Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2017, 9(4):108-113

Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Synthesis and Characterization of Dihydro-1H-Benzimidazole-8-Carboxylic Acids as a Potential Antimicrobial Agents

Mohana Rao Anguru, Ashok Kumar Taduri and Rama Devi Bhoomireddy*

Department of Chemistry, Jawaharlal Nehru Technological University Hyderabad, College of Engineering, Kukatpally, Hyderabad (T.S.), India

---

ABSTRACT

The structure activity relationships of 1H-benzimidazole-8-carboxylic acids (9a-9f), 1H-benzimidazole-8-carboxylates (8a-8f) are described. Characteristic compounds showed improved demanding activity over a previously recognized 2-aminobenzimidazole series. In the lead optimization process and structure–activity relationship (SAR) of separate inhibitors based on modification of the lead molecule (E)-1,6-dimethyl-9-oxo-2-styryl-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylic acids, identified from a high-throughput-screen. Noticeably, 4-chlorobenzyl, 4-fluorobenzyl benzimidazole derivatives 9c, 9e gave remarkable antimicrobial activities against saccharomyces cerevisiae, *MRSA and* bacillus proteus *with MIC values of 1, 2 and 4 μg/mL, respectively. Experimental research revealed that compound* ***5c*** *could effectively intercalate into calf thymus DNA to form compound* ***5c*** *DNA complex which might block DNA replication and thus exert antimicrobial activities.*

Keywords: OPDA; Benzimidazole; Acetic acid; 1H-benzimidazole-8-carboxylate; Antimicrobial activity

---

INTRODUCTION

Benzimidazoles and its derivatives represent one of the most biologically active classes of compounds, possessing a wide spectrum of activities and these are well-documented in literature.

Microbial drug resistance is a serious issue, especially as increasing numbers of strains are becoming resistant to multiple antimicrobial agents, with some bacteria now being resistant to all available antibiotics. There is thus a critical need to develop new drugs with novel mechanism of action. However, the investment available for such development is frequently lower than the required level. The development of new drug entities is hampered by several issues, notably the high cost and length of time required, as well as the logistical and regulatory challenges of performing the necessary clinical evaluations across multiple geographical areas. Therefore, a few new classes of antimicrobials have been developed since the late 1980s, [1-3] and much research has focused only on the chemical modification of existing drugs to improve their potency and/or ability to overcome antibiotic resistance mechanisms. Even if this approach does not improve antimicrobial activity directly, it may lead to derivatives that can usefully inhibit virulence mechanisms [4]. Compounds having benzimidazole as a structural motif have been widely used in medicinal chemistry drug development, and researchers are actively seeking new uses and applications of this heterocycle [5]. In the past few decades, benzimidazole and its derivatives have grasped much attention due to their chemotherapeutic values [6]. Furthermore, the pharmacological properties as well as therapeutic applications of benzimidazole depend upon the pattern of substitution and recently they are reported to possess many pharmacological activities. Benzimidazole-containing compounds have numerous medical and biological activities, such as antitumor [7] antibacterial, [8-11] antifungal, [12] antiviral, [13-17] anticonvulsant, [18] antidepressant, [19] analgesic, [20] anti-inflammatory, [21] anthelmintic [22] and antidiabetic properties [23]. Therefore it was enabled that compounds containing benzimidazole nucleus would result in interesting biological activities. In the present

study 2-substituted benzimidazoles were synthesized by treating o-phenylenediamine with different carboxylic acids. They were then subjected to nitration at room temperature to get 5-nitro-2-substituted benzimidazole derivatives (Figures 1 and 2).

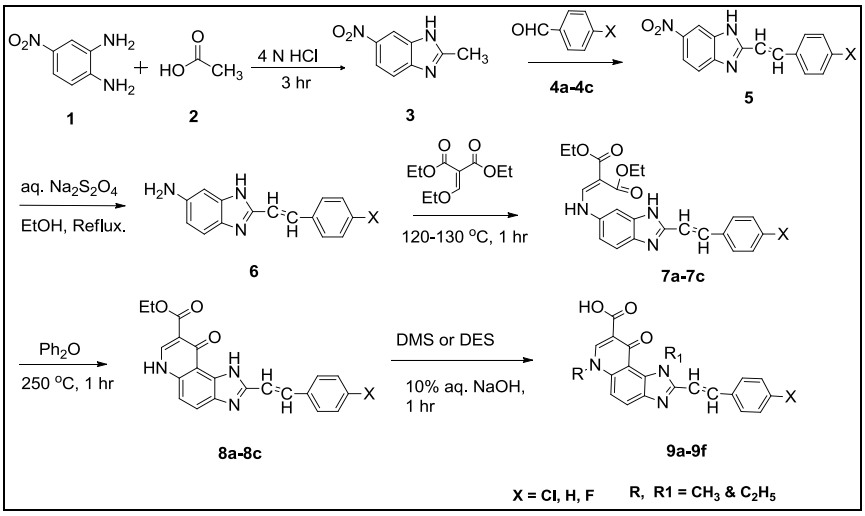

Figure 1: Scheme-synthesis of target compounds (9a-9f)

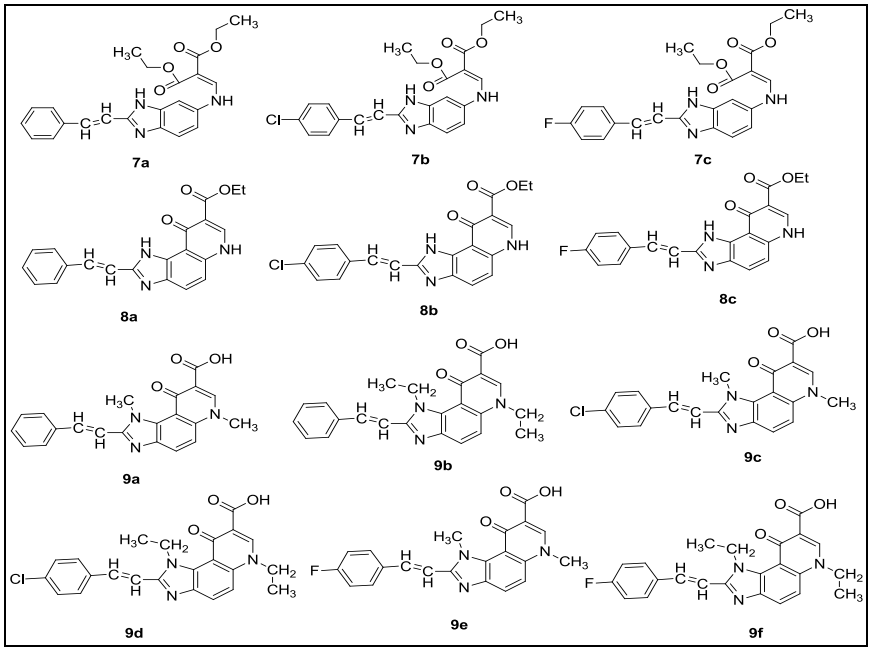

Figure 2: Synthesis of target compounds (9a-9f)

EXPERIMENTAL SECTION

Preparation of 5-nitro-2-methylbenzimidazole (3)

A mixture of 4-nitro-o-phenyldiamine (1) (10 mmol), acetic acid (2) (10 mmol) and 4 N HCl (50 mL) was refluxed for 3 hrs at 100°C. The progress of the reaction was monitored by TLC analysis. After completion of the reaction, the mixture was cooled to room temperature and neutralized using aq. ammonia solution to pH ≥ 8.0. The separated solid was filtered, washed with ice cold water (2 × 30 mL) to remove any salts present and dried to obtain crude product. The latter was recrystallized using ethanol as solvent to obtain pure 5-nitro-2-methyl benzimidazole (3).

Compound 3 on fusion with substituted benzaldehydes (4a-4c), under neat conditions at 160-180°C for 2-3 hr yielded known 5-nitro-2-styrylbenzimidazole derivatives 5. Compound 5 was treated with aq. $Na_2S_2O_4$ in ethanol and refluxed for 30 min gave a reduced product 5-amino-2-styryl benzimidazole derivative 6 which on reaction with

ethoxymethylene malonic acids ester (for 1 hr at 120-130°C gave an enamine i.e., (E)-diethyl-2-(((2-(4-substitutedstyryl)-1*H*-benzimidazol-6-yl)amino)methylene)malonate 7. Compound 7 on thermal cyclization using diphenyl ether under refluxing conditions at 250 ^{o}C for about 1 hr yielded (*E*)-ethyl 9-oxo-2-styryl-6,9-dihydro-1*H*-imidazo[4,5-f]quinoline-8-carboxylate 8. Compound 8 on reaction with alkyalting agents such as DMS or DES in 10% aq. NaOH and refluxing on water bath for about 30 min resulted (*E*)-1,6-dialkyl-9-oxo-2-styryl-6,9-dihydro-1*H*-imidazo[4,5-f]quinoline-8-carboxylic acid (9).

Preparation of (E)-diethyl-2-(((2-(4-substitutedstyryl)-1H-benzimidazol-6-yl)amino)methylene)malonate(7) from 5 and 6:

A mixture of **5** (10 mmol), aq. $Na_2S_2O_4$ (30 mmol) in ethanol (25 mL) was allowed to reflux on water bath for 30 min. The completion of the reaction was monitored by checking TLC. At the end of the period, the reaction mixture was cooled to RT and ice cold water (3 × 20 mL) was added to reaction mixture and extracted by using ethyl acetate. The organic layer separated and concentrated to obtain crude 6. The mixture of compound **6** (10 mmol), EMME (10 mmol) and ethanol (20 mL) was allowed to reflux at 120-130°C for 1 hr and the reaction was monitored by checking TLC. At the end of this period, the reaction mixture was concentrated under reduced pressure to obtain **7**.

(E)-diethyl-2-(((2-styryl-1H-benzo[d]imidazol-6-yl)amino)methylene)malonate (7a):

Yield = 2.8 gm (75%), M. P. = 180°C, IR (KBr): 3000-3409 cm^{-1} (broad, medium, -NH stretching), 1707 cm^{-1} (strong, sharp, C=O stretching), 1620 cm^{-1} (strong, sharp, C=O stretching); ^{1}H NMR spectrum (DMSO/d_6/TMS, 400 MHz): δ 1.2-1.4 (m, 6H, 2-O-CH-CH_3), 4.2-4.4 (m, 4H, 2-O-CH_2-CH), 6.9 (dd, 2H, Ar-H), 7.2 (s, 1H, Ar-H), 7.32 (d, 1H, vinylic –CH=CH, *J* = 16 Hz), 7.34 (d, 1H, vinylic –CH=CH, *J* = 16 Hz), 8.0-8.1 (m, 5H, phenylic protons), 8.4 (s, 1H, enamine proton), 8.42 (s, 1H, enamine –NH), 11.0 (s, 1H, imidazole –NH), MS: *m/z* 406.2 ($M^{+\cdot}$).

(E)-diethyl 2-(((2-(4-chlorostyryl)-1H-benzo[d]imidazol-6-yl)amino)methylene)malonate7b:

Yield = 3.0 gm (70%), M. P. = 190°C, IR (KBr): 3267 cm^{-1} (broad, medium, -NH of enamine), 3467 cm^{-1} (broad, medium, -NH stretching of imidazole ring), 2937-2980 cm^{-1} (small, sharp, due to –C=N stretching), 1735 cm-1 (strong, sharp, -C=O stretching), 1680 cm^{-1} (strong, sharp, -O-C=O stretching); ^{1}H – NMR spectrum (DMSO/d_6/TMS, 400 MHz): δ 1.2-1.4 (m, 6H, 2-O-CH-CH_3), 4.2-4.4 (m, 4H, 2-O-CH_2-CH), 6.9 (dd, 2H, Ar-H), 7.2 (s, 1H, Ar-H), 7.32 (d, 1H, vinylic –CH=CH, *J* = 16 Hz), 7.34 (d, 1H, vinylic –CH=CH, *J* = 16 Hz), 7.4-7.6 (m, 4H, phenylic protons), 8.4 (s, 1H, enamine proton), 8.42 (s, 1H, enamine –NH), 11.0 (s, 1H, imidazole –NH), MS: *m/z* 443.14 ($M^{+\cdot}$).

(E)-diethyl-2-(((2-(4-fluorostyryl)-1H-benzo[d]imidazol 6yl)amino)methylene)malonate(7c):

Yield = 2.8 gm (70%), M. P. = 160°C, IR (KBr): 3267 cm^{-1} (broad, medium, -NH of enamine), 3467 cm^{-1} (broad, medium, -NH stretching of imidazole ring), 2937-2980 cm^{-1} (small, sharp, due to –C=N stretching), 1735 cm^{-1} (strong, sharp, -C=O stretching), 1680 cm^{-1} (strong, sharp, -O-C=O stretching), ^{1}H – NMR spectrum (DMSO/d_6/TMS, 400 MHz): δ 1.2-1.4 (m, 6H, 2-O-CH-CH_3), 4.2-4.4 (m, 4H, 2-O-CH_2-CH), 6.9 (dd, 2H, Ar-H), 7.2 (s, 1H, Ar-H), 7.32 (d, 1H, vinylic –CH=CH, *J* = 16 Hz), 7.34 (d, 1H, vinylic –CH=CH, *J* = 16 Hz), 7.4-7.6 (m, 4H, phenylic protons), 8.4 (s, 1H, enamine proton), 8.42 (s, 1H, enamine –NH), 11.0 (s, 1H, , imidazole –NH), MS: *m/z* 424.16 ($M^{+\cdot}$).

Preparation of (E)-ethyl-9-oxo-2-styryl-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylate(8)form(E)-diethyl-2-(((2-(4-substitutedstyryl)-1H-benzimidazol-6-yl)amino)methylene)malonate (7) :

A mixture of 7 (10 mmol) and diphenyl ether (25 mL) was allowed to reflux at 250°C for 1 hr. At the end of the period, the reaction mixture was cooled to rt and treated with hexane (25 mL) and separated solid was filtered and again washed with diethyl ether (25 mL), dried to obtain 8.

(E)-ethyl 9-oxo-2-styryl-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylate (8a):

Yield = 2.3 gm (65%), M. P. = 160°C, IR (KBr): 2982 cm^{-1} (small, broad. –NH of quinolone ring), 3239 cm^{-1} (broad, -NH stretching of imidazole ring), 1721 cm^{-1} (small, sharp, -C=O stretching), 1686 cm^{-1} (small, sharp, due to –O-C=O stretching), ^{1}H – NMR spectrum (DMSO/d_6/TMS, 400 MHz): δ 1.3 (t, 3H, -O-CH_2-CH_3), 4.3 (q, 2H, -O-CH_2-CH_3), 7.15 (d, 1H, Ar-H), 7.3 (d, 1H, Ar-H), 7.68 (d, 1H, vinylic proton, *J* = 12 Hz), 7.93 (s, 1H, phenyl proton), 7.96 (d, 1H, vinylic proton, *J* = 12 Hz), 8.14 (m, 4H, phenyl protons), 8.93 (s, 1H, enamine –CH), 12.8 (s, 1H, quinolone –NH), 13.9 (s, broad, 1H, imidazole –NH), ^{13}C NMR (DMSO-d_6, 100 MHz) δ in ppm: 15.0, 33, 62, 65, 68, 103, 105, 107, 108, 112, 118, 120, 123,126, 128, 130, 132, 134, 136, 140, 142, 146, 166, 174, MS: *m/z* 362.15 ($M^{+\cdot}$).

(E)-Ethyl-2-(4-chlorostyryl)-9-oxo-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylate (8b):
Yield = 2.3 gm (60%), M. P. = >240°C, IR (KBr): 2982 cm^{-1} (small, broad. –NH of quinolone ring), 3239 cm^{-1} (broad, -NH stretching of imidazole ring), 1721 cm^{-1} (small, sharp, -C=O stretching), 1686 cm^{-1} (small, sharp, due to –O-C=O stretching), ^{1}H – NMR spectrum (DMSO/d_6/TMS, 400 MHz): δ 1.29 (t, 3H, -O-CH_2-CH_3), 4.2 (q, 2H, -O-CH_2-CH_3), 6.6 (d, 1H, Ar-H), 6.95 (d, 1H, vinylic proton, *J* = 16 Hz), 7.0 (d, 1H, vinylic proton, *J* = 16 Hz), 7.5 (d, 1H, Ar-H), 7.6-7.8 (m, 4H, phenyl protons), 8.93 (s, 1H, enamine –CH), 12.8 (s, 1H, quinolone –NH), 13.9 (s, broad, 1H, imidazole –NH), MS: *m/z* 396.11 ($M^{+\cdot}$).

(E)-ethyl-2-(4-fluorostyryl)-9-oxo-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylate (8c):
Yield = 2.1 gm (58%), M. P. = >240°C, IR (KBr): 2982 cm^{-1} (small, broad. –NH of quinolone ring), 3239 cm^{-1} (broad, -NH stretching of imidazole ring), 1721 cm^{-1} (small, sharp, -C=O stretching), 1686 cm^{-1} (small, sharp, due to –O-C=O stretching), ^{1}H – NMR spectrum (DMSO/d_6/TMS, 400 MHz): δ 1.29 (t, 3H, -O-CH_2-CH_3), 4.2 (q, 2H, -O-CH_2-CH_3), 6.6 (d, 1H, Ar-H), 6.95 (d, 1H, vinylic proton, *J* = 16 Hz), 7.0 (d, 1H, vinylic proton, *J* = 16 Hz), 7.5 (d, 1H, Ar-H), 7.6-7.8 (m, 4H, phenyl protons), 8.93 (s, 1H, enamine –CH), 12.8 (s, 1H, quinolone –NH), 13.9 (s, broad, 1H, imidazole –NH), MS: *m/z* 378.12 ($M^{+\cdot}$).

Preparation of (E)-1,6-dialkyl-9-oxo-2-styryl-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylic acid (9) from(E)-ethyl 9-oxo-2-styryl-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylate (8) :
A mixture of 76 (10 mmol), DMS or DES (20 mmol) and 10% NaOH was allowed to heat on water bath for 30 min. The completion of the reaction was monitored by checking TLC. At the end of the period, the reaction mixture was cooled to rt and the reaction mixture was acidified using Conc. HCl till the P^H paper turns to pink (~4.0). The separated solid was filtered and dried to obtain 9.

(E)-1,6-dimethyl-9-oxo-2-styryl-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylicacid:
9a: Yield = 1.6 gm (48%), M. P. = >240°C, IR (KBr): 3020-3300 cm^{-1} (broad, medium, due to –OH), 1735 cm^{-1} (strong, sharp, due to –C=O stretching), 1620 cm^{-1} (small, sharp, due to –O-C=O stretching), ^{1}H – NMR spectrum (DMSO/d_6/TMS, 400 MHz): δ 2.69 (s, 3H, quinolone N-CH_3), 2.9 (s, 3H, imidazole N-CH_3), 6.6 (d, 1H, Ar-H), 6.9 (d, 1H, vinylic proton, *J* =16 Hz), 7.3-7.6 (m, 5H, phenyl protons), 7.7 (s, 1H, Ar-H), 7.9 (d, 1H, 6.6 (d, 1H, Ar-H), 6.9 (d, 1H, vinylic proton, *J* =16 Hz), 9.2 (s, 1H, enamine –CH), 14.0 (s, 1H, -OH), MS: *m/z* 360.13 ($M^{+\cdot}$).

(E)-1,6-diethyl-9-oxo-2-styryl-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylicacid:
9b: Yield = 1.9 gm (50%), M. P. = >240°C, IR (KBr): 3020-3300 cm^{-1} (broad, medium, due to –OH), 1735 cm^{-1} (strong, sharp, due to –C=O stretching), 1620 cm^{-1} (small, sharp, due to –O-C=O stretching), ^{1}H – NMR spectrum (DMSO/d_6/TMS, 400 MHz): δ 1.29-1.31 (m, 6H, 2-N-CH_2-CH_3), 4.3-4.6 (m, 4H, 2-N-CH_2-CH_3), 6.6 (d, 1H, Ar-H), 6.9 (d, 1H, vinylic proton, *J* =16 Hz), 7.3-7.6 (m, 5H, phenyl protons), 7.7 (s, 1H, Ar-H), 7.9 (d, 1H, vinylic proton, *J* =16 Hz), 9.2 (s, 1H, enamine –CH), 14.0 (s, 1H, -OH), MS: *m/z* 388.16 ($M^{+\cdot}$).

(E)-2-(4-chlorostyryl)-1,6-dimethyl-9-oxo-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylic acid 9c:
Yield = 2.1 gm (55%), M. P. = >240°C, IR (KBr): 3020-3300 cm^{-1} (broad, medium, due to –OH), 1735 cm^{-1} (strong, sharp, due to –C=O stretching), 1620 cm^{-1} (small, sharp, due to –O-C=O stretching), ^{1}H – NMR spectrum (DMSO/d_6/TMS, 400 MHz): δ 3.4 (s, 3H, quinolone -N-CH_3), 3.8 (s, 3H, imidazole –N-CH_3), 6.6 (d, 1H, Ar-H), 6.9 (d, 1H, vinylic proton, *J* =16 Hz), 7.4-7.6 (m, 4H, phenyl protons), 7.9 (d, 1H, vinylic proton, *J* =16 Hz), 8.4 (s, 1H, enamine –CH), 14.0 (s, 1H, -OH), MS: *m/z* 394.09 ($M^{+\cdot}$).

(E)-2-(4-chlorostyryl)-1,6-diethyl-9-oxo-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylic acid 9d:
Yield = 2.1 gm (50%), M. P. = >240°C, IR (KBr): 3020-3300 cm^{-1} (broad, medium, due to –OH), 1735 cm^{-1} (strong, sharp, due to –C=O stretching), 1620 cm^{-1} (small, sharp, due to –O-C=O stretching), ^{1}H – NMR spectrum (DMSO/d_6/TMS, 400 MHz): δ 1.29-1.31 (m, 6H, 2-N-CH_2-CH_3), 4.3-4.6 (m, 4H, 2-N-CH_2-CH_3), 6.6 (d, 1H, Ar-H), 6.9 (d, 1H, vinylic proton, *J* =16 Hz), 7.4-7.6 (m, 4H, phenyl protons), 7.7 (s, 1H, Ar-H), 7.9 (d, 1H, vinylic proton, *J* =16 Hz), 9.2 (s, 1H, enamine –CH), 14.0 (s, 1H, -OH), MS: *m/z* 422.12 ($M^{+\cdot}$).

(E)-2-(4-fluorostyryl)-1,6-dimethyl-9-oxo-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylic acid 9e:
Yield = 1.8 gm (45%), M. P. = >240°C, IR (KBr): 3020-3300 cm^{-1} (broad, medium, due to –OH), 1735 cm^{-1} (strong, sharp, due to –C=O stretching), 1620 cm^{-1} (small, sharp, due to –O-C=O stretching), ^{1}H – NMR spectrum (DMSO/d_6/TMS, 400 MHz): δ 3.4 (s, 3H, quinolone -N-CH_3), 3.8 (s, 3H, imidazole –N-CH_3), 6.6 (d, 1H, Ar-H), 6.9

(d, 1H, vinylic proton, *J* =16 Hz), 7.4-7.6 (m, 4H, phenyl protons), 7.9 (d, 1H, vinylic proton, *J* =16 Hz), 8.4 (s, 1H, enamine –CH), 14.0 (s, 1H, -OH), MS: *m/z* 378.14 ($M^{+\cdot}$).

(E)-1,6-diethyl-2-(4-fluorostyryl)-9-oxo-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylic acid 9f:

Yield = 2.0 gm (50%), M. P. = > 240°C, IR (KBr): 3020-3300 cm^{-1} (broad, medium, due to –OH), 1735 cm^{-1} (strong, sharp, due to –C=O stretching), 1620 cm^{-1} (small, sharp, due to –O-C=O stretching), ^1H – NMR spectrum (DMSO/d_6/TMS, 400 MHz): δ 1.29-1.31 (m, 6H, 2-N-CH_2-CH_3), 4.3-4.6 (m, 4H, 2-N-CH_2-CH_3), 6.6 (d, 1H, Ar-H), 6.9 (d, 1H, vinylic proton, *J* =16 Hz), 7.4-7.6 (m, 4H, phenyl protons), 7.7 (s, 1H, Ar-H), 7.9 (d, 1H, vinylic proton, *J* =16 Hz), 9.2 (s, 1H, enamine –CH), 14.0 (s, 1H, -OH), MS: *m/z* 406.15 ($M^{+\cdot}$) (Table 1).

Antimicrobial Activity [24]

The synthesized compounds were tested for antimicrobial activity by disc diffusion method. They were dissolved in DMSO and sterilized by filtering through 0.45 µm millipore filter. Final inoculums of 100 µL suspension containing 108 CFU/ml of each bacterium and fungus used. Nutrient agar (antibacterial activity) and sabouraud's dextrose agar medium (antifungal activity) was prepared and sterilized by an autoclave (121°C and 15 Ibs for 20 min) and transferred to previously sterilized petridishes (9 cm in diameter). After solidification, petriplates were inoculated with bacterial organisms in sterile nutrient agar medium at 45°C, and fungal organisms in sterile sabouraud's dextrose agar medium at 45°C in aseptic condition. Sterile Whatmann filter paper discs (previously sterilized in U.V. lamp) were impregnated with synthesized compounds at a concentration of 25; 100 mg/disc were placed in the organism-impregnated petriplates under sterile condition. The plates were left for 30 min to allow the diffusion of compounds at room temperature. Antibiotic discs of ciprofloxacin (100 µg/disc) and ketaconazole (100µg /disc) were used as positive control, while DMSO used as negative control. Then the plates were incubated for 24 hr at 37 ± 1°C for antibacterial activity and 48 hr at 37 ± 1°C for antifungal activity. The zone of inhibition was calculated by measuring the minimum dimension of the zone of no microbial growth around each disc.

Table 1: The zone of inhibition in µg/mL of the target compounds (9a-9f)

Entry	PV	KP	BC	EF	AN	AF
7a	10	12	16	15	10	8
7b	12	18	16	12	11	10
7c	10	---	14	---	9	8
8a	15	12	12	18	11	10
8b	18	19	20	21	15	13
8c	20	16	15	11	10	9
9a	22	25	26	---	13	12
9b	23	27	22	20	12	10
9c	---	15	16	18	8	7
9d	28	26	29	22	16	14
9e	15	29	22	20	17	12
9f	18	15	---	12	10	9
Norfloxacin	30	31	28	25	---	---
Ketoconazole	---	---	---	---	18	16

PV: *proteus vulgaris* (NCTC 4635), KP: *klesibella pneumonia* (ATCC 29655), *BC: bacillus cereus* (NL98), *EF: enterococcus faecium* (ATCC 29212), *AN: aspergillus niger, AF: aspergillus fumigatus, ---:* No Zone of Inhibition

RESULTS AND DISCUSSION

The synthesized compounds were evaluated for *in vitro* antibacterial activity against gram negative bacteria *Proteus vulgaris* (NCTC 4635), *klesibella pneumonia* (ATCC 29655) and gram positive bacteria *bacillus cereus* (NL98), *enterococcus faecium* (ATCC 29212). These are the agents which commonly causes urinary tract infection, nosocomial infection, biliary tract infection. The gram negative organism *klesibella pneumonia* causes pneumonia, bronco pneumonia and bronchitis infection. The gram negative organisms *bacillus cereus* and *enterococcus faecium* cause endocarditis, bacteremia, meningitis and septicaemia. From the biological data, it was evident that the compound 9d was found to be more active against *bacillus cereus* (NL98), *proteus vulgaris* (NCTC 4635); whereas compound 9e, 9b was found to be more active aganist *klesibella pneumonia* (ATCC 29655) with zone of inhibition 29, 27 mm values respectively when compared to reference compound (norfloxacin). Further, the compound 9a showed better acivity against microorganisms *klesibella pneumonia* (ATCC 29655), *bacillus cereus* (NL98) with 25, 26 mm values respectively. Compound 9e, 9d was found to be more active against *aspergillus niger* with zone of inhibition of 17, 16 mm values and active against fungal strain *aspergillus fumigatus* 12, 14 mm values. However the antimicrobial activity of the synthesized compounds against the tested organisms was found to be more active

than that of respective standard drug at tested dose level. In future study the activity of the compounds may be manipulated by introducing unsaturation or heterocyclic ring at C_2 of benzimidazole.

CONCLUSION

With an aim of developing potent antimicrobial agent, a series of novel (E)-1,6-dimethyl-9-oxo-2-styryl-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylic acid and its derivatives were synthesized from 4-nitro-o-phenyldiamine by the multistep reaction synthesis and characterized by FT-IR, 1H-NMR, mass spectroscopy and elemental analysis. All the title compounds were screened for their *in vitro* antimicrobial activity by the agar streak dilution method, and its MIC was determined against various strains of microorganisms. Results revealed that compounds containing an electron-withdrawing group such as floro,at the phenyl group attached to C–4 of 2-styryl displayed superior antimicrobial activity. Moreover, the unsubstituted derivatives displayed moderate activity. Among several tested compounds (E)-2-(4-fluorostyryl)-1,6-dimethyl-9-oxo-6,9-dihydro-1H-imidazo[4,5-f]quinoline-8-carboxylic acid (9e) showed better activity. Hence, this compound may serves as a lead molecule to obtain clinically useful antimicrobial agent.

ACKNOWLEDGEMENTS

The authors (AM) are thankful to the authorities, Department of Chemistry, College of Engineering, Jawaharlal Nehru Technological University Hyderabad for providing financial support and laboratory facilities.

REFERENCES

[1] BE Franco; MA Martínez; MS Rodríguez; AI Wertheimer. *Infect Drug Resist*. **2009**, 2, 1-11.
[2] CT Barrett; JF Barrett. *Curr Opin Biotechnol*. **2003**, 14, 621-626.
[3] ST Cole. *Philos Trans R Soc B Biol Sci*. **2014**, 369.
[4] F Imperi; F Massai; M Facchini; E Frangipani; D Visaggio; L Leoni; A Bragonzi; P Visca. *Proc Natl Acad Sci*. **2013**, 110, 7458-7463.
[5] M Wang; XHan; Z Zhou. *Expert Opin Ther Pat*. **2015**, 25, 595-612.
[6] KJ Soderlind; B Gorodetsky; AK Singh; NR Bachur; GG Miller; JW Lown. *Anticancer Drug Des*. **1999**, 14, 19-36.
[7] K Kumar; D Awasthi; SY Lee; JE Cummings; SE Knudson; RA Slayden; I Ojima. *Bioorg Med Chem*. **2013**, 21, 3318-3326.
[8] E Mentese; H Bektas; S Ulker; O Bekircan; B Kahveci. *J Enzyme Inhib Med Chem*. **2014**, 29, 64-68.
[9] J Velík; V Baliharová; J Fink-Gremmels; S Bull; J Lamka; L Skálová. *Res Vet Sci*. **2004**, 76, 95-108.
[10] D Pathak; N Siddiqui; B Bhrigu;W Ahsan; M Shamsher Alam. *Der Pharmacia Lettre*. **2010**, 2(2), 27-34.
[11] R Janupally; VU Jeankumar; KA Bobesh; V Soni; PB Devi; VK Pulla, P Suryadevara; KS Chennubhotla; P Kulkarni; P Yogeeswari; D Sriram. *Bioorg Med Chem*. **2014**, 22, 5970-5987.
[12] Y Ke; X Zhi; X Yu; G Ding; C Yang; H Xu. *Comb Chem High Throughput Screen*. **2014**, 17, 89-95.
[13] M Tonelli; G Paglietti; V Boido; F Sparatore; F Marongiu; E Marongiu; P La Colla; R Loddo. *Chem Biodivers*. **2008**, 5, 2386-2401.
[14] G Vitale; P Corona; M Loriga; A Carta, G Paglietti; C Ibba; G Giliberti; R Loddo; E Marongiu; P la Colla. *Med Chem*. **2010**, 6, 70-78.
[15] M Tonelli; M Simone; B Tasso, F Novelli, V Boido; F Sparatore; G Paglietti; S Pricl; G Giliberti; S Blois; C Ibba. *Bioorg Med Chem*. **2010**, 18, 2937-2953.
[16] G Vitale; P Corona; M Loriga; A Carta; G Paglietti; G Giliberti; G Sanna; P Farci; ME Marongiu; P la Colla. *Eur J Med Chem*. **2012**, 53, 83-97.
[17] M Tonelli; F Novelli; B Tasso; I Vazzana; A Sparatore, V Boido; F Sparatore; P la Colla; G Sanna; G Giliberti; B Busonera. *Bioorg Med Chem*. **2014**, 22, 4893-4909.
[18] RV Shingalapur; KM Hosaman; RS Keri; MH Hugar. *Eur J Med Chem*. **2010**, 45, 1753–1759.
[19] NS Andalip; S Bawa; R Ali; O Afzal; MJ Akhtar; B Azad; R Kumar. *J Pharm Bioallied Sci*. **2011**, 3, 194-212.
[20] PA Datar; SA Limaye. *Med Chem*. **2015**, 14, 35-46.
[21] KCS Achar; KM Hosamani; HR Seetharamareddy. *Eur J Med Chem*. **2010**, 45, 2048-2054.
[22] K Sreena; R Ratheesh; M Rachana; M Poornima; C Shyni. *Hygeia*. **2009**, 1(1), 21-22.
[23] Y Bansal; O Silakari. *Bioorg Med Chem*. **2012**, 20, 6208-6236.
[24] B Vijaya; K Rajeev; G Varadaraj; G Gautham. *Asian J Research Chem*. **2009**, 2(2), 162-167.