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Synthesis of some novel Pyrimidine derivatives and compared to available antimicrobial (*MTCC inhibitors*) drugs.

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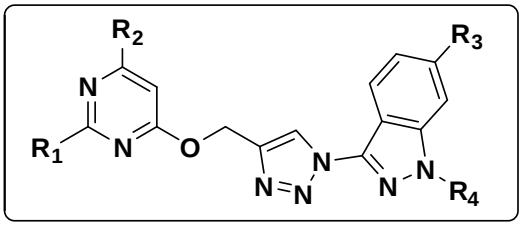
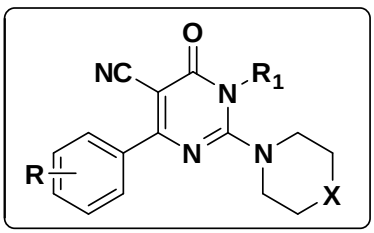
Abstract - A new series of pyrimidine derivatives have been synthesized by reacting substituted pyrimidine carboxylic acid with substituted aniline to get substituted pyrimidine amide **(3),** which was de-methylated using BBr3 to form hydroxy derivative of substituted pyrimidine amide **(4).** This was further alkylated with respective alkyl halide to synthesize substituted pyrimidine derivatives **(5)**.The synthesized compounds were screened for their *in vitro* growth inhibiting activity against different strains.
Key words: Multi-component reactions, Pyrimidine derivative, antimicrobial activity, minimum inhibitory concentration (MIC).

I. INTRODUCTION

The medicinal value of pyrimidine derivatives is significant among various heterocycles, as they are found to possess antineoplastic,[1-3] antiviral,[4-6] antibiotic[7] and anti-inflammatory[8] including other biological activities. Since benzothiazoles, pyrimidines and thiourea all possess diverse biological activities[9-11]; the aim of this study was to synthesize some new derivatives incorporating these nuclei and evaluate the prepared compounds for antibacterial activity.Communicable disease due to gram-positive bacteria such as methicillin resistant Staphylococcus aureus (MRSA), vancomycin resistant Enterococcus faecium (VREF), and penicillin resistant Streptococcus pneumoniae are the prominent cause of indisposition and impermanence to the civicat present [12]. For the period of last few years an escalation of intrusive microbial and fungal contagion has been perceived, principally in immune- inhibited patients, which are now the cause of indisposition and impermanence as well. Therefore, there is exigentrequisite to develop new antimicrobial agents[13].Heterocyclic compounds have attracted attention due to their diverse biological and pharmacological properties. Pyrimidine derivatives are significant moieties in medicinal chemistry because of their wide range of biological activities like antibacterial[14], antifungal [15-16].Furlani, R. E.; Yeagley, A. A.; Melander, C., Eur. J. Med. Chem., 2013, 62: 59. (Srtucture-1) comprise a very interesting class of compounds because of their significant and versatile biological and pharmacological activities, such as antimicrobial, antimalarial[17].Basavaraja H S, Jayadevaiah K V, Mumtaz M. Hussain,a Vijay Kumar M M J, BasavarajPadmashali, J. Pharm. Sci. & Res., Vol.2(1), 2010, 5-12(Structure- 2).



Structure-1



Structure-2

II. RESULTS AND DISCUSSION

In the contemporaneousstudyPyrimidine derivatives was chosen for synthesis of novel molecules as antimicrobial agents due to testified antimicrobial activity of pyrimidine nucleus. 2- & 6-substituted Arylalkoxy and amide derivatives were synthesized using multistep procedure by following substituted pyrimidine carboxylic acid was coupled with substituted aniline to get substituted pyrimidine amide**,** which was de-methylated using BBr3 to form hydroxy derivative of substituted pyrimidine amide**.** This was further alkylated with respective alkyl halide in presence of potassium carbonate to synthesize substituted pyrimidine derivatives. Synthetic pathways for the synthesis of targets compounds are shown in general scheme with the hope of discovering new antimicrobial agents. In comparison with several control drugs available in market in different categories. Newly synthesized derivatives have been evaluated for antibacterialand antifungal activity against standard strains. Their Structures were elucidated with ^{1}H NMR, and mass spectroscopy. Mass spectra of the compoundsshowed [M^+ + H] peaks, since the electrospray ionization method was employed. The structures of all derivatives were confirmed by spectral analysis and results are presented in the experimental section. In the ^{1}H NMR spectra of the compounds, the signals of aromatic protons were observed at δ9.56–6.75, as singlet bands, doublet bandsand multiplet bands.

Compounds were evaluated for the antimicrobial and compared with the activity of standard drugs available. All the derivatives synthesized were tested in vitro for antibacterial activity against *E. coli***MTCC442**, *P. aerugenosa***MTCC441**, *S. aureus***MTCC96** and *S. Pyogenus***MTCC443.** All the derivatives were tested for the antifungal activity against *C. albicans***MTCC227**, *A. niger***MTCC282** and *A. clavatus***MTCC1323**.The activity of antibacterial drugs depends upon its concentration *in vitro* characterization of antibacterial activity commonly includes the determination of minimum inhibitory concentration (MIC)[18-21]. Each synthesized drugs was diluted with DMSO obtaining 2000 µg/ml concentrations as a stock solution for biological screening. For primary screening 1000 µg/ml to 250 µg/ml concentrations and for secondary screening 200 µg/ml to 6.25 µg/ml concentrations was used. The standard drugs employed while assessing antimicrobial activities were Gentamycin, Chloramphenicol, Ampicillin, Ciprofloxacin and Norfloxacin for antibacterialactivity; Nystatin and Greseofulvin for antifungal activity. We have used the Broth Dilution Method[22]to evaluate the antibacterial activity. This archetypal method produces a quantifiable result for the amount of antimicrobial agents that is needed to inhibit growth of unambiguous microorganisms.

Some compounds exhibits broad antibacterial activity with MIC values of 12.5 – 500 µg/ml against *E. coli*, *P. aerugenosa*, *S. aureus* and *S. Pyogenus* and its isolate except for derivative **MT-21** that had a MIC value of 12.5 µg/mlagainst *S. aureus* stains and compound **MT-24** had a MIC values of 25µg/ml against *P. aerugenosa* (Table- 1).

In the antifungal assay compounds exhibits broad antifungal activity with MIC values of 125 – >1000µg/ml against *C. albicans*, *A. niger* and *A. clavatus* and its isolate except for derivative **MT-21** that had a MIC value of 125 µg/ml against *C. albicans*, and *A. niger,*and 100 µg/ml against*A. clavatus.* All the derivatives had lower antifungal activities than the standard drugs (Table-2).

III. EXPERIMENTAL SECTION

3.1 Materials & Methods

The chemicals and solvents were purchased from Sigma-Aldrich Co. (Taufkirchen, Munich, Germany), Merck Lifescience Pvt. Ltd. (Vikhroli, Mumbai, India) and Fisher Scientific (Pittsburgh, PA, USA) and usedwithout further purification. Silica gel (with Mesh size 230-400) was used for column chromatography and TLCplates were purchased from Merck Lifescience Pvt. Ltd. (Vikhroli, Mumbai, India) and ethyl acetate: hexaneswere used as mobile phase. NMR spectra were recorded on Bruker 400 MHz NMR spectrometer in CDCl3and DMSO; tetramethylsilane (TMS) was used as an internal standard. The mass spectra were recorded o Waters ZQ Micromass LC-MS spectrometer (Milford, MA, USA) usingthe ESI(+) method.

3.2 General Procedure

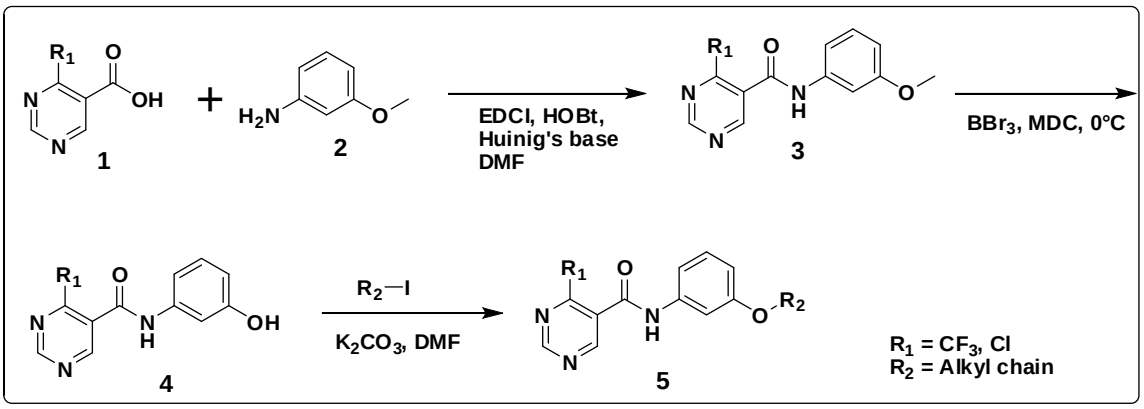
Series A:

Step-1: Synthesis of 4-(trifluoromethyl)-N-(3-methoxyphenyl) pyrimidine-5-carboxamide:

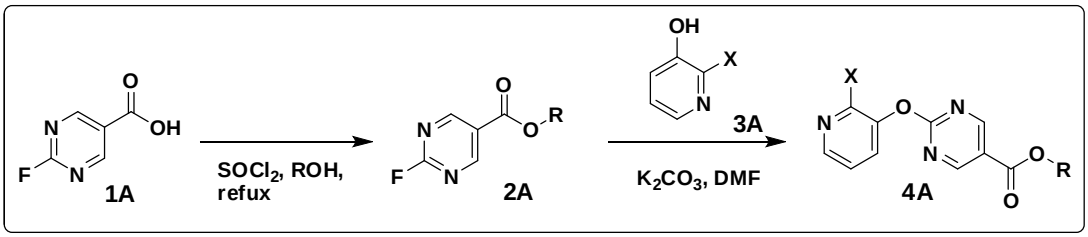
Compound 1 (1.0 Eq.) was taken in a RBF with compound 2 (1.1 Eq.), 1.5 eq. of huinig's base and N, N- Dimethylformamide at room temperature and stirred for 10 min. Now EDCI-HCl (1.0 eq.) and HOBt (1.0 eq.) were added under N_2 atmosphere and reaction mixture was allowed to stirred at rt, till completion of reaction. After that it was diluted with ethyl acetate and organic layer was washed with water, sodium bicarbonate solution and then with brine solution. Organic layer was concentrated and purified by column chromatography to afford desired product**(3). 1 H NMR (4-(trifluoromethyl)-N-(3-methoxyphenyl) pyrimidine-5-carboxamide) CDCl3:** 9.51 (1H, bs); 9.23 (1H, s); 7.42 (1H, bs); 7.31-7.29 (2H, m); 7.05-7.03 (1H, d, *J*= 8.0 Hz); 6.79-6.77 (1H, dd, *J*= 6.4 Hz, 1.6 Hz); 4.54 (3H, s).

Step-2: Synthesis of 4-(trifluoromethyl)-N-(3-hydroxyphenyl)pyrimidine-5-carboxamide:
To the stirred solution of compound **3** (1.0 Eq.) in MDC at 0°C, BBr3 (1.2 eq.) was added dropwise under N2-atmosphere and reaction mixture was allowed to stirred at 80 deg. C till completion of reaction. After that it was diluted with MDC and organic layer was washed with ice-water and then with brine solution. Organic layer was concentrated and purified by column chromatography to afford desired product (**4**). **1H NMR (4-(trifluoromethyl)-N-(3-hydroxyphenyl)pyrimidine-5-carboxamide):** 9.51 (1H, bs); 9.23 (1H, s); 7.42 (1H, bs); 7.31-7.29 (2H, m); 7.05-7.03 (1H, d, *J*= 8.0 Hz); 6.79-6.77 (1H, dd, *J*= 6.4 Hz, 1.6 Hz); 4.01 (1H, s).
Step-3: Synthesis ofN-(3-Isopropoxyphenyl)-4-(trifluoromethyl)pyrimidine-5-carboxamide:
To the stirred solution of compound **4** (1.0 Eq.) in DMF was added1.5 eq. of potassium carbonate and N, N-Dimethylformamide at room temperature and stirred for 10 min. Now Isopropyl Iodide (1.1 Eq.) was added under N_2 atmosphere and reaction mixture was allowed to stirr at room temperature, till completion of reaction. After that it was diluted with ethyl acetate and organic layer was washed with water, sodium bicarbonate solution and then with brine solution. Organic layer was concentrated and purified by column chromatography to afford desired product (**5**). **1H NMR (N-(3-Isopropoxyphenyl)-4-(trifluoromethyl)pyrimidine-5-carboxamide):**9.46 (1H, bs); 9.15 (1H, s); 7.45 (1H, bs); 7.30-7.28 (2H, m); 7.04-7.02 (1H, d, *J*= 8.0 Hz); 6.77-6.75 (1H, dd, *J*= 6.4 Hz, 1.6 Hz); 4.62-4.54 (2H, m); 1.36-1.35 (6H, d, *J*= 6.0 Hz).
Series B:
Step-1: Synthesis of ethyl 2-chloropyrimidine-5-carboxylate (3):
To the stirred solution of compound 1 (1.0 Eq.) in EtOH at 0O C under N2 atmosphere and flowed by dropwise addition of thionyl chloride. Now reaction mixture was allowed to stir at reflux for 10 hrs. After that it was quenched with ice-water and extracted with ethyl acetate and organic layer was concentrated under reduced pressure. Crude product was purified by column chromatography to afford compound (4). **1 H NMR (Ethyl 2-chloropyrimidine-5-carboxylate) CDCl3:** 8.48 (2H, s); 4.33-4.27 (2H, q, J= 7.2 Hz); 1.27-1.23 (3H, t, J = 7.2 Hz).
Step-2: Synthesis of Ethyl 2-(2-chloropyridin-3-yloxy) pyrimidine-5-carboxylate (4):
To the stirr solution of compound 3 (1.0 Eq.) in a RBF with 1.5 eq. of potassium carbonate and N, N-Dimethylformamide at room temperature, compound 2 (1.0 Eq.) was added under N2 atmosphere and reaction mixture was allowed to stirred at 80 deg. C, till completion of reaction. After that it was diluted with ethyl acetate and organic layer was washed with water, sodium bicarbonate solution and then with brine solution. Organic layer was concentrated and purified by column chromatography to afford desired product (3). **1H NMR (Ethyl 2-(2-chloropyridin-3-yloxy) pyrimidine-5-carboxylate) CDCl3**: 8.69-8.68 (1H, dd, J= 3.2 Hz, 1.2 Hz); 8.47 (2H, s); 7.67-7.64 (1H, m); 7.61-7.58 (1H, m); 4.33-4.27 (2H, q, J= 7.2 Hz); 1.27-1.23 (3H, t, J = 7.2 Hz).

4. Figures & Tables

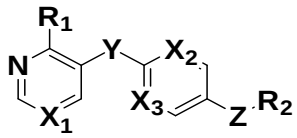


Scheme-1



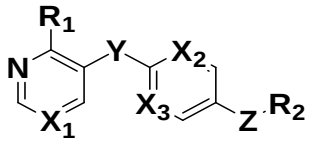
Scheme-2

Table-1: Invitro antibacterial activity of novel pyrimidine derivatives in comparision with standard drugs:



No.	Sample ID	X1	X2	X3	R1	R2	Y	Z	MTCC442	MTCC441	MTCC96	MTCC443
1	MT-19	N	CH	CH	CF3	Me	CONH-	O	500	500	250	250
2	MT-20	N	CH	CH	CF3	Et	CONH-	O	250	500	500	500
3	MT-21	N	CH	CH	CF3	iPr	CONH-	O	50	25	12.5	25
4	MT-22	N	CH	CH	Cl	Me	CONH-	O	250	250	200	125
5	MT-23	N	CH	CH	Cl	Et	CONH-	O	125	200	100	100
6	MT-24	N	CH	CH	Cl	iPr	CONH-	O	100	25	125	100
7	MT-25	CH	N	N	Cl	Et	O	COO-	250	125	250	250
8	MT-26	CH	N	N	Cl	C3H7	O	COO-	125	200	500	250
9	MT-27	CH	N	N	Cl	C2H4CF3	O	COO-	125	200	500	250
10	MT-28	CH	N	N	I	Et	O	COO-	100	200	200	250
11	MT-29	CH	N	N	I	C3H7	O	COO-	500	500	250	250
12	MT-30	CH	N	N	I	C2H4CF3	O	COO-	500	500	250	250
Reference												
Gentamycin									0.05	1	0.25	0.5
Chloramphenicol									50	50	50	50
Ciprofloxacin									25	25	50	50
Norfloxacin									10	10	10	10

Table-2: Invitro antifungal activity of novel pyrimidine derivatives in comparision with standard drugs:



No.	Sample ID	X1	X2	X3	R1	R2	Y	Z	MTCC227	MTCC282	MTCC1323
1	MT-19	N	CH	CH	CF3	Me	CONH-	O	125	>1000	500
2	MT-20	N	CH	CH	CF3	Et	CONH-	O	150	1000	500
3	MT-21	N	CH	CH	CF3	iPr	CONH-	O	125	125	100
4	MT-22	N	CH	CH	Cl	Me	CONH-	O	1000	1000	500
5	MT-23	N	CH	CH	Cl	Et	CONH-	O	500	>1000	500
6	MT-24	N	CH	CH	Cl	iPr	CONH-	O	500	500	1000
7	MT-25	CH	N	N	Cl	Et	O	COO-	1000	500	500
8	MT-26	CH	N	N	Cl	C3H7	O	COO-	>1000	500	500
9	MT-27	CH	N	N	Cl	C2H4CF3	O	COO-	500	500	1000
10	MT-28	CH	N	N	I	Et	O	COO-	>1000	1000	1000
11	MT-29	CH	N	N	I	C3H7	O	COO-	1000	1000	500
12	MT-30	CH	N	N	I	C2H4CF3	O	COO-	500	1000	1000
Reference											
Nystatin									100	100	100
Gresiofulvin									500	100	100

IV. CONCLUSION

In conclusion,a new class of pyrimidine derivatives synthesized and evaluated as antibacterial and antifungal agents. Few derivatives showed good antibacterial activity against *E. coli*, *P. aerugenosa*, *S. aureus* and *S. Pyogenus***,** and antifungal activity against *C. albicans*, *A. niger*and *A. clavatus*, but less as compared to the standard drugs. So, It can be concluded that these class of compounds certainly holds greatpromise towards good active leads in medicinal chemistry. A further study to acquire more information concerning pharmacological activity is in progress.

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REFRENCES

[1]. Giller, S. A., Zhuk, R. A., Lidak, M.I. U., Dokl. Akad. Nauk.SSR. **1967**, 176, 332-335.
[2]. Curreri, A. R., Ansfield, R. J., McIver, F. H., Waisman, H. A., Hedelberger., C. *Cancer Res.*, **1958**, 18, 478-484.
[3]. Chaudhary, N. K., Montag, B. J., Heidelberger., C. *Cancer Res.* **1958**, 18, 318-328.
[4]. Malik,V., Singh, P., Kumar, S., *Tetrahedron.* **2006**, 62, 5944-5951.
[5]. AntonínHolý, Ivan Votruba and Milena Masojídková,*J. Med. Chem.* **2002**, 45, 1918–1929.
[6]. Andres, P., Marhold, A., *J. Fluorine Chem.* **1996**, 77, 93-95.
[7]. Reddick, J. J., Saha, S., Lee, J., Melnick, J. S., Perkins, J., Begley, T. P., *Bioorg. Med. Chem. Lett.* **2001**, 11, 2245-2248.
[8]. Gauthier, B., *Ann. Pharm. Fr*. **1963**, 21, 655- 666.
[9]. Gowda F. E., Alaa A. M. Abdel Aziz, Omer A. Attef; *Bioorganic and Medicinal Chemistry*; 12. **2004,** 1845-1852.
[10]. Sanna, M. E.;Wahid, M. B.; Laila, I. I. Egypt J. Pharm. Sci. **1998**, 39, 197.
[11]. Carla, M. S. M.;Carlos, M. R. S. A.;Carlos, R. R.; Eliezer, J. B. Journal of Molecular Structure (Theochem)**2002,** 579, 31.
[12]. Moustafa M A, Gineinah M M, Nasr M N & Bayoumi W A, *Arch Pharm*, 337,**2004,** 427.
[13]. Andriole V T, *J Antimicrob Chemother*, 44, **1999**, 151.
[14]. Panwar H., Verma R. S., Srivastava V. K., Kumar A., *India J Chem* 45B, **2006**, 2099.
[15]. Abele E, Abele R, Dzenitis O, Lukevics, *E.Chem Heterocycl Compd* 39, **2003**, 3.
[16]. Skii N. M. P., Magedov I. V., Drozed V. N., *Chem Heterocycle Compd* 33, **1997**, 1475.

[17]. Sato, Y.;Shimogi, Y.;Kumakura, S.;Takagi, H. Ja pan, Kokai 75,151,896;Appl. 74,61,149; Chem. Abstr. 1976, 84, 164771.
[18]. Clinical microbiology procedure handbook, vol. II, chapter 5 pages no 5.0.1, Henry d. Isenberg, 2nd edition.
[19]. Desai N C, Shihora P N & Moradia D L, "Synthesis and characterization of new quinazolines as potential antimicrobial agent" *Indian journal of chemistry, Sec-b,* 46b, **2007**, 550-553.
[20]. Shadomy, S. In Manual of Clinical Microbiology; Albert, B. Ed.; ASM Press: Washington, DC, **1991**, 1173.
[21]. V. M. Barot, B. G. Rathod, *Asian Journal of Biochemical and Pharmaceutical Research*, Issue 1(3), **2013**, 80-85.
[22]. Donal R. Stalons & Clyde Thornsberry, "Broth-dilution method for determining the antibiotic susceptibility of anaerobic bacteria", *Antimicrobial Agents & Chemotherapy*, 7(1), **1975**, 15-21.

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