Asian Journal of Chemistry; Vol. 31, No. 4 (2019), 965-970

ASIAN JOURNAL OF CHEMISTRY

https://doi.org/10.14233/ajchem.2019.21831

Synthesis and Preliminary Evaluation of Benzofuran-Oxadiazole Conjugates as Potential Antitubercular Agents

VEERABHADRAYYA S. NEGALURMATH[1], OBELANNAVAR KOTRESH[1,*] and MAHANTESHA BASANAGOUDA[2,]

[1]Department of Chemistry, Karnatak Science College, Dharwad-580001, India
[2]P.G. Department of Chemistry, P.C. Jabin Science College, Hubballi-580031, India

*Corresponding author: E-mail: kotresh.obelannavar@gmail.com

Received: 23 November 2018; *Accepted*: 28 January 2019; *Published online*: 27 February 2019; AJC-19313

In the present study, a series of benzofuran-oxadiazole conjugates **7(a-o)** was designed, synthesized and characterized through IR, ^{1}H NMR, ^{13}C NMR and mass spectral data. All the compounds were screened for preliminary antitubercular activity against *Mycobacterium phlei* and *Mycobacterium tuberculosis* H_{37}RV. Among all the target compounds, the compound possessing chlorine (**7k**, MIC 1.56 µg/mL) and bromine (**7m**, MIC 1.56 µg/mL) on 6th position of benzofuran showed highest activity against *Mycobacterium phlei*. Whereas, bromine on either 5th position (**7l**, MIC 3.125 µg/mL) or 6th position (**7m** MIC 3.125 µg/mL) on benzofuran exhibited highest activity for *Mycobacterium tuberculosis* (H_{37} RV).

Keywords: Antituberculosis, Benzofuran, Oxadiazole, SAR.

INTRODUCTION

Tuberculosis (TB) is an old disease, which is caused by *Mycobacterium tuberculosis* (Mtb). Worldwide, tuberculosis is one of the top 10 causes of death and the leading cause from a single infectious agent (above HIV/AIDS). Millions of people continue to fall sick with tuberculosis each year [1]. During the last decade there has been an increased interest for research on tuberculosis by international and national organizations, pharmaceutical companies. The innovative focus on tuberculosis has partly been prompted by the persistent larger number of tuberculosis case studies in developing countries and partly by the increased occurrence of multidrug and extensively drug resistant tuberculosis (MDR- and XDR-TB) [2,3]. According to WHO 2018 report, in 2017, tuberculosis caused an estimated 1.3 million deaths among HIV-negative people and there were an additional 300 000 deaths from tuberculosis among HIV-positive people. Globally, the best estimate is that 10 million people developed tuberculosis disease in 2017: 5.8 million men, 3.2 million women and 1.0 million children [1]. Hence, the need for search the new and efficient antituberculosis agents with a new mechanism of action remains a crucial task [4,5].

Benzofuran and its derivatives are important basis for drug discovery and possessing broad spectrum of biological and pharmaceutical activities [6-11]. The 3-substituted benzofurans have attracted the medicinal chemists due to their exciting biological properties such as antifungal and antitubercular agents [12], antiviral and antitumor [13], cytotoxicity [14], hepatitis C virus inhibitors [15], inhibitors of mycobacterium protein tyrosine phosphatase [16], dual 5-HT_{1A} receptor and serotonin transporter affinity [17], bone morphogenetic protein-2 up-regulators [18], glycogen synthase kinase 3β inhibitors [19], calcium activated chloride channel inhibitors [20], inhibition of Aβ neurotoxicity, cholinesterase activity and β-amyloid aggregation [21], ischemic cell death inhibitors [22], orally bioavailable GPR40 agonist [23]. The potent antitubercular properties exhibited by some compounds with benzofuran [24-30] and 3-substituted benzofuran moieties [31-36] are reported in the literature.

Recently, there are many reports pertaining to biological activity of 1,3,4-oxadiazole as antitubercular [37], anticonvulsant [38], antiallergic [39], antiepileptic [40], cytotoxic and antimicrobial [41] and anticancer [42] agents.

The continued attempt of our group on designing oxygen based heterocycles as biologically effective molecules [43-

45], here we are reporting the design and synthesis of benzofuran-oxadiazole conjugates **7(a-o)** with the aim to study structure activity relationships and thereby provide novel compounds as potential antitubercular agents against *Mycobacterium phlei* and *Mycobacterium tuberculosis* $H_{37}RV$.

EXPERIMENTAL

The melting points were determined by open capillary method and are uncorrected. The IR spectra (KBr disc) were recorded on a Thermo Fisher Nicolet-6700 FT-IR spectrophotometer. ^1H NMR and ^{13}C NMR spectra were recorded on 500 MHz Bruker spectrometer using dimethylsulfoxide (DMSO-d_6) as solvent and tetramethylsilane (TMS) as an internal standard. The chemical shifts were expressed in δ ppm and coupling constant (*J*) values were given in Hertz. The mass spectra were recorded using Shimadzu GCMS-QP2010S instrument. The elemental analysis was carried out using Heraus CHN rapid analyzer. Progress of the reaction was monitored by TLC using aluminium sheets precoated with UV fluorescent silica gel Merck 60 F254 and were visualized by using UV lamp. All the chemicals of analytical grade were purchased from Sigma-Aldrich Chemicals (India) and S.D. Fine Chemicals (India) and were used without further purification unless otherwise stated.

General procedure for synthesis of benzofuran-oxadiazole conjugates 7(a-o): The required benzofuran-3-yl-acetic acid hydrazides **6(a-o)** were prepared according our earlier report [45]. A mixture of carbohydrazide **6(a-o)** (10 mmol) and triethyl orthoformate (2 mL) in toluene (60 mL) was heated under reflux for 15 h. The excess solvent was evaporated and the reaction mass was cooled to room temperature. The resultant solid **7(a-o)** was filtered and recrystallized from ethanol.

2-((5-Methylbenzofuran-3-yl)methyl)-1,3,4-oxadiazole (7a): Colourless solid; m.p. 147-148 °C; (74 %); IR (KBr, ν_{max}, cm^{-1}): 1606 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 2.41 (*s*, 3H, 5-CH_3), 3.76 (*s*, 2H, C3-CH_2), 7.14-7.16 (*dd*, *J* = 8.5 Hz, 1.0 Hz, 1H, C6-H), 7.41 (*s*, 2H, C4-H and oxadiazole-H), 7.45 (*d*, *J* = 8.5 Hz, C7-H), 7.80 (*s,* 1H, C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): δ 20.12, 21.68, 111.82, 113.84, 120.18, 124.42, 125.56, 134.74, 143.28, 155.42, 170.90; GCMS *m/z*: 214 [M+]; Anal. calcd. for $C_{12}H_{10}N_2O_2$; C, 67.28; H, 4.71; ; N, 13.08; Found: C, 67.26; H, 4.71; N, 13.07.

2-((6-Methylbenzofuran-3-yl)methyl)-1,3,4-oxadiazole (7b): Colourless solid; m.p. 173-174 °C; yield (76 %); IR (KBr, ν_{max}, cm^{-1}): 1606 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 2.40 (*s*, 3H, 6-CH_3), 3.73 (*s*, 2H, C3-CH_2), 7.06-7.08 (*d*, *J* = 8.0 Hz, 1H, C5-H), 7.35 (*s*, 2H, C7-H and oxadiazole-H), 7.43-7.45 (*d*, *J* = 8.0 Hz, 1H, C4-H), 7.79 (*s,* 1H, C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): δ 19.29, 21.63, 111.78, 113.78, 120.03, 124.39, 125.54, 134.66, 143.25, 155.39, 170.87; GCMS *m/z*: 213 [M^+-1]; Anal. calcd. for $C_{12}H_{10}N_2O_2$; C, 67.28; H, 4.71; N, 13.08; Found: C, 67.27; H, 4.71; N, 13.07.

2-((4,6-Dimethylbenzofuran-3-yl)methyl)-1,3,4-oxadiazole (7c): Colourless; m.p. 167-168 °C, yield (72 %); IR (KBr, ν_{max}, cm^{-1}) 1619 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 2.35 (*s*, 3H, CH_3), 2.46 (*s*, 3H, CH_3), 3.75 (*s*, 2H, C3-CH_2), 6.80 (*s,* 1H, C5-H), 7.19 (*s,* 1H, C7-H), 7.28 (s, 1H, oxadiazole-H), 7.82 (*s,* 1H, C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): 19.08, 21.48, 21.94, 110.94, 113.65, 120.14, 124.44, 126.02, 134.98, 143.56, 155.99, 164.72, 171.10; GCMS *m/z*: 228 [M+]; Anal. calcd. for $C_{13}H_{12}N_2O_2$; C, 68.41; H, 5.30; N, 12.27; Found: C, 68.40; H, 5.30; N, 12.26.

2-((6,7-Dimethylbenzofuran-3-yl)methyl)-1,3,4-oxadiazole (7d): Colourless solid; m.p. 154-155 °C; yield (70 %); IR (KBr, ν_{max}, cm^{-1}): 1620 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 2.34 (*s*, 3H, CH_3), 2.45 (*s*, 3H, CH_3), 3.78 (*s*, 2H, C3-CH_2), 7.08-7.10 (*d*, *J* = 8.0 Hz, 1H, C5-H), 7.24-7.26 (*d*, *J* = 8.0 Hz, 1H, C4-H), 7.45 (*s*, 1H, oxadiazole-H), 7.78 (*s*, 1H, C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): δ 18.65, 19.94, 23.24, 110.63, 113.21, 123.10, 125.27, 125.64, 130.60, 142.11, 155.67, 166.92, 171.06; GCMS *m/z*: 228 [M+]; Anal. calcd. for $C_{13}H_{12}N_2O_2$; C, 68.41; H, 5.30; N, 12.27; Found: C, 68.39; H, 5.30; N, 12.25.

2-((5-*iso*-Propylbenzofuran-3-yl)methyl)-1,3,4-oxadiazole (7e): Colourless solid; m.p. 142-143 °C; yield (72 %); IR (KBr, ν_{max}, cm^{-1}): 1630 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 1.27 (*d*, 6H, isopropyl-CH_3, *J* = 6 Hz), 2.95-3.07 (*m*, 1H, isopropyl-CH), 3.76 (*s*, 2H, C3-CH_2), 7.11 (*dd*, *J* = 8.5 Hz, 1.5 Hz, 1H, C6-H), 7.40-7.45 (*m*, 2H, C4-H and C7-H), 7.60 (*s,* 1H, oxadiazole-H), 7.75 (*s,* 1H, C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): δ 23.10, 29.52, 34.06, 111.16, 114.58, 122.26, 123.45, 129.34, 135.18, 145.12, 156.54, 167.63, 171.20; GCMS *m/z*: 242 [M+]; Anal. calcd. for $C_{14}H_{14}N_2O_2$; C, 69.38; H, 5.82; N, 11.54; Found: C, 69.36; H, 5.82; N, 11.53.

2-((5-*tert*-Butylbenzofuran-3-yl)methyl)-1,3,4-oxadiazole (7f): Colourless solid; m.p. 163-164 °C; yield (75 %); IR (KBr, ν_{max}, cm^{-1}): 1620 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 1.38 (*s*, 9H, CH_3), 3.84 (*s*, 2H, C3-CH_2), 7.10 (*dd*, *J* = 8.5 Hz, 1.5 Hz, 1H, C6-H), 7.44-7.50 (*m*, 2H, C4-H, C7-H and oxadiazole-H), 7.81 (*s,* 1H, C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): 29.40, 31.56, 44.56, 114.15, 117.32, 121.45, 126.94, 128.68, 131.68, 144.36, 157.14, 167.38. 171.33; GCMS *m/z*: 256 [M+]; Anal. calcd. for $C_{15}H_{16}N_2O_2$; C, 70.29; H, 6.29; N, 10.93; Found: C, 70.27; H, 6.29; N, 10.92.

3-((1,3,4-Oxadiazol-2-yl)methyl)benzofuran-6-ol (7g): Colourless solid; m.p. 150-151 °C; yield (73 %); IR (KBr, ν_{max}, cm^{-1}): 1632 (C=N), 3256 (Broad, OH); ^1H NMR (500 MHz, DMSO-d_6): δ 3.86 (*s*, 2H, C3-CH_2), 6.76 (*dd*, *J* = 8.4 Hz, 2.0 Hz, 1H, C5-H), 6.86-6.88 (*d*, *J* = 2.0 Hz, 1H, C7-H), 7.36 (*d*, *J* = 8.4 Hz, 1H, C4-H), 7.43 (*m*, 2H, oxadiazole-H and C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): δ 29.50, 99.74, 114.30, 116.66, 120.24, 123.92, 144.84, 158.30, 158.64, 167.80, 172.18; GCMS *m/z:* 216 [M+]; Anal. calcd. for $C_{11}H_8N_2O_3$; C, 61.11; H, 3.73; N, 12.96; Found: C, 61.09; H, 3.73; N, 12.95.

2-((5-Methoxybenzofuran-3-yl)methyl)-1,3,4-oxadiazole (7h): Beige solid; m.p. 146-147 °C; yield (82 %); IR (KBr, ν_{max}, cm^{-1}): 1618 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 3.58 (*s*, 3H, 5-OCH_3), 3.89 (*s*, 2H, C3-CH_2), 6.85-6.91 (*dd*, *J* = 8.0 Hz, 2.0 Hz, 1H, C6-H), 7.25 (*d*, *J* = 2.0 Hz, 1H, C4-H), 7.41 (*d*, *J* = 8.0 Hz, 1H, C7-H), 7.56 (*s,* 1H, oxadiazole-H), 7.83 (*s,* 1H, C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): δ 29.84, 57.12, 106.18, 115.10, 115.63, 115.79, 128.12, 144.58, 151.35, 156.94, 168.87, 172.33; GCMS *m/z*: 230 [M+]; Anal. calcd.

for $C_{12}H_{10}N_2O_3$; C, 62.60; H, 4.38; N, 12.17; Found: C, 62.54; H, 4.38; N, 12.16.

2-((6-Methoxybenzofuran-3-yl)methyl)-1,3,4-oxadiazole (7i): Grey solid; m.p. 171-172 °C; yield (80 %); IR (KBr, ν_{max}, cm^{-1}): 1627 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 3.59 (*s*, 3H, 6-OCH_3), 3.89 (*s*, 2H, C3-CH_2), 6.86 (*dd*, *J* = 9.0 Hz, 2.0 Hz, 1H, C5-H), 7.14 (*d*, *J* = 2.0 Hz, 1H, C7-H), 7.44 (*d*, *J* = 9.0 Hz, 1H, C4-H), 7.61 (*s*, 1H, oxadiazole-H), 7.78 (*s*, 1H, C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): δ 21.70, 56.14, 104.55, 105.34, 112.98, 114.14, 125.38, 144.17, 144.54, 169.05, 172.06; GCMS *m/z*: 230 [M+]; Anal. calcd. for $C_{12}H_{10}N_2O_3$; C, 62.60; H, 4.38; N, 12.17; Found: C, 62.58; H, 4.38; N, 12.16.

2-((5-Chlorobenzofuran-3-yl)methyl)-1,3,4-oxadiazole (7j): Colourless solid; m.p. 149-150 °C; yield (69 %); IR (KBr, ν_{max}, cm^{-1}): 1639 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 3.92 (*s*, 2H, C3-CH_2), 7.36 (*d*, *J* = 9.0 Hz, 1H, C6-H), 7.59 (*d*, *J* = 9.0 Hz, 1H, C7-H), 7.64 (*s*, 2H, C4-H and oxadiazole-H), 7.90 (*s*, 1H, C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): 29.71, 114.18, 115.92, 121.44, 125.56, 128.01, 131.35, 145.98, 155.33, 168.12, 171.32; GCMS *m/z*: 234, 236 [M+, M+2]; Anal. calcd. for $C_{11}H_7N_2O_2Cl$; C, 56.31; H, 3.01; N, 11.94; Found: C, 56.30; H, 3.01; N, 11.93.

2-((6-Chlorobenzofuran-3-yl)methyl)-1,3,4-oxadiazole (7k): Colourless solid; m.p. 158-159 °C; yield (67 %); IR (KBr, ν_{max}, cm^{-1}): 1631 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 3.90 (*s*, 2H, C3-CH_2), 7.09-7.11 (*d*, *J* = 8.0 Hz, 1H, C5-H), 7.42 (*s*, 1H, C7-H), 7.44-7.46 (*d*, *J* = 8.0 Hz, 1H, C4-H), 7.51 (*s*, 1H, oxadiazole-H), 7.76 (*s*, 1H, C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): δ 29.98, 115.33, 116.54, 121.98, 125.74, 128.45, 132.44, 147.58, 156.10, 166.41, 172.43; GCMS *m/z*: 234, 236 [M+, M+2]; Anal. calcd. for $C_{11}H_7N_2O_2Cl$; C, 56.31; H, 3.01; N, 11.94; Found: C, 56.30; H, 3.01; N, 11.93.

2-((5-Bromobenzofuran-3-yl)methyl)-1,3,4-oxadiazole (7l): Colourless solid; m.p. 168-169 °C; yield (66 %); IR (KBr, ν_{max}, cm^{-1}): 1635 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 3.91 (*s*, 2H, C3-CH_2), 7.41-7.44 (*dd*, *J* = 9.0 Hz, 2.0 Hz, 1H, C6-H), 7.53 (*d*, *J* = 9.0 Hz, 1H, C7-H), 7.86 (*d*, *J* = 2.0 Hz, 1H, C4-H), 7.88-7.90 (*m,* 2H, oxadiazole-H and C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): δ 29.52, 114.04, 115.98, 116.07, 124.53, 128.33, 132.84, 146.78, 155.62, 164.47, 172.14; GCMS *m/z*: 277, 279 [M+, M+2]; Anal. calcd. for $C_{11}H_7N_2O_2Br$; C, 47.34; H, 2.53; N, 10.04; Found: C, 47.33; H, 2.53; N, 10.04.

2-((6-Bromobenzofuran-3-yl)methyl)-1,3,4-oxadiazole (7m): Colourless solid; m.p. 134-135 °C; yield (60 %); IR (KBr, ν_{max}, cm^{-1}): 1629 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 3.91 (*s*, 2H, C3-CH_2), 7.08-7.10 (*d*, *J* = 8.0 Hz, 1H, C5-H), 7.39 (*s*, 1H, C7-H), 7.42-7.44 (*d*, *J* = 8.0 Hz, 1H, C4-H), 7.60 (*s*, 1H, oxadiazole-H), 7.75 (*s*, 1H, C2-H); ^{13}C NMR (125 MHz, DMSO-d_6): 29.98, 115.11, 116.47, 121.52, 127.45, 129.82, 135.84, 148.44, 156.87, 169.33, 172.70; GCMS *m/z*: 277, 279 [M+, M+2]; Anal. calcd. for $C_{11}H_7N_2O_2Br$; C, 47.34; H, 2.53; N, 10.04; Found: C, 47.32; H, 2.53; N, 10.04.

2-(Naphtho[2,1-b]furan-1-ylmethyl)-1,3,4-oxadiazole (7n): Beige solid; m.p. 201-202 °C; yield (72 %); IR (KBr, ν_{max}, cm^{-1}): 1618 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 3.81 (*s*, 2H, C3-CH_2), 7.46-7.49 (*t*, *J* = 7.5 Hz, 1H, Ar-H), 7.52-7.55 (*t*, *J* = 7.5 Hz, 1H, Ar-H), 7.78-7.80 (*d*, *J* = 9.0 Hz, 1H, Ar-H), 7.86-7.88 (*d*, *J* = 9.0 Hz, 1H, Ar-H), 8.05-8.10 (*m*, 2H, oxadiazole-H and Ar-H), 8.10 (*s*, 1H, C2-H), 8.14-8.15 (*d*, *J* = 8.5 Hz, 1H, Ar-H); ^{13}C NMR (125 MHz, DMSO-d_6): δ 22.56, 114.12, 114.64, 121.28, 123.87, 126.44, 126.82, 127.60, 127.98, 129.87, 131.74, 145.33, 154.82, 164.74, 172.10; GCMS *m/z*: 250 [M+]; Anal. calcd. for $C_{15}H_{10}N_2O_2$; C, 71.99; H, 4.03; N, 11.19; Found: C, 71.97; H, 4.03; N, 11.18.

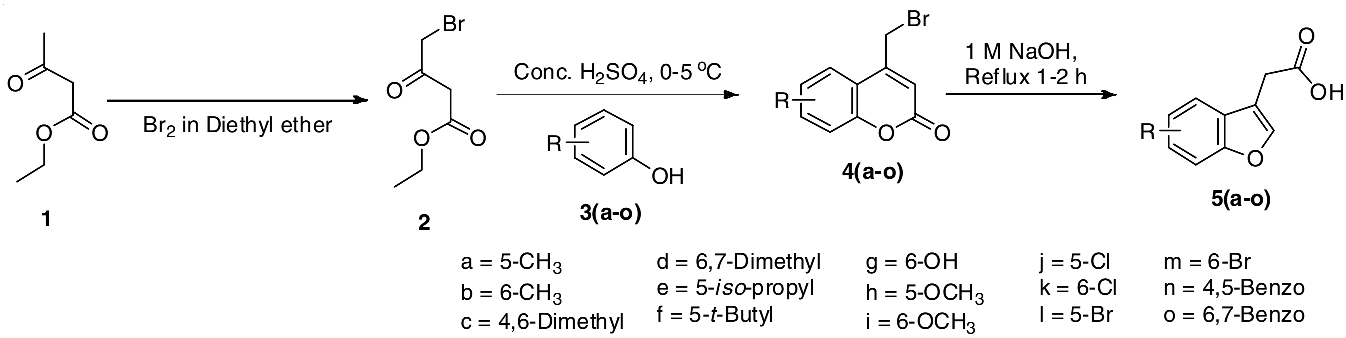
2-(Naphtho[1,2-b]furan-3-ylmethyl)-1,3,4-oxadiazole (7o): Brown solid; m.p. 161-162 °C; yield (70 %); IR (KBr, ν_{max}, cm^{-1}): 1652 (C=N); ^1H NMR (500 MHz, DMSO-d_6): δ 3.86 (*s*, 2H, C3-CH_2), 7.46-7.49 (*m*, 1H, Ar-H), 7.61-7.63 (*m*, 1H, Ar-H), 7.65-7.67 (*d*, *J* = 8.4 Hz, 1H, Ar-H), 7.70-7.72 (*d*, *J* = 8.8 Hz, 1H, Ar-H), 8.02 (*m*, 3H, oxadiazole and Ar-H), 8.18 (*d*, *J* = 8.0 Hz, 1H, Ar-H); ^{13}C NMR (125 MHz, DMSO-d_6): δ 29.68, 116.68, 119.82, 119.99, 121.82, 124.56, 124.18, 126.63, 128.10, 128.64, 133.18, 145.68, 149.98, 169.15, 172.44; GCMS *m/z*: 250 [M+]; Anal. calcd. for $C_{15}H_{10}N_2O_2$; C, 71.99; H, 4.03; N, 11.19; Found: C, 71.98; H, 4.03; N, 11.19.

Antitubercular activity: The antitubercular activity of titled compounds **7(a-o)** were performed against *M. phlei* and *M. tuberculosis* $H_{37}RV$ using well known procedure microplate alamar blue assay (MABA) [46]. This methodology is non-toxic, uses stable reagents and shows good correlation with proportional and BACTEC radiometric method. In brief, 200 µL of sterile deionized water was added to all outer perimeter wells of sterile 96-well plate to minimize evaporation of medium in the test wells during incubation. The 96-well plate received 100 µL of the Middlebrook 7H9 broth and serial dilution of compounds was made directly on plate. The final drug concentrations tested were 100-0.1 µg/mL. Plates were covered and sealed with parafilm and incubated at 37 °C for 5 days. After this time, 25 µL of freshly prepared 1:1 mixture of Alamar Blue reagent and 10 % Tween 80 were added to the plate and incubated for 24 h. A blue colour in the well was interpreted as no bacterial growth and pink colour was scored as growth. The MIC was defined as the lowest drug concentration that prevented the colour change from blue to pink.

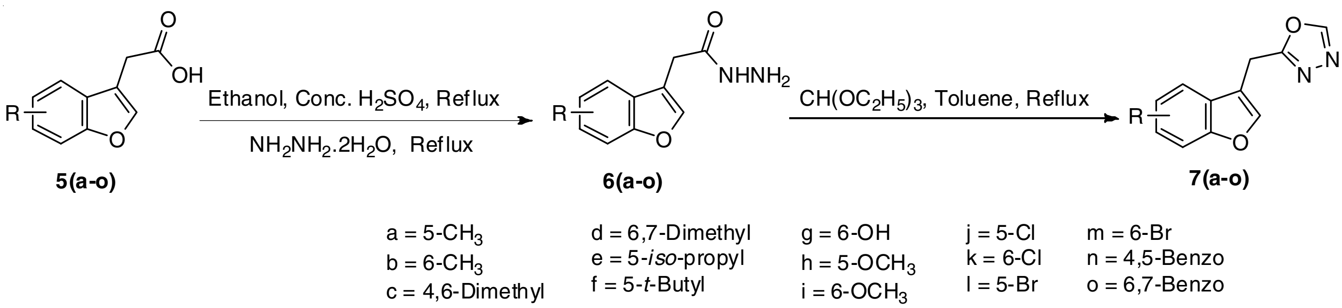
RESULTS AND DISCUSSION

The concise synthetic route used to synthesize the intermediates and titled compounds **7(a-o)** are outlined in **Schemes I** and **II**. Benzofuran-3-yl-acetic acids **5(a-o)** were converted into corresponding ethyl esters by refluxing with absolute ethanol in presence of conc. sulphuric acid. The resulting mixture, was converted into the acid hydrazides **6(a-o)** by the reaction with hydrazine hydrate at reflux temperature [45]. Further, these acid hydrazides **6(a-o)** were reacted with triethyl orthoformate in toluene at reflux temperature for 15 h afforded title compounds **7(a-o)**. All the synthesized compounds **7(a-o)** were characterized by elemental analysis, IR, ^1H NMR, ^{13}C NMR and mass spectral data.

The IR spectrum of an intermediate (6-methyl-benzofuran-3-yl)-acetic acid hydrazide (**6b**), showed a strong peak at 1643 cm^{-1} for carbonyl group whereas 3303 cm^{-1} for $NHNH_2$ stretching. The ^1H NMR spectrum exhibited δ 2.41 (*s*, 3H, 6-CH_3), 3.63 (*d, J* = 1.0 Hz, 2H, C3-CH_2), 4.22 (*s,* br, 2H, NH_2,



Scheme-I: Synthesis of benzofuran acetic acids **5(a-o)**



Scheme-II: Synthesis of benzofuran-oxadiazole conjugates **7(a-o)**

D_2O exchangeable), 7.06 (*d*, *J* = 8.0 Hz, 1H, C5-H), 7.33 (*s*, 1H, C7-H), 7.43 (*d*, *J* = 8.0 Hz, 1H, C4-H), 7.73 (*s*, 1H, C2-H), 9.26 (*s*, br, 1H, NH, D_2O exchangeable); which was further confirmed by ^{13}C NMR spectrum agrees with the number of carbons and by its mass spectrum that showed the molecular ion peak *m/z* 204 (M+), confirms the molecular weight of the compound [45].

The IR spectrum of representative compound in the series 2-((6-methylbenzofuran-3-yl)methyl)-1,3,4-oxadiazole (**7b**), showed absence of carbonyl group and $NHNH_2$ stretching frequencies. Further, new band appeared at 1606 (C=N) indicate the cyclization. The ^1H NMR spectrum exhibited δ 2.40 (*s*, 3H, 6-CH_3), 3.73 (*s*, 2H, C3-CH_2), 7.06-7.08 (*d*, *J* = 8.0 Hz, 1H, C5-H), 7.35 (*s*, 2H, C7-H and oxadiazole-H), 7.43-7.45 (*d*, *J* = 8.0 Hz, 1H, C4-H), 7.79 (*s*, 1H, C2-H); Here, the disappearance of NH_2 protons at 4.22 (*s*, br, 2H, NH_2, D_2O exchangeable) in the precursor compound indicates the cyclization to the compound **7b**. Futher, all the carbons are resonated in the expected regions. The formation of the compound **7b** is confirmed by its mass spectrum which showed molecular ion peak at 213.

Antitubercular evaluation: The *in vitro* antitubercular activity against *Mycobacterium phlei* and *Mycobacterium tuberculosis* $H_{37}RV$ was carried out by using standard procedure, microplate alamar blue assay (MABA) [46]. The MIC values of all the title compounds **7(a-o)** along with standard drugs pyrazinamide and streptomycin for the comparison are summarized in Table-1. The MIC ranges in between 1.56 and > 100 μg/mL.

TABLE-1
RESULTS OF ANTITUBERCULAR ACTIVITY OF COMPOUNDS **7(a-o)** MICs (μg/mL)

Compound	R	*Mycobacterium phlei*	*Mycobacterium tuberculosis* $H_{37}RV$
7a	5-CH_3	> 100	> 100
7b	6-CH_3	> 100	> 100
7c	4,6-di-CH_3	> 100	> 100
7d	6,7-di-CH_3	> 100	> 100
7e	5-*iso*-Propyl	> 100	50
7f	5-*t*-Butyl	> 100	50
7g	6-OH	12.5	12.5
7h	5-OCH_3	12.5	25
7i	6-OCH_3	12.5	25
7j	5-Cl	3.125	6.25
7k	6-Cl	1.56	6.25
7l	5-Br	3.125	3.125
7m	6-Br	1.56	3.125
7n	4,5-Benzo	> 100	25
7o	6,7-Benzo	> 100	25
Pyrazinamide	–	3.125	3.125
Streptomycin	–	6.25	6.25

***Mycobacterium phlei*:** In the series of compounds **7(a-o)**, the chlorine or bromine on 6th position of benzofuran exhibited equipotent activity (**7k**, **7m**: MIC 1.56 μg/mL), which were more potent than standard drugs pyrazinamide (MIC 3.125 μg/mL) and streptomycin (MIC 6.25 μg/mL). Whereas, decrease in activity were observed by change in the position of chlorine or bromine from 6th to 5th position *i.e.*, **7j**, **7l**: MIC 3.125 μg/mL. Further, decrease in activity were observed on varying the substituent by hydroxy (**7g**), methoxy (**7h**, **7i**) groups to MIC 12.5 μg/mL. The least activity were observed for alkyl (**7a-7f**), benzo (**7n**, **7o**) derivatives with MIC > 100 μg/mL.

***Mycobacterium tuberculosis* $H_{37}RV$:** In the series of compounds **7(a-o)**, the bromine either 5th position or 6th position on benzofuran exhibited equipotent activity (**7l**, **7m**: MIC 3.125 μg/mL), were equal to standard drug pyrazinamide (MIC 3.125 μg/mL) and more potent to streptomycin (MIC 6.25 μg/mL). Whereas, decrease in activity were observed by changing the bromine by chlorine *i.e.*, **7j**, **7k**: MIC 6.25 μg/mL. The hydroxy derivative **7g** exhibited with MIC 12.5 μg/mL, was more active than methoxy (**7h**, **7i**) and benzo (**7n**, **7o**) derivatives with MIC 25 μg/mL. Branched alkyl derivatives (**7e**, **7f**) were exhibited MIC 50 μg/mL, whereas mono-methyl (**7a**,**7b**) and dimethyl (**7c**,**7d**) derivatives were least active in the series with MIC > 100 μg/mL.

Conclusion

In conclusion, a series of novel benzofuran-oxadiazole conjugates **7(a-o)** prepared were proved to be potential anti-tubercular agents against *Mycobacterium phlei* and *Mycobacterium tuberculosis* $H_{37}RV$. The compounds bearing chlorine (**7j**, **7k**) and bromine (**7l**, **7m**) substituent of the benzofuran ring were found to be the most potent in the series. Among them, compound **7m** bearing bromine on 6^{th} position on benzofuran was found to be most potent against both *Mycobacterium phlei* and *Mycobacterium tuberculosis* $H_{37}RV$. Further biological investigation is under progress in our laboratory and will be reported in due course.

ACKNOWLEDGEMENTS

The authors thank to the SAIF, Indian Institute of Technology Madras, Chennai, India for the ^1H NMR and ^{13}C NMR spectra; and USIC Karnatak University, Dharwad, India for IR, GCMS spectral data.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

REFERENCES

1. WHO Global Tuberculosis Report (2018).
2. V. Bhowruth, L.G. Dover and G.S. Besra, *Prog. Med. Chem.*, **45**, 169 (2007); https://doi.org/10.1016/S0079-6468(06)45504-1.
3. H. Lang, G. Quaglio and O. Olesen, *Tuberculosis*, **90**, 1 (2010); https://doi.org/10.1016/j.tube.2009.10.002.
4. L.G. Dover and G.D. Coxon, *J. Med. Chem.*, **54**, 6157 (2011); https://doi.org/10.1021/jm200305q.
5. Beena and D.S. Rawat, *Med. Res. Rev.*, **33**, 693 (2013); https://doi.org/10.1002/med.21262.
6. K.M. Dawood, *Expert Opin. Ther. Pat.*, **23**, 1133 (2013); https://doi.org/10.1517/13543776.2013.801455.
7. R. Naik, D.S. Harmalkar, X. Xu, K. Jang and K. Lee, *Eur. J. Med. Chem.*, **90**, 379 (2015); https://doi.org/10.1016/j.ejmech.2014.11.047.
8. A. Radadiya and A. Shah, *Eur. J. Med. Chem.*, **97**, 356 (2015); https://doi.org/10.1016/j.ejmech.2015.01.021.
9. H. Khanam and Shamsuzzaman, *Eur. J. Med. Chem.*, **97**, 483 (2015); https://doi.org/10.1016/j.ejmech.2014.11.039.
10. R.J. Nevagi, S.N. Dighe and S.N. Dighe, *Eur. J. Med. Chem.*, **97**, 561 (2015); https://doi.org/10.1016/j.ejmech.2014.10.085.
11. A. Hiremathad, M.R. Patil, C. K. R, K. Chand, M.A. Santos and R.S. Keri, *RSC Adv.*, **5**, 96809 (2015); https://doi.org/10.1039/C5RA20658H.
12. V.N. Telvekar, A. Belubbi, V.K. Bairwa and K. Satardekar, *Bioorg. Med. Chem. Lett.*, **22**, 2343 (2012); https://doi.org/10.1016/j.bmcl.2012.01.067.
13. S.A. Galal, A.S. Abd El-All, M.M. Abdallah and H.I. El-Diwani, *Bioorg. Med. Chem. Lett.*, **19**, 2420 (2009); https://doi.org/10.1016/j.bmcl.2009.03.069.
14. L. Bigler, C. Spirli, R. Fiorotto, A. Pettenazzo, E. Duner, A. Baritussio, F. Follath and H.R. Ha, *Eur. J. Med. Chem.*, **42**, 861 (2007); https://doi.org/10.1016/j.ejmech.2006.12.031.
15. S. He, P. Jain, B. Lin, M. Ferrer, Z. Hu, N. Southall, X. Hu, W. Zheng, B. Neuenswander, C.H. Cho, Y. Chen, S.A. Worlikar, J. Aube, R.C. Larock, F.J. Schoenen, J.J. Marugan, T.J. Liang and K.J. Frankowski, *ACS Comb. Sci.*, **17**, 641 (2015); https://doi.org/10.1021/acscombsci.5b00101.
16. Y. He, J. Xu, Z.H. Yu, A.M. Gunawan, L. Wu, L. Wang and Z.Y. Zhang, *J. Med. Chem.*, **56**, 832 (2013); https://doi.org/10.1021/jm301781p.
17. A.M. Venkatesan, O. Dos Santos, J. Ellingboe, D.A. Evrard, B.L. Harrison, D.L. Smith, R. Scerni, G.A. Hornby, L.E. Schechter and T.H. Andree, *Bioorg. Med. Chem. Lett.*, **20**, 824 (2010); https://doi.org/10.1016/j.bmcl.2009.12.093.
18. H.F. Guo, H.Y. Shao, Z.Y. Yang, S.T. Xue, X. Li, Z.Y. Liu, X.B. He, J.D. Jiang, Y.Q. Zhang, S.Y. Si and Z.R. Li, *J. Med. Chem.*, **53**, 1819 (2010); https://doi.org/10.1021/jm901685n.
19. I.N. Gaisina, F. Gallier, A.V. Ougolkov, K.H. Kim, T. Kurome, S. Guo, D. Holzle, D.N. Luchini, S.Y. Blond, D.D. Billadeau and A.P. Kozikowski, *J. Med. Chem.*, **52**, 1853 (2009); https://doi.org/10.1021/jm801317h.
20. S. Kumar, W. Namkung, A.S. Verkman and P.K. Sharma, *Bioorg. Med. Chem.*, **20**, 4237 (2012); https://doi.org/10.1016/j.bmc.2012.05.074.
21. S. Rizzo, C. Riviere, L. Piazzi, A. Bisi, S. Gobbi, M. Bartolini, V. Andrisano, F. Morroni, A. Tarozzi, J.P. Monti and A. Rampa, *J. Med. Chem.*, **51**, 2883 (2008); https://doi.org/10.1021/jm8002747.
22. J. Suh, K.Y. Yi, Y.S. Lee, E. Kim, E.K. Yum and S. Yoo, *Bioorg. Med. Chem. Lett.*, **20**, 6362 (2010); https://doi.org/10.1016/j.bmcl.2010.09.102.
23. N. Negoro, S. Sasaki, S. Mikami, M. Ito, M. Suzuki, Y. Tsujihata, R. Ito, A. Harada, K. Takeuchi, N. Suzuki, J. Miyazaki, T. Santou, T. Odani, N. Kanzaki, M. Funami, T. Tanaka, A. Kogame, S. Matsunaga, T. Yasuma and Y. Momose, *ACS Med. Chem. Lett.*, **1**, 290 (2010); https://doi.org/10.1021/ml1000855.
24. J.C. Sacchettini, A. Aggarwal and M.K. Parai, Substituted Benzofuran Derivatives as Novel Antimycobacterial Agents, Patent WO 2016172498 A1 (2016).
25. K. Manna and Y.K. Agrawal, *Eur. J. Med. Chem.*, **45**, 3831 (2010); https://doi.org/10.1016/j.ejmech.2010.05.035.
26. J. Renuka, K.I. Reddy, K. Srihari, V.U. Jeankumar, M. Shravan, J.P. Sridevi, P. Yogeeswari, K.S. Babu and D. Sriram, *Bioorg. Med. Chem.*, **22**, 4924 (2014); https://doi.org/10.1016/j.bmc.2014.06.041.
27. B.R. Thorat, B. Nazirkar, V.B. Thorat, M. Mandewale, A. Nagarsekar and R.S. Yamgar *J. Chem. Sci. Photon*, **110**, 279 (2016).
28. B. Nazirkar, U. Patil, B. Thorat, M. Mandewale, H. Gaokar, A. Pandhare and R. Yamgar, *Der Pharm. Chemica*, **9**, 45 (2017).
29. S. Santoshkumar, N.D. Satyanarayana, R. Anantacharya and P. Sameer, *Int. J. Pharm. Pharm. Sci.*, **9**, 260 (2017); https://doi.org/10.22159/ijpps.2017v9i5.17564.
30. T. Aboul-Fadl, H.A. Abdel-Aziz, M.K. Abdel-Hamid, T. Elsaman, J. Thanassi and M.J. Pucci, *Molecules*, **16**, 7864 (2011); https://doi.org/10.3390/molecules16097864.
31. B. Zhou, Y. He, X. Zhang, J. Xu, Y. Luo, Y. Wang, S.G. Franzblau, Z. Yang, R.J. Chan, Y. Liu, J. Zheng and Z.-Y. Zhang, *Proc. Natl. Acad. Sci. USA*, **107**, 4573 (2010); https://doi.org/10.1073/pnas.0909133107.
32. L. Encinas, H. O'Keefe, M. Neu, M.J. Remuinan, A.M. Patel, A. Guardia, C.P. Davie, N. Perez-Macias, H. Yang, M.A. Convery, J.A. Messer, E. Perez-Herran, P.A. Centrella, D. Alvarez-Gomez, M.A. Clark, S. Huss, G.K. O'Donovan, F. Ortega-Muro, W. McDowell, P. Castaneda, C.C. Arico-Muendel, S. Pajk, J. Rullas, I. Angulo-Barturen, E. Alvarez-Ruiz, A. Mendoza-Losana, L.B. Pages, J. Castro-Pichel and G. Evindar, *J. Med. Chem.*, **57**, 1276 (2014); https://doi.org/10.1021/jm401326j.
33. S. Iqbal, M. Kesherwani, S. Dixit, D. Velmurugan and G. Krishnasamy, *Int. J. Innov. Res. Computer Commun. Eng.*, **3**, 11428 (2015).
34. P. Karunakar, C.R. Girija, V. Krishnamurthy, V. Krishna and K.V. Shivakumar, *Tuberc. Res. Treat.*, **2014**, Article ID 697532 (2014); https://doi.org/10.1155/2014/697532.

35. B.R. Thorat, B. Nazirkar, V.B. Thorat, K. More, R. Jagtap and R. Yamgar, *Asian J. Res. Chem*, **9**, 116 (2016); https://doi.org/10.5958/0974-4150.2016.00021.3.
36. B.R. Thorat, B. Nazirkar, V.B. Thorat, K. More, R. Jagtap and R. Yamgar, *Asian J. Chem.*, **28**, 2346 (2016); https://doi.org/10.14233/ajchem.2016.19894.
37. M.J. Ahsan, J.G. Samy, C.B. Jain, K.R. Dutt, H. Khalilullah and M.S. Nomani, *Bioorg. Med. Chem. Lett.*, **22**, 969 (2012); https://doi.org/10.1016/j.bmcl.2011.12.014.
38. K.P. Harish, K.N. Mohana, L. Mallesha and B.N. Prasanna Kumar, *Eur. J. Med. Chem.*, **65**, 276 (2013); https://doi.org/10.1016/j.ejmech.2013.04.054.
39. D.R. Guda, S.J. Park, M.W. Lee, T.J. Kim and M.E. Lee, *Eur. J. Med. Chem.*, **62**, 84 (2013); https://doi.org/10.1016/j.ejmech.2012.12.035.
40. H. Rajak, B. Singh Thakur, A. Singh, K. Raghuvanshi, A.K. Sah, R. Veerasamy, P.C. Sharma, R. Singh Pawar and M.D. Kharya, *Bioorg. Med. Chem. Lett.*, **23**, 864 (2013); https://doi.org/10.1016/j.bmcl.2012.11.051.
41. N.C. Desai, N. Bhatt, H. Somani and A. Trivedi, *Eur. J. Med. Chem.*, **67**, 54 (2013); https://doi.org/10.1016/j.ejmech.2013.06.029.
42. J. Sun, H. Zhu, Z.M. Yang and H.L. Zhu, *Eur. J. Med. Chem.*, **60**, 23 (2013); https://doi.org/10.1016/j.ejmech.2012.11.039.
43. M. Neelgundmath, K.R. Dinesh, C.D. Mohan, F. Li, X. Dai, K.S. Siveen, S. Paricharak, D.J. Mason, J.E. Fuchs, G. Sethi, A. Bender, K.S. Rangappa, O. Kotresh and Basappa, *Bioorg. Med. Chem. Lett.*, **25**, 893 (2015); https://doi.org/10.1016/j.bmcl.2014.12.065.
44. S.M. Hiremath, A. Suvitha, N.R. Patil, S.S. Khemalapure, C.S. Hiremath, S.K. Pattanayak, V.S. Negalurmath, K. Obelannavar, S.J. Armakovic and S. Armakovic, *Spectrochim. Acta A Mol. Biomol. Spectrosc.*, **205**, 95 (2018); https://doi.org/10.1016/j.saa.2018.07.003.
45. V.S. Negalurmath, S.K. Boda, O. Kotresh, P.V. Anantha Lakshmi and M. Basanagouda, *Chemical Data Collections*, **19**, 100178 (2019); https://doi.org/10.1016/j.cdc.2019.100178.
46. M.C.S. Lourenco, M.V.N. de Souza, A.C. Pinheiro, M.L. Ferreira, R.S.B. Goncalves, T.C.M. Nogueira and M.A. Peralta, *ARKIVOC*, **15**, 181 (2007); https://doi.org/10.3998/ark.5550190.0008.f18.