International Standard Serial Number (ISSN): 2249-6807

International Journal of Institutional Pharmacy and Life Sciences 5(5): September-October 2015

INTERNATIONAL JOURNAL OF INSTITUTIONAL PHARMACY AND LIFE SCIENCES

Pharmaceutical Sciences

Research Article……!!!

Received: 21-06-2015; Revised: 18-09-2015; Accepted: 19-09-2015

SYNTHESIS, CHARACTERIZATION, BIOLOGICAL EVALUATION AND DOCKING STUDIES OF PYRAZOLE-5-ONE DERIVATIVES OF PIRIMIDINES SORTASE A STAPHYLOCOCCUS INHIBITORS

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Keywords:

Pyrazole 5-one, pirimidines, Antimicrobial and Antifungal activity, docking studies of sortase A Staphylococcus inhibitors

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ABSTRACT

New novel derivatives of (4-diethylamine/4-pyrrolidine/4-pyronphenyl)-2(oxo/thiones/imines)1,2,3,4-tetrahydropyrimidine-5 carbonyl)-3-(trifluromethyl)-1H-pyrazol-5(4H)-one (3a-i). containing various heterocyclic substituent's were synthesized, characterized by elemental analysis,IR,[1]HNMR and [13]CNMR spectra and evaluated for in antimicrobial and antifungal activity. Molecular docking studies were performed to calculate docking scores and to propose the binding mode of pirimidines.

INTRODUCTION

The 4-aryl-1,2,3,4-tetrahydropyrimidine-2(1H) ones/thiones/imines -5 carbonyl)-3-(triflouro methyl)-1H-pyrazole-5(4H)-one are an important class of compounds exhibiting broad spectra of pharmacological activities such as anti-microbial and antihyper tensives[1-2]. These compounds have a huge interest in the medicinal chemistry community in recent years.

Moreover, this class of heterocycles revealed other pharmacological activities such as anti inflammatory[3],calcium channel modulators[4] antifungal,antibactireal[5],melanin concentrating hormone receptors (MCHI-R) antagonists[6], chemical modulators of heat shock protein 70(Hsp 70)[7],hepatitis B replication inhibitors[8] and inhibitors of the fatty acid transporters[9].

A good deal of importance was given to pirimidines and their derivatives in the field of heterocyclic chemistry due to their unique biological applications [10]. In view of the above observations, we synthesized pyrazole 5-one possessing pirimidines and screening for possible biological, pharmacological activities and docking sortase A staphylococus inhibitory activity by silico methods.

EXPERIMENTAL SECTION

MATERIALS AND METHODS

All the chemicals used in the present investigation were purchased from sigma-Aldrich chemicals company, Inc.USA. And used without further purification. TLC was performed on aluminum sheet of silica gel $60F_{254,}$ E-Merk, Germany using iodine as visualizing agent. Melting points were determined in open capillary tubes on Mel –temp apparatus and are uncorrected. Column chromatography was performed on silica gel with different solvent systems as eluents to afford the pure compounds. The IR Spectra were recorded as KBr pellets on Perkin –Elmer 1000units, instruments. All H and C-NMR spectra were recorded on a Varian XL-300 Spectrometer operating at 400MHz for ^{1}H-NMR and 75 MHz for ^{13}C-NMR. Mass spectral data was recorded on a Carlo Erba 1108 elemental analyser, Central drug Research Institute, Lucknow, India.

Docking method

Docking was carried out using GOLD (Genetic Optimization of Ligand Docking) software which is based on genetic algorithm (GA). This method allows as partial flexibility of protein and full flexibility of ligand. The compounds are docked to the active site of the Sortase A. The interaction of these compounds with the active site residues are thoroughly studied using molecular mechanics calculations. The parameters used for GA were population size (100), selection pressure (1.1), number of operations (10,000), number of island (1) and niche size

(2). Operator parameters for crossover, mutation and migration were set to 100, 100 and 10 respectively. Default cutoff values of 3.0 A° (dH-X) for hydrogen bonds and 6.0 A° for vanderwaals were employed. During docking, the default algorithm speed was selected and the ligand binding site in the Sortase A was defined within a 10 A° radius with the centroid as CE atom of PHE136. The number of poses for each inhibitor was set 100, and early termination was allowed if the top three bound conformations of a ligand were within 1.5A° RMSD. After docking, the individual binding poses of each ligand were observed and their interactions with the protein were studied. The best and most energetically favorable conformation of each ligand was selected.

RESULTS AND DISCUSSION

Synthesis of (4-diethylphenyl/4-pyrrolidine/4-pyranphenyl)-2(oxo/thiones/imines) 1,2,3,4-tetrahydropyrimidine-5-carbohydrate (2a-i).

A solution of 1(a) and hydrazine hydrate in ethanol was refluxed for 5Hours. The reaction mixture was cooled and poured on to ice cold water with stirring. The progress of the reaction was monitored by TLC with acetone: ethyl acetate (7:3) as elutent. The separated solid was filtered, washed with water and recrystallized from ethanol to afford (4-diethylphenyl phenyl)-2(oxo/thiones/imines) 1,2,3,4-tetrahydropyrimidine-5-carbohydrate (2a).

The similar procedure was adopted to synthesize (2b-i) by the reaction between 2(a-i) with hydrazine hydrate. The structure of these newly synthesized compounds 2(a-i) were characterized by their elemental analysis and spectral data (^{1}H-NMR and IR).

Synthesis of (4-diethylphenyl/4-pyrrolidine/4-pyran phenyl)-2(oxo/ thiones/imines)1,2,3,4-tetrahydropyrimidine-5carbonyl)-3-(trifluromethyl) -1H-pyrazol-5(4H)-one (3a-i).

In a solution of (4-diethylphenylphenyl)-2(oxo/thiones/imines) 1,2,3,4-tetrahydropyrimidine-5-carbohydrate (2a) in ethanol, and ethyl 4-trifluoro-3 oxobutanonate were added and the mixture was refluxed for 12 hours in presence of catalytical amount glacial acetic acid.Exess of ethanol was removed by distillation and crystalline residue obtained was filtered, washed with ethanol, dried and recrystallized to get the compound Synthesis of (4-diethylphenylphenyl)-2(oxo/thiones/imines)1,2,3,4-tetrahydropyrimidine-5 carbonyl)-3-(trifluromethyl)-1H-pyrazol-5(4H)-one (3a) in good yields.

The structure of these newly synthesized compounds (3a-i) was based on the characterized by their elemental analysis and spectral data (^{1}HNMR, IR, and Mass).

Scheme: Synthesis of (4-diethylphenyl/4-pyrrolidine/4-pyranphenyl)-2(oxo/thiones/imines) 1,2,3,4-tetrahydropyrimidine-5 carbonyl)-3-(trifluromethyl)-1H-pyrazol-5(4H)-one (3a-i).

COMPOUND NO	R	X
3a		O
3b		S
3c		NH
3d		O
3e		S
3f		NH
3g		O
3h		S
3i		NH

Physical, analytical and spectral data of compounds (2a-i)

Synthesis of (4-diethylaminophenyl)-2(oxo) 1,2,3,4-tetrahydropyrimidine-5-carbohydrate (2a):

Yield (65%); m.p (143-146); IR(KBr,cm^{-1}) 3594(OH),3378(NH),3040 (Ar-H), 1655(C=O),; ^{1}HNMR(400MHz,DMSO-d_6):δppm1.15(t,6H, 2(-CH_3) groups),3.21 (q, 4H, CH_2-N-CH_2),4.20(s,2H,NH_2),5.13(s,1H,CHof pyrimidine ring),5.35(s,1H,-OH),6.15(s, 1H, NH-C=Oofpyrimidine ring),6.22(s,1H,Ar-H),6.30(d,1H,Ar-H),6.85(s,1H,NHof pyrimidine ring), 6.94(d,1H, Ar-H),7.93(s,1H,=CHof pyrimidine ring),8.72 (s,1H,NH-C=O); Anal.calcd (%) for $C_{15}H_{21}N_5O_3$: C 51.94%,H 4.59% and N 15.94% Found: C 51.92%, H 4.55% and N 15.91%.

Synthesis of (4-diethylaminophenyl)-2(thiones)1,2,3,4-tetrahydropyrimidine-5-carbohydrate (2b):

Yield (70%); m.p (162-164); IR (KBr,cm^{-1}) 3600(OH), 3385(NH) ,3040 (Ar-H), 1655(C=O); ^{1}HNMR(400MHz,DMSO-d_6):δppm1.15(t,6H,2(-CH_3)groups),3.21(q,4H,CH_2-N-CH_2),4.00 (s,1H,CHofpyrimidinering),4.20(s,2H,NH_2),5.35(s,1H,-OH),6.15(s,1H, NH-C=O of pyrim idinering),6.22(s,1H,Ar-H),6.30(d,1H,Ar-H),6.53 (s,1H,NH of pyrimidine ring), 6.70 (s,1H,=CH of pyrimidine ring),6.95(d,1H,Ar-H),8.72(s,1H,NH-C=O), 13.76(s,1H,NH of pyrimidine ring).; Anal.calcd (%) for $C_{15}H_{21}N_5O_2S$: C 52.70%,H 4.43% and N 15.38% Found: C 52.50%, H 4.23% and N 15.08%.

Synthesis of (4-diethylaminophenyl)-2(imines)1,2,3,4-tetrahydropyrimidine-5-carbohydrate (2c):

Yield (70%); m.p (148-150); IR (KBr,cm^{-1}) 3591(OH),3381(NH),3040 (Ar-H), 1655(C=O); ^{1}HNMR(400MHz,DMSO-d_6):δppm1.15(t,6H,2(-CH_3)groups),3.21(q,4H,CH_2-N-CH_2),4.10 (s,1H, CH ofpyrimidine ring),4.20(s,2H, NH_2) ,5.35 (s,1H,-OH), 6.15 (s,1H,NH-C=Oofpyrimidinering),6.22(s,1H,Ar-H),6.30(d,1H,Ar-H),6.50(s,1H,NH ofpyrimidinering), 6.70(s,1H, =CHofpyrimidinering),6.95(d,1H,Ar-H),8.75 (s, 1H,NH-C=O),10.2(s,1H,=NH),13.76 (s, 1H,NH of pyrimidine ring).; Anal.calcd (%) for $C_{15}H_{22}N_6O_2$: C 52.05%,H 4.83% and N 19.17% Found: C 51.75%, H 4.53% and N 19.07%.

Synthesis of (4-pyrrolidinephenyl)-2(oxo)1,2,3,4-tetrahydropyrimidine-5-carbohydrate (2d):

Yield (70%); m.p (175-177); IR (KBr,cm^{-1}) 3365(NH), 3040 (Ar-H), 1659 (C=O); ^{1}HNMR(400MHz,DMSO-d_6):δppm1.92(t,4H,2CH_2ofpyrolidine ring),3.44(t,4H,CH_2-N-CH_2 of pyrolidine ring),4.20(s,2H,NH_2), 5.13 (s,1H ,CH of pyrimidine ring),6.15(s,1H,NH of

pyrimidine ring),6.55(s,1H,NH of pyrimidine ring),6.45-6.65(m,4H,Ar-H),7.93(s,1H,=CH of pyrimidine ring),8.75(s,1H,O=C-NH); Anal.calcd (%) for $C_{15}H_{19}N_5O_2$: C 59.79%,H 6.36% and N 23.24% Found: C 59.59%, H 6.06% and N 23.04%.

Synthesis of (4-pyrrolidinephenyl)-2(thiones) 1,2,3,4-tetrahydropyrimidine-5-carbohydrate (2e):

Yield (60%); m.p (143-145); IR (KBr,cm^{-1})3392(NH), 3040 (Ar-H), 1698 (C=O),1276 (C=S); ^{1}HNMR (400MHz,DMSO-d_6):δppm1.92(t,4H,2CH_2 of pyrolidine ring),3.44(t,4H,CH_2-N-CH_2ofpyrolidinering),4.00(s,1H,CHofpyrimidinering),4.20(s,2H,NH_2) ,6.52(s,1H, NHof pyrimidine ring),6.42-6.65(m,4H,Ar-H),6.70(s,1H,=CH of pyrimidine ring), 8.75 (s,1H, O=C-NH),13.76(s,1H, NH of pyrimidine ring); Anal.calcd (%) for $C_{15}H_{19}N_5OS$: C 56.76%,H 6.03% and N 22.06% Found: C 56.56%, H 5.73% and N 21.76%.

Synthesis of (4-pyrrolidinephenyl)-2(imines)1,2,3,4-tetrahydropyrimidine-5-carbohydrate (2f):

Yield (68%); m.p (161-163); IR (KBr,cm^{-1})3375(NH), 3040 (Ar-H),1623 (C=N); ^{1}HNMR(400MHz,DMSO-d_6):δppm1.92(t,4H,2CH_2ofpyrolidine ring),3.44(t,4H,CH_2-N-CH_2 of pyrolidine ring),4.10 (s,1H,CH of pyrimidine ring),4.20(s,2H,NH_2), 6.50(s,1H,NH of pyrimidine ring), 6.42-6.65(m,4H,Ar-H),6.70(s,1H,=CH of pyrimidine ring),8.75 (s,1H,O=C-NH),9.89(s,1H,=NH),13.76(s,1H, NH of pyrimidine ring).; Anal.calcd (%) for $C_{15}H_{20}N_6O$: C 59.98%,H 6.71% and N 27.98% Found: C 59.68%, H 6.61% and N 27.68%.

Synthesis of (4-pyronphenyl)-2(oxo)1,2,3,4-tetrahydropyrimidine-5-carbohydrate (2g):

Yield (65%); m.p (151-153); IR (KBr,cm^{-1})3395(NH), 3040 (Ar-H), 1620 (C=N); ^{1}HNMR(400MHz,DMSO-d_6):δppm1.85(t,4H,2CH_2 of pyron ring),3.65(t,4H,CH_2-O-CH_2ofpyronring),4.20(s,2H,NH_2),5.13(s,1H,CH of pyrimidine ring),6.15(s,1H,NH of pyrimidine ring),6.64-6.65(m,4H,Ar-H),6.85(s,1H,NH of pyrimidine ring) ,7.93 (s,1H,=CH of pyrimidine),8.75 (s, 1H,O=C-NH); Anal.calcd (%) for $C_{16}H_{20}N_4O_3$: C 60.75%,H 6.37% and N 17.71% Found: C 60.55%, H 6.07% and N 17.51%.

Synthesis of (4-pyronphenyl)-2(thiones)1,2,3,4-tetrahydropyrimidine-5-carbohydrate (2h):

Yield (75%); m.p (191-192); IR (KBr,cm^{-1})3386(NH), 3040 (Ar-H), 1698 (C=O),1269 (C=S); ^{1}HNMR(400MHz,DMSO-d_6):δppm1.85(t,4H,2CH_2 of pyron ring),3.65(t,4H,CH_2-O-CH_2 of pyron ring),4.00(s,1H,CH of pyrimidine),4.20(s,2H,NH_2), 6.45-6.65(m,4H,Ar-H),6.50(s,1H,NHofpyrimidinering),6.70(s,1H, =CH of pyrimidine),8.75(s,1H,O=C-

NH),13.76 (s,1H,NH of pyrimidine).; Anal.calcd (%) for $C_{16}H_{20}N_4O_2S$: C 57.81%,H 6.06% and N 16.85% Found: C 57.51%, H 5.86% and N 16.1165%.

Synthesis of (4-pyronphenyl)-2(imines) 1,2,3,4-tetrahydropyrimidine-5-carbohydrate (2i):

Yield (65%); m.p (151-153); IR (KBr,cm^{-1})3378(NH), 3040 (Ar-H), 1698 (C=O),1618 (C=N); ^{1}HNMR(400MHz,DMSO-d_6):δppm 1.85(t,4H,2CH_2 of pyron ring),3.65(t,4H,CH_2-O-CH_2 of pyron ring),4.10(s,1H,CH of pyrimidine),4.20(s,2H,NH_2), 6.42-6.65(m,4H,Ar-H),6.50(s,1 H, NH of pyrimidine ring),6.70 (s,1H,=CH of pyrimidine),8.75(s,1H,O=C-NH),13.75(s,1H,NH of pyrimidine); Anal.calcd (%) for $C_{16}H_{21}N_5O_2$: C 60.94%,H 6.71% and N 22.21% Found: C 60.90%, H 6.51% and N 22.01%.

Physical, analytical and spectral data of compounds (3a-i)

Synthesis of (4-diethylmethylphenyl)-2(oxo) 1,2,3,4-tetrahydropyrimidine-5carbonyl)-3-(trifluro methyl) -1H-pyrazol-5(4H)-one (3a):

Yield (70%); m.p (174-176); IR (KBr,cm^{-1}) 3594(OH),3328(N-H),3040(strecthing of Ar-H), 1655(C=O),1597(C=N);^{1}HNMR(400MHz,DMSO-d_6):δppm1.15(t,6H,2(-CH_3) groups),2.62 (s,2H,CH_2 of pyrazole ring),3.21(q,4H,CH_2-N-CH_2),5.13(s,1H, CH of pyrimidine ring),5.35(s,1H,-OH),6.15 (s,1H,NH-C=O of pyrimidine ring),6.22(s,1H,Ar-H),6.30(d,1H,Ar-H),6.85(s,1H,NH of pyrimidine ring),6.94(d,1H,Ar-H),7.93(s,1H,=CH of pyrimidine ring);^{13}CNMR(75MHz,DMSO-d_6δppm):150.2,48.4,116.0,133.7,112.3,129.2,106.3,149.0, 98.8, 154.9,12.9,47.1,166.3,163.0,22.0,155.6, 126.2, and these signals are due to C_1, C_2 , C_3, C_4, C_5 C_6 ,C_7, C_8, C_9,C_{10}, C_{11}&C_{14}, C_{12}& C_{13}, C_{15}, C_{16}, C $_{17}$, C_{18} and C_{19} Carbon atoms respectively; Anal.calcd (%) for $C_{19}H_{20}F_3N_5O_4$: C 48.89%,H 4.50% and N 12.87% Found: C 48.59%, H 4.30% and N 12.57%.

Synthesis of (4-diethylmethylphenyl)-2(thiones) 1,2,3,4-tetrahydropyrimidine-5carbonyl)-3-(trifluro methyl) -1H-pyrazol-5(4H)-one (3b):

Yield (70%); m.p (185-187); IR (KBr,cm^{-1})3600(OH),3343(N-H),3040(strecthing of Ar-H), 1691(C=O),1592(C=N);^{1}HNMR(400MHz,DMSO-d_6):δppm1.15(t,6H,2(-CH_3)groups),2.62(s ,2H,CH_2ofpyrazolering),3.21(q,4H,CH_2-N-CH_2),4.00(s,1H,CH of pyrimidine ring),5.35(s,1H,-OH),6.15(s, 1H, NH-C=O of pyrimidine ring),6.22(s,1H,Ar-H),6.30(d,1H,Ar-H),6.53(s,1H,NH of pyrimidine ring),6.70(s,1H,=CHofpyrimidinering),6.95 (d,1H,Ar-H), 13.76(s,1H,NH of pyrimidine ring).;^{13}CNMR(75MHz,DMSO-d_6δppm):174.1,53.5,113.8,146.7,129.2,105.3,149.0, 98.8,154.9, 112.3,

12.9,47.1,166.3,163.0,22.0,155.6,126.2, and these signals are due to C_1, C_2 , C_3, C_4, C_5 C_6 ,C_7, C_8, C_9,C_{10}, C_{11}&C_{14}, C_{12}& C_{13}, C_{15}, C_{16}, C $_{17}$, C_{18} and C_{19} Carbon atoms respectively; Anal.calcd (%) for $C_{19}H_{20}F_3N_5O_3S$: C 48.89%,H 4.50% and N 12.87% Found: C 48.59%, H 4.30% and N 12.57%.

Synthesis of (4-diethylmethylphenyl)-2(imines) 1,2,3,4-tetrahydropyrimidine-5carbonyl)-3-(trifluro methyl) -1H-pyrazol-5(4H)-one (3c):

Yield (75%); m.p (128-130); IR (KBr,cm^{-1}) 3186(N-H),3040(strecthing of Ar-H), 2940 and 2895(aliphatic –CH(str) of CH_3&CH_2 groups), 1664(C=O),1256(P=O), 1041(C-O),and 755cm^{-1}(P-O);^{1}HNMR(400MHz, DMSO-d_6):δppm 1.15(t,6H,2(-CH_3) groups),3.21(q,4H,CH_2-N-CH_2),4.10(s,1H,CH of pyrimidine ring),4.20(s,2H, NH_2),5.35 (s,1H,-OH), 6.15 (s,1H,NH-C=Oof pyrimidine ring),6.22(s,1H,Ar-H),6.30(d,1H,Ar-H),6.50(s,1H,NH of pyrimidine ring), 6.70(s,1H ,=CHofpyrimidine ring),6.95(d,1H,Ar-H),8.75 (s,1H,NH-C=O), 10.2(s,1H,=NH),13.76(s,1H,NH ofpyrimidinering);^{13}CNMR(75MHz,DMSO-d_6δppm):150.2,54.6,116.0,133.7,126.1,112.7, 150.8, 132.8, 50.6, 25.5,166.3,163.0,22.0,155.6,126.2, and these signals are due to C_1, C_2 , C_3, C_4, C_5,& C_9 C_6& C_8 ,C_7, C_{10}, C_{11}&C_{14}, C_{12}& C_{13}, C_{15}, C_{16}, C $_{17}$, C_{18} and C_{19} Carbon atoms respectively. Anal.calcd (%) for $C_{19}H_{21}F_3N_6O_3$: C 52.05%, H 4.83% and N 19.17% Found: C 51.75%, H 4.63% and N 19.07%.

Synthesis of (4-pyrrolidinephenyl)-2(oxo) 1,2,3,4-tetrahydropyrimidine-5carbonyl)-3-(trifluro methyl) -1H-pyrazol-5(4H)-one (3d):

Yield (69%); m.p (180-182); IR (KBr,cm^{-1}) 3186(N-H),3040(strecthing of Ar-H), 2940 and 2895(aliphatic –CH(str) of CH_3&CH_2 groups), 1664(C=O),1256(P=O), 1041(C-O),and 755cm^{-1}(P-O);^{1}HNMR(400MHz,DMSO-d_6):δppm1.92(t,4H,2CH_2ofpyrolidinering),2.62(s,2H, CH_2 of pyrazole ring), 3.44(t,4H,CH_2-N-CH_2 of pyrolidine ring), 5.13(s,1H,CH of pyrimidine ring),6.15 (s,1H,NH of pyrimidine ring),6.55(s, 1H,NHofpyrimidine ring),6.45-6.65(m,4H,Ar-H),7.93(s,1H,=CH of pyrimidine ring);^{13}CNMR (75MHz, DMSO-d_6 δppm) :174.1,59.7,113.8,146.7,126.1,112.1,150.8,50.6,25.5, 166.3, 163.0, 22.0,155.6,126.2, and these signals are due to C_1, C_2 , C_3, C_4, C_5,& C_9 C_6& C_8 ,C_7, C_{10}, C_{11}&C_{14}, C_{12}& C_{13}, C_{15}, C_{16}, C $_{17}$, C_{18} and C_{19} Carbon atoms respectively.; Anal.calcd (%) for $C_{19}H_{18}F_3N_5O_3$: C 54.16%,H 4.31% and N 16.62% Found: C 54.06%, H 4.01% and N 16.32%.

Synthesis of (4-pyrrolidinephenyl)-2(thiones) 1,2,3,4-tetrahydropyrimidine-5carbonyl)-3-(trifluro methyl) -1H-pyrazol-5(4H)-one (3e):

Yield (65%); m.p (175-176); IR (KBr,cm^{-1}) 3186(N-H),3040(strecthing of Ar-H), 2940 and 2895(aliphatic –CH(str) of CH_3&CH_2 groups), 1664(C=O),1256(P=O), 1041(C-O),and 755cm^{-1}(P-O);^{1}HNMR(400MHz,DMSO-d_6):δppm1.92(t,4H,2CH_2ofpyrolidinering), 2.62(s,2H,CH_2 of pyrazole ring),3.44 (t,4H,CH_2-N-CH_2 of pyrolidinering),4.00(s,1H,CH of pyrimidine ring),4.20 (s, 2H, NH_2), 6.52 (s,1H, NH of pyrimidinering),6.42-6.65(m,4H,Ar-H),6.70 (s,1H, =CH of pyrimidine ring), 13.76(s,1H, NH of pyrimidine ring).;^{13}CNMR (75MHz, DMSO-d_6δppm): 174.1,59.7,113.8,146.7,126.1,112.1,150.8,50.6,25.5,166.3,163.0,22.0,155.6,126.2, and these signals are due to C_1, C_2 , C_3, C_4, C_5,& C_9 C_6& C_8 ,C_7, C_{10}, C_{11}&C_{14}, C_{12}& C_{13}, C_{15}, C_{16}, C_{17}, C_{18} and C_{19} Carbon atoms respectively; Anal.calcd (%) for $C_{19}H_{18}F_3N_5O_2S$: C 52.17%,H 4.15% and N 16.01% Found: C 52.07%, H 4.05% and N 15.81%.

Synthesis of (4-pyrrolidinephenyl)-2(imines)1,2,3,4-tetrahydropyrimidine-5carbonyl)-3-(trifluro methyl) -1H-pyrazol-5(4H)-one (3f):

Yield (75%); m.p (204-206); IR (KBr,cm^{-1}) 3186(N-H),3040(strecthing of Ar-H), 2940 and 2895(aliphatic –CH(str) of CH_3&CH_2 groups), 1664(C=O),1256(P=O), 1041(C-O),and 755cm^{-1}(P-O);^{1}HNMR(400MHz,DMSO-d_6):δppm1.92(t,4H,2CH_2ofpyrolidinering), 2.62(s,2H, CH_2 of pyrazole ring),3.44(t,4H,CH_2-N-CH_2 of pyrolidine ring), 4.10(s,1H,CH of pyrimidine ring), 6.50(s,1H,NH of pyrimidine ring),6.42-6.65(m,4H,Ar-H),6.70(s,1H,=CH of pyrimidine ring), 9.89(s,1H,=NH),13.76(s,1H, NH of pyrimidine ring); ^{13}C-NMR (75MHz, DMSO-d_6 δppm):150.2,54.6,116.0,133.7,127.0,127.9,139.0,140.6,33.0,66.6,45.7,166.3,163.0, 22.0, 155.6, 126.2, and these signals are due to C_1, C_2 , C_3, C_4, C_5,& C_9 C_6& C_8 ,C_7, C_{10}, C_{11}&C_{14}, C_{12}& C_{13}, C_{15}, C_{16}, C_{17}, C_{18} and C_{19} Carbon atoms respectively; Anal.calcd (%) for $C_{19}H_{19}F_3N_6O_2$: C 54.28%,H 4.56% and N 19.99% Found: C 54.08%, H 4.36% and N 19.69%.

Synthesis of (4-pyronphenyl)-2(oxo) 1,2,3,4-tetrahydropyrimidine-5carbonyl)-3-(trifluro methyl) -1H-pyrazol-5(4H)-one (3g):

Yield (65%); m.p (168-170); IR (KBr,cm^{-1}) 3186(N-H),3040(strecthing of Ar-H), 2940 and 2895(aliphatic –CH(str) of CH_3&CH_2 groups), 1664(C=O),1256(P=O), 1041(C-O),and 755cm^{-1}(P-O);^{1}HNMR(400MHz,DMSO-d_6):δppm1.85(t,4H,2CH_2ofpyron ring), 2.62(s,2H,CH_2 of pyrazole ring),3.65(t,4H,CH_2-O-CH_2 of pyron ring), 5.13(s,1H,CH of pyrimidine ring),6.15(s,1H,NH of pyrimidine ring),6.64-6.65(m,4H,Ar-H),6.85(s,1H,NH of

pyrimidine ring),7.93(s,1H,=CH of pyrimidine);^{13}CNMR (75MHz,DMSO-d_6 δppm):150.2,54.6,116.0,133.7, 127.0,127.9,139.0,140.6,33.0, 66.6,45.7,166.3,163.0,22.0,155.6,126.2, and these signals are due to C_1, C_2 , C_3, C_4, $C_{5,&}$ C_9 $C_{6&}$ C_8 ,C_7, C_{10}, C_{11}&C_{14}, C_{12}& C_{13}, C_{15}, C_{16}, C $_{17}$, C_{18} and C_{19} Carbon atoms respectively.; Anal.calcd (%) for $C_{20}H_{19}F_3N_4O_4$: C 48.89%,H 4.50% and N 12.87% Found: C 48.59%, H 4.30% and N 12.57%.

Synthesis of (4-pyron phenyl)-2(thiones) 1,2,3,4-tetrahydropyrimidine-5carbonyl)-3-(trifluro methyl) -1H-pyrazol-5(4H)-one (3h):

Yield (65%); m.p (157-159); IR (KBr,cm^{-1}) 3186(N-H),3040(strecthing of Ar-H), 2940 and 2895(aliphatic –CH(str) of CH_3&CH_2 groups), 1664(C=O),1256(P=O), 1041(C-O),and 755cm^{-1}(P-O);^{1}HNMR(400MHz,DMSO-d_6):δppm1.85(t,4H,2CH_2 of pyron ring), 2.62(s,2H,CH_2 of pyrazole ring),3.65(t,4H,CH_2-O-CH_2 of pyron ring),4.00(s,1H,CH of pyrimidine),6.45-6.65(m,4H,Ar-H),6.50(s,1H,NH of pyrimidinering),6.70(s,1H,=CHofpyrimidine),13.76(s,1H,NH of pyrimidine) ;^{13}CNMR (75MHz, DMSO-d_6 δppm):174.6,59.7,113.8,146.7,127.0,127.9,139.0, 140.6, 33.0, 66.6,45.7,166.3,163.0,22.0,155.6,126.2, and these signals are due to C_1, C_2 , C_3, C_4, $C_{5,&}$ C_9 $C_{6&}$ C_8 ,C_7, C_{10}, C_{11}&C_{14}, C_{12}& C_{13}, C_{15}, C_{16}, C $_{17}$, C_{18} and C_{19} Carbon atoms respectively; Anal.calcd (%) for $C_{20}H_{19}F_3N_4O_3S$: C 53.09%,H 4.23% and N 12.38% Found: C 52.78%, H 4.03% and N 12.08%.

Synthesis of (4-pyronphenyl)-2(imines)1,2,3,4-tetrahydropyrimidine-5carbonyl)-3-(trifluro methyl) -1H-pyrazol-5(4H)-one (3i):

Yield (65%); m.p (190-192); IR (KBr,cm^{-1}) 3186(N-H),3040(strecthing of Ar-H), 2940 and 2895(aliphatic –CH(str) of CH_3&CH_2 groups), 1664(C=O),1256(P=O), 1041(C-O),and 755cm^{-1}(P-O);^{1}HNMR(400MHz,DMSO-d_6):δppm1.85(t,4H,2CH_2 of pyron ring), 2.62(s,2H,CH_2 of pyrazole ring),3.65(t,4H,CH_2-O-CH_2 of pyron ring),4.10(s,1H,CH of pyrimidine), 6.42-6.65(m,4H,Ar-H),6.50(s,1H,NH of pyrimidine ring),6.70 (s,1H, =CH of pyrimidine),13.75(s, 1H,NH of pyrimidine);^{13}CNMR(75MHz,DMSO-d_6δppm): 153.3,50.2,113.8,146.7,127.0,127.9, 139.0, 140.6,33.0,66.6,45.7,166.3,163.0,22.0,155.6,126.2, and these signals are due to C_1, C_2 , C_3, C_4, $C_{5,&}$ C_9 $C_{6&}$ C_8 ,C_7, C_{10}, C_{11}&C_{14}, C_{12}& C_{13}, C_{15}, C_{16}, C $_{17}$, C_{18} and C_{19} Carbon atoms respectively. Anal.calcd (%) for $C_{20}H_{20}F_3N_5O_3$: C 55.17%, H 4.63% and N 16.08% Found: C 55.07%, H 4.33% and N 15.78%.

Biological activity:

Antimicrobial activity of these newly synthesized compounds was performed according to disc diffusion method, as recommended by the national committee of clinical laboratory. The synthesized compounds were used at the concentration of 250 µg/ml DMF as a solvent.

Antimicrobial activity:

Antibacterial activity the newly synthesized Organo pyrazole 5-one pirimidines (**3a-i)** exhibit moderate antibacterial activity against the *Staphylococcus aureus* NCCS 2079, *BacillusCerus* NCCS 2106, *Escherichia coli* NCCS 2065 and *Pseudomonas aeruginosa* NCCS 2200 at the concentration of 250µg/disc. In this series structures consisting of of **3d, 3g** and **3i** have shown increased effect on their antibacterial activity. The decreasing Oder of antibacterial activity of **(3a-i)** is as follows **"3d>3g>3i>3e=3h>3f>3c>3a=3b".**

Table 1: Antibacterial activity of newly synthesized compounds 3(a-i)

Comp no	**R**	**Zone of inhibition (mm)**			
		Staphylococu saureusNCCS2079250(µg/ml)	***BacillusCerus NCCS2106 250(µg/ml***	***Escherichia ColiNCCS2065 250(µg/ml)***	***Pseudomonas aeruginosa NCCS2200250(µg/ml)***
3a		19	12	17	18
3b		18	13	16	17
3c		20	13	17	18
3d		22	16	18	20
3e		20	15	17	18

3f		19	14	15	17
3g		21	16	18	19
3h		19	14	16	18
3i		18	13	14	17
Amoxicillin		27	24	22	25

Antifungal activity:

Antifungal activity pyrazole 5-one pirimidines (3**a-i)** exhibited moderate antifungal activity against the *Aspergillus niger* NCCS1196 and *Candida albicans* NCCS 3471 at the concentration of 250µg/disc. In this series structures consisting of **3d, 3g** and **3e** have shown increased effect on their antibacterial activity. The decreasing Oder of antibacterial activity of **(3a-i)** is as follows **"3d>3g=3e>3a>3f=3h>3i>3b>3c".**

Table 2: Antifungal activity of newly synthesized compounds 3a-i

Com no	**R**	**Zone of inhibition (mm)**	
		Aspergillus niger NCCS1196250(µg/ml)	***Canadidaalbicans NCCS 3471 250(µg/ml)***
3a		18	16
3b		13	12
3c		11	9
3d		20	18

3e		19	16
3f		17	14
3g		19	17
3h		17	15
3i		14	13
Ketoconazole		22	25

Docking studies Of Pyrazole5-one pirimidines with sortaseA

Synthesis and characterization of Synthesis of (4-diethylamine/4-pyrrolidine/4-pyronphenyl)-2(oxo/thiones/imines)1,2,3,4- tetrahydropyrimidine-5 carbonyl)-3-(trifluromethyl)-1H-pyrazol-5(4H)-one (3a-i).

Table3: Docking results of 3(a-i) on sortase A staphylococcus protein

Com no	R	X	Fittness	S(hb-ext)	S(vdw-ext)	S(hb-int)	S(int)
3a		O	20.96	11.31	23.42	0.00	-22.56
3b		S	20.58	0.52	28.99	0.00	-19.79
3c		NH	24.53	2.85	29.08	0.00	-18.30
3d		O	39.90	0.00	31.26	0.00	-3.08
3e		S	34.66	2.00	26.43	0.00	-3.68
3f		NH	30.94	5.02	20.04	0.00	-1.64
3g		O	37.53	0.38	28.04	0.00	-1.41
3h		S	34.55	0.00	27.55	0.00	-3.33
3i		NH	35.09	7.14	21.43	0.00	-1.53

RESULTS AND DISCUSSIONS

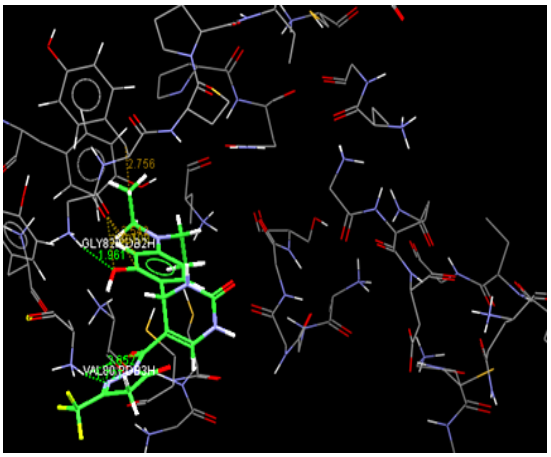
The docking studies of 3(a-i) were carried out on sortase A staphylococcus (PDB ID: 1T2P). The docking ligands were found to have some interations between an oxygen atom of the ligands and sortase A staphylococcus protein. Moreover, these docked conformations formed hydrogen bond interactions with the active site of the protein. Bind pocket, common hydrogen bonding interactions were for formed between all the docked ligands and TYR89, CYS77, GLY82PDB, MET84, GLN364PDB, VAL80PDB.The order of protein-ligand hydrogen bond score is3a>3i>3f>3c>3e>3b>3d=3h.Besides hydrogen bonding interaction between ligand-protein,the vanderwalls interations between ligand-protein were also noticed. The order of protein-ligand vanderwaals score 3d>3c>3b>3g>3h>3e>3a>3f of interaction with the protein. However the ligands fails to exhibit intramolecular hydrogen bonding with the ligand. The ligands exhibit minimum intramolecular strain. Finally, all the ligands exhibit moderate to good antimicrobial activity with sortaseA staphylococuus protein. The order of gold score fitness value of the ligands is 3d>3g>3i>3e>3h>3f>3c>3a>3b According to gold score fitness value ligand 3d,3g exhibits high binding activity with the protein and ligand 3b showed leads binding activity with the the protein.

Table 4: Hydrogen bonding interactions of compounds (3a-i) with sortase A staphylococcus

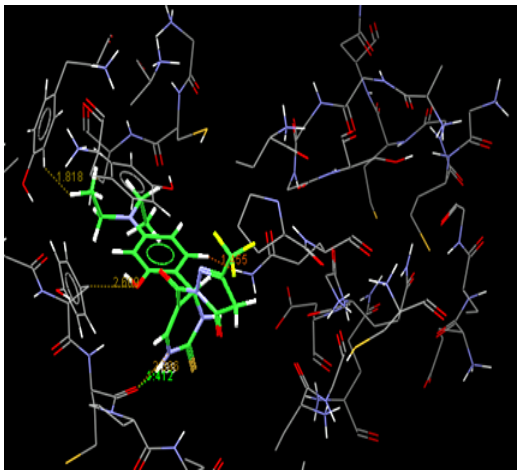
COMP NO	R	X	Number ofhydrogen bonds	Atom		Bond Length (A^0)	Fitness
				protein	Atom		
3a		O	2	GLY82PDB VAL80PDB	2(C=O) 9(C=O)	1.961 2.852	20.96
3b		S	2	GLY82PDB VAL80PDB	2(C=O) 9(C=O)		20.58
3c		NH	2	GLY82PDB CYS77	2(C=O) 9(C=O)	2.154 2.474	24.53
3d		O	1	MET840	2(C=O)	-	39.90
3e		S	2	MET84 ILE86	2(C=O) 9(C=O)	2.098 2.534	34.66

3f		N H	1	GLY82PDB	2(C=O) 9(C=O)	2.236 1.877	30.94
3g		O	2	LYR67 MET84	2(C=O) 9(C=O)	2.611 2.636	37.53
3h		S	1	LE312	2(C=O)	2.224	34.55
3i		N H	2	GLY82PDB GLY82	2(C=O) 9(C=O)	2.381 1.405	35.09

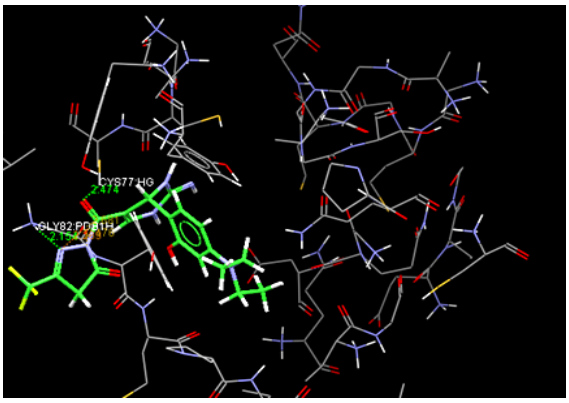
3a.



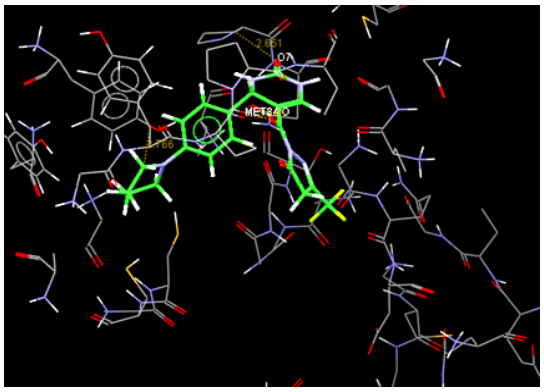
3b.



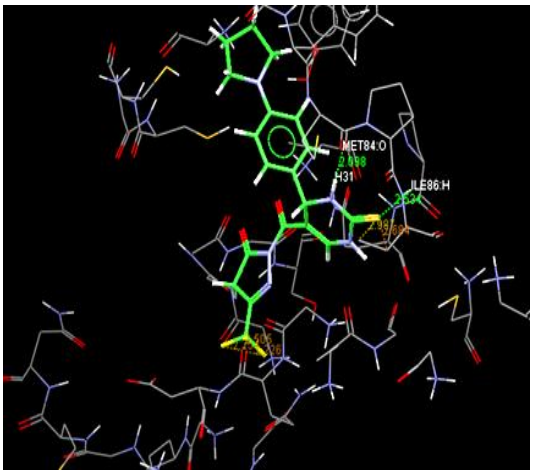
3c.



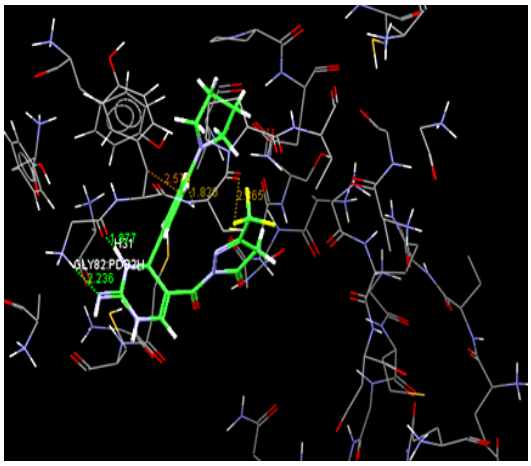
3d.



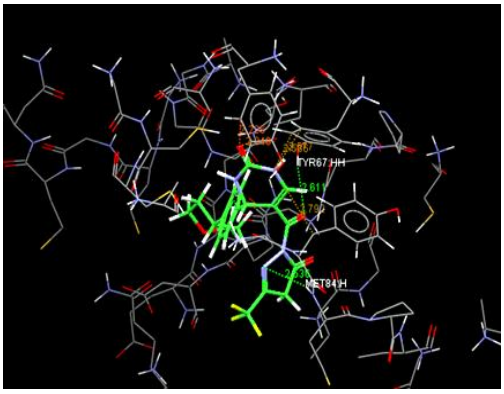
3e.


MET84:O
H31
ILE86:H

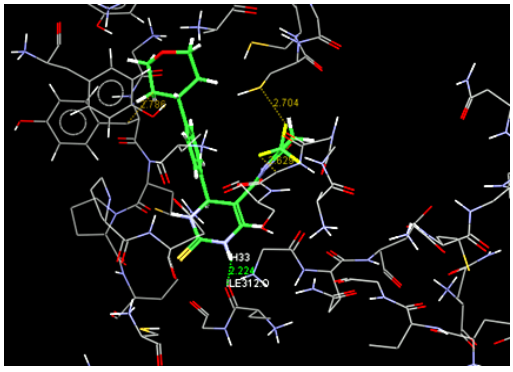
3f.


H31
GLY82:PDB3H

3g.


TYR67:HH
MET84:H

3h.


2.704
H33
ILE312:O

3i.

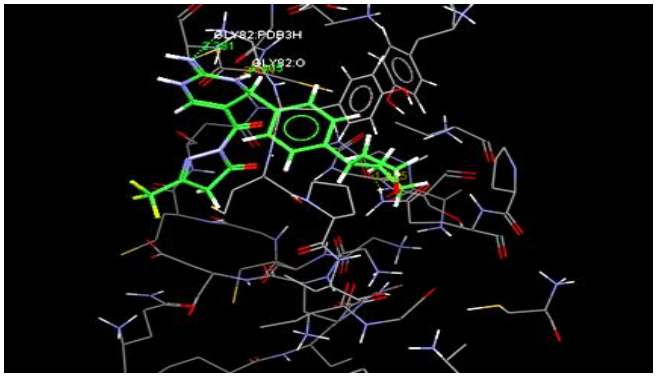

GLY82:PDB3H
GLY82:O

Figure :Docking images of compounds 3(a-i)

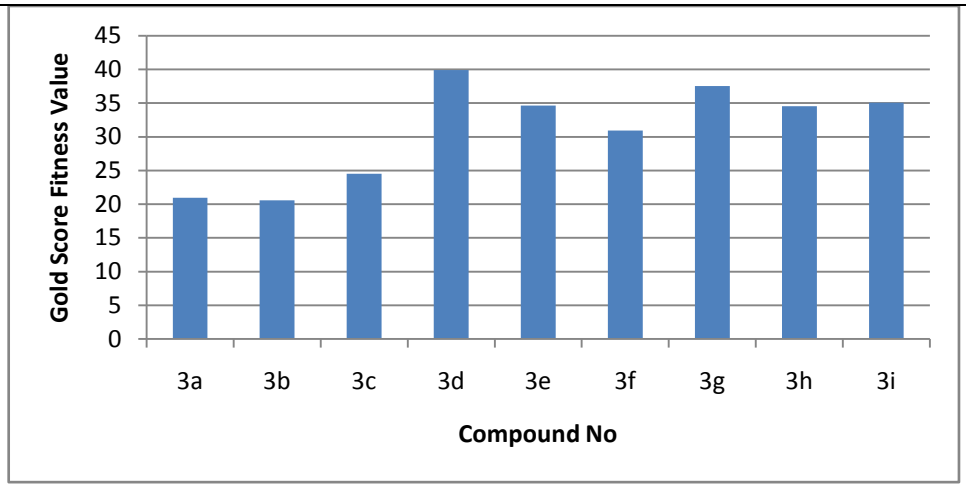


Figure :comparative Gold score fitness values for compounds 3(a-i)

CONCLUSION

The newly synthesized compounds pyrazole 5-one containing pirimidines 3(a-i) were found to be active in study of anti-bacterial, anti-fungal activity. It can be concluded that this class of compounds certainly holds great promise to discover novel classes of antimicrobial agents.

ACKNOWLEDGEMENTS

The authors are grateful to Pro.L.K.Ravindranath professor in deportment of chemistry, S.K. University, Anantapur for providing necessary facilities for this work. They are also thanks to IICT Hyderabad and CDRI Lucknow for spectral and analytical data**.**

REFERENCES

1. Kappe, C.O. *Tetrahydron*, 1993, *49*, 6937.
2. Grover, G.I.; Dzwonczyk, S.; McMullen, D.E,; Parham,C.S.; Sleph, P.G.; Moreland, S. *J. Cardiovasc, Pharmacol.* 1995, 26, 289.
3. Bahekar, S. S.; Shinde, D. B.; *Bioorg. Med. Chem. Lett*. 2004,*14*,1733.
4. Sujatha, K.; Shanmugam, P.; Perumal, P. T.; Muralidharan, D.; Rajendran, M. *Bioorg. Med. Chem. Lett*. 2006, *16*, 4893.
5. Ashok, M.; Holla, B. S.; Kumari, N. S.; *Eur. J. Med. Chem*. 2007,*42*,380.
6. Basso, A. M.; Bratcher, N. A.; Gallagher, K. B.; Cowart, M. D.; Zhao, C.; Sun, M.; Esbenshade, T. A.; Brune, M. E.; Fox, G. B.; Schmidt, M.; Collins, C. A.; Souers, A. J.; Iyengar, R.; Vasudevan, A.; Kym, P. R.; Hancock, A. A.; Rueter, L. E.; *Eur. J. Pharmacol*. 2006, *540*, 115.
7. Wisen, S.; Androsavich, J.; Evans, C. G.; Gestwicki, J. E.; *Bioorg. Med. Chem. Lett*. 2008, *18*, 60.
8. Deres, K.; Schröder, C. H.; Paessens, A.; Goldmann, S.; Hacker, H. J.; Weber, O.; Krämer, T.; Niewöhner, U.; Pleiss, U.; Stoltefuss, J.; Graef, E.; Koletzki, D.; Masantschek, R. N. A.; Reimann, A.; Jaeger, R.; Groβ, R.; Beckermann, B.; Schlemmer, K.-H.; Haebich, D.; Rübsamen-Wagmann, H.; *Science* 2003, *299*, 893.
9. Blackburn, C.; Guan, B.; Brown, J.; Cullis, C.; Condon, S. M.; Jenkins, T. J.; Peluso, S.;Ye, Y.; Gimeno, R. E.; Punreddy, S.; Sun, Y.; Wu, H.; Hubbard, B.; Kaushik, V.; Tummino, P.; Sanchetti, P.; Sun, D. Y.; Daniels, T.; Tozzo, E.; Balani, S. K.; Raman, P.; *Bioorg. Med. Chem. Lett*. 2006, *16*, 3504.
10. Armarego, W. L. F.; Perrin, D. D. *Purification of Laboratory Chemicals*, 4th edn., Butterworth, Heinemann, Oxford, 1997, *OX2*, 8DP.
11. Umamaheswari Devi, P.; Usha Rani, N.R.; Reddanna. P. *Eur. J. Plant. Path*. 2000, 106, 857.
12. Colle, J. G.; Duguid, J. P.; Firaser, A. G.; Mannion, B. P. *Mackie & Mecartney Practical Medicinal Microbiology*, 13th edn; Churchil : Edinburgh and London. 1989, Vol. 2.