Journal of Saudi Chemical Society (2016) **20**, S606–S614

King Saud University

Journal of Saudi Chemical Society

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ORIGINAL ARTICLE

Synthesis of novel (*E*)-*N*′-(2-chloropyrimidin-4-yl)-*N*-(5-cyano-2-hydroxy-6-phenylpyrimidin-4-yl) formamidine derivatives and their antimicrobial activity

C. Mallikarjunaswamy [a], **D.G. Bhadregowda** [a,*], **L. Mallesha** [b]

[a] *Department of Chemistry, Yuvaraja's College, University of Mysore, Mysore 570005, India*
[b] *PG Department of Chemistry, JSS College of Arts, Commerce and Science, Ooty Road, Mysore 570025, India*

Received 28 January 2013; accepted 18 April 2013
Available online 7 May 2013

KEYWORDS

Pyrimidines;
Schiff bases;
Antimicrobial activity;
Antifungal activity

Abstract A series of novel (*E*)-*N*′-(2-chloropyrimidin-4-yl)-*N*-(5-cyano-2-hydroxy-6-phenylpyrimidin-4-yl) formamidine derivatives were synthesized by the reaction of different aldehydes with 2-chloropyrimidin-4-amine and *in vitro* antimicrobial activity was evaluated. The synthesized compounds were characterized by elemental analyses, FT-IR, ^{1}H NMR and LC–MS spectral studies. Antimicrobial data revealed that among all the compounds screened, compounds **7l** and **7m** were found to have promising antimicrobial activity against all the selected pathogenic bacteria and fungi.

1. Introduction

Heterocyclic molecules are of biological interest due to their potential physical and chemical properties (Brown, 1998). Among these, the pyrimidine compounds occupy a unique position in pharmaceutical chemistry, as they are components of nucleic acids. Nucleosides are the most frequently used effective class of antiviral agents, with over twenty drugs currently approved for the treatment of viral diseases and a number of candidates in the clinical trials (Mansour and Storer 1997; Bergman et al., 2004). Consequently, the intense search for new nucleoside derivatives attracted extensive attention. Being bioactive molecules, pyrimidines are important components of the biological macromolecules, such as DNA and RNA. So introducing the pyrimidines into nucleoside derivatives may result in the discovery of a number of novel derivatives with potential antitumor and antiviral activities. The explosion of new approaches for their synthesis and most importantly, their selective synthesis is an interesting subject of organic and bioorganic chemistry.

The treatment of bacterial infections remains a challenging therapeutic problem because of emerging infectious diseases and the increasing number of multidrug-resistant microbial pathogens. Despite many antibiotics and chemotherapeutics available, the emergence of old and new antibiotic-resistant bacterial strains in the last decades leads to a substantial need

* Corresponding author. Tel.: +91 9449264945.
E-mail address: dgbycm@gmail.com (D.G. Bhadregowda).
Peer review under responsibility of King Saud University.

Production and hosting by Elsevier

http://dx.doi.org/10.1016/j.jscs.2013.04.005

for new classes of anti-bacterial agents (Inca et al., 2006). Pyrimidine and their derivatives are continuously attracting attention of the medicinal chemists in view of their profound range of biological activities, anti-HIV (Rawal et al., 2007), analgesic agents (Chhabria et al., 2007), antiproliferative (Schenone et al., 2008) and antitumor (Abbas et al., 2011). The pyrimidine nucleus has increasingly attracted the attention of synthetic chemists. Though the antimicrobial activity of pyrimidine derivatives has been extensively studied and well documented in the literature (Holla et al., 2006; Chambhare et al., 2003; Narayana et al., 2009), however, relatively few reports were available on the anti-inflammatory activity of the pyrimidine derivatives (Ramesh et al., 2010; Ballesteros et al., 1995; O'Hare et al., 2011; Shishoo et al., 1999; Papadakis and Targan, 2000). In connection with such studies, the present paper reported on the synthesis, antibacterial and antifungal activities of (*E*)-*N*′-(2-chloropyrimidin-4-yl)-*N*-(5-cyano-2-hydroxy-6-phenylpyrimidin-4-yl) formamidine derivatives **7a–n**.

2. Materials and methods

2.1. General

All solvents and reagents were purchased from Sigma Aldrich Chemicals. Melting points were determined on an electrically heated VMP-III melting point apparatus. The elemental analyses of the compounds were performed on a Perkin Elmer 2400 Elemental Analyzer. The FT-IR spectra were recorded using KBr discs on a FT-IR 4100 Infrared spectrophotometer. The NMR spectra were recorded using a Bruker DRX 400 spectrometer at 400 MHz for ^{1}H NMR with tetramethylsilane as the internal standard. Mass spectral data were obtained by LC/MSD Trap XCT. Silica gel for column chromatography was performed using Merck 7734 silica gel and Merck-made TLC plates. (*E*)-*N*′-(2-chloropyrimidin-4-yl)-*N*-(5-cyano-2-hydroxy-6-phenylpyrimidin-4-yl) formamidine derivatives **7a–n,** were prepared by the method summarized in Scheme 1.

2.2. Chemistry

2.2.1. General method for the synthesis of 4-amino-2-hydroxy-6-phenylpyrimidine-5-carbonitrile derivatives (*4a–n*)

The reaction of aromatic aldehyde **1a–n** (10 mmol), malononitrile (10 mmol) and phosphorus pentoxide (3.54 mmol) is stirred mechanically for 10 minutes in 25 ml absolute ethanol and then thiourea (20 mmol) is added and mixed thoroughly. The resulting reaction mixture was heated at reflux and it was poured on the crushed ice (about 200 gm) after the completion of the reaction monitored by TLC. The solid was filtered, washed with petroleum ether, dried and recrystallized by using ethanol.

2.2.2. General procedure for the synthesis of N-(5-cyano-2-hydroxy-6-phenylpyrimidin-4-yl) formamidine derivatives (*5a–n*)

A solution of compound **4a–n** (5.0 mmol) in formic acid (20 mL) was refluxed for 1 h. The solvent was evaporated and the residue was crystallized from ethanol to give compound **5a–n** in good yield.

2.2.3. General procedure for the synthesis of (*E*)-*N*′-(2-chloropyrimidin-4-yl)-*N*-(5-cyano-2-hydroxy-6-phenylpyrimidin-4-yl) formamidine derivatives (*7a–n*)

The Schiff base was prepared by reaction of equimole of **5a–n** and 2-chloropyrimidin-4-amine. Each reactant was dissolved in a minimum amount of ethanol, then mixed together and followed by addition of 2 ml glacial acetic acid. The solution was refluxed for 8 h then cooled to room temperature and poured into ice cold water. The solid product was collected through filtration and then dried using drying oven at 80 °C. The product was redissolved in ethanol for recrystallization and then dried to give a product.

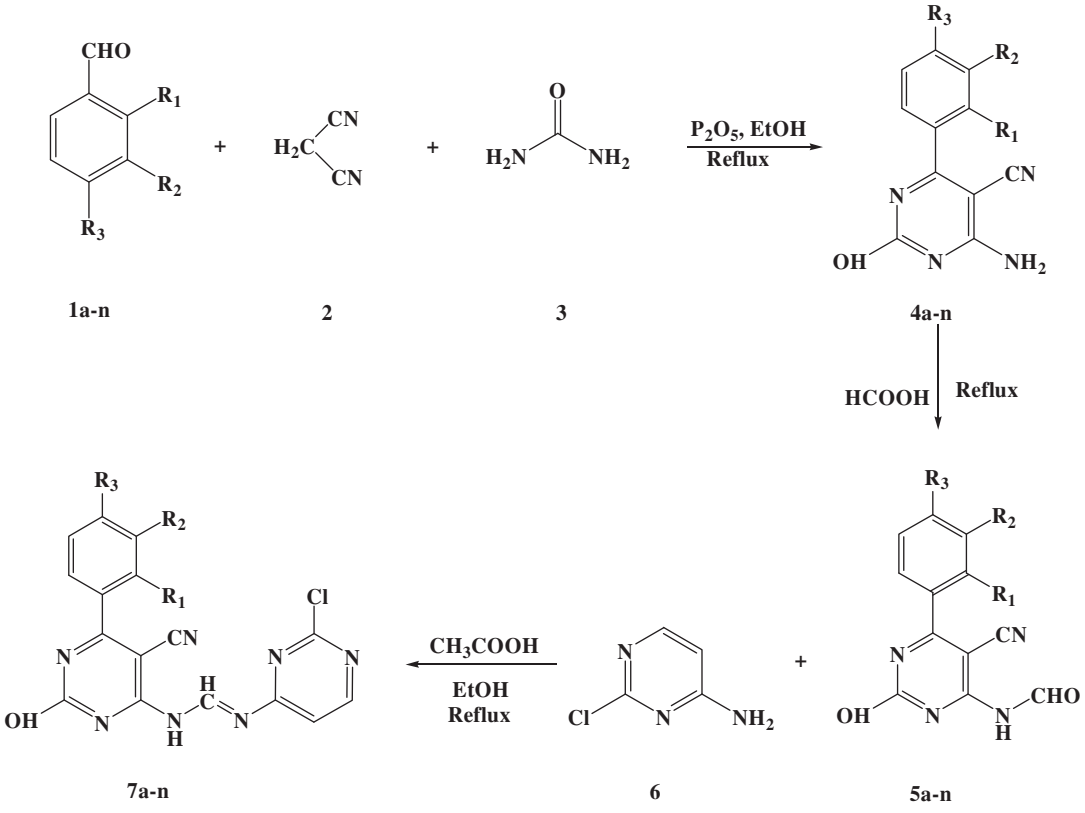
*2.2.3.1. (E)-N′-(2-chloropyrimidin-4-yl)-N-(5-cyano-2-hydroxy-6-phenylpyrimidin-4-yl)formamidine (**7a**).* Recrystallization from ethanol afforded 80. FT-IR (KBr, cm^{-1}) *v*: 3560 (O–H), 3271 (N–H), 2220 (CN), 1697 (C=N). ^{1}H NMR (DMSO-d$_6$, 400 MHz) *δ*: 7.15–7.57 (m, 5H, Ar–H), 7.70–7.91 (d, 2H. Pyrimidine), 8.54 (s, 1H, N=CH), 8.93 (s, 1H, NH), 10.67 (bs, 1H, OH). MS (ESI) *m*/*z*: 352. Anal. Calcd. For $C_{16}H_{10}ClN_7O$ (in%): C, 54.63; H, 2.87; N, 27.87. Found. C, 54.61; H, 2.82; N, 27.95.

*2.2.3.2. (E)-N-(6-(4-chlorophenyl)-5-cyano-2-hydroxypyrimidin-4-yl)-N′-(2-chloropyrimidin-4-yl) formamidine (**7b**).* Recrystallization from ethanol afforded 82%. FT-IR (KBr, cm^{-1}) *v*: 3563 (O–H), 3276 (N–H), 2226 (CN), 1694 (C=N). ^{1}H NMR (DMSO-d$_6$, 400 MHz) *δ*: 7.31–7.51 (m, 4H, Ar–H), 7.72–7.93 (d, 2H. Pyrimidine), 8.52 (s, 1H N=CH), 8.92 (s, 1H NH), 10.67 (bs, 1H OH). MS (ESI) *m*/*z*: 386. Anal. Calcd. For $C_{16}H_9Cl_2N_7O$ (in%): C, 49.76; H, 2.35; N, 25.39. Found C, 49.71; H, 2.36; N, 25.32.

*2.2.3.3. (E)-N-(6-(4-bromophenyl)-5-cyano-2-hydroxypyrimidin-4-yl)-N′-(2-chloropyrimidin-4-yl) formamidine (**7c**).* Recrystallization from ethanol afforded 70%. FT-IR (KBr, cm^{-1}) *v*: 3566 (O–H), 3269 (N–H), 2219 (CN), 1690 (C=N). ^{1}H NMR (DMSO-d$_6$, 400 MHz) *δ*: 7.32–7.44 (m, 4H, Ar–H), 7.74–7.91 (d, 2H. Pyrimidine), 8.57 (s, 1H N=CH), 8.94 (s, 1H NH), 10.65 (bs, 1H OH). MS (ESI) *m*/*z*: 431. Anal. Calcd. For $C_{16}H_9BrClN_7O$ (in%): C, 44.62; H, 2.11; N, 22.77. Found C, 44.69; H, 2.15; N, 22.71.

*2.2.3.4. (E)-N′-(2-chloropyrimidin-4-yl)-N-(5-cyano-2-hydroxy-6-o-tolylpyrimidin-4-yl)formamidine (**7d**).* Recrystallization from ethanol afforded 78%. FT-IR (KBr, cm^{-1}) *v*: 3569 (O–H), 3271 (N–H), 2221 (CN), 1692 (C=N). ^{1}H NMR (DMSO-d$_6$, 400 MHz) *δ*: 2.50 (s, 3H, Ar–CH$_3$), 7.33–7.48 (m, 4H, Ar–H), 7.73–7.95 (d, 2H. Pyrimidine), 8.46 1H N=CH), (s, 8.95 (s, 1H NH), 10.67 (bs, 1H OH). MS (ESI) *m*/*z*: 366. Anal. Calcd. For $C_{17}H_{12}ClN_7O$ (in%): C, 55.82; H, 3.31; N, 26.81. Found C, 55.87; H, 3.36; N, 26.85.

*2.2.3.5. (E)-N-(6-(3-chlorophenyl)-5-cyano-2-hydroxypyrimidin-4-yl)-N′-(2-chloropyrimidin-4-yl) formamidine (**7e**).* Recrystallization from ethanol afforded 83%. FT-IR (KBr, cm^{-1}) *v*: 3560 (O–H), 3279 (N–H), 2218 (CN), 1695 (C=N). ^{1}H NMR (DMSO-d$_6$, 400 MHz) *δ*: 7.35–7.51 (m, 4H, Ar–H), 7.71–7.92 (d, 2H. Pyrimidine), 8.49 (s, 1H N=CH), 8.91 (s, 1H NH), 10.67 (bs, 1H OH). MS (ESI) *m*/*z*: 386. Anal. Calcd. For $C_{16}H_9Cl_2N_7O$ (in%): C, 49.76; H, 2.35; N, 25.39. Found C, 49.71; H, 2.38; N, 25.32.



a: $R_1=R_2=R_3=H$

b: $R_1=R_2=H$, $R_3=Cl$

c: $R_1=R_2=H$, $R_3=Br$

d: $R_1=R_2=H$, $R_3=CH_3$

e: $R_1=R_3=H$, $R_2=Cl$

f: $R_1=R_3=H$, $R_2=Br$

g: $R_1=R_3=H$, $R_2=CH_3$

h: $R_1=R_2=H$, $R_3=OCH_3$

i: $R_1=R_2=R_3=CH_3$

j: $R_1=H$, $R_2=R_3=CH_3$

k: $R_2=H$, $R_1=R_3=CH_3$

l: $R_2=H$, $R_1=Cl$, $R_3=Br$

m: $R_1=R_2=H$, $R_3=NO_2$

n: $R_2=H$, $R_1=R_3=OCH_3$

Scheme 1 Synthesis of intermediates and title compounds.

*2.2.3.6. (E)-N-(6-(3-bromophenyl)-5-cyano-2-hydroxypyrimidin-4-yl)-N′-(2-chloropyrimidin-4-yl) formamidine (**7f**).* Recrystallization from ethanol afforded 75%. FT-IR (KBr, cm^{-1}) *v*: 3569(O–H), 3273 (N–H), 2223 (CN), 1699 (C═N). ^1H NMR (DMSO-d_6, 400 MHz) *δ*: 7.32–7.46 (m, 4H, Ar–H), 7.70–7.92 (d, 2H. Pyrimidine), 8.49 (s, 1H N═CH), 8.95 (s, 1H NH), 10.67 (bs, 1H OH). MS (ESI) *m*/*z*: 431. Anal. Calcd. For $C_{16}H_9BrClN_7O$ (in%): C, 44.62; H, 2.11; N, 22.77. Found C, 44.66; H, 2.15; N, 22.72.

*2.2.3.7. (E)-N′-(2-chloropyrimidin-4-yl)-N-(5-cyano-2-hydroxy-6-m-tolylpyrimidin-4-yl)formamidine (**7g**).* Recrystallization from ethanol afforded 70%. FT-IR (KBr, cm^{-1}) *v*: 3562 (O–H), 3279 (N–H), 2227 (CN), 1692 (C═N). ^1H NMR (DMSO-d_6, 400 MHz) *δ*: 2.51 (s, 3H, Ar–CH_3), 7.35–7.51 (m, 4H, Ar–H), 7.71–7.95 (d, 2H. Pyrimidine), 8.50 (s, 1H N═CH), 8.94 (s, 1H NH), 10.67 (bs, 1H OH). MS (ESI) *m*/*z*: 366. Anal. Calcd. For $C_{17}H_{12}ClN_7O$ (in%): C, 55.82; H, 3.31; N, 26.81. Found C, 55.86; H, 3.37; N, 26.84.

*2.2.3.8. (E)-N′-(2-chloropyrimidin-4-yl)-N-(5-cyano-2-hydroxy-6-(4-methoxyphenyl)pyrimidin-4-yl) formamidine (**7h**).* Recrystallization from ethanol afforded 76%, mp: 205–207 °C. FT-IR (KBr, cm^{-1}) *v*: 3560 (O–H), 3278 (N–H), 2228 (CN), 1693(C═N). ^1H NMR (DMSO-d_6, 400 MHz) *δ*: 2.20 (s, 3H, Ar–CH_3), 7.32–7.52 (m, 4H, Ar–H), 7.69–7.91 (d, 2H. Pyrimidine), 8.44 (s, 1H N═CH), 8.90 (s, 1H NH), 10.67 (bs, 1H OH). MS (ESI) *m*/*z*: 382. Anal. Calcd. For $C_{17}H_{12}ClN_7O_2$ (in%): C, 53.48; H, 3.17; N, 25.68. Found C, 53.41; H, 3.13; N, 25.64.

*2.2.3.9. (E)-N′-(2-chloropyrimidin-4-yl)-N-(5-cyano-2-hydroxy-6-(2, 3, 4-trimethylphenyl) pyrimidin-4-yl) formamidine (**7i**).* Recrystallization from ethanol afforded 84. FT-IR (KBr, cm^{-1}) *v*: 3559 (O–H), 3269 (N–H), 2217 (CN), 1689 (C═N). ^1H NMR (DMSO-d_6, 400 MHz) *δ*: 2.20–2.61 (s, 9H, Ar–CH_3), 7.35–7.46 (m, 2H, Ar–H), 7.72–7.92 (d, 2H. Pyrimidine), 8.57 (s, 1H N═CH), 8.96 (s, 1H NH), 10.67 (bs, 1H OH). MS

(ESI) *m*/*z*: 394. Anal. Calcd. For $C_{19}H_{16}ClN_7O$ (in%): C, 57.94; H, 4.09; N, 24.90. Found C, 57.91; H, 4.12; N, 24.97.

*2.2.3.10. (E)-N′-(2-chloropyrimidin-4-yl)-N-(5-cyano-2-hydroxy-6-(3, 4-dimethylphenyl) pyrimidin-4-yl) formamidine (**7j**).* Recrystallization from ethanol afforded 81%. FT-IR (KBr, cm^{-1}) *v*: 3563 (O–H), 3276 (N–H), 2226 (CN), 1694 (C═N). ^{1}H NMR (DMSO-d_6, 400 MHz) *δ*: 2.31–2.70 (s, 6H, Ar–CH_3), 7.33–7.46 (m, 3H, Ar–H), 7.71–7.95 (d, 2H. Pyrimidine), 8.58 (s, 1H N═CH), 8.93 (s, 1H NH), 10.67 (bs, 1H OH). MS (ESI) *m*/*z*: 380. Anal. Calcd. For $C_{18}H_{14}ClN_7O$ (in%): C, 56.92; H, 3.72; N, 25.82. Found C, 56.96; H, 3.73; N, 25.87.

*2.2.3.11. (E)-N′-(2-chloropyrimidin-4-yl)-N-(5-cyano-2-hydroxy-6-(2,4-dimethylphenyl)pyrimidin-4-yl)formamidine (**7k**).* Recrystallization from ethanol afforded 79%. FT-IR (KBr, cm^{-1}) *v*: 3566 (O–H), 3272 (N–H), 2228 (CN), 1691 (C═N). ^{1}H NMR (DMSO-d_6, 400 MHz) *δ*: 2.11–2.52 (s, 6H, Ar–CH_3), 7.34–7.49 (m, 3H, Ar–H), 7.72–7.90 (d, 2H. Pyrimidine), 8.48 (s, 1H N═CH), 8.97 (s, 1H NH), 10.67 (bs, 1H OH). MS (ESI) *m*/*z*: 380. Anal. Calcd. For $C_{18}H_{14}ClN_7O$ (in%): C, 56.92; H, 3.72; N, 25.82. Found C, 56.97; H, 3.75; N, 25.81.

*2.2.3.12. (E)-N-(6-(4-bromo-2-chlorophenyl)-5-cyano-2-hydroxypyrimidin-4-yl)-N′-(2 chloropyrimidin-4-yl) formamidine (**7l**).* Recrystallization from ethanol afforded 83%. FT-IR (KBr, cm^{-1}) *v*: 3569 (O–H), 3278 (N–H), 2223 (CN), 1695 (C═N). ^{1}H NMR (DMSO-d_6, 400 MHz) *δ*: 7.33–7.45 (m, 3H, Ar–H), 7.72–7.94 (d, 2H. Pyrimidine), 8.53 (s, 1H N═CH), 8.95 (s, 1H NH), 10.67 (bs, 1H OH). MS (ESI) *m*/*z*: 465. Anal. Calcd. For $C_{16}H_8BrCl_2N_7O$ (in%): C, 41.32; H, 1.73; N, 21.08. Found C, 41.37; H, 1.74; N, 21.12.

*2.2.3.13. (E)-N′-(2-chloropyrimidin-4-yl)-N-(5-cyano-2-hydroxy-6-(4-nitrophenyl)-pyrimidin-4-yl) formamidine (**7m**).* Recrystallization from ethanol afforded 85%. FT-IR (KBr, cm^{-1}) *v*: 3565 (O–H), 3271 (N–H), 2228 (CN), 1692 (C═N). ^{1}H NMR (DMSO-d_6, 400 MHz) *δ*: 7.36–7.47 (m, 3H, Ar–H), 7.71–7.96 (d, 2H. Pyrimidine), 8.55 (s, 1H N═CH), 8.91 (s, 1H NH), 10.67 (bs, 1H OH). MS (ESI) *m*/*z*: 397. Anal. Calcd. For $C_{16}H_8Cl_2N_8O_3$ (in%): C, 48.44; H, 2.29; N, 28.24. Found C, 48.46; H, 2.21; N, 28.26.

*2.2.3.14. (E)-N′-(2-chloropyrimidin-4-yl)-N-(5-cyano-2-hydroxy-6-(2,4-imethoxyphenyl)pyrimidin-4-yl) formamidine (**7n**).* Recrystallization from ethanol afforded 70%. FT-IR (KBr, cm^{-1}) *v*: 3272 (N–H), 3562 (O–H), 2227 (CN), 1691 (C═N). ^{1}H NMR (DMSO-d_6, 400 MHz) *δ*: 2.72–2.78 (s, 6H, Ar–CH_3), 7.33–7.45 (m, 3H, Ar–H), 7.72–7.92 (d, 2H. Pyrimidine), 8.46 (s, 1H N═CH), 8.93 (s, 1H NH), 10.67 (bs, 1H OH). MS (ESI) *m*/*z*: 412. Anal. Calcd. For $C_{18}H_{14}ClN_7O_3$ (in%): C, 52.50; H, 3.43; N, 23.81. Found C, 52.55; H, 3.48; N, 23.86.

2.3. In vitro antibacterial activity

Antibacterial activity of the synthesized compounds was determined against Gram-positive bacteria (*Bacillus subtilis* MTCC 121, *Staphylococcus aureus* MTCC 7443) and Gram-negative bacteria (*Xanthomonas campestris* **MTCC 7908** and *Escherichia coli* MTCC 7410) in DMF by the disc diffusion method on nutrient agar medium (Bauer et al., 1966). The sterile medium (Nutrient Agar Medium, 15 ml) in each petriplates was uniformly smeared with cultures of Gram positive and Gram negative bacteria. Sterile discs of 10 mm diameter (Hi-Media) were placed in the petriplates, to which 50 µl (1 mg/ml i.e., 50 µg/disc) of different synthesized compounds was added. The treatments also included 50 µl of DMF as negative, Bacteriomycin and Gentamycin as positive control for comparison. For each treatment, three replicates were maintained. The plates were incubated at 37 ± 2 °C for 24 h and the zone of inhibition was determined.

2.4. In vitro antifungal activity

The synthesized compounds were screened for their antifungal activity against *Fusarium oxysporum* MTCC 2480 in DMF by the poisoned food technique (Satish et al., 2007). Potato Dextrose Agar (PDA) media was prepared and about 15 ml of PDA was poured into each petriplate and allowed to solidify. 5 mm disc of 7 days old culture of the test fungi was placed at the centre of the petriplates and incubated at 26 °C for 7 days. After incubation the percentage inhibition was measured and three replicates were maintained for each treatment. Nystatin was used as standard. All the synthesized compounds were tested (at the dosage of 500 µl of the novel compounds/petriplates, where concentration was 0.1 mg/ml) by the poisoned food technique.

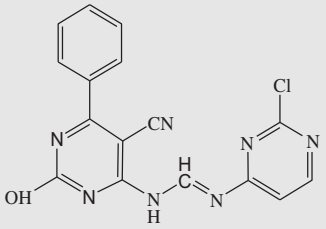
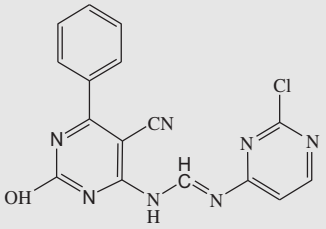
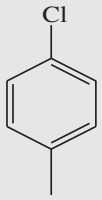
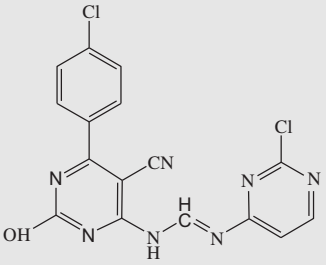
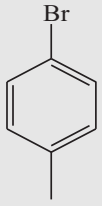
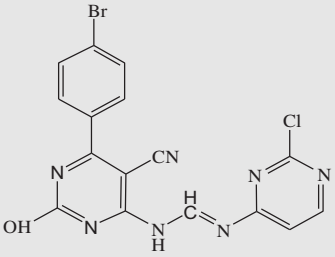
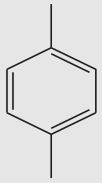
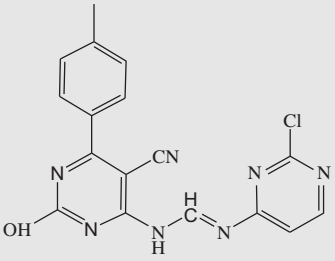
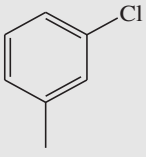
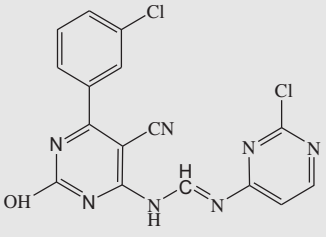
3. Results and discussion

3.1. Chemistry

The subject of this work was the synthesis of heterocyclic (*E*)-*N*′-(2-chloropyrimidin-4-yl)-*N*-(5-cyano-2-hydroxy-6-phenylpyrimidin-4-yl) formamidine derivatives **7a–n**. The performed reactions are shown in Scheme 1. Synthesized compounds were characterized by elemental analyses, FT-IR, ^{1}H NMR and Mass spectral studies. Compounds were purified by the recrystallization method using ethanol. The elemental analyses data showed good agreement between the experimentally determined values and the theoretically calculated values within ±0.3%. Physicochemical data of the synthesized compounds were reported in Table 1. The compounds show very good activity towards antimicrobial and antifungal activities. So, in regard to enhancing the biological activity of these derivatives we have synthesized compounds **7a–n** via formylation and Schiff base of compounds **4a–n** (Dipti et al., 2010).

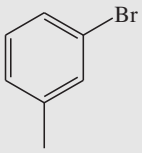
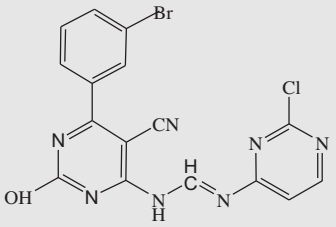
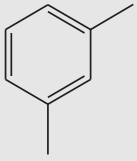
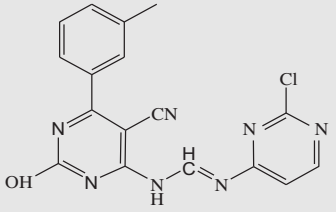
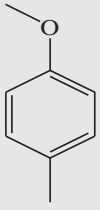
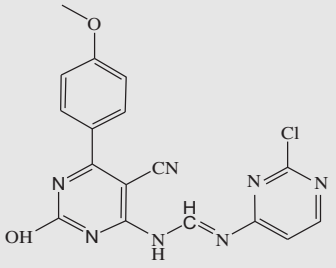
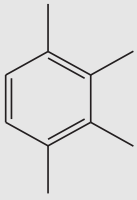
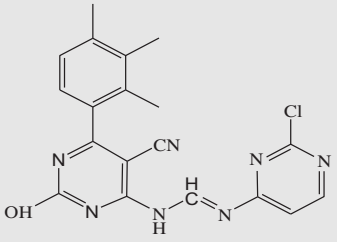
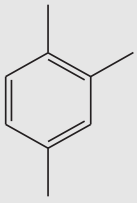
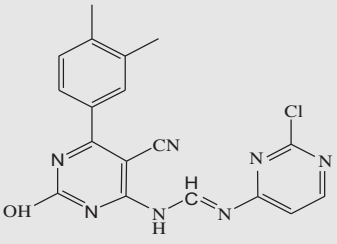
The absence of NH_2 absorption bands in **4a–n** at 2956 cm^{-1} in the IR spectra confirmed that the synthesized compounds were obtained. The absorption above 3200 cm^{-1} in compounds, **7a–n** confirms the N–H stretching vibrations and the appearance of medium to strong absorption bands around 1700 cm^{-1} due to a stretching vibration of the C═N bond formation in the synthesized compounds. The proton spectral data agree with respect to the number of proton and their chemical shift with the proposed structure. In all the compounds **7a–n**, the disappearance of CHO and NH_2 signals at *δ* 9.80 ppm and 4.10 ppm respectively and appearance of singlet CH proton at *δ* 8.40–8.95 ppm are evident for synthesized compounds. The

Table 1 Physicochemical data of the synthesized compounds **7a–7n**.

Compound	R	Structure	mp (°C)
7a			175–178
7b			185–187
7c			225–227
7d			191–193
7e			189–191

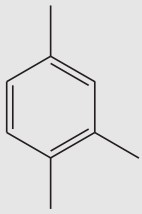
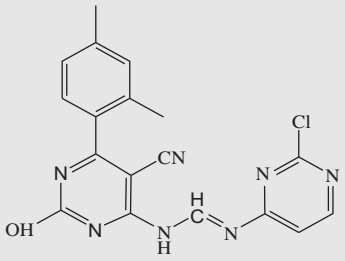
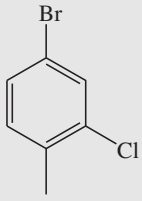
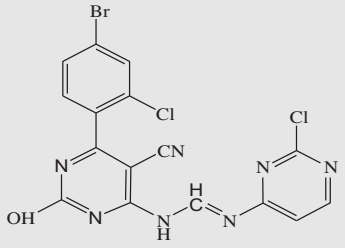
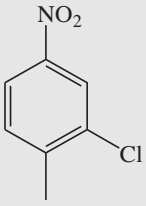
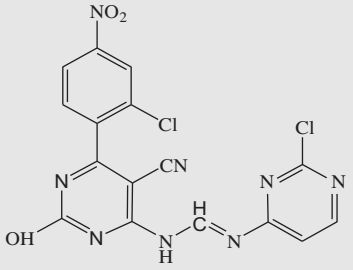
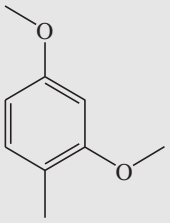
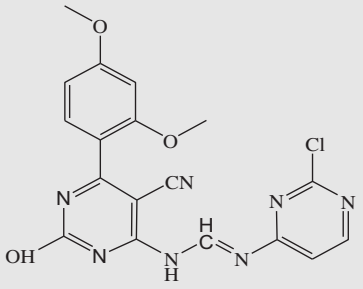
(*continued on next page*)

Table 1 *(continued)*

Compound	R	Structure	mp (°C)
7f			209–211
7g			182–184
7h			205–207
7i			201–203
7j			186–188

(*continued on next page*)

Table 1 *(continued)*

7k			214–216
7l			207–209
7m			179–181
7n			217–219

synthesized compounds were further confirmed by the appearance of molecular ion peak in mass spectra, mass spectra of all the newly synthesized compounds showed M^+ fragmentation peak in agreement with their molecular formula.

3.2. Antimicrobial activity

In order to search for the potent compounds from these newly synthesized compounds **7a–n** they were evaluated for *in vitro* antibacterial and antifungal activity against various Gram negative, Gram positive and fungal strains. The investigation of antibacterial screening data revealed that all tested compounds showed antibacterial activity against four pathogenic bacterial strains. Among the series **7a–n**, compounds **7l** and **7m** exhibited an elevated antibacterial activity against Gram positive (zone of inhibition 29–33 mm) and Gram negative (zone of inhibition 32–33 mm) bacteria. Compounds **7c**, **7e** and **7f** showed good antibacterial activity against all the tested organisms. All the remaining compounds showed moderate inhibitory activity. The results were compared with standard drugs Bacteriomycin and Gentamycin as depicted in Table 2. The compounds **7a–n** showed antimicrobial activity in the order: **7l > 7m > 7c** > **7e** > **7f > 7b > 7a > 7h > 7n > 7j > 7i > 7d > 7g > 7k** against tested microbial strains.

Table 2 *In vitro* antibacterial and antifungal activities of **7a–n.**

Compound	Zone of inhibition in diameter (mm)				% Inhibition
	B. subtilis	*S. aureus*	*X. campestris*	*E. coli*	*F. oxysporum*
7a	27	25	28	29	69.5
7b	26	24	27	28	71.5
7c	31	23	29	29	78.6
7d	28	26	27	28	66.1
7e	30	27	30	30	78.4
7f	29	28	28	29	77.3
7g	29	24	26	27	64.3
7h	30	27	30	30	68.4
7i	31	26	29	29	66.7
7j	29	24	30	29	67.1
7k	28	27	26	30	61.2
7l	33	26	32	29	97.1
7m	31	29	30	32	96.9
7n	29	27	30	29	67.4
Bacteriomycin	–	–	34	–	–
Gentamycin	35	30	–	35	–
Nystatin	–	–	–	–	100

The *in vitro* antifungal activity of the synthesized compounds **7a–n** was studied against *F. oxysporum*. The results were compared with the standard drug Nystatin as in Table 2. Compounds **7l** and **7m** showed good antifungal activity with 97.1% and 96.9% inhibition when compared with other compounds in the series against *F. oxysporum* respectively. The good inhibition by compounds **7l** and **7m** could be attributed to the presence of electron withdrawing groups. Antimicrobial activity of some pyrimidine derivatives has been reported (Keche et al., 2012). Compounds **7c**, **7e** and **7f** have demonstrated good antifungal activity against *F. oxysporum*. Compounds **7a**, **7b, 7e**, **7d**, **7g, 7h, 7i, 7j, 7k** and **7n** were found to be moderately active against tested fungal strain. From the results it is evident that most of the compounds showed moderately active and few showed good activity. The results of the antibacterial and antifungal activities are collected in Table 2. As in Table 2 compounds, **7l** and **7m** were found to be more active than other compounds in the series. Especially, compounds **7l** was found to be more active while **7m** exhibited similar activity with respect to the standard drugs. Compounds **7c**, **7e** and **7f** found to have good activity. The remaining compounds exhibited moderate antimicrobial activity. The pyrimidine derivatives containing Cl, NO_2 and OCH_3 of the benzene ring were found to be effective antimicrobial agents among the series.

4. Conclusion

In conclusion, the objectives of the present study were to synthesize and investigate the antimicrobial activities of some new heterocycles with the hope of discovering new structure leads serving as potent antimicrobial agents. The obtained results clearly revealed that compounds **7l** and **7m** derived from pyrimidine exhibited better antimicrobial activity while compounds **7a**–**k** and **7n** were found to be moderate antimicrobial agents with functionalities such as Br, OCH_3 and Cl groups on the benzene ring which were found to have strong relevance to the antimicrobial activity.

Acknowledgements

One of the authors (C. Mallikarjunaswamy) is grateful to St. Philomena's College, Mysore, for providing laboratory facility and thanks to the University of Mysore.

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