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SYNTHESIS, BIOLOGICAL EVALUATION AND INSILICO STUDIES OF THIENO[2,3-d]PYRIMIDINE-1,2,3-TRIAZOLES

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Abstract: The design and development of novel therapeutic candidates is an ever-demanding area to tackle various communicable and non-communicable diseases. By considering the pharmaceutical importance of thienopyrimidines and 1,2,3-triazoles, in the present study we have attempted the synthesis of thieno[2,3-d]pyrimidine tethered 1,2,3-triazoles(**6a-j**) through click-chemistry approach in reliable yields (80-92%) under Cu (I) promoted azide-alkyne cycloaddition (CuAAC). Further all the synthesized compounds were successfully established through ^{1}H, ^{13}C NMR and Mass spectral characterization. Finally, we presume that our study could set a stage for the identification of potential hits which could facilitate the development a wide array of renowned therapeutic classes with a promising clinical significance.

Keywords: Click-chemistry, Azides Heterocycles, Thienopyrimidine-1, 2, 3-triazole.

Introduction:

Heterocyclic compounds are serving as crucial templates for several potent and efficacious drug classes. Even in the field of agricultural, heterocyclic derivatives have been employing as insect repellents, crop protection agents, and fungicides respectively. Systematic structural manipulation through synthesis is one of the most important processes in the journey of drug discovery and development resulting in clinical drug candidates or drug-like molecules [I, II] physicochemical/ pharmacological properties, Click-chemistry is the most gifted strategy which involves coupling of two different substrates with a choice of selective heterocyclic biomolecules and has some important advantages like simplicity in reaction handling, wide choice of substrates, insensitivity to oxygen/water, solvent/solvent-free reactions, stable end-products with high yields that are free of by-products, and easy product isolation [III-V]. The Huisgen's 1, 3-cycloaddition of azides and alkynes, yielding1,2,3-triazole is a key achievement in the azole chemistry[VI] which is extended to make complex natural products and a variety of heterocycles[VII]. In recent years, Cu (I) catalyzed azide-alkyne cycloaddition (CuAAC) has emerged as a powerful approach for the synthesis of new pharmacologically active compounds[VIII-X].

Among the various heterocyclic scaffolds, thienopyrimidine and 1,2,3-triazole are considered as pharmaceutically important scaffolds (**fig.1**) to exhibit biological activities i.e., antimicrobial[XI] anti-cancer[XII, XIII], anti-malaria[XIV, XV], anti-bacterial[XVI], anti-alzheimer[XVII, XVIII], anti-tubercular[XIX, XX], anti-viral[XXI, XXII], anti-microbial[XXIII], antiviral[XXIV], anti-inflammatory[XXV], antidiabetic[XXVI], antioxidant[XXVII], anti-fungal[XXVIII], bronchodilator[XXXIX] and anxiolytic[XXX] activities respectively. Inspired by the above facts, the present study was aimed to synthesize a series of thieno[2, 3-d]pyrimidine-1,2,3-triazole derivatives (**6a-j**) by integrating two important pharmacophores viz., 1,2,3-triazoles and thienopyrimidines through click-reaction.

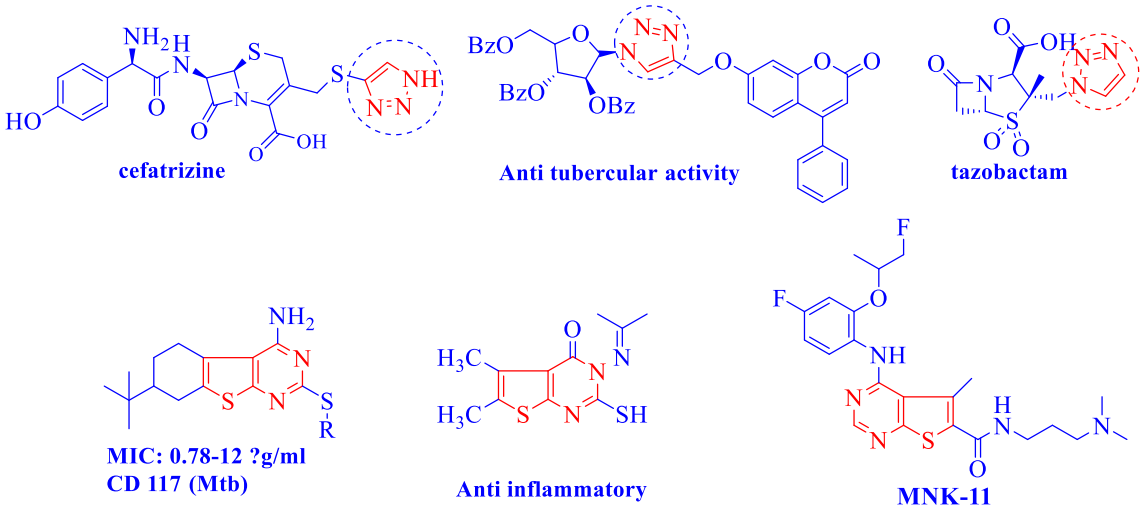


Fig. 1. Structures of some biologically potent thieno[2,3-d]pyrimidine and 1,2,3-triazole group containing drugs

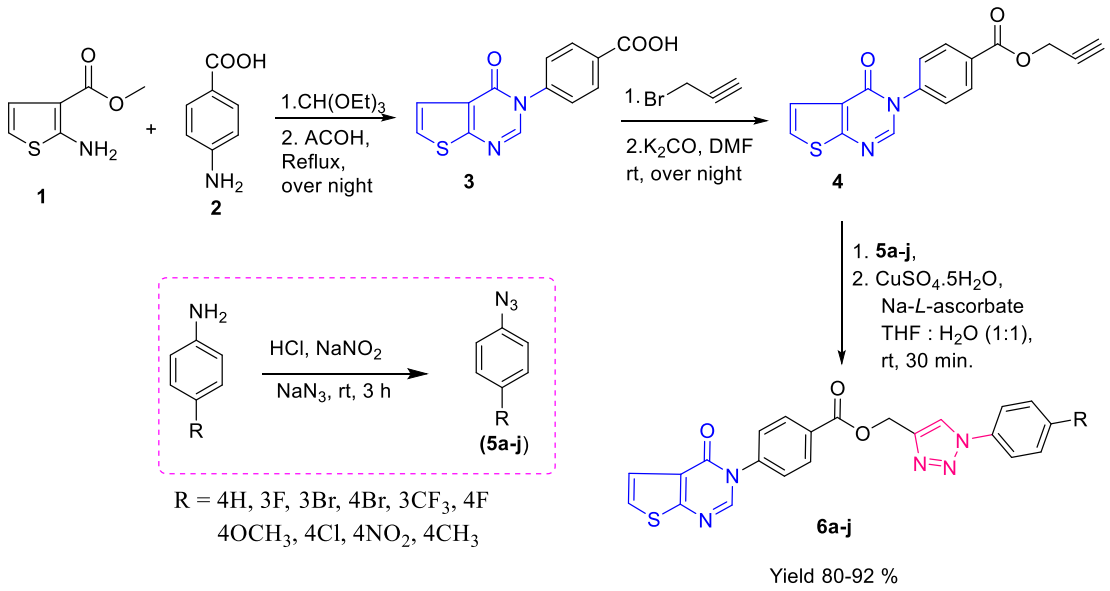
2. Results and discussion

2.1. Chemistry

We herein reported the feasible procedure for the expedient synthesis of thieno[2,3-d]pyrimidine tethered to 1,2,3-triazoles. The current synthesis involved the conventional processes of Huisgen's 1,3-cycloaddition, which generates 1,2,3-triazole in the presence of Cu (I) catalyst under mild conditions (**Scheme 1**). Moreover, **CuAAC** promoted azide-alkyne cycloaddition has emerged as a powerful approach in pharmaceutical applications for creating new biologically active heterocycles.

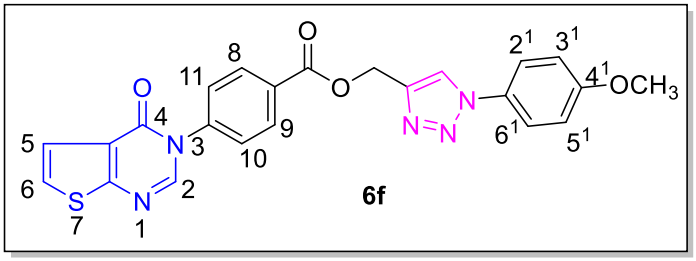
The starting material ethyl 3-cyanothiophene-2-carboxylate **1** was treated with para aminobenzoic acid **2** and triethyl orthoformate in the presence of acetic acid and refluxed at 110^0C which resulted in the formation of compound **3** which on treatment with propargyl bromide and K_2CO_3 in DMF at room temperature for 12 hours formed compound **4**. The respective aromatic azides **5a-j** were obtained on treatment of substituted anilines with sodium nitrite and sodium azide in conc. HCl through constant stirring ice-bath for 2 hrs. The reaction progress was monitored with the TLC under UV and I_2 vapors. The finally compounds **6a-j** were attained by the click-reaction of compound **4** with **5a-j** in the presence of $CuSO_4.5H_2O$, sodium-ascorbate in THF: H_2O (1:1) at rt over 1hr reaction time. The structures of newly synthesized thieno[2,3-d]pyrimidine-1,2,3-triazole derivatives (**6a-j**) were characterized by IR, LC-Mass, ^{1}H and ^{13}C NMR spectral studies.

Scheme 1



Scheme 1: synthesis of thieno[2,3-d]pyrimidine and 1,2,3-triazole derivatives (**6a-j**).

Spectral interpretation:



The structures of the newly synthesized (1H-1,2,3-triazol-4-yl) methyl 4-(4-oxothieno[2,3-d] pyrimidin-3(4H)-yl) benzoate derivatives **6a-j** were identified using ^{1}H & ^{13}C NMR, mass and IR spectral data. ^{1}H NMR spectra of **6a-j** showed singlets in the regions of 8.80 and 8.85 ppm, correspond to the protons of the pyrimidine CH and triazole ring CH groups, respectively. Aromatic protons didn't appear in the expected region, which was between 6.5 and 8.85 ppm. The mass spectra of the compounds showed (M+1) peaks and matched their chemical formulae. The IR spectra of compounds **6a-j** showed the specific absorption bands for the corresponding functional groups in the structure.

IR spectrum of compound **6f** showed characteristic band at 3016 (Ar-C-H, str), 1754 (C=O, str), 1664 (C=O, lactam), 1528 (C=N), groups respectively. The ^{1}H NMR spectra showed singlets at δ 8.83 & 8.81 corresponding to pyrimidine ring and triazole ring CH groups respectively. Among four A_2B_2 doublets two are for *p*-substituted benzoate ring one at δ 7.86 for H-8 & H-9 and other at δ 7.24 for H-10 & H-11 and another two are for triazole ring at δ 7.80 for H-2^{1}& H-6^{1} and δ 6.89 for H-3^{1} & H-5^{1}. Two doublets at δ 7.54 & 7.56 respectively are for H-6 & H-7 of thiophene ring and two singlets at δ 3.83 & 5.42 are corresponding to -OCH_3 and O-CH_2- groups respectively. ^{13}C NMR spectra of 165.38, 160.59, 159.83, 149.80, 143.13, 137.25, 131.06, 130.63, 130.33, 128.55, 125.74, 124.44, 123.47, 122.32, 119.09, 116.80, 115.32, 58.09, 56.00 ppm; LC-MS (Positive ion mode): m/z = 459 $(M+H)^+$ with its molecular formula ($C_{23}H_{17}N_5O_4S$).

ANTIMICROBIAL ACTIVITY:

The invitro antibacterial activity was tested using the agar well diffusion method[XXXI] against a series of two Gram-positive and Gram-negative bacteria, such as *Staphylococcus aureus*,

Bacillus subtilis, *Pseudomonas aeruginosa* and *Escherichia coli*. Streptomycin was used as the standard reference drug. Out of all the synthesized derivatives, according to **Table 1** results from the screening of anti-bacterial data, compounds **6g** (R=4-Cl) and **6j** (R=4-F) showed excellent activity against all of the tested bacterial strains, while compounds **6a** (R=4-H), **6d** (R=4-Br) and compound **6f** (R=4-OMe) demonstrated good activity in comparison to the standard drug streptomycin. The above-mentioned all microorganisms were tested and none of them showed any activity against **6b**, **6c**, **6e**, **6h** and **6i**. Activity was measured in zones exhibiting complete inhibition (mm) and growth inhibition calculations were done with reference to a positive control by taking each sample three times.

Table 1: Anti-bacterial of compounds **6a-j**

Compounds	Substituents R	Zone of Inhibition (mm)			
		Gram-positive (50 µg/ml)		Gram-negative (50 µg/ml)	
		B. subtilis	*S. aureus*	*E. coli*	*P. aeruginosa*
6a	-H	0.5	0.7	0.9	1.2
6b	-3F	-	-	-	-
6c	-3Br	-	-	-	-
6d	-4Br	**0.8**	**1.0**	**1.5**	**1.3**
6e	$-3CF_3$	-	-	-	-
6f	$-OCH_3$	0.5	0.8	1.0	0.9
6g	-4Cl	**1.1**	**1.1**	**1.8**	**3.1**
6h	$-4NO_2$	-	-	-	-
6i	$-4CH_3$	-	-	-	-
6j	-4F	**0.9**	**1.2**	**1.7**	**2.5**
Streptomycin		0.7	3.2	3.0	3.4

Bacillus subtilis, Staphylococcus aureus, Pseudomonas aeruginosa and Escherichia coli.

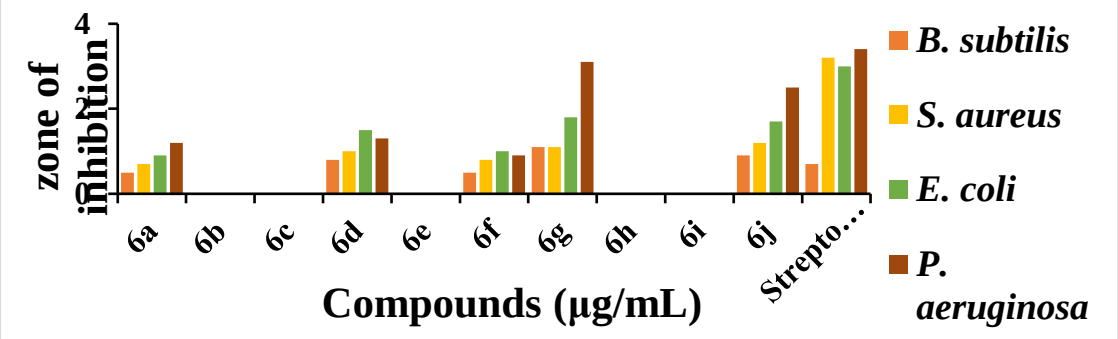


Figure: 2 Graphical representation of antimicrobial activity of compounds 6a-j

Molecular Docking Simulations

The docking simulations were performed using CB-Dock[XXXII] a reliable and free online tool. It mainly operates by automatically identifying the binding pockets and performs the docking through Auto Dock Vina. This tool works with accuracy by target's binding site prediction through curvature-based cavity detection approach. The two-dimensional (2D) structures of all the 10 query ligands were drawn using ChemBioDraw Ultra 14.0 and saved as MDLSD file (*.sdf) type. The selected co-crystal structure of type IV topoisomerase (PDB ID: 3RAE) with a resolution of 2.90A^{o} was downloaded and saved in PDB format. Further, after selecting the "dock" option in the home page, the files of the respective query ligands and the protein were uploaded in the space provided and submitted for docking. The number of cavities for docking was set as 5 in default. The output was generated as a set of five vina scores based on the decreasing order of the cavity sizes. Among those the first and foremost score should be taken into consideration as it has the highest negative value. The ciprofloxacin, an established drug of fluroquinolone class was used as a reference.

Molecular docking studies: The in-silico screening of all newly synthesized molecules were carried out by using CB-DOCK through Auto Dock Vina tools against the *Bacillus subtilis* bacteria. The majority of the new ligands have shown interactions with the target molecules of *Bacillus subtilis*.

Table2: Docking results of anti-microbial of compounds of 6a-j

S. No	Ligands	Binding energy	Binding site
1	**6a**	**-12.2**	SER79, ARG756, ARG117
2	6b	-10.7	SER79, ARG456, ASP506, ASP508
3	6c	-10.0	SER79, ASP506, ASP508
4	**6d**	**-12.2**	SER79, ARG456, ARG117
5	6e	-10.4	SFR79, ARG456
6	**6f**	**-11.7**	SER79, ARG117, GLY582
7	**6g**	**-12.6**	SER79, ARG117, ARG456, ASP506, ASP508, HIS76
8	6h	-10.7	SER79, ARG456,
9	6i	-10.3	SER79, ASP506, ASP508
10	**6j**	**-12.4**	SER79, ARG117, ARG456, ASP506, ASP508, GLY582, SER457
Ciprofloxacin		-9.2	R456, G457,

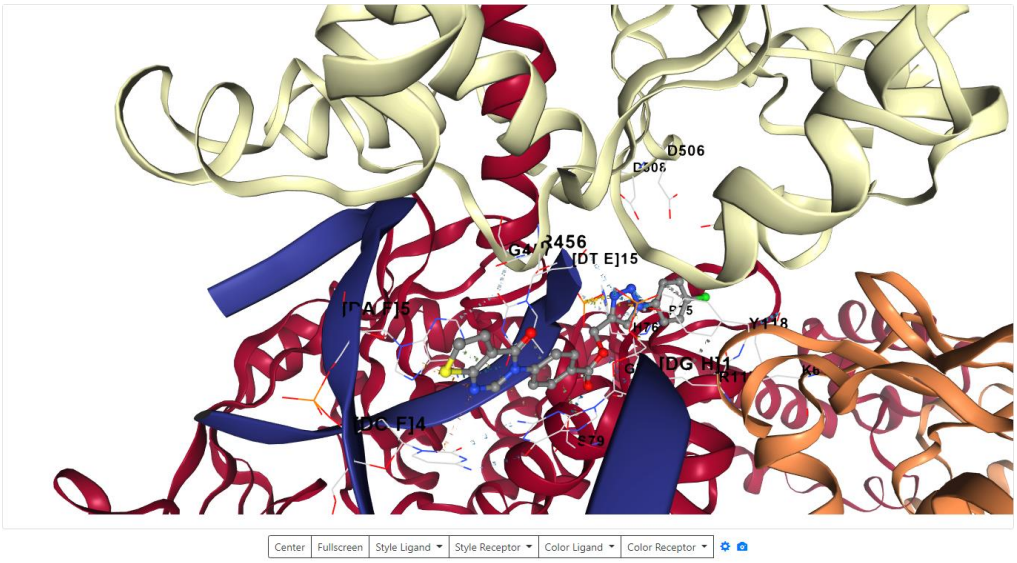


Figure: 2 Docking interaction of compound **6g** selected co-crystal structure of type IV topoisomerase (PDB ID: 3RAE).

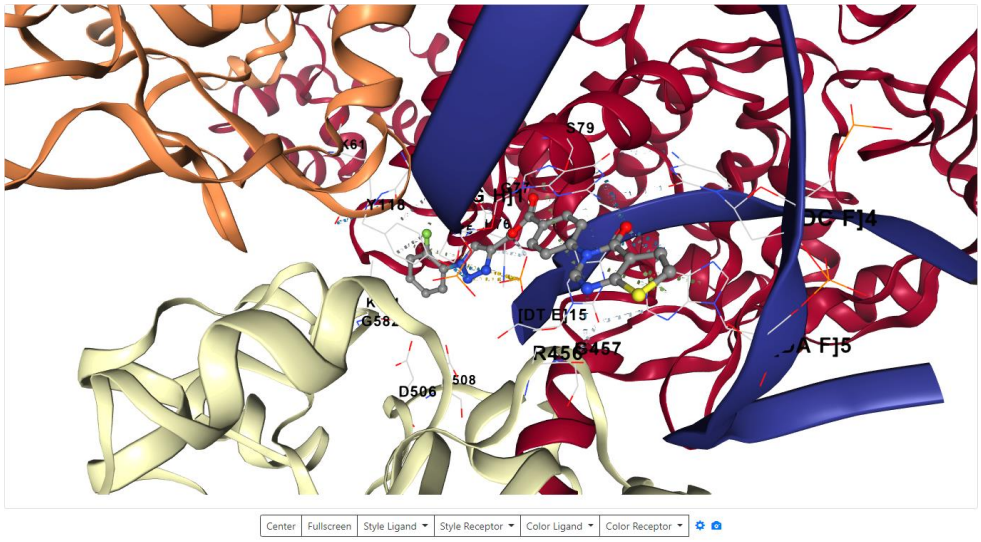


Figure: 3 Docking interaction of compound **6j** selected co-crystal structure of type IV topoisomerase (PDB ID: 3RAE).

Conclusion:

Here, in the present study we were able to synthesize ten new 1,2,3-triazole integrated thieno[2,3-d]pyrimidine derivatives **6a-j** through click-reaction in good yields 80-92%. Compounds **6g** and **6j** were potential towards anti-microbial activity which was also confirmed by molecular docking studies.

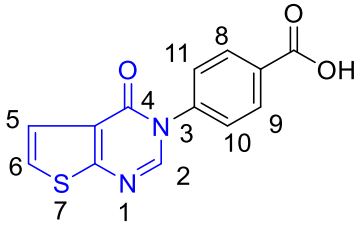
Experimental:

Melting points (^{o}C, uncorrected) were checked in open glass capillary tube by using auto melting point apparatus and found uncorrected. All the chemicals were analytical grade and purchased from Sigma Aldrich supplies used without further purification. The reactions were monitored by thin layer chromatography (TLC) on silica gel and column chromatography was performed on silica-gel (60-120 and 100-200 mesh) packed in a glass column. The ^{1}H and ^{13}C NMR were recorded in $CDCl_3$ on Jeol 400 MHz spectrometers using TMS as the internal

standard. FT-IR spectra were recorded on using KBr pellets. LCMS spectra were recorded on a micro-mass ESI QToF Premier instrument (UK).

General procedure for the synthesis of 4-(4-oxothieno[2,3-d]pyrimidin-3(4H)-yl) benzoic acid:

Compound **3** was obtained by the reaction of methyl 2-aminothiophene-3-carboxylate (1 gm, 6.37 mmol) **1** was treated with para aminobenzoic acid (0.87 mg, 6.37 mmol) **2** and triethyl orthoformate (12 ml) in the presence of acetic acid (4 ml) refluxing at 110^0C. After completion of reaction the mixture was poured into ice cold water and filtered to obtain solid.

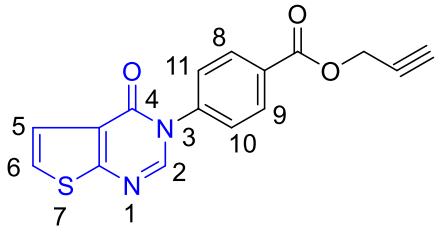


4-(4-Oxothieno [2, 3-d]pyrimidin-3(4H)-yl)benzoic acid

IR (KBr, ν, (cm $^{-1}$): 3400-2400 (O-H, str), 3016 (Ar-C-H), 1730-1700 (C=O, str), 1660 (C=O, lactam), 1617 (Ar- C=C), 1536 (C=N); ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 12 (s, 1H, -COOH), 8.02 (d, 2H, Ar-H_8, H_9), 8.52 (s, 1H, pyrimidine -CH), 7.52 (d, 1H, thiophene ring -H_6), 7.50 (d, 1H, thiophene ring -H_7), 7.35 (d, 2H, Ar-H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ 168.00, 162.01, 154.12, 141.80, 145.48, 134.52, 130.64, 126.71, 124.39, 123.07, 120.45.

Protocol for the synthesis of prop-2-yn-1-yl 4-(4-oxothieno[2,3-d]pyrimidin-3 (4H)-yl) benzoate

In a DMF (10ml) solution of compound **4** (0.500 mg, 1.84 mmol), K_2CO_3 (0.76 mg, 5.52 mmol) and 3-bromoprop-1-yne (0.24 mg, 2.02 mmol) were added sequentially and stirred for overnight at room temperature. After completion of the reaction, the reaction mixture was poured into crushed ice and the resulting solid was filtered and purified by column chromatography utilizing ethyl acetate and pet ether as eluents.



Prop-2-yn-1-yl 4-(4-oxothieno[2,3-d]pyrimidin-3(4H)-yl)benzoate

IR (KBr, ν, (cm $^{-1}$): 3018 (Ar-C-H), 1765 (C=O, str), 1661 (C=O, lactam), 1537 (C=N), ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 8.56 (s, 1H, pyrimidine -CH), 7.86 (d, 2H, Ar-H_8, H_9, J = 8 Hz), 7.55 (d, 1H, thiophene ring -H_6), 7.53 (d, 1H, thiophene ring -H_5), 7.28 (d, 2H, Ar-H_{10}, H_{11}, J = 8 Hz), 4.54 (s, 2H, -CH_2-OEt), 3.33 (s, 1H, alkyne -CH) ppm; ^{13}C NMR ($CDCl_3$, 100 MHz): δ165.62, 160.00, 157.76, 145.28, 142.32, 133.16, 130.32, 127.81, 125.81, 124.54, 120.15, 78.27, 76.53, 53.62 ppm.

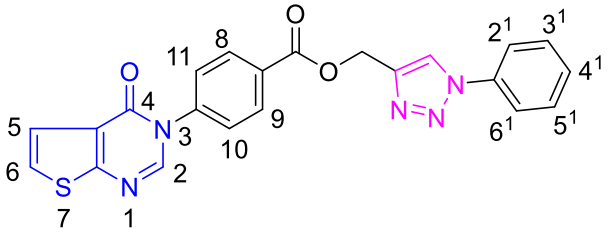
Synthesis of aryl azide (5a-j)

Respective substituted aniline (9.1 mmol) was dissolved in Conc. HCl (9.1 mmol) at room temperature and then cooled to 0^0C, before addition a sodium nitrite solution (45.5 ml). The reaction mixture was agitated for 10 min at 0-5^0C, followed by the addition of sodium azide (27.3 mmol), the mixture was continued stirring for another 2 hrs at room temperature. After completion of the reaction, it was extracted thrice (10 ml each) with pet-ether. The organic layer was dried over anhydrous sodium sulphate and the solvent was vacuum distilled to get the azide derivatives **(5a-j).**

General procedure for 6a-j

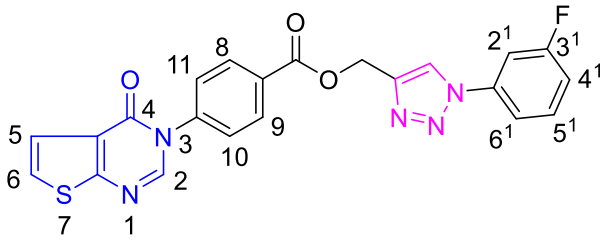
Compound **4** (0.500 mg, 1.61 mmol) and compound **5a-j** (0.31 mg, 1.61 mmol) were dissolved in 5ml of THF: H_2O (1:1 mixture) solvent. Next sodium ascorbate (0.95 mg, 4.83 mmol) and $CuSO_4.5H_2O$ (1.20 gm, 4.83 mmol) were added and the reaction mixture was stirred for 1 hr. The reaction mixture was poured into crushed ice and the obtained solid was filtered and dried. All the compounds were subjected to silica gel (60-120 mesh) column chromatography purification through gradient elution by a combination of pet-ether and ethyl acetate.

(1-Phenyl-1H-1,2,3-triazol-4-yl) methyl 4-(4-oxothieno[2,3-d]pyrimidin-3(4H)-yl)benzoate (6a)



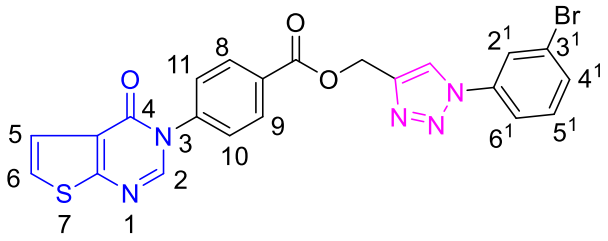
Light cream solid, Yield: 85%, mp: 110-111 °C, IR (KBr, ν, (cm $^{-1}$): 3016 (Ar-C-H), 1763 (C=O, str), 1632 (C=O, lactam), 1530 (C=N), ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 8.56 (s, 1H, pyrimidine -CH), 8.45 (s, 1H, triazolering -CH), 7.83 (d, 2H, Ar-H_8, H_9, J = 8 Hz), 7.62 (d, 2H, Ar-H_2^1, H_6^1, J = 8 Hz), 7.53 (d, 3H, Ar-H_3^1, H_5^1, -thiophene ring -H_6, J = 8 Hz,), 7.52 (d, 1H, thiophene ring -H_5), 7.49 (t, 1H, Ar-H_4^1), 7.25 (d, 2H, Ar-H_{10}, H_{11}, J = 8 Hz), 5.38 (s, 2H, -O-CH_2-) ppm; ^{13}C NMR ($CDCl_3$, 100 MHz): δ 165.65, 160.03, 157.53, 147.21, 144.36, 141.67, 136.75, 133.15, 130.21, 128.67, 128.66, 127.8, 124.32, 125.57, 120.48, 120.29, 119.17, 61.09 ppm; LC-MS (Positive ion mode): m/z = 430 $(M+H)^+$ for $C_{22}H_{15}N_5O_3S$.

1-(3-Fluorophenyl)-1H-1,2,3-triazol-4-yl) methyl 4-(4-oxothieno[2,3-d]pyrimidin-3(4H)-
yl)benzoate (6b)



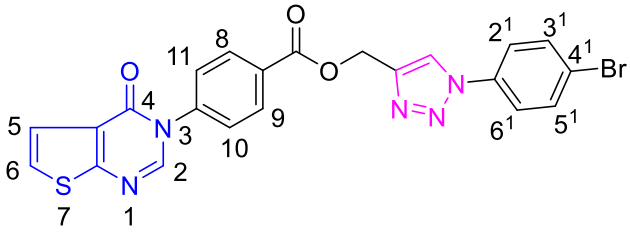
Light brown solid, Yield: 83%, mp: 130-131 °C, IR (KBr, ν, (cm $^{-1}$): 3015 (Ar-C-H), 1766 (C=O, str), 1635 (C=O, lactam), 1532 (C=N), ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 8.54 (s, 1H, pyrimidine -CH), 8.37 (s, 1H, triazole ring -CH), 7.81 (d, 2H, Ar-H_8, H_9, J = 8Hz), 7.54 (d, 1H, thiophene ring -H_6), 7.51 (d, 1H, thiophene ring -H_5), 7.39 (d, 1H, Ar-H_6^1), 7.28 (t, 1H, Ar-H_5^1), 7.26 (d, 2H, Ar-H_{10}, H_{11}, J = 8 Hz), 7.02 (s, 1H, Ar-H_4^1), 6.98 (d, 1H, Ar-H_2^1), 5.29 (s, 2H, -O-CH_2-) ppm; ^{13}C NMR ($CDCl_3$, 100 MHz): δ 165.63, 160.17, 162.86, 157.56, 147.23, 144.29, 141.75, 133.27, 130.15, 130.17, 129.25, 127.58, 125.67, 125.19, 124.26, 120.21, 115.63, 115.51, 119.15, 61.21 ppm; LC-MS (Positive ion mode): m/z = 448 $(M+H)^+$ for $C_{22}H_{14}FN_5O_3S$.

(1-(3-Bromophenyl)-1H-1,2,3-triazol-4-yl) methyl 4-(4-oxothieno[2,3-d]pyrimidin-3(4H)-yl)benzoate (6c)



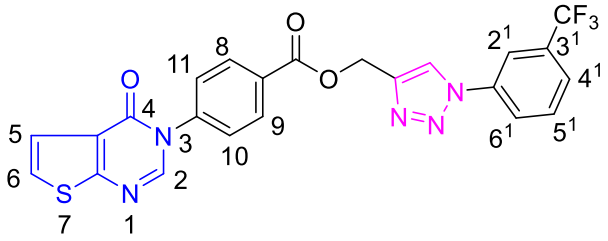
Light brown solid, Yield: 79%, mp: 155-156 °C, IR (KBr, ν, (cm $^{-1}$): 3018 (Ar-C-H), 1763 (C=O, str),1645 (C=O, lactam), 1534 (C=N), ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 8.86 (s, 1H, pyrimidine -CH), 8.82 (s, 1H, triazole ring -CH), 7.79 (d, 2H, Ar-H_8, H_9, J = 8Hz), 7.68 (d, 1H, Ar-$H_6{}^1$, J = 8Hz), 7.55 (d, 2H, -thiophene ring -H_6, $H_4{}^1$), 7.53 (d, 1H, -thiophene ring -H_5), 7.49 (s, 1H, Ar-$H_2{}^1$), 7.43 (t, 1H, Ar-$H_5{}^1$, J = 8Hz), 7.24 (d, 2H, Ar-H_{10}, H_{11}, J = 8Hz), 5.36 (s, 2H, -O-CH_2-) ppm; ^{13}C NMR ($CDCl_3$, 100 MHz): δ 165.38, 160.60, 143.87, 143.27, 136.71, 136.17, 133.22, 131.48, 131.08, 130.64, 130.19, 128.56, 125.76, 124.42, 123.47, 122.57, 121.90, 119.11, 116.90, 58.04 ppm; LC-MS (Positive ion mode): m/z = 508 $(M+H)^+$ for $C_{22}H_{14}BrN_5O_3S$.

(1-(4-Bromophenyl)-1H-1,2,3-triazol-4-yl)methyl 4-(4-oxothieno[2,3-d]pyrimidin-3(4H)-yl)benzoate (6d)



Light brown solid, Yield: 87%, mp: 150-151 °C, IR (KBr, ν, (cm $^{-1}$): 3017 (Ar-C-H), 1764 (C=O, str),1635 (C=O, lactam), 1534 (C=N), ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 8.85 (s, 1H, pyrimidine -CH), 8.80 (s, 1H, triazole ring -CH),7.77 (d, 2H, Ar-H_8, H_9, J = 8Hz), 7.65 (d, 2H, Ar-$H_3{}^1$, $H_5{}^1$, J = 8Hz), 7.57 (d, 2H, Ar-$H_2{}^1$, $H_6{}^1$, J = 8Hz), 7.54 (d, 1H, -thiophene ring -H_6), 7.53 (d, 1H, -thiophene ring -H_5), 7.26 (d, 2H, Ar-H_{10}, H_{11}, J = 8Hz), 5.31 (s, 2H, -O-CH_2-) ppm; ^{13}C NMR ($CDCl_3$, 100 MHz): δ 165.36, 160.61, 143.82, 143.25, 136.70, 136.15, 133.21, 131.43, 131.05, 130.45, 130.70, 128.55, 123.45, 122.53, 121.89, 119.12, 116.92, 58.07 ppm; LC-MS (Positive ion mode): m/z = 508 $(M+H)^+$ for $C_{22}H_{14}BrN_5O_3S$.

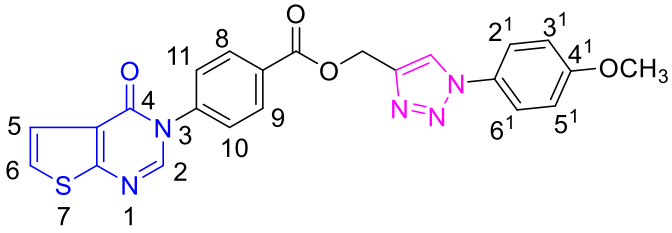
(1-(3-(Trifluoromethyl) phenyl)-1H-1,2,3-triazol-4-yl)methyl 4-(4-oxothieno[2,3-d]pyrimidin-3(4H)-yl)benzoate (6e)



Light brown solid, Yield: 75%, mp: 160-161 °C, IR (KBr, ν, (cm $^{-1}$): 3018 (Ar-C-H), 1768 (C=O, str),1643 (C=O, lactam), 1536 (C=N), ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 8.84 (s, 1H, pyrimidine -CH), 8.80 (s, 1H, triazole ring -CH), 7.93 (s, 1H,Ar-$H_2{}^1$), 7.73 (d, 2H, Ar-H_8, H_9, J = 8Hz), 7.62 (d, 1H, $H_6{}^1$, J = 8Hz), 7.55 (d, 2H, -thiophene ring -H_6, Ar-$H_4{}^1$), 7.52 (d, 1H, -thiophene ring -H_5), 7.27 (d, 3H, Ar-H_{10}, H_{11}, $H_5{}^1$, J = 8 Hz), 5.39 (s, 2H, -O-CH_2-) ppm; ^{13}C

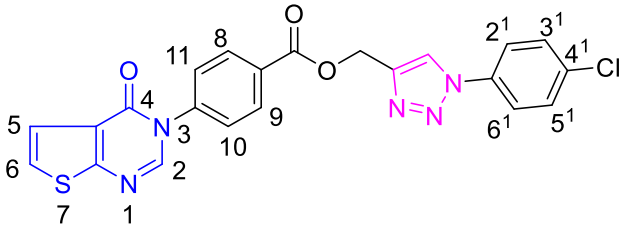
NMR ($CDCl_3$, 100 MHz): δ 165.31, 160.58, 157.52, 143.87, 144.31, 141.58, 133.32, 131.25, 131.07, 130.63, 130.19, 128.53, 125.71, 125.68, 123.72, 122.18, 120.15, 119.13, 116.89, 60.17 ppm; LC-MS (Negative ion mode): m/z = 496 $(M-H)^+$ for $C_{23}H_{14}F_3N_5O_3S$.

(1-(4-Methoxyphenyl)-1H-1,2,3-triazol-4-yl)methyl 4-(4-oxothieno[2,3-d]pyrimidin-3(4H)-yl)benzoate (6f)



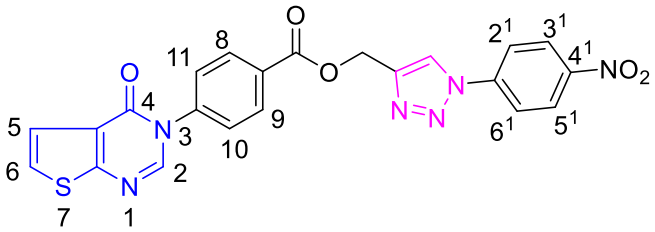
Light brown solid, Yield: 90%, mp: 170-171 °C, IR (KBr), ν, (cm^{-1}): 3016 (Ar-C-H), 1754 (C=O, str),1664 (C=O, lactam), 1528 (C=N), ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 8.83 (s, 1H, pyrimidine -CH), 8.81 (s, 1H, triazole ring -CH), 7.86 (d, 2H, Ar-H_8, H_9, J = 8 Hz), 7.80 (d, 2H, Ar-H_2^1, H_6^1, J = 8 Hz), 7.56 (d, 1H, thiophene ring -H_6), 7.54 (d, 1H, thiophene ring -H_5), 7.24 (d, 2H, Ar-H_{10}, H_{11}, J = 8Hz), 6.89 (d, 2H, Ar-H_3^1, H_5^1, J = 8Hz), 3.83 (s, , 3H, -OCH_3), 5.42 (s, 2H, -O-CH_2-) ppm; ^{13}C NMR ($CDCl_3$, 100 MHz): δ 165.38, 160.59, 159.83, 149.80, 143.13, 137.25, 131.06, 130.63, 130.33, 128.55, 125.74, 124.44, 123.47, 122.32, 119.09, 116.80, 115.32, 58.09, 56.00 ppm; LC-MS (Positive ion mode): m/z = 460 $(M+H)^+$ for $C_{23}H_{17}N_5O_4S$.

(1-(4-Chlorophenyl)-1H-1,2,3-triazol-4-yl)methyl 4-(4-oxothieno[2,3-d]pyrimidin-3(4H)-yl)benzoate (6g)



Light brown solid, Yield: 85%, mp: 155-157 °C, IR (KBr, ν, (cm^{-1}): 3018 (Ar-C-H), 1763 (C=O, str), 1648 (C=O, lactam), 1536 (C=N), ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 8.79 (s, 1H, pyrimidine -CH), 8.75 (s, 1H, triazole ring -CH),7.92 (d, 2H, Ar-H_8, H_9, J = 8Hz), 7.58 (d, 1H, -thiophene ring -H_6), 7.56 (d, 3H, 1H, -thiophene ring -H_5, Ar-H_2^1, H_6^1), 7.30 (d, 2H, Ar-H_3^1, H_5^1, J = 8 Hz), 7.25 (d, 2H, Ar-H_{10}, H_{11}, J = 8Hz), 5.45 (s, 2H, -O-CH_2-) ppm; ^{13}C NMR ($CDCl_3$, 100 MHz): δ 165.35, 160.48, 157.78, 147.68, 143.35, 137.46, 131.09, 130.72, 130.31, 128.51, 125.70, 124.40, 123.45, 122.38, 119.07, 115.96, 115.38, 56.46 ppm; LC-MS (Positive ion mode): m/z = 464 $(M+H)^+$ for $C_{22}H_{14}ClN_5O_3S$.

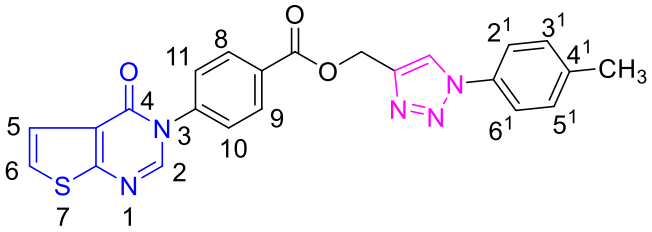
(1-(4-Nitrophenyl)-1H-1,2,3-triazol-4-yl)methyl 4-(4-oxothieno[2,3-d]pyrimidin-3(4H)-yl)benzoate (6h)



Light brown solid, Yield: 75%, mp: 135-136 °C, IR (KBr, ν, (cm^{-1}): 3016 (Ar-C-H), 1766 (C=O, str),1645 (C=O, lactam), 1534 (C=N), ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 8.84 (s, 1H, pyrimidine -CH), 8.80 (s, 1H, triazole ring -CH), 8.21 (d, 2H, Ar-H_3^1, H_5^1, J = 8 Hz), 8.01 (d,

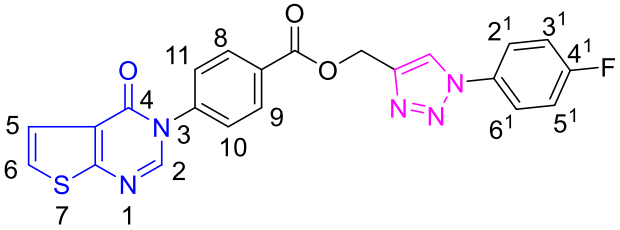
2H, Ar-H_2^1, H_6^1, *J* = 8Hz), 7.58 (d, 1H, thiophene ring -H_6), 7.55 (d, 1H, thiophene ring -H_5), 7.84 (d, 2H, Ar-H_8, H_9, *J* = 8Hz), 7.27 (d, 2H, Ar-H_{10}, H_{11}, *J* = 8Hz), 5.42 (s, 2H, -O-CH_2-) ppm; ^{13}C NMR ($CDCl_3$, 100 MHz): δ 165.52, 160.60, 157.62, 147.83, 143.78, 143.18, 138.73, 136.27, 136.46, 133.89, 131.69, 130.68, 128.21, 125.47, 124.74, 123.67, 122.56, 58.13 ppm; LC-MS (Positive ion mode): m/z = 475 $(M+H)^+$ for $C_{22}H_{14}N_6O_5S$.

(1-(p-Tolyl)-1H-1,2,3-triazol-4-yl)methyl 4-(4-oxothieno[2,3-d]pyrimidin-3(4H)-yl)benzoate(6i)



Light brown solid, Yield: 90%, mp: 120-121 °C, IR (KBr, ν, (cm $^{-1}$): 3016 (Ar-C-H), 1765 (C=O, str), 1643 (C=O, lactam), 1538 (C=N), ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 8.95 (s, 1H, pyrimidine -CH), 8.85 (s, 1H, triazole ring -CH), 7.91 (d, 2H,Ar-H_8, H_9, *J* = 8 Hz), 7.84 (d, 2H, Ar-H_2^1, H_6^1, *J* = 8 Hz), 7.66 (d, 2H, Ar-H_3^1, H_5^1, *J* = 8 Hz), 7.59 (d, 1H, thiophene ring -H_6), 7.44 (d, 1H, thiophene ring -H_5), 7.29 (d, 2H, Ar-H_{10}, H_{11}, *J* = 8 Hz), 2.45 (s, 3H, Ar-CH_3), 5.42 (s, 2H, -O-CH_2-) ppm; ^{13}C NMR ($CDCl_3$, 100 MHz): δ 165.53, 160.07, 155.03, 143.21, 142.03, 139.01, 138.48, 136.52, 134.85, 131.15, 130.76, 128.59, 126.34, 124.55, 123.40, 120.58, 119.19, 58.39, 21.09, ppm; LC-MS (Positive ion mode): m/z = 444 $(M+H)^+$ for $C_{23}H_{17}N_5O_3S$.

(1-(2-Fluorophenyl)-1H-1,2,3-triazol-4-yl)methyl 4-(4-oxothieno[2,3-d]pyrimidin-3(4H)-yl)benzoate (6j)



Light brown solid, Yield: 76%, mp: 145-146 °C, IR (KBr, ν, (cm $^{-1}$): 3018 (Ar-C-H), 1763 (C=O, str), 1646 (C=O, lactam), 1536 (C=N), ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 8.86 (s, 1H, pyrimidine -CH), 8.83 (s, 1H, triazole ring -CH), 7.85 (d, 2H,Ar-H_8, H_9, *J* = 8 Hz), 7.67 (d, 2H, Ar-H_2^1, H_6^1, *J* = 8 Hz),7.57 (d, 1H, thiophene ring -H_6), 7.55 (d, 1H, thiophene ring -H_5), 7.27 (d, 2H, Ar-H_{10}, H_{11}, *J* = 8 Hz), 7.23 (d, 2H, Ar-H_3^1, H_5^1, *J* = 8 Hz),5.46 (s, 2H, -O-CH_2-) ppm; ^{13}C NMR ($CDCl_3$, 100 MHz): δ 165.72, 162.53, 160.43, 157.14, 144.32, 141.67, 139.28, 137.32, 134.84, 132.32, 122.73, 128.23, 129.38, 125.68, 120.43, 119.18, 115.16, 60.19, ppm; LC-MS (Positive ion mode): m/z = 444 $(M+H)^+$ for $C_{23}H_{17}N_5O_3S$.

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