7.14-8.08 (14H, m, Ar- H); **^{13}C-NMR δ:** 81.3, 113.5, 113.6, 116.3, 118.3, 121.6, 126.1, 127.1 (2C), 127.2 (2C), 127.3, 127.8 (2C), 130.1, 131.2, 137.1, 140.3, 147.6, 161.3, 162.4; **MS (m/z):** 314.11, 315 $[M+1]^+$, 296.13, 238.09, 220.09, 144.06.; **Elemental analysis:** Calcd. for $C_{21}H_{15}FN_2$: C; 80.24, H; 4.81, F; 6.04, N; 8.91%. Found: C; 80.24, H; 4.79, F; 6.01, N; 8.89 %.

4-(4'-Chlorophenyl)-2-(4''-nitrophenyl)-1*H*-benzo[*b*][1,5]diazepine (5B)

Brown crystals; **UV (λ_{max}) (DMSO):** 356.0 nm; **FTIR (KBr)**: 3377.12 (N-H, str.), 3058.89 (C-H, str.), 1596.51 (C=N, str.), 1582.95 (C=C, str.), 1272.64 (C-N, str.), 842.83

(C-N, str. for NO_2), 750.26 cm^{-1} (C-Cl, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ:** 4.17 (1H, s, N-H, D_2O exchangeable), 7.23-8.64 (13H, m, Ar- H).^{13}C-NMR δ: 81.1, 115.4, 119.1, 120.0, (2C), 121.1, 125.6, 126.1, 127.0, 127.2, 128.0, 129.5 (2C), 130.2, 134.3, 137.1, 139.1, 140.2, 145.3, 147.6, 163.1. MS (m/z): 375.06, 376 $[M+H]^+$, 341.11, 330.09, 296.12, 265.09, 254.06, 144.06; **Elemental analysis:** Calcd. for $C_{21}H_{14}ClN_3O_2$: C; 67.12, H; 3.75, Cl; 9.43, N; 11.18. Found: C; 67.10, H; 3.72, Cl; 9.41, N; 11.15%.

2-(2'-Chlorophenyl)-4-(4''-chlorophenyl)-1*H*-benzo[*b*][1,5]diazepine (6B)

Dark brown crystals; **UV (λ_{max}) (DMSO):** 406.0 nm; **FTIR (KBr)**: 3386.12 (N-H, str.), 3058.89 (C-H, str.), 1596.22 (C=N, str.), 1536.95 (C=C, str.), 1270.64 (C-N, str.), 771.47 cm^{-1} (C-Cl, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ:** 4.22 (1H, s, N-H, D_2O exchangeable), 7.25-8.61(13H, m, Ar- H); **^{13}C-NMR δ**: 80.8, 115.6, 119.2, 121.6, 125.4, 125.6, 126.4, 127.1 (3C), 127.3, 128.7, 129.1 (2C), 130.0, 134.1, 135.1, 137.1, 139.8, 147.7, 162.8; **MS (m/z):** 364.01,365 $[M+1]^+$, 330.09, 296.13, 220.10, 144.07, 265.09; **Elemental analysis:** Calcd. for $C_{21}H_{14}Cl_2N_2$: C; 69.05, H; 3.86, Cl; 19.41, N; 7.67%. Found: C; 69.03, H; 3.83, Cl; 19.39, N; 7.65%.

4-(4'-Aminophenyl)-2-(3'', 4'', 5''-trimethoxyphenyl)-1*H*benzo[*b*][1,5] diazepine (7B)

Dark brown crystals; **UV (λ_{max}) (DMSO)**: 220.0 nm; **FTIR (KBr)** 3404.63 (N-H, str.), 3059.82 (C-H, str.), 2933.53 (C-H, str. in CH_3), 1591.15 (C=N, str.), 1525.02 (C=C, str.), 1280.00 (C-N, str.), 1156.50 cm^{-1} (C-O-C, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ**: 3.30(9H, s, OCH_3), 6.21 (2H, s, NH_2, D_2O exchangeable), 4.41 (1H, s, N-H, D_2O exchangeable), 6.96-8.09 (11H, m, Ar- H); **^{13}C-NMR δ**: 54.8 (2C), 55.1, 80.6, 101.8 (2C), 115.1, 115.2, 115.3, 119.0, 121.2, 121.3, 126.3, 127.2, 128.0, 128.6, 136.3, 136.4, 140.1, 147.6, 148.3, 148.6(2C), 161.9; **MS (m/z)**: 401.16, 402 $[M+H]^+$, 386.44, 371.16, 341.41, 311.38, 310.35, 235.28, 220.10, 144.07; **Elemental analysis:** Calcd. for $C_{24}H_{23}N_3O_3$: C; 71.80, H; 5.77, N; 10.47. Found: C; 71.78, H; 5.75, N; 10.45%.

4-(4'-Chlorophenyl)-2-(3''-nitrophenyl)-1*H*-benzo[*b*][1,5]diazepine(8B)

Brown crystals; **UV (λ_{max}) (DMSO)**: 304.0 nm; **FTIR (KBr)**: 3430.42 (N-H, str.), 3080.17 (C-H, str.), 1551.96 (C=N, str.), 1538.21 (C=C, str.), 1263.86 (C-N, str.), 853.43 (C-N, str. for NO_2), 672.42 cm^{-1} (C-Cl, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ**: 3.47 (1H, s, N-H, D_2O exchangeable), 7.41-8.08 (13H, m, Ar- H); **^{13}C-NMR δ**: 81.2, 115.5, 118.6, 120.4, 121.2, 122.8, 125.3, 126.1, 126.8, 127.2 (2C), 127.6, 130.8, 131.8, 132.5, 134.2, 137.1, 139.8, 146.5, 148.2, 163.1; **MS (m/z):** 375.74, 376 $[M+1]^+$, 296.13, 220.10, 144.07, 265.09; **Elemental analysis:** Calcd. for $C_{21}H_{14}ClN_3O_2$: C; 67.12, H; 3.75, Cl; 9.43, N; 11.18. Found: C; 67.10, H; 3.71, Cl; 9.41, N; 11.15%.

4-(4'-Chlorophenyl)-2-(3'', 4'', 5''-trimethoxyphenyl)-1*H*-benzo[*b*][1,5]diazepine (9B)

Yellow amorphous solid; **UV (λ_{max}) (DMSO):** 364.0 nm **FTIR (KBr)**: 3395.41 (N-H, str.), 3095.12 (C-H, str.), 2984.42 (C-H, str. in CH_3), 1585.42 (C=N, str.), 1548.32 (C=C, str.), 1295.53 (C-N, str.), 1049.98 (C-O-C, str.), 756.08 cm^{-1} (C- Cl, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ**: 3.40 (9H, s, OCH_3), 4.17 (1H, s, N-H, D_2O exchangeable), 7.23-8.64 (11H, s, Ar-H); **^{13}C-NMR δ**: 55.3(2C), 55.8, 80.8, 104.8 (2C), 115.4, 118.6, 121.2, 126.2(2C), 126.6, 126.8, 127.2 (2C), 131.2, 132.2, 137.0, 139.8, 140.2, 147.4, 148.4(2C), 162.8; **MS (m/z):** 420.15, 421 $[M+1]^+$, 386.44, 390.11, 360.10, 330.09, 310.35, 254.06, 144.07; **Elemental analysis**: Calcd. for $C_{24}H_{21}ClN_2O_3$: C; 68.49, H; 5.03, Cl; 8.42, N; 6.66. Found: C; 68.43, H; 5.01, Cl; 8.40, N; 6.63%.

2-(3',4',5'-Trimethoxyphenyl)-4-phenyl-1*H*-benzo[*b*][1,5]diazepine (10B)

Brown crystals; **UV (λ_{max}) (DMSO)**: 304.0 nm; **FTIR (KBr)**: 3393.43 (N-H, str.), 3072.22 (C-H, str.), 2982.11(C-H, str. in CH_3), 1592.33 (C=N, str.), 1540.32 (C=C, str.), 1295.53 (C-N, str.), 1046.67 cm^{-1} (C-O-C, str. for OCH_3); **^{1}H-NMR (DMSO-d_6, ppm) δ**: 3.10 (9H, s, OCH_3), 3.96 (1H, s, N-H, D_2O exchangeable), 7.25-8.61 (12H, m, Ar- H); **^{13}C-NMR δ:** 55.2 (2C), 55.3, 81.0, 104.8 (2C), 116.2, 118.8, 121.7, 125.1, 125.2, 126.2 (2C), 126.8, 127.1, 127.2, 133.2, 136.8, 139.7, 140.8, 148.2, 149.8 (2C), 163.2; **MS (m/z):** 386.15, 387 $[M+1]^+$, 356.15, 326.14, 310.13, 296.13, 220.10, 144.07; **Elemental analysis:** Calcd. for $C_{24}H_{22}N_2O_3$: C; 74.59, H; 5.74, N; 7.25% Found: C; 74.55, H; 5.71, N; 7.23%.

2-(2'-Chlorophenyl)-4-phenyl-1*H*-benzo[*b*][1,5]diazepine (11B)

Light brown crystals; **UV (λ_{max}) (DMSO):** 254.0 nm; **FTIR (KBr)**: 3379.23 (N-H, str.), 3074.55 (C-H, str.), 1599.88 (C=N, str.), 1531.21 (C=C, str.), 1332.33 (C-N, str.), 753.87 cm^{-1} (C- Cl, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ**: 3.82(1H, s, N-H, D_2O exchangeable), 7.22 – 8.08 (14H, m, Ar-H); **^{13}C-NMR δ:** 81.2, 113.1, 116.2, 118.8, 121.8, 125.2 (2C), 125.8, 126.8, 127.0, 127.2 (2C), 128.6, 130.2, 131.2, 131.2, 133.2 (2C), 137.2, 140.2, 147.8; **MS (m/z):** 330.09, 331 $[M+1]^+$, 296.13, 254.08, 220.09, 144.06; **Elemental analysis:** Calcd. for $C_{21}H_{15}ClN_2$: C; 76.24, H; 4.57, Cl; 10.72, N; 8.47%. Found: C; 76.22, H; 4.53, Cl; 10.69, N; 8.45%.

4-(4'-Methoxyphenyl)-2-(3'', 4'', 5''-trimethoxyphenyl)-1*H*-benzo[*b*][1,5] diazepine (12B)

Brown crystals; **UV (λ_{max}) (DMSO):** 329.0 nm; **FTIR (KBr)**: 3392.24 (N-H, str.), 3071.21 (C-H, str.), 2986.32 (C-H, str. in CH_3), 1582.32 (C=N, str.), 1547.32 (C=C, str.), 1292.09 (C-N, str.), 1039.88 cm^{-1} (C-O-C, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ:** 3.30 (12H, s, OCH_3), 4.31 (1H, s, N-H, D_2O exchangeable), 6.96- 8.06 (11H, m, Ar- H); **^{13}C-NMR δ:** 54.2 (2C), 55.6 (2C), 81.2, 105.2 (2C), 112.6 (2C), 116.2, 118.2, 121.2, 124.6, 125.4, 125.5, 125.8, 127.1, 136.8, 140.2 (2C), 147.8, 148.8 (2C), 157.9, 163.2; **MS (m/z):** 416.38, 417 $[M+1]^+$, 386.16, 356.15, 326.14, 310.13, 250.11, 220.10, 144.07; **Elemental analysis**: Calcd. for $C_{25}H_{24}N_2O_4$: C; 72.10, H; 5.81, N; 6.73. Found: C; 72.08, H; 5.78, N; 6.70%.

4-(4'-Aminophenyl)-2-(2''-hydroxyphenyl)-1*H*-benzo[*b*][1,5]diazepine (13B)

Black crystals; **UV (λ_{max}) (DMSO):** 308.0 nm; **FTIR (KBr):** 3595.40 (O-H, str.), 3486.12 (N-H, str.), 3070.63 (C-H, str.), 1535.42 (C=N, str.), 1523.01 (C=C, str.), 1269.28 cm^{-1} (C-N, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ:** 6.14 (2H, s, NH_2, D_2O exchangeable), 3.47 (1H, s, N-H, D_2O exchangeable), 7.14-8.08(13H, m, Ar-H), 10.4 (1H, s, OH, D_2O exchangeable); **^{13}C-NMR δ:** 80.6, 107.8, 114.2(2C), 114.6, 116.6, 118.1, 120.2, 121.2, 122.1, 126.4, 128.2 (2C), 137.2, 140.1, 147.6, 147.8, 157.2, 162.8; **MS (m/z):** 328.38, 329 $[M+1]^+$, 311.13, 296.13, 220.10, 144.07; **Elemental analysis:** Calcd. for $C_{21}H_{18}N_3O$: C; 77.04, H; 5.23, N; 12.84. Found: C; 77.01, H; 5.20, N; 12.81%.

4-(3'-Nitrophenyl)-2-(4''-nitrophenyl)-1*H*-benzo[*b*][1,5]diazepine (14B).

Black crystals; **UV (λ_{max}) (DMSO):** 303.0 nm; **FTIR (KBr)**: 3390.42 (N-H, str.), 3082.08 (C-H, str.), 1547.26 (C=N, str.),

1531.42 (C=C, str.), 1346.22 (C-N, str.), 854.00 cm^{-1} (C-N, str. for NO_2); **^{1}H-NMR (DMSO-d_6, ppm) δ**: 4.17 (1H, s, N-H, D_2O exchangeable), 7.23-8.64 (12H, m, Ar- H); **^{13}C-NMR δ**: 81.2, 115.8, 118.8, 119.6, 120.0, 121.4, 121.8, 123.1, 126.1, 126.2, 127.0, 127.9, 132.8, 133.8, 137.2, 138.8, 140.2, 146.2, 146.5, 147.8, 163.2; **MS (m/z):** 386.33,387 $[M+1]^+$, 341.11, 296.13, 220.10, 144.07; **Elemental analysis**: Calcd. for $C_{21}H_{14}N_4O_4$: C; 65.28, H; 3.65, N; 14.50. Found: C; 65.26, H; 3.62, N; 14.47%.

4-(4'-Methoxyphenyl)-2-(4''-nitrophenyl)-1*H*-benzo[*b*][1,5]diazepine (15B)

Dark Brown crystals; **UV (λ_{max}) (DMSO):** 253.0 nm; **FTIR (KBr)**: 3405.41 (N-H, str.), 3095.12 (C-H, str.), 2984.42 (C-H, str. in CH_3), 1586.42 (C=N, str.), 1548.32 (C=C, str.), 1295.53 (C-N, str.), 1049.98 (C-O-C, str.), 855.43 cm^{-1} (C-N, str. for NO_2); **^{1}H-NMR (DMSO-d_6, ppm) δ**: 3.30(3H, s, OCH_3), 4.06 (1H, s, N-H, D_2O exchangeable), 7.25-8.61 (13H, m, Ar- H);**^{13}C-NMR δ:** 54.9, 80.9, 113.4 (2C), 115.8, 118.8, 120.0 (2C), 121.2, 124.1, 126.1 (2C), 126.8, 128.2 (2C), 137.1, 138.8, 140.2, 146.1, 147.8, 161.8, 163.2; **MS (m/z)**: 371.37. 372 $[M+1]^+$, 326.14, 296.13, 220.10, 144.07; **Elemental analysis**: Calcd. for $C_{22}H_{17}N_3O_3$: C; 71.15, H; 4.61, N; 11.31. Found: C; 71.13, H; 4.59, N; 11.28%.

2-(2'-Hydroxyphenyl)-4-(4''-methoxyphenyl)-1*H*benzo[*b*][1,5]diazepine (16B)

Light black amorphous solid; **UV (λ_{max}) (DMSO)**: 307.0 nm; **FTIR (KBr)**: 3586.42 (O-H, str.), 3402.24 (N-H, str.), 3071.21 (C-H, str.), 2986.32 (C-H, str. in CH_3), 1587.32 (C=N, str.), 1543.32 (C=C, str.), 1292.09 (C-N, str.), 1039.88 cm^{-1} (C-O-C, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ**: 3.10 (3H, s, OCH_3), 3.82 (1H, m, N-H, D_2O exchangeable), 7.22-8.04(13H, m, Ar- H), 9.92 (1H, s, OH, D_2O exchangeable); **^{13}C-NMR δ:** 55.4, 81.8, 107.2, 112.8 (2C), 114.2, 116.1, 118.2, 120.1, 121.2, 124.5, 126.1, 127.1, 128.2 (3C), 137.1, 139.6, 147.8, 157.1, 162.1, 163.2; **MS (m/z)**: 342.38,343.0 $[M+1]^+$, 326.14, 296.13, 220.10, 144.07; **Elemental analysis:** Calcd. for $C_{22}H_{18}N_2O_2$: C; 77.17, H; 5.30, N; 8.18. Found: C; 77.15, H; 5.28, N; 8.16%.

2-(2'-Chlorophenyl)-4-(3''-nitrophenyl)-1*H*-benzo[*b*][1,5] diazepine (17B)

Light brown crystals; **UV (λ_{max}) (DMSO):** 229.0 nm; **FTIR (KBr)**: 3346.22 (N-H, str.), 3012.12 (C-H, str.), 1534.12 (C=N, str.), 1517.11 (C=C, str.), 1343.11 (C-N, str.), 848.12 (C-N, str. for NO_2), 632.98 cm^{-1} (C-Cl, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ**: 4.31(1H, s, N-H, D_2O exchangeable), 6.96-8.09(13H, s, Ar- H); **^{13}C-NMR δ:** 81.2, 116.1, 118.8, 121.8 (2C), 123.0, 125.6, 126.5, 126.8, 127.8, 128.2, 130.1, 133.0, 134.2 (2C), 137.1 (2C), 140.2, 147.1, 147.6, 163.2; **MS (m/z):** 375.06, 376 $[M+1]^+$, 341.11, 330.09, 296.12, 265.09, 254.06, 144.06; **Elemental analysis:** Calcd. for $C_{21}H_{14}ClN_3O_2$: C; 67.12, H; 3.75, Cl; 9.43, N; 11.18. Found: C; 67.10, H; 3.71, Cl; 9.40, N; 11.15%.

4-(2',5'-Dihydroxyphenyl)-2-(4''-nitrophenyl)-1*H*-benzo[*b*][1,5]diazepine(18B).

Dark yellow amorphous solid; **UV (λ_{max}) (DMSO):** 302.0 nm; **FTIR (KBr)**: 3587.41 (O-H, str.), 3395.41 (N-H, str.), 3095.12 (C-H, str.), 1598.32 (C=N, str.), 1548.42 (C=C, str.), 1295.53 (C-N, str.), 855.43 cm^{-1} (C-N, str. for NO_2); **^{1}H-NMR (DMSO-d_6, ppm) δ**: 3.47 (1H, s, N-H, D_2O exchangeable), 7.14-8.08 (12H, m, Ar-H), 10.40 (2H, s, OH, D_2O exchangeable); **^{13}C-NMR δ:** 81.4, 114.2, 115.8, 116.2, 117.8, 118.2, 119.1, 119.5 (2C), 121.2, 126.2 (2C), 127.1, 137.1, 139.2, 140.2, 146.1, 147.8, 150.1, 152.6, 163.4; **MS (m/z)**: 373.35, 374 $[M+1]^+$, 328.12, 312.13, 296.13, 220.10, 144.07; **Elemental analysis**: Calcd. for $C_{21}H_{15}N_3O_4$: C; 67.56, H; 4.05, N; 11.25. Found: C; 67.56, H; 4.01, N; 11.22%.

2-(2'-Chlorophenyl)-4-(4''-methoxyphenyl)-1*H*-benzo[*b*][1,5]diazepine (19B)

Light yellow crystals; **UV (λ_{max}) (DMSO):** 256.0 nm; **FTIR (KBr)**: 3385.41 (N-H, str.), 3095.12 (C-H, str.), 2984.42 (C-H, str. in CH_3), 1588.42 (C=N, str.), 1536.32 (C=C, str.), 1295.53 (C-N, str.), 1049.93 (C-O-C, str.), 756.08 cm^{-1} (C-Cl, str.); **^{1}H-NMR (DMSO-d_6, ppm) δ**: 3.10 (3H, s, OCH_3), 4.17 (1H, s, N-H, D_2O exchangeable), 7.23-8.64 (13H, m, Ar- H); **^{13}C-NMR δ:** 54.2, 80.9, 113.1 (2C), 118.6, 122.1, 124.1, 125.4, 126.2, 126.9, 127.6, 128.2 (2C), 129.1, 130.1, 134.1, 137.2, 140.2, 148.2, 162.1, 162.8; **MS (m/z)**: 360.12, 361 $[M+1]^+$, 326.14, 296.13, 220.10, 144.07; **Elemental analysis**: Calcd. for $C_{22}H_{17}ClN_2O$: C; 73.23, H; 4.75, Cl; 9.83, N; 7.76. Found: C; 73.19, H; 4.72, Cl; 9.80, N; 7.74%.

Anthelmintic activtiy

Anthelmintic studies were carried out against *Megascoplex konkanensis* (ICARBC 211) and *Eudrilus sp.* (ICARBC 042) of earthworms by Garg and Atal method [15] at 2 mg/ml concentrations. Suspension of samples were prepared by triturating synthesized compounds (100 mg) with Tween 80 (0.5%) and distilled water and the resulting mixtures were stirred using a mechanical stirrer for 30 minutes. Suspension of reference drug mebendazole was prepared with same concentration in similar way. Three sets of five earthworms of almost similar sizes (3 inch in length) were placed in petri plates of 4 inch diameter containing 50mL of suspension of test sample and reference drug at room temperature. Another set of five earthworms was kept as control in 50 ml suspension of distilled water and Tween 80 (0.5%). The paralyzing and death times were noted and their mean was calculated for triplicate sets. The death time was ascertained by placing the earthworms in warm water (50ºC), which stimulate the movement, if the worm was alive.

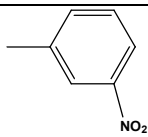
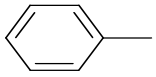
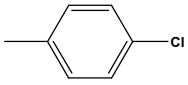
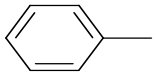
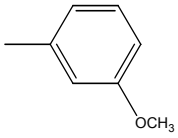
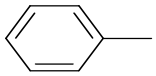
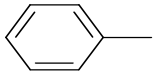
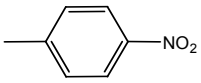
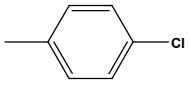
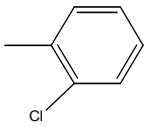
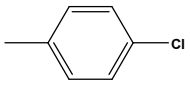
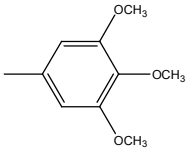
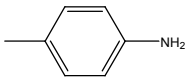
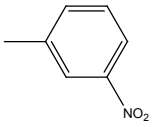
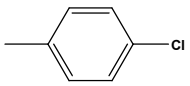
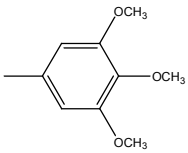
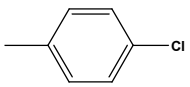
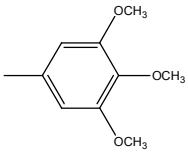
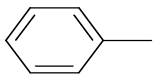
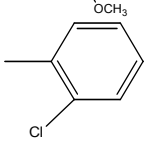
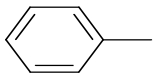
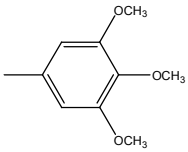
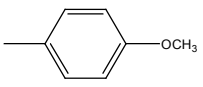
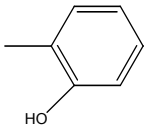
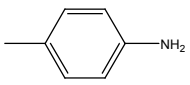
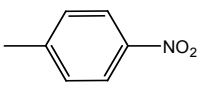
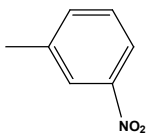
Anticancer activity

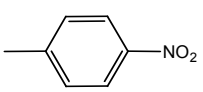
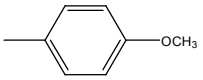
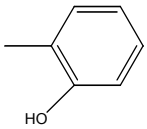
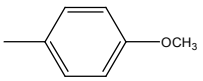
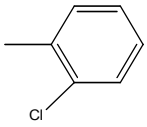
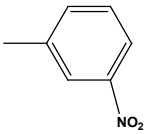
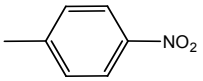
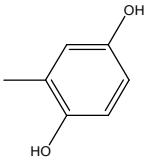
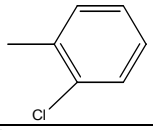
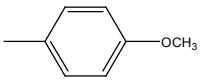
Anticancer activities of the synthesized compounds were assessed by determining the percentage inhibition of DLA, EAC and HEP-2 cell line cells by tryphan blue dye exclusion technique according to the standard procedure. We checked anticancer activity of all the synthesized compounds at the concentration of 500, 250, 125, 62.5, 31.25 µg/ml. The percentage growth inhibition was calculated by using the following formula: % Growth inhibition = [(Total cells – Live cells) × 100]/Total cells. The CTC_{50} values were calculated by plotting the graph between concentration *versus* percentage growth inhibition and by bisecting concentration at the 50% growth inhibition. The synthesized 2, 4-disubstituted 1, 5 benzodiazepines and their CTC_{50} values are as shown in **Table-3.** 5-Fluorouracil was used as standard drug.

Antibacterial activity

The synthesized benzodiazepine derivatives were screened for their antibacterial activity against two gram positive bacterial strains *Staphylococcus aureus* (ATCC 6538)**,** *Bacillus subtilis* (NCIM 2063) and two gram negative bacterial strains *Escherichia coli* (ATCC 35210)**,** *Pseudomonas aeruginosa* (ATCC 27853) by using modified Kirby-Bauer disc diffusion method.

Table 1: Comparative data of the synthesized compounds

Comp. no.	R_1	R_2	Reaction Time		Yield (%)[a]		M.P. (°C)	R_f value
			MW[b] (min)	Conventional (h)	MW	Conventional		
1B			5.01	14.00	78.00	35.00	129-130	0.59
2B			5.05	13.40	64.48	45.00	116-117	0.65
3B			6.00	13.00	68.00	32.35	60-61	0.55
4B			6.01	8.00	78.00	24.11	78-79	0.62
5B			5.08	13.30	77.45	37.54	53-54	0.63
6B			6.00	15.00	69.58	31.00	59-60	0.55
7B			5.05	10.00	81.01	31.00	98-99	0.60
8B			5.05	10.00	75.24	28.00	138-139	0.61
9B			6.00	15.00	61.90	35.71	73-74	0.58
10B			5.00	11.10	80.69	35.11	63-64	0.59
11B			5.05	14.40	59.47	21.23	77-78	0.54
12B			6.00	10.30	73.47	23.47	88-89	0.60
13B			9.00	8.00	91.75	41.26	58-59	0.63
14B			8.00	11.00	48.67	26.21	69-70	0.56

15B			7.00	10.00	71.37	34.50	94-95	0.58
16B			5.05	9.00	73.86	32.44	48-49	0.54
17B			8.00	9.00	78.07	31.11	67-68	0.52
18B			10.00	11.00	69.08	26.11	56-57	0.64
19B			8.00	10.00	70.53	32.95	51-52	0.60

[a] Isolated yield.
[b] Microwave irradiation (LG^{TM} domestic microwave oven).

Table 2: Anthelmintic data of the synthesized compounds against two Earthworm species

Compound	Earthworm species			
	M. konkanensis		*Eudrilus sp.*	
	Mean paralysing time (min)[a]	Mean death time (min)[a]	Mean paralyzing time (min)[a]	Mean death time (min)[a]
1B	13.68 ± 0.52	21.58 ± 0.59	14.78 ± 0.48	25.70 ± 1.00
2B	11.54 ± 0.83	20.18 ± 1.28	12.58 ± 0.50	22.50 ± 0.50
3B	08.44 ± 0.56	16.49 ± 0.72	13.61 ± 0.62	21.56 ± 0.51
4B	14.10 ± 0.23	22.13 ± 0.92	14.70 ± 0.50	26.16 ± 0.76
5B	11.22 ± 0.37	20.56 ± 0.72	13.60 ± 1.00	23.16 ± 0.76
6B	24.66 ± 1.46	33.22 ± 0.94	19.00 ± 1.00	32.66 ± 1.52
7B	24.22 ± 0.67	33.54 ± 0.90	16.00 ± 1.00	25.16 ± 0.76
8B	13.78 ± 0.34	22.12 ± 0.87	14.83 ± 0.76	24.16 ± 0.76
9B	23.44 ± 0.56	32.49 ± 0.72	19.50 ± 0.50	32.50 ± 0.50
10B	34.78 ± 0.21	60.48 ± 0.91	17.05 ± 0.55	30.50 ± 1.00
11B	14.67 ± 0.17	22.48 ± 1.95	14.60 ± 0.50	24.16 ± 0.76
12B	20.22 ± 0.32	26.32 ± 0.34	22.40 ± 1.00	32.70 ± 1.00
13B	10.99 ± 0.84	18.12 ± 0.31	13.80 ± 0.28	24.00 ± 1.00
14B	21.12 ± 0.13	38.56 ± 0.81	19.33 ± 1.52	33.66 ± 2.08
15B	10.56 ± 0.19	17.16 ± 0.29	13.70 ± 2.50	23.66 ± 1.52
16B	33.12 ± 0.72	38.15 ± 0.12	15.50 ± 1.32	27.00 ± 1.00
17B	22.34 ± 0.65	26.75 ± 0.67	18.00 ± 1.00	29.50 ± 0.50
18B	14.34 ± 1.12	23.45 ± 2.23	14.60 ± 1.00	26.00 ± 1.00
19B	11.12 ± 0.23	19.83 ± 1.23	13.16 ± 0.76	24.16 ± 0.76
Control	-	-	-	-
Mebendazole	13.22 ± 0.54	21.12 ± 0.76	14.54 ±1.00	25.05 ± 1.00

[a] Data are given as mean ± S.D (n=3)

MIC values of test compounds were determined by tube dilution technique. All the synthesized compounds were dissolved separately to prepare a stock solution of 1 mg ml^{-1} using DMF. Stock solution was aseptically transferred and suitably diluted with sterile broth medium to have seven different concentrations of each test compound ranging from 200 to 3.1 µg ml^{-1} in different test tubes. All the tubes were inoculated with one loopful of one of the test bacteria. The process was repeated with different test bacteria and different samples. Tubes inoculated with bacterial cultures were incubated at 37°C for 18 h and the presence/absence of growth of the bacteria was observed. From these results, MIC of each test compound was determined against each test bacterium. A spore suspension in sterile distilled water was prepared from five-days-old culture of the test bacteria growing on nutrient broth media. About 20 ml of the growth medium was transferred into sterilized petri plates and inoculated with 1.5 ml of the spore suspension (spore concentration 6×10^4 spores ml^{-1}). Filter paper disks of 6 mm diameter and 2 mm thickness were sterilized by autoclaving at 121°C (15 psi) for 15 min. Each petri plate was divided into five equal portions along the diameter to place one disc. Three discs of test sample were placed on three portions together with one disc with reference drug ciprofloxacin and a disk impregnated with the solvent (DMF) as negative control. Test sample and reference drugs were tested at the concentration of 10 µg ml^{-1}. The petri plates inoculated with bacterial cultures were incubated at 37°C for 18 h. Diameters of the zones of inhibition (mm) were measured and the average diameters for test sample were calculated in triplicate sets. The diameters obtained for the test sample were

compared with that produced by the standard drug ciprofloxacin.

Antifungal activity

The newly synthesized compounds were tested *in-vitro* for their antifungal activity using disc diffusion method. The following fungal strains were used: *Candida albicans (*ATCC 10261) and *Aspergillus niger* (ATCC 9643). In the disc-diffusion method, disc impregnated with compounds dissolved in DMF at concentration 25, 50 and 100 µg ml^{-1} were used. Disc impregnated with DMF were used as solvent control for antifungal activity because of free solubility of test compounds. The microorganism culture was spread over nutrient agar media in petri dishes, and then the disc impregnated with the solution was placed on the surface of the media inoculated with the fungal strain. The plates were incubated at 25ºC for 48 hours for fungal strains. After incubation, the growth inhibiting zones around the disc were observed. Growth inhibiting zone indicate that the compounds inhibit growth of microorganism. Each experiment is done in triplicate. Griseofulvin at concentration 50 µgml^{-1} was used as standard drug for antifungal activity. Results were interpreted in terms of diameter (mm) of zone of inhibition.

RESULTS AND DISCUSSION

The synthesis of 1, 5-benzodiazepines was accomplished as presented in **Scheme-1**. Substituted benzaldehydes on reaction with substituted ketones in the presence of ethanolic sodium hydroxide yielded corresponding chalcones **(1A-19A)**. The latter were converted to desired 2, 4-disubstituted 1, 5-benzodiazepine **(1B-19B)** by the treatment with o-phenylenediamine in glacial acetic acid. The synthesis was ascertained from spectral, physicochemical and elemental analysis.

Table 3: Anticancer data of the synthesized compounds (1B-19B)

Compound	[a]CTC_{50} (µg/ml) Cell lines		
	DLA	EAC	HEp-2
1B	33.21	78.01	29.00
2B	27.04	48.06	25.00
3B	107.04	142.00	114.00
4B	21.12	35.15	18.00
5B	>200	>200	>325
6B	105.06	130.19	119.00
7B	149.34	164.89	103.00
8B	75.29	103.00	99.00
9B	106.11	159.00	199.32
10B	122.47	184.05	122.47
11B	137.21	196.04	176.00
12B	108.00	167.02	197.00
13B	23.50	55.50	24.50
14B	135.00	175.15	210.00
15B	109.63	191.22	219.63
16B	145.00	168.28	145.00
17B	121.20	180.34	160.34
18B	150.02	170.21	150.00
19B	25.13	63.05	27.25
Control	-	-	-
5-Fluorouracil	37.36	90.55	31.76

[a] The cytotoxic concentration (which inhibited the growth of 50% of total cells)

Table 4: Data of antibacterial data of the synthesized compounds

Compound	Diameter of zone of inhibition (mm) Bacterial strains			
	Gram (+ve)		Gram (-ve)	
	S. aureus	*B. subtilis*	*E. coli*	*P. aeruginosa*
1B	14.43 (25)	14.33 (25)	14.93 (25)	14.73(25)
2B	12.73 (100)	13.76 (25)	13.76 (25)	13.33 (50)
3B	13.36 (50)	14.13 (25)	10.90 (100)	12.13 (100)
4B	12.26 (100)	13.96 (25)	14.63 (25)	13.36 (50)
5B	14.33 (25)	14.20 (25)	10.56 (100)	14.26 (25)
6B	14.13 (25)	12.20 (100)	14.20 (25)	13.60 (100)
7B	12.06 (100)	12.46 (50)	14.43 (25)	13.50 (50)
8B	13.23 (100)	10.20 (100)	14.76 (25)	12.46 (100)
9B	12.16 (100)	14.06 (25)	13.56 (50)	13.20 (50)
10B	13.16 (50)	12.56 (100)	14.36 (25)	14.33 (25)
11B	12.53 (100)	12.20 (100)	13.46 (50)	13.13 (50)
12B	13.40 (50)	14.33 (25)	13.56 (50)	12.93 (100)
13B	13.20 (50)	11.36 (100)	12.86 (50)	13.56 (50)
14B	14.33 (25)	13.63 (50)	13.03 (50)	12.13 (100)
15B	12.96 (100)	14.26 (25)	13.46 (100)	14.23 (25)
16B	13.16 (50)	13.83 (25)	14.49 (25)	13.96 (25)
17B	12.26 (100)	12.26 (100)	14.16 (25)	12.10 (100)
18B	13.26 (50)	13.00 (50)	13.23 (50)	13.30 (50)
19B	12.10 (100)	12.86 (100)	12.93 (100)	14.43 (25)
Control	-	-	-	-
Ciprofloxacin	13.46 (25)	13.45 (6)	13.50 ±(12.5)	13.70 (25)

Values in bracket are MIC value (µg ml^{-1})

The novel benzodiazepines were synthesized successfully by conventional and microwave assisted methods. The newly synthesized compounds were identified on the basis of R_f value, melting point range, solubility studies, FTIR, ^{1}H-NMR, ^{13}C-NMR, MASS Spectral data and elemental analysis. The ^{1}H-NMR spectrum showed the presence of N-H Protons at δ: 3.00-5.00 ppm and aromatic protons at 6.00-8.00 ppm. FTIR spectrum showed the presence of characteristic peak of N-H at 3200-3450 cm^{-1} and C=N peak at 1500-1600 cm^{-1}. The comparative data of the synthesized compounds are provided in **Table 1**. The reaction time for the synthesis of benzodiazepine derivatives by conventional method was 8-15 h in comparison with the microwave heating (5-10 min), which reduced the time duration many fold. Approximately 96 to 98% reaction time was reduced and the yield was increased by 30 to 50%. Thus, this methodology becomes an efficient strategy for the rapid synthesis of 2, 4 disubstituted 1, 5 benzodiazepines.

Pharmacological Activity

All the newly synthesized compounds **(1B-19B)** were screened for anthelmintic activity against *Megascoplex konkanensis* (ICARBC 211) and *Eudrilus sp.* (ICARBC 042) at 2 mg ml^{-1} concentration and all the 2, 4 Disubstituted 1, 5 benzodiazepines showed moderate to good activity. The

synthesized compounds were effective by killing and paralyzing the worms. The compounds produced by variation at the 2- and 4- position were highly active for anthelmintic activity. Comparison of anthelmintic data (**Table 2**) revealed that derivative **2B, 3B, 5B, 13B, 15B** and **19B** were found to be most active against *Megascoplex konkanensis* (ICARBC 211) and *Eudrilus sp.* (ICARBC 042) in comparison to standard drug Mebendazole. Compound **1B, 4B, 8B, 11B** and **18B** showed comparable anthelmintic activity. It was observed that introduction of OCH_3 group in the phenyl ring attached to 1, 5 benzodiazepine as shown in compound **3B** & **15B** makes the compound more active then mebendazole. Compound **13B** having hydroxyl group which is responsible for inhibition of respiration & blocking glucose absorption by the intestinal adult worms. Compound **5B** having chloro group (Electron withdrawing group) which is electronegative in nature makes the compound lipophilic and responsible for good anthelmintic activity.

Table 5:Data of antifungal activity of the synthesized compounds

Compound	Diameter of zone of inhibition (mm) Fungal strains	
	C. albicans	*A. niger*
1B	16.53 (25)	16.54 (25)
2B	16.23 (50)	18.12 (100)
3B	13.13 (100)	14.12 (100)
4B	15.04 (100)	16.60 (100)
5B	17.07 (25)	16.12 (100)
6B	16.14 (50)	14.15 (100)
7B	13.43 (100)	15.02 (100)
8B	16.43 (25)	17.63 (25)
9B	15.54 (100)	14.10 (100)
10B	17.21 (25)	18.12 (25)
11B	16.65 (25)	17.08 (50)
12B	14.71 (100)	15.32 (100)
13B	14.06 (100)	16.56 (50)
14B	14.35 (100)	17.42 (25)
15B	15.18 (100)	15.94 (100)
16B	17.46 (25)	15.06 (100)
17B	15.67 (100)	16.27 (100)
18B	14.19 (100)	15.49 (100)
19B	15.64 (100)	16.97 (50)
Control	-	-
Griseofulvin	16.43 (12.5)	17.23 (25)

Values in bracket are MIC value (μg ml^{-1}).

The synthesized compounds were tested for the antiproliferation activity against three cell lines using DLA, EAC and HEP-2 cell lines by SRB assay listed in **Table 3**. Compounds **1B, 2B, 4B, 13B** and **19B** were found to be active against all the three cell lines. Compound **4B** which contains a fluoro substituent at para position was found to be significantly active against all the three cell lines.

The newly synthesized compounds were tested *in vitro* for their antibacterial activity (**Table 4**). The antibacterial activities of all the synthesized compounds were conducted against the pathogenic bacterias: *Staphylococcus aureus* (ATCC 6538)**,** *Bacillus subtilis (*NCIM 2063) (**gram positive**) and *Escherichia coli* (ATCC 35210)**,** *Pseudomonas aeruginosa* (ATCC 27853) (**gram negative**). The zone of inhibition was measured by antibiotic zone reader. The results revealed that the newly synthesized compound **1B, 2B, 4B, 6B, , 7B, 8B, 10B, 16B** and **17B** showed significant antibacterial activity against *Escherichia coli* (ATCC 35210), compound **1B, 2B, 3B, 4B, 5B, 9B, 12B, 15B** and **16B** showed good antibacterial activity against *Bacillus subtilis* (NCIM 2063), compound **1B, 5B, 10B, 15B, 16B** and **19B** showed significant antibacterial activity against *Pseudomonas aeruginosa* (ATCC 27853) and compound **1B, 5B, 6B** and **14B** showed good antibacterial activity against *Staphylococcus aureus* (ATCC 6538) when given at a concentration of 50 $\mu g ml^{-1}$ and ciprofloxacin was used as a standard drug.

The newly synthesized compounds were tested *in vitro* for their antifungal activity (**Table-5**). The fungal strains used were: *Candida albicans (*ATCC 10261) *and Aspergillus niger* (ATCC 9643). The results revealed that the newly synthesized compound **1B, 5B, 8B, 10B, 11B** and **16B** showed significant antifungal activity against *Candida albicans (*ATCC 10261). Whereas compound **2B, 8B, 10B** and **14B** also showed good antifungal activity against *Aspergillus niger* (ATCC 9643) when given at a concentration of 50μg mL^{-1} and griseofulvin was used as a standard drug.

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