Design and Synthesis of Novel Quinoline-based Heterocyclic Schiff Bases for their Anti-microbial and Anti-tuberculosis activity

V. Rajashakar [1*], Y. Naresh [2], M. ChinnaEswaraiah[3]

[1]Department of Pharmaceutical Chemistry, Anurag Pharmacy College, Ananthagiri, kodad Suryapet- 508206, Telangana, India

[2]Department of Pharmaceutical Chemistry, Anurag Pharmacy College, Ananthagiri, kodad Suryapet- 508206, Telangana, India

[3]Department of Pharmacognosy, Anurag Pharmacy College, Ananthagiri, kodad Suryapet- 508206, Telangana, India

*Corresponding author: V. Rajashakar

E-mail:shekar864@gmail.com

Mobile No.: 8008072838

ABSTRACT

The potent and broad activity of Quinoline has established it as one of the important biologically important scaffold. The present work describes the synthesis of series of quinoline-based heterocyclic schiff bases by the reaction of various substituted 2-chloro-3-formylquinolines with substituted aminopyridines and Isoniazide. The starting compound *i.e* 2-chloro-3-formylquinolines was synthesized through Vilsmeier-Haack reaction from corresponding acetanilides. All the compounds were characterized by spectroscopic techniques like ^{1}H NMR, IR and mass spectra. The newly synthesized compounds were tested for their antimicrobial and anti-tuberculosis activities. The primarily biological screening results shows that compound **15d** showed highest activity against *S. auresus* and will be emerge as potential anti-microbial agent. The compound **16d** displayed the highest anti-tuberculosis activity and presumably will be a potential candidate for further biological studies.

Keywords: Quinoline, Schiff bases, Vilsmeier-Haack reaction, Antimicrobial activity, Anti-tuberculosis activity.

INTRODUCTION

Schiff's bases are substances with an imine or an azomethine(-C=N-) functional group. Hugo Schiff published the initial research on these, which are the condensation by products of primary amines with carbonyl compounds.[1-3] Due to a variety of biological actions including anti-inflammatory[4–7],

analgesic [5-8], antibacterial[9–10], antitubercular[11], and cytotoxic[12]. Schiff bases have become more significant in the medical and pharmaceutical fields.

Quinolines are present in a variety of natural products and show exceptional antimalaria[13], antibiotic[14], and anti-tubercular[15] properties. Given the biological significance of quinoline-based heterocyclic Schiff bases, this effort is being done to create new heterocyclic Schiff bases of 2-Chloro-3-formylquinolines using various heterocyclic amines. The thorough literature search found that newly developed Schiff bases of various heterocyclic amines with quinoline aldehydes have not been produced and demonstrated for their anti-tuberculosis, antibacterial action. Therefore, the ultimate goal of the current effort is to design and create the Schiff bases of 2-chloro-3-formyl using heterocyclic amines.

EXPERIMENTAL

Material and Methods

All the required chemicals used were obtained from sigma Aldrich and Sd-fine chemicals. All the solvents used were of laboratory grade. Each reaction was monitored by TLC by using appropriate solvent system. Precoated TLC plates (0.25 mm silica gel) were obtained from E.Merck. Visualization of the spots on TLC plates was achieved by exposure to U.V. light. All the synthesized compounds were purified by column chromatography. Melting points were determined on CINTEX programmable melting point apparatus. Infrared (IR) spectra were recorded in KBr on a Perkin Elmer FT-IR instrument and frequencies are given in cm^{-1}. ^1H NMR spectra were recorded on Bruker Avance 300 MHz. The samples were made in CDCl3 / DMSO/CF_3COOD and TMS used as the internal standard and are given in δ scale. Mass spectra were recorded on ESI mass spectrometer.

General procedure for synthesis of Acetanilide (13a-13g)

Different anilines (0.05 mol) were dissolved in glacial acetic acid (0.05 mol), and acetic anhydride was then added. After stirring, this reaction mixture was placed into ice and left to stand for two hours. The precipitate was obtained and then filtered before being dried, washed with water multiple times, and re-crystallized using methanol.

General procedure for synthesis of 2-chloro-3-formylquinoline (14a-14d)

In a 500 ml three-necked round bottom flask, combine 9.1 gm (9.6 ml, 0.125 mol) of DMF with 0 °C for duration of one hour and 53.7 gm (32.2 ml, 0.35 mol) of $POCl_3$ for the same time period. A full

hour of stirring was permitted at 0 °C. During a 30-minute period at 0 °C, 6.75 gm (0.05 mol) of acetanilide was added in small amounts. The reaction mixture then refluxed at 85 °C for 16 hours while being tracked by TLC. After the reaction was finished, 300 g of ice was added to the reaction mixture, which was then agitated for 30 minutes at 0 °C. A light yellow solid was then obtained by extracting it with ethyl acetate and evaporating the top layer.

2-Chloro-3-formylquinoline (14a)

Yield: 80%; pale yellow solid; m.p.: 142- 143 °C; FT-IR (KBr) cm^{-1}: 750 (C-Cl), 1667 (C=O) ; ^{1}H NMR (300 MHz, $CDCl_3$): δ 9.1 (1H, d, *J* = 3.96 Hz), 8.9 (1H, d, *J* = 2.45 Hz), 8.5 (1H, d, *J* = 8.2 Hz), 8.4 (1H, d, *J* = 2.45, 9.25 Hz), 7.9 (1H, s), 7.6 (1H, m). ESI (MS): 222 [M+1].

2-chloro-3-formylquinoline (14b)

Yield: 80% ; pale yellow solid; m.p.: 142-143 °C; FT-IR (KBr) cm^{-1}: 769 (C-Cl), 1687 (C=O); ^{1}H NMR (300 MHz, $CDCl_3$): δ 9.7 (1H, d, *J* = 3.32 Hz), 9.02 (1H, d *J* =2.20 Hz), 7.66 (1H, d, *J* = 2.36 Hz), 7.51 (1H d, *J* =3.36 Hz), 2.35 (3H, s); ESI (MS) : 222 [M+1].

8-methyl 2-chloro-3-formylquinoline (14c)

Yield: 73% ; pale yellow solid; m.p.: 123- 125 °C;); FT-IR (KBr) cm^{-1} : 720 (C-Cl), 1670 (C=O); ^{1}H NMR (300 MHz, $CDCl_3$) : δ 9.8 (1H, d *J* = 3.32 Hz), 9.1(1H, d, *J* = 2.20 Hz), 7.66 (1H, d, *J* =2.36 Hz) 7.51 (1H, d, *J* =3.36 Hz) 2.35 (3H, s); ESI (MS) : 254 [M+1].

6-methoxy 2-chloro-3-formylquinoline (14d)

Yield: 73%; pale yellow solid; m.p.: 142-145 °C; FT-IR (KBr) cm^{-1} : 725(C-Cl), 1682(C=O); ^{1}H NMR (300 MHz, $CDCl_3$): δ 9.73 (1H, d, *J* =2.16 Hz), 7.92 (1H, d, *J* =2.63 Hz), 7.66(1H, d, *J* =3.36 Hz), 2.21 (3H, s); ESI (MS): 257 [M+1].

General procedure for synthesis of (Z)-N-((2-chloroquinolin-3-yl)methylene)pyridin-2-amine (15a-15e)

2-Chloro-3-Formylquinolines 1.91 gm (0.01 mol) and 2-Aminopyridine 0.94 gm (0.01 mol) were dissolved in ethanol and catalytic amounts of sulphuric acid were added to a 100 ml three-necked round-bottomed flask. After that, it was allowed to reflux for 4 hours while TLC kept an eye on it.

The desired product was obtained by recrystallizing the reaction mixture with ethanol once the reaction was complete by pouring the reaction mixture into ice, filtering, washing the solid mass several times with water, drying, and filtering again. 5.3.7 gm (32.2 ml, 0.35 mol) POCl3 for an hour at 0 °C in a 500 ml three-necked round-bottomed flask with 9.1 gm (9.6 ml, 0.125 mol) of DMF. The mixture was left to stir at 0 °C.

(Z)-N-((2-chloroquinolin-3-yl)methylene)pyridin-2-amine (15a)

Yield: 71 %; yellow solid; m.p.: 285 $^{\circ}$C; FT-IR (KBr) cm^{-1} : 2998 (Ar-CH), 1669 (C=N), 1219 (C-N), 765 (C- Cl); ^{1}H-NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 8.32 (1H, s), 7.64-7.62 (2H, d, *J* = 8.2 Hz), 7.49 (5H, s), 7.37-7.32 (1H, d, *J* = 8.2 Hz), 7.31(1H, s), 7.16-7.12(1H, t, *J* = 8.2 Hz); ESI (MS): *m/z* 268[M+1].

Synthesis of (Z)-N-((2-chloroquinolin-3-yl)methylene)pyridin-3-amine (15b)

Yield: 68 %; yellowish brown solid; m.p.: 267 $^{\circ}$C; FT-IR (KBr) cm$^{-1:}$ 2999 (Ar-CH), 1667 (C=N), 1219 (C-N), 765 (C- Cl); ^{1}H-NMR (300 MHz, CF_3COOD-$CDCl_3$): δ 9.5 (1H, s), 8.32 (1H, s), 7.62-7.60 (1H, d, *J*= 7.2 Hz), 7.50-7.47 (2H, t, *J* = 7.2 Hz), 7.37 (3H, s), 7.31(1H, s), 7.15-7.12 (1H, t, *J* = 7.2 Hz); ESI (MS): *m/z* 268 [M+1].

Synthesis of (Z)-N-((2-chloroquinolin-3-yl)methylene)pyridin-4-amine(15c)

Yield: 70 %; pale yellow solid; m.p.: 297 $^{\circ}$C; FT-IR (KBr) cm^{-1}: 2880 (Ar-CH), 1668 (C=N), 1258 (C-N), 739 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$): δ 10.33 (1H, s), 8.32 (1H, s), 7.62-7.60 (1H, d, *J* = 7.2 Hz), 7.4-7.47 (2H, t, *J* = 7.2 Hz), 7.37 (3H, s), 7.31 (1H, s), 7.15-7.12 (1H, t, *J* = 7.2 Hz); ESI (MS) : *m/z* 268 [M+1].

(E)-5-bromo-N-((2-chloroquinolin-3-yl) methylene) pyridin-2-amine (15d)

Yield: 65 % ; pale yellow solid; m.p.: 218 $^{\circ}$C; FT-IR (KBr) cm^{-1}: 2970 (Ar-CH), 1668 (C=N), 1239 (C-N), 754 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 9.7 (1H, s), 8.2 (1H, s), 7.5-7.60 (1H, d, *J* = 7.2 Hz), 7.4-7.47 (2H, t, *J* = 7.2 Hz), 7.37 (3H, s), 7.31 (1H, s); ESI (MS): *m/z* 347 [M+1].

(E)-6-bromo-N-((2-chloroquinolin-3-yl) methylene) pyridin-3-amine (15e)

Yield 68 %; pale yellow solid; m.p.: 239 °C; FT-IR (KBr) cm^{-1}: 2998 (Ar-CH), 1668 (C=N), 1219 (C-N), 784 (C- Cl); ^{1}H-NMR (300 MHz, CF_3COOD-$CDCl_3$: δ 8.32 (1H, s), 7.64-7.62 (2H, d, *J* = 8.2 Hz), 7.49 (4H, s), 7.37-7.32 (1H, d, *J* = 8.2 Hz), 7.31(1H, s); ESI (MS): *m/z* 347 [M+1].

General procedure for synthesis of (E)-N-((2-chloro-6-methylquinolin-3-yl) methylene) pyridin-2-amine (16a-16e).

6-methyl 2-chloro-3-formylquinoline, 2-Aminopyriridine, and 0.94 gm (0.01 mol) of 6-methyl-2-chloro-3-formylquinoline were dissolved in ethanol and catalytic amounts of sulphuric acid were added to a 100 ml three-necked round-bottomed flask. After that, it was allowed to reflux for 4 hours while TLC kept an eye on it. After the reaction was complete, the mixture was dumped into ice, and the solid mass that was separated off was filtered, washed with water several times, dried, and recrystallized with ethanol to get the desired product.

(E)-N-((2-chloro-6-methylquinolin-3-yl) methylene) pyridin-2-amine (16a)

Yield: 78 %; pale yellow solid; m.p.: 257-259 °C, FT-IR (KBr) cm^{-1}: 2958 (Ar-CH), 1631 (C=N), 1090 (C-N), 778 (C- Cl); ESI (MS): *m/z* 282 [M+1].

(E)-N-((2-chloro-6-methylquinolin-3-yl) methylene) pyridin-3-amine (16b)

Yield; 75 %; Yellow solid, m.p.: 265-269°C, FT-IR (KBr) $cm^{-1:}$ 2922 (Ar-CH), 1621 (C=N), 1205 (C-N), 787 (C- Cl); ESI (MS): *m/z* 282 [M+1].

(E)-N-((2-chloro-6-methylquinolin-3-yl) methylene) pyridin-4-amine (16c)

Yield: 77 %; pale yellow solid, m.p.: 319-324 °C; FT-IR (KBr) cm^{-1} : 2869 (Ar-CH), 1660 (C=N), 1128 (C-N), 778 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 10.33 (1H, s), 8.32 (1H, s), 7.4-7.47 (2H, t, *J* = 7.2 Hz), 7.37 (3H, s), 7.31(1H, s), 7.15-7.12 (1H, t, *J* = 7.2 Hz), 2.9 (3H, s); ESI (MS) : *m/z* 282 [M+1].

(E)-5-bromo-N-((2-chloro-6-methylquinolin-3-yl) methylene) pyridin-2-amine (16d)

Yield: 75 %; pale yellow solid; m.p.: 272-276 °C; FT-IR (KBr) cm^{-1}: 2934 (Ar-CH), 1640 (C=N), 1198(C-N), 788 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$): δ 10.33 (1H, s), 8.32 (1H, s), 7.4-7.47 (2H, t, *J* = 7.2 Hz), 7.37 (2H, d), 7.31 (1H, s), 7.15-7.12 (1H, t, *J* = 7.2 Hz), 2.9 (3H, s); ESI (MS): *m/z* 361 [M+1].

(E)-6-bromo-N-((2-chloro-6-methylquinolin-3-yl) methylene) pyridin-3-amine (16e)

Yield: 78 %; pale yellow solid; m.p.: 296-299 °C; FT-IR (KBr) cm^{-1} : 2899 (Ar-CH), 1605 (C=N), 1156(C-N), (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 9.8 (1H, s), 8.2 (1H, s), 7.4-7.47 (2H, t, *J* = 7.2 Hz), 7.37 (2H, d), 7.41(2H, d), 7.15-7.12(1H, t, *J* = 7.2 Hz; ESI (MS): *m/z* 361 [M+1].

General procedure for synthesis of (E)-N-((2-chloro-8-methylquinolin-3-yl) methylene) pyridin-2-amine (17a-17e)

. 8-methyl 2-chloro-3-formylquinoline, 0.94 g of 2-Aminopyridine, and 2.05 g of this compound (0.01 mol) were dissolved in ethanol and catalytic amounts of sulphuric acid were added to a 100 ml three necked round bottomed flask. TLC continued to observe it as it refluxed over the following four hours. The required product was obtained by pouring the reaction mixture into ice once it had finished reacting. The solid mass that had been separated off was then filtered, thoroughly washed with water, dried, and recrystallized with ethanol.

(E)-N-((2-chloro-8-methylquinolin-3-yl) methylene) pyridin-2-amine (17a)

Yield: 78 %; pale yellow solid; m.p.: 296-299 °C; FT-IR (KBr) cm^{-1}: 2925 (Ar-CH), 1621 (C=N), 1090 (C- N), 788 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 9.9 (1H, s), 8.9 (1H, s), 7.4-7.47 (2H, t, *J* = 7.2 Hz), 7.37 (3H, s), 7.31(1H, s), 7.15-7.12(1H, t, *J* = 7.2 Hz), 2.9 (3H, s); ESI (MS): *m/z* 282 [M+1].

(E)-N-((2-chloro-8-methylquinolin-3-yl) methylene) pyridin-3-amine (17b)

Yield: 78 %; pale yellow solid; m.p.: 296-299 °C; FT-IR (KBr) cm^{-1}: 2925 (Ar-CH), 1621 (C=N), 1090 (C-N), 788 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 10.33 (1H, s), 8.1 (1H, s), 7.4-7.47 (2H, t, *J* =7.2 Hz), 7.37 (3H, s), 7.31(1H, s), 6.95(1H, t, *J* = 7.2 Hz), 3.1(3H, s); ESI (MS): *m/z* 282 [M+1].

.

(E)-N-((2-chloro-8-methylquinolin-3-yl) methylene) pyridin-4-amine (17c)

Yield: 75 %; pale yellow solid; m.p.: 312-315 °C; FT-IR (KBr) cm^{-1}: 2909 (Ar-CH), 1615 (C=N), 1201(C- N), 802 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 10.2 (1H, s), 8.32 (1H, s), 7.5 (2H, t, *J* = 7.2 Hz), 7.2(3H, s), 7.19 (1H, s), 7.15-7.12(1H, t, *J* = 7.2 Hz),3.1(3H,s); ESI (MS): *m/z* 282 [M+1].

(E)-5-bromo-N-((2-chloro-8-methylquinolin-3-yl) methylene) pyridin-2-amine (17d)

Yield: 75 %; pale yellow solid; m.p.: 276-278°C °C; FT-IR (KBr) cm^{-1}: 2890 (Ar-CH), 1596 (C=N), 1198 (C-N) 766(C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 10.1 (1H, s), 8.2 (1H, s), 7.8 (2H, t, *J* = 7.2 Hz), 7.37 (2H, d), 7.41(2H, d), 7.15-7.12 (1H, t, *J* = 7.2 Hz),3 (3H, s); ESI (MS) : *m/z* 361 [M+1].

(E)-6-bromo-N-((2-chloro-8-methylquinolin-3-yl) methylene) pyridin-3-amine (17e)

Yield: 75 %; pale yellow solid; m.p.: 262-266 °C; FT-IR (KBr) cm^{-1}: 2924 (Ar-CH), 1620 (C=N), 1198(C-N), 798 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$): δ 10.21 (1H, s), 8.5 (1H, s), 7.4-7.36 (2H, t, *J* = 7.2 Hz), 7.37 (2H, d), 7.31(1H, s), 7.12 (1H, t, *J* = 7.2 Hz), 3.05 (3H, s); ESI (MS): *m/z* 361 [M+1].

General procedure for synthesis of (E)-N-((2-chloro-6-methoxyquinolin-3-yl) methylene) pyridin-2-amine (18a-18e)

4-Aminopyridine and 6-methoxy-2-chloro-3-formylquinoline were dissolved in ethanol and catalytic amounts of sulphuric acid were added to a 100 ml three necked round bottomed flask. After that, it was allowed to reflux for 4 hours while TLC kept an eye on it. After the reaction was complete, the mixture was dumped into ice, and the solid mass that was separated off was filtered, washed with water several times, dried, and recrystallized with ethanol to get the desired product.

(E)-N-((2-chloro-6-methoxyquinolin-3-yl) methylene) pyridin-2-amine (18a)

Yield: 75 %; pale yellow solid; m.p.: 297-300 °C; FT-IR (KBr) cm^{-1}: 2963 (Ar-CH), 1720 (C=N), 1198(C-N), 793 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 10.9 (1H, s), 8.32 (1H, s), 7.5 (2H, t, *J* = 7.2 Hz), 7.2 (3H, s), 7.19 (1H, s), 7.15-7.12 (1H, t, *J* = 7.2 Hz),3.0 (3H, s); ESI (MS): *m/z* 298 [M+1].

(E)-N-((2-chloro-6-methoxyquinolin-3-yl) methylene) pyridin-3-amine (18b)

Yield: 79 %; yellow solid; m.p.: 275-281 °C; FT-IR (KBr) cm^{-1}: 2950 (Ar-CH), 1650 (C=N), 1198 (C-N), 788 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$): δ 9.9 (1H, s), 8.9 (1H, s), 7.4-7.47 (2H, t, *J* = 7.2 Hz), 7.37 (3H, s), 7.31 (1H, s), 7.15-7.12 (1H, t, *J* = 7.2 Hz), 2.9 (3H, s); ESI (MS): *m/z* 298 [M+1].

(E)-N-((2-chloro-6-methoxyquinolin-3-yl) methylene) pyridin-4-amine (18c)

Yield: 75 %; pale yellow solid; m.p.: 252-256 °C; FT-IR (KBr) cm^{-1}: 2956 (Ar-CH), 1620 (C=N), 1120 (C-N), 790 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$): δ 10.25 (1H, s), 8.4 (1H, s), 7.3 (2H, t, *J* = 7.2 Hz) 7.27 (3H, s), 7.31 (1H, s), 7.15-7.12 (1H, t, *J* = 7.2 Hz), 3.0 (3H, s); ESI (MS): *m/z* 298 [M+1].

(E)-5-bromo-N-((2-chloro-6-methoxyquinolin-3-yl) methylene) pyridin-2-amine (18d)

Yield: 81 %; pale yellow solid; m.p.: 289-292°C; FT-IR (KBr) cm^{-1}: 2900 (Ar-CH), 1623 (C=N), 1198(C-N), 780 (C- Cl) ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 10.21 (1H, s), 8.5 (1H, s), 7.4-7.36 (2H, t, *J* = 7.2 Hz), 7.37 (2H, d), 7.31(1H, s), 7.15-7.12(1H, t, *J* = 7.2 Hz), 3.05(3H, s); ESI (MS): *m/z* 377 [M+1].

(E)-6-bromo-N-((2-chloro-6-methoxyquinolin-3-yl) methylene) pyridin-3-amine (18e)

Yield: 75 %; Pale yellow solid; m.p.: 262-266 °C; FT-IR (KBr) cm^{-1}: 2924 (Ar-CH), 1620 (C=N), 1198 (C-N), 798 (C- Cl); ESI (MS): *m/z* 377 [M+1].

Synthesis of Isonicotinic acid hydrazide (INH) (19)

In a 100 ml three-necked round-bottomed flask, 4-Cyanopyridine was hydrolized at the C-4 position to create 4-pyridine carboxylic acid. The resultant product was then treated with hydrazine hydrate in the presence of NaOH and refluxed for 7 hours at 100 °C to produce isoniazide.

General procedure for synthesis of (E)-N'-((2-chloroquinolin-3-yl) methylene) isonicotinohydrazide (20a-20d)

2-Chloro-3-Formylquinolines and isoniazide were dissolved in ethanol and catalytic amounts of sulphuric acid were added to a 100 ml three necked round bottomed flask. After that, it was permitted to reflux for 4 hours while TLC kept an eye on it. After the reaction was complete, the reaction mixture was dumped into ice, and the solid mass that was separated off was filtered, washed with water several times, dried, and recrystallized with ethanol to get the desired product.

(E)-N'-((2-chloroquinolin-3-yl) methylene) isonicotinohydrazide (20a)

Yield: 65 %; Pale yellow solid; m.p.: 258 °C; FT-IR (KBr) cm^{-1}: 2988 (Ar-CH), 1668 (C=N), 1219 (C-N), 764 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 10.1 (1H, s), 8.32 (1H, s), 8.1 (1H, s),

7.62-7.60 (1H, d, *J* = 7.2 Hz), 7.4-7.47 (2H, t, *J* = 7.2 Hz), 7.37 (3H, s), 7.31 (1H, s), 7.15-7.12 (1H, t, *J* = 7.2 Hz); ESI (MS) : *m/z* 311 [M+1].

(E)-N'-((2-chloro-6-methylquinolin-3-yl) methylene) isonicotinohydrazide (20b)

Yield: 65 %; Pale yellow solid; m.p.: 245 °C; FT-IR (KBr) cm^{-1} : 2998 (Ar-CH), 1668 (C=N), 1219 (C-N), 764 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 10.1 (1H, s), 8.32 (1H, s), 8.1 (1H, s), 7.62-7.60 (1H, d, *J* = 7.2 Hz), 7.4-7.47 (2H, t, *J* = 7.2 Hz), 7.37 (3H, s), 7.31 (1H, s), 7.15-7.12 (1H, t, *J* = 7.2 Hz),3.1 (3H, s); ESI (MS) : *m/z* 325 [M+1].

(E)-N'-((2-chloro-8-methylquinolin-3-yl) methylene) isonicotinohydrazide (20c)

Yield: 65 %; pale yellow solid; m.p.: 245 °C; FT-IR (KBr) cm^{-1}: 2998 (Ar-CH), 1668 (C=N), 1219 (C-N), 764 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 10.2 (1H, s), 9.1 (1H, s), 8.1(1H,s), 7.62-7.60 (1H, d, *J* = 7.2 Hz), 7.4-7.47 (2H, t, *J* = 7.2 Hz), 7.37 (3H, s), 7.31 (1H, s), 7.15-7.12 (1H, t, *J* = 7.2 Hz), 3.1(3H, s).

(E)-N'-((2-chloro-6-methoxyquinolin-3-yl) methylene) isonicotinohydrazide (20d)

Yield: 65 %; pale yellow solid; m.p.: 245 °C; FT-IR (KBr) cm^{-1}: 2998 (Ar-CH), 1668 (C=N), 1219 (C-N), 764 (C- Cl); ^{1}H NMR (300 MHz, CF_3COOD-$CDCl_3$) : δ 10.2 (1H, s), 9.1 (1H, s), 8.1 (1H, s), 7.62-7.60 (1H, d, *J* = 7.2 Hz), 7.4-7.47 (2H, t, *J* = 7.2 Hz), 7.37 (3H, s), 7.31 (1H, s), 7.15-7.12 (1H, t, *J* = 7.2 Hz), 3.1 (3H, s).

Antimicrobial activity

Microbial strains

Bacillus subtilis, S. aureus (gramme +ve), E. coli, and P. vulgaris were among the microorganisms utilised in this investigation (gram -ve). The nutrient agar medium was used to cultivate the bacteria, which were then incubated for 24 hours at 37°C. The bactericidal activity of each synthetic substance was examined. Applying the Holder and Boyce's agar well diffusion assay technique. On nutrient agar medium, the tested organisms were subcultured (Oxoid Laboratories, U.K.). As a positive control for bacterial strains, ciprofloxacin was utilised. The plates were made in three copies. Bacterial cultures were cultured for 24 hours at 37°C. By measuring the zone of inhibition, antimicrobial activity was identified.

Determination of MIC

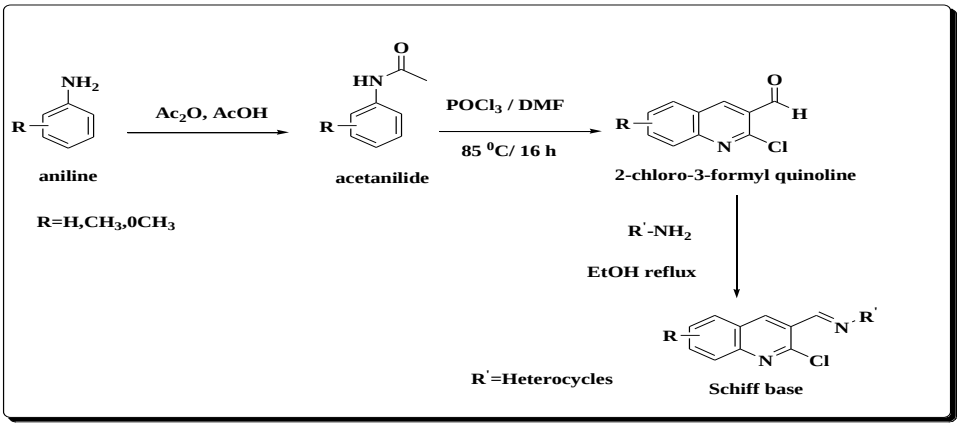
The compounds that showed positive antimicrobial activity against the majority of the microorganisms tested in the disc diffusion bioassay were further tested for the determination of minimum inhibitory concentration, even though the results of the disc diffusion assay cannot always be compared to the MIC data (Njenga et al., 2005). (MIC). For each of the examined species, the MIC of the synthesised samples was calculated in triplicates. Following the addition of nutrition broth and varying sample concentrations (10–100 um), a loopful of the test organism that had been diluted to 0.5 McFarland turbidity standard was added to the tubes. The test organisms were sown in a tube containing only broth media as a control. After that, test organism cultures were cultured in tubes for 24 hours at 37°C. The tubes were then checked for turbidity in order to look for growth.

Anti-tuberculosis activity

MTT Assay (anti- mycobacterial Activity)

Methanol was used to dissolve the chemicals. Using Mycobacterium smegmatis, the compounds were examined for their anti-mycobacterial activity. In triplicates, Mycobacterium smegmatis is grown in Middlebrook 7H9 media and seeded at 5x105 to 1x106 O.D. 600 with varying concentrations of the compounds. The culture is then incubated at 37 °C for 24 days. MTT (3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide) is then added, and the mixture is incubated for 4 hours at 37 °C. At 540 nm, the absorbance was measured while the formazan crystals were dispersed in DMSO.

RESULTS AND DISCUSSION



Scheme:I Synthesis of Quinoline-based Heterocyclic Schiff bases

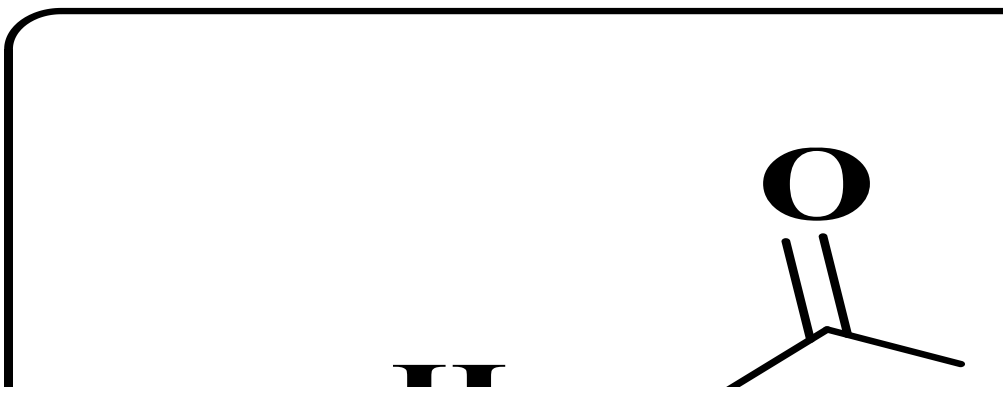
Step-1: Synthesis of Acetanilide

Different anilines were dissolved in glacial acetic acid and treated with acetic anhydride. This reaction mixture was poured into ice to obtain respective acetanilides. The compounds **13a**, **13b**, **13c**, **13d, 13e, 13f** and **13g** are known compounds and confirm by their melting points and IR spectra with authentic values were shown in Table-1.

Table-1: Melting points and IR spectral data of Compounds (13a-13g)

Compound	**C=O (cm^{-1})**	**C-N (cm^{-1})**	**C-X (cm^{-1})**	**N-H (cm^{-1})**	**Al-CH (cm^{-1})**	**Mass value** [m/z]	**% Yield**	**M.P ^{0}C (Lit M.P ^{0}C**
13a	1665	1315	_	3295	3021	135 [M^+1]	82	110 ^{0}C (113^0C)
13b	1690	1310	760	3350	3000	170 [M^+1]	78	178 ^{0}C (176 ^{0}C)
13c	1667	1206	1206	3306	2879	153 [M^+1]	75	180 ^{0}C (182 ^{0}C)
13d	1669	1311	_	3352	2912	150 [M^+1]	84	108 ^{0}C (110^0C)
13e	1671	1255	_	3321	3001	166 [M+1]	90	120^0C 125^0C
13f	1615	1296	_	3297	3025	150 [M^+1]	89	88 ^{0}C (90 ^{0}C)

Step-2: Synthesis of 2-Chloro-3-formylquinoline (14a-14d)



The 2-chloro-3-formylquinoline were prepared by using literature method [16]. Acetanilide undergoes cyclization with $POCl_3$ in DMF to afford corresponding 2-chloro-3-formylquinoline. The confirmation regarding the formation of compounds **14a, 14b, 14c,** & **14d** was obtained from IR, Mass and NMR spectroscopic methods.

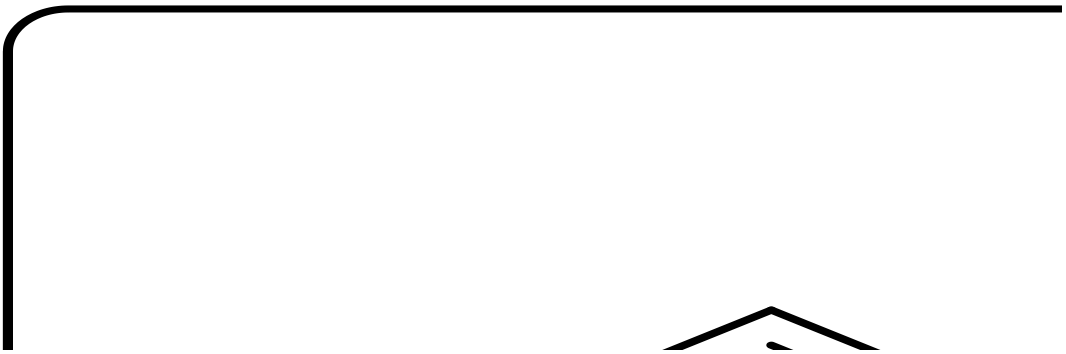
Step-3: Synthesis of 2-chloro 3-formylquinoline Schiff bases (15a-15e)

2-Chloro-3-formylquinolines was treated with different aminopyridine by using ethanol as a solvent and sulphuric acid as catalyst. Desired products were obtained in good yield. The compounds **15a, 15b, 15c, 15d & 15e** were confirmed by IR, Mass and NMR spectral data.

Step-3: Synthesis of 6-methyl 2-chloro-3-formylquinoline Schiff bases (16a-16e)

6-methyl-2-Chloro-3-formylquinolines was treated with different aminopyridine by using ethanol as a solvent and sulphuric acid as catalyst. Desired products were obtained in good yield. The compounds **16a, 16b, 16c, 16d & 16e** were obtained was characterized by their IR, Mass and NMR spectral data.

Step-3: Synthesis of 8-methyl 2-chloro 3-formyl quinoline Schiff bases (17a-e)



8-methyl-2-Chloro-3-formylquinolines was treated with different aminopyridine by using ethanol as a solvent and sulphuric acid as catalyst. Desired products were obtained in good yield. The compounds **17a, 17b, 17c, 17d & 17e** were characterized by their IR, Mass and NMR spectral data.

Step-3: Synthesis of 6-methoxy-2-chloro 3-formylquinoline Schiff bases (18a-e)

6-methoxy-2-Chloro-3-formylquinolines was treated with different aminopyridine by using ethanol as a solvent and sulphuric acid as catalyst. Desired products were obtained in good yield. The compounds **18a, 18b, 18c, 18d & 18e** were confirmed by their IR, Mass and NMR spectral data. Characteristics peaks are shown in Table-6.

Step-4: Synthesis of Isonicotinic acid hydrazide (INH) (19)

4-Cyanopyridine was hydrolized at C-4 position to form 4-pyridine carboxylic acid; the resulting product was treated with hydrazine hydrate in presence of NaOH and refluxed for 7 h at 100 ^{0}C to obtain Isoniazide. The product is confirmed by its IR, NMR and mass sprecal analysis. The mass spectra is showing the following fragments pattern ESI-MS m/z [M+1] =138.

Step-5: Synthesis of 2-chloro-3-formylquinoline with INH linked Hydrazones (20a-20d)

2-Chloro-3-formylquinolines was treated with INH (19) by using ethanol as a solvent and sulphuric acid as catalyst. Desired products were obtained in good yield. The compounds **20a, 20b, 20c, 20d & 20e** were confirmed by their IR, Mass and NMR spectral data.

Biological Activity

Antimicrobial activity

Few of the newly synthesized compounds were examined for their in vitro antibacterial activity by using pour plate method with two gram +ve bacteria i.e, *Bacillus subtilis* & *S. aureus* and two gram -ve bacteria i.e., *E. Coli* & *P. vulgaris*. Ciprofloxacin were used in assay as a standard control drug. DMSO was used as diluents which is ineffective to the growth of microbes. The antimicrobial activity was tested at 50 µM concentration.

Table-2: Antimicrobial activity Diameter of Zone of inhibition (cm)

S. No	Comp. no.	Control (DMSO) (5%)	Standard (Ciprofloxacin) (5µg)	Concentration of samples	Gram (+ve)		Gram (-ve)	
					Bacillus Subtilis	*S. aureus*	*E. Coli*	*P. vulgaris*
1	20 b	-	31	50 µM	13	13	16	13
2	15d	-	31	50 µM	25	31	27	29
3	16d	-	31	50 µM	25	26	29	20
4	15e	-	31	50 µM	26	28	28	28
5	16c	-	31	50 µM	13	16	11	18

Table-3: Minimum Inhibitory Concentration (MIC) data for active compounds

Compound	*E. Coli*	*B. subtilis*	*P. vulguris*	*S. aureus*
15d	20 µM	20 µM	20 µM	20 µM
16d	30 µM	20 µM	30 µM	20 µM
15e	50 µM	30 µM	50 µM	50 µM

Antibacterial activity

The antibacterial activity of newly synthesised Schiff bases **20b, 15d, 16d, 15e** and **16c** were tested against Gram-positive (*Streptococcus pneumonia*, *Bacillis subtilis*) and Gram-negative (*Pseudomonas aeruginosa*, *Escherichia coli*) bacteria and results are summarised in the Table 2 and 3. The results were compared with antibacterial activity of reference drug Ciprofloxacin. All the tested compounds

20b, 15d, 16d, 15e and **16c** demonstrated high inhibition against the tested Gram-positive microorganisms (*S. pneumonia* and *B. subtilis*) (inhibition zones varied from 11.0–30.2 mm when compared with Ampicillin), and against the Gram-negative *P. aeruginosa* bacteria (inhibition zones varied from 10.2–20.3 mm). The SAR shows that the compound **15d** (Schiff base of 2-chloro-3-formylquinoline with 2-amino-5-bromopyridine) is found to be as active as standard drug ciprofloxacin against *S. aureus*. The compounds **16d** & **15e** are showing good activity against *S. aureus* where as the compounds **16c** and **20b** are moderately active. In the case of *B. Subtilis* the **15d**, **15e** & **16d** are exhibiting good activity where as the compounds **16c** & **20b** are moderately active. Similar trends were obtained when these compounds were tested against gram (-) bacteria for example: In the case of *E .coli* the **16d** is exhibiting highest followed by **15e**, **15d**. The compounds **16c** & **20b** are comparatively less active. The compound 15 d is found to be most active against *P. vulgaris* followed by 15e. The compounds 16d, 16e and 20b are found to be lest active against *P. vulgaris*.

The minimum inhibitory concentration results (MIC) values were determined for most active compounds **15d**, **16d** & **15e**. As per the MIC results it is found that the compound 15d is showing an MIC of 20 μM against all four microbes i.e., Gram-positive (*Streptococcus pneumonia*, *Bacillis subtilis*) and Gram-negative (*P. aeruginosa*, *Escherichia coli*) bacteria. The compound 16d is an MIC of 20 μM against *B. subtilis* & *S. Aureus*. While against *E. Coli* & *P. vulguris* it is showing an MIC of 30 μM. The compound 15e is showing a MIC of 30 μM against *S. subtilis*. Where as in the case of *S. Aureus, E. Coli* & *P.* vulguris it is showing an MIC of 50 μM.

Anti-tuberculosis activity

MTT Assay (Anti- Mycobacterial Activity):

Few newly synthesised compounds were tested for their anti-tuberculosis activity by using MTT Assay methods. The results show that the compounds16d is found to be exhibiting extremely good anti-tuberculosis activity as its give 91.67426 % of death at a concentration of 12.5 μg/ml. The compound 15e showing an 85.47554 % of death, whereas the compound 15d is exhibiting a 73.77697 % of death. The compound 16c is displaying least activity amongst all the above tested compounds (62.57976% of death).

Table-4: Anti-tuberculosis activity by using MTT Assay methods

Name of compound	Conc.	% of Death
15d	12.5 µg/ml	73.77697
16d	12.5 µg/ml	**91.67426**
15e	12.5 µg/ml	**85.47554**
16c	12.5 µg/ml	62.57976

CONCLUSION

In view of the biological importance of quinoline derivatives the design and synthesis of quinoline incorporated heterocyclic Schiff's bases is undertaken. The newly synthesised compounds were tested for their anti tuberculosis & antimicrobial activity. Synthesis of various substituted quinoline based Schiff's bases have been performed using reported synthetic procedures. The compounds were confirmed by their melting points, ^{1}H-NMR, infrared (IR) & Mass spectra. The primarily biological screening results shows that compound **15d** showed highest activity against *S. auresus* and will be emerge as potential anti-microbial agent. The compound **16d** displayed the highest anti-tuberculosis activity and presumably will be a potential candidate for further biological studies.

ACKNOWLEDGEMENT

We are Greatful to the management of Anurag Pharmacy College, Kodad for providing facilities to carryout synthesis, *in vitro* anti-microbial and anti-tuberculosis activity. Thanks to NIPER, Hyderabad for providing Mass and ^{1}HNMR spectral data.

CONFLICT OF INTEREST

No conflict of interests regarding the publication of this article.

REFERENCES

1. H. Schiff. Mittheilungen aus dem Universitätslaboratorium in Pisa: Eine neue Reihe organischer Basen. Justus Liebigs Justus Liebigs Annalen der Chemie, 1864; 131: 118-119. DOI: 10.1002/jlac.18641310113.
2. D.N. Dhar and C.L. Taploo. Schiff bases and their applications. Journal of Scientific and Industrial Research 198); 41: 501-506. DOI: 10.22270/ajprd.v8i5.837.
3. B. S. Sathe, E. Jaychandran, V. A. Jagtap, and G. M. Sreenivasa. Synthesis Characterization and Anti-Inflammatory Evaluation of New Fluorobenzothiazole Schiff's Bases. International Journal of Pharmaceutical Research and Development. 2011; 3:164–169. DOI: 10.1155/2013/893512.
4. S. M. Sondhi, N. Singh, A. Kumar, O. Lozach, and L. Meijer. Synthesis, anti-inflammatory, analgesic and kinase (CDK-1, CDK-5 and GSK-3) inhibition activity evaluation of benzimidazole/benzoxazole derivatives and some Schiff's bases. Bioorganic and Medicinal Chemistry. 2006; 14: 3758–3765. DOI: 10.1016/j.bmc.2006.01.054.
5. A. Pandey, D. Dewangan, S. Verma, A. Mishra, and R. D. Dubey. Synthesis of Schiff Bases of 2-Amino-5-aryl-1,3,4-thiadiazole and Its Analgesic, Anti-Inflammatory, Anti-Bacterial and Anti-Tubercular Activity. International Journal of ChemTech Research, 2011; 3: 178–184. DOI: 10.1155/2012/145028.
6. C. Chandramouli, M. R. Shivanand, T. B. Nayanbhai, B. Bheemachari, and R. H. Udupi. Synthesis and Biological Screening of Certain New Triazole Schiff Bases and Their Derivatives Bearing Substituted Benzothiazole Moiety. Journal of Chemical and Pharmaceutical Research, 2012; 4: 1151–1159. DOI: 10.4236/ijoc.2020.101001.
7. R. P. Chinnasamy, R. Sundararajan, and S. Govindaraj. Synthesis, characterization, and analgesic activity of novel schiff base of isatin derivatives. Journal of Advanced Pharmaceutical Technology and Research, 2010; 1: 342–347. DOI: 10.4103/0110-5558.72428.
8. K. Mounika, B. Anupama, J. Pragathi, and C. Gyanakumari. Synthesis Characterization and Biological Activity of a Schiff Base Derived from 3-Ethoxy Salicylaldehyde and 2-Amino Benzoic acid and its Transition Metal Complexes. Journal of Scientific Research. 2010; 2: 513–524. DOI: 10.4236/ijoc.2014.41002.

9. P. Venkatesh. Synthesis, Characterization and Antimicrobial Activity of Various Schiff Bases Complexes of Zn (II) and Cu (II) Ions. Asian Journal of Pharmaceutical and Health Sciences 2011; 1: 8–11. DOI: 10.1016/j.rechem.2019.100006.

10. T. Aboul-Fadl, F. A. Mohammed, and E. A. Hassan, Synthesis, antitubercular activity and pharmacokinetic studies of some schiff bases derived from 1- alkylisatin and isonicotinic acid hydrazide (inh). Archives of Pharmacal Research, 2003; 26: 778–784. DOI: 10.1007/BF02980020.

11. R. Miri, N. Razzaghi-asl, and M. K. Mohammadi. QM Study and Conformational Analysis of an Isatin Schiff Base as a Potential Cytotoxic Agent. Journal of Molecular Modeling. Journal of Molecular Modeling. 2013; 19: 727–735.
DOI: 10.1007/s00894-012-1586-x.
12. V. Rajashakar, K. Saisree, M. Sikender, S. Naveen, B. Madhava Reddy and V. Harinadha Babu Synthesis of Pyrazolyl Thiobarbituric Acids and their Cytotoxic and Antimicrobial Evaluation. Asian Journal of Organic & Medicinal Chemistry. 2016; 1: 83–86. DOI: 10.14233/ajomc.2016.AJOMC-P23.
13. Pretorius SI, Breytenbach WJ, de Kock C Smith PJ. Synthesis, characterization and antimalarial activity of quinoline-pyrimidine hybrids. Bioorg Med Chem. 2013; 21: 269-77. DOI: 10.3390 molecules22122268.
14. Yeh-Long C, Hsien-Ming H, Chih-Ming L , Kuang- Chieh L, Cherng-Chyi T. Design, synthesis, and biological evaluation of new quinoline-based heterocyclic derivatives as novel antibacterial agents. Bio Org Med Chem. 2004; 12: 6539 DOI: 10.1007/s00706-020-02686-3.
15. Lilienkampf A, Mao J, Wan B, Wang Y, Franzblau SG, Kozikowski AP. Structure-activity relationships for a series of quinoline-based compounds active against replicating and nonreplicating Mycobacterium tuberculosis. J Med Chem. 2009; 52:2109.
DOI: 10.1021/jm900003c.
16. P. G. Avaji, C. H. Vinod Kumar, S. A. Patil, K. N. Shivananda, and C. Nagaraju. Synthesis, Spectral Characterization, In-Vitro Microbiological Evaluation and Cytotoxic Activities of Novel Macrocyclic Bishydrazone. European Journal of Medicinal Chemistry. 2009; 44: 3552–3559. DOI:10.1016/j.ejmech.2009.03.032.