Heterocyclic Letters
Vol. 10| No.3|509-515|May-July|2020
ISSN : (print) 2231–3087 / (online) 2230-9632
CODEN: HLEEAI
http://heteroletters.org

A FACILE SYNTHESIS AND DOCKING STUDIES OF PYRIDINE CONTAINING 1*H*-THIENO[3,2-C]PYRAZOLE

Rajeshwari Madipelly

Telangana University#Dichpally, Nizamabad, Telangana 503322
Email: rajeshwarimadipelly79@gmail.com

Abstract
A simple and efficient route is proposed for the synthesis of title compound starting from Bromothiophene-2-carboxylic acid. The newly synthesized compounds **4-9**were characterized by spectroscopic investigation. Docking studies for the target molecule was also presented.

Introduction

Pyrazole containing compounds have practical applications in the medicinal and agrochemical field and the biological activity of pyrazoles[i,ii] and its derivatives are well documented. The pyrazole ring has shown to be the basic moiety for a number of dyes and drugs[iii,iv]. Substituted pyrazolopyrimidinones are found to be useful as cardiotonic,[v]herbicidal[vi] and antiviral[vii] agents. Literature survey reveals that substituted pyrazolopyrimidinones are potent and selective inhibitors of type 5 cyclic guanosine-3′, 5′-monophosphate phosphodiesterase (cGMP) PDE-5[viii,ix] and as such, have utility in the treatment of male erectile dysfunction (MED) and female sexual dysfunction (FSD)[x]. C-6 substituted pyrimidinone and pyrimidindione derivatives have shown selective antitumor,[xi]antiviral,[xii]antitubercular[xiii] and antifungal activity[xiv].
In view of the importance of thienopyrazole and pyrimidine derivatives, we herein report synthesis and docking studies of the title compounds.

Experimental Section

Thin layer chromatography was run on silicagel-G and visualization were done using UV light or iodine. ^{1}H NMR were recorded with a Varian Mercury plus 400 MHz instrument in DMSO-d_6 solvent using trimethylsilane as internal standard. All the chemical shifts were reported in δ (ppm) using TMS as an internal standard. The ^{1}H NMR chemical shifts and coupling constants were determined assuming first-order behavior. Multiplicity is indicated by one or more of the following: s (singlet), d (doublet), t (triplet), q (quartet), m (multiplet), br (broad). Jeol-JMS D-300 spectrometer was used to record mass spectra.

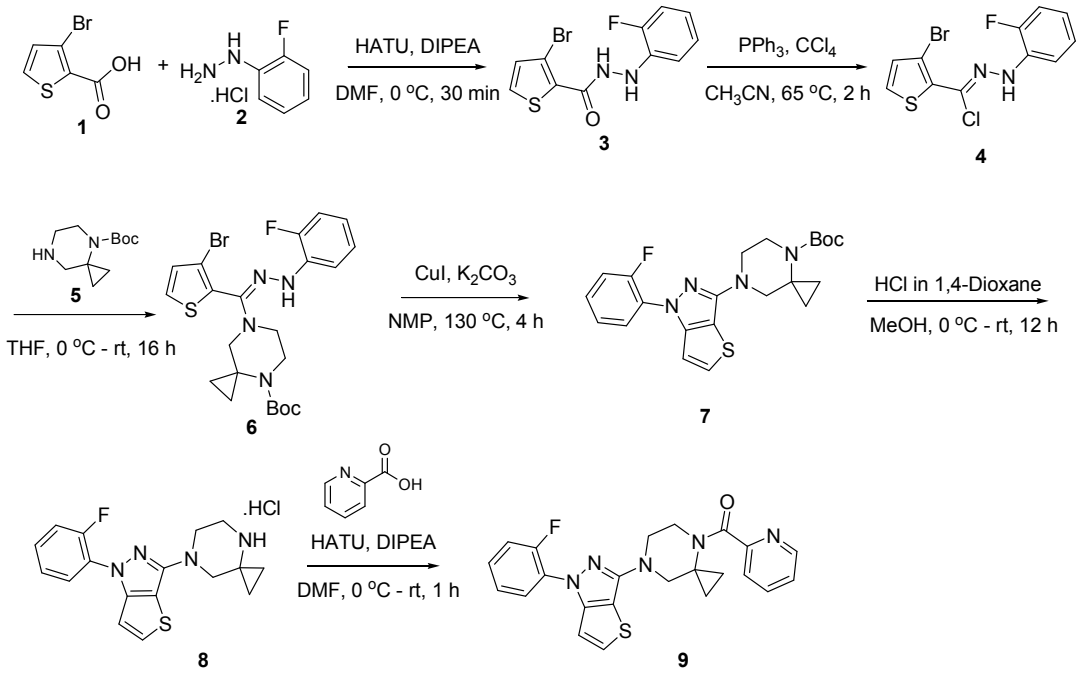
Result and Discussion

3-Bromothiophene-2-carboxylic acid (**1**), 2-Flourophenylhydrazine hydrochloride (**2**) react each other in presence of DMF to form 3-bromo-N'-(2-fluorophenyl)thiophene-2-carbohydrazide **(3)**, which waschlorinated with CCl_4 in presence of PPh_3. Further the chlorine is protected and cyclised in presence of CuI. Finally, the piperzine derivative coupled with Pyridine-2-carboxylic acid to form compound **9**.

Docking Studies

The protein 1jff (tubulin) was downloaded from RSC PDB (Research Collaboratory for Structural BioinformaticsProtein Data Bank)and was docked. Compound **9** was the most efficient for inhibiting the structural protein. Least inhibiton was seen by the compound 9 as shown in table 1 and 2. The major aminoacids which were involved in the binding of the compounds were tyrosine, asparagines, alanine, glutamine, glutamic acid, leucine, serine (Figures 1)

Scheme



3-bromo-N'-(2-fluorophenyl)thiophene-2-carbohydrazide: HATU was added to a solution of 3-Bromothiophene-2-carboxylic acid (**1**),2-Flourophenylhydrazinehydrochloride (**2**) and N, N-Diisopropylethylamine in DMF (250 mL) at 0 °C and stirred ~~at 0 °C~~ for 30 min. The reaction progress was monitored by TLC.After completion of reaction, the reaction mixture was poured into ice water;solids were filtered, washed with pentane and dried under vacuum for 5 h to get PRODUCT (80%).

^{1}H NMR (400 MHz, DMSO-d_6):10.2(s,1H),7.95(s,1H),7.87-7.86(d,1H), 7.24-7.22 (d, 1H), 7.12-7.07 (m, 1H), 7.05-7.01(t,1H),6.93-6.89(t,1H),6.77-6.73(m,1H).Mass *m/z* 316 [M+H], 317 [M+2H].

3-Bromo-N'-(2-fluorophenyl)thiophene-2-carbohydrazonoyl chloride: Carbon tetrachloride was added to a mixture of compound-**3** and triphenylphosphinein acetonitrile at 65 °Cand stirred at same temperature for 2h.The reaction progress was monitored

byTLC,after completion of reaction,the reaction mixture was concentrated under reduced pressure to get 160g ofcrude product. The crude product was purified by column chromatographyby eluting 2% ethyl acetate in pet ether and finished with 10% ethyl acetate in pet ether to afford compound-**4** (45%).

^{1}H NMR (400 MHz, DMSO-d_6): 9.11(s,1H),7.80-7.79(d,1H),7.62-7.57(t,1H),7.26-7.16 (m, 2H), 7.00-6.94 (m, 1H).Mass *m/z* 334 [M+H], 335 [M+2H]

tert-Butyl-7-((3-bromothiophen-2-yl)(2-(2-fluorophenyl)hydrazono)methyl)-4,7-diazaspiro[2.5]octane-4-carboxylate: Compound-**5** was added to a ice cold solution of compound-**4**and triethylamine in anhydrous THF and stirred at rt for 16 h. The progress of the reaction was monitored by TLC. After completion of reaction, the reaction mixture was filtered and the filtrate was concentrated to get crude product.The crude as such used for the next step without further purification.

***tert*-Butyl-7-(1-(2-fluorophenyl)-1*H*-thieno[3,2-c]pyrazol-3-yl)-4,7-diazaspiro[2.5] octane-4-carboxylate:**A mixture of compound-**6**, Copper iodide and potassium carbonatein NMP was stirred at 130 ^{o}C for 4h. The reaction was monitored by LCMS.After completion of reaction, the reaction mixture was poured in ice water and extracted with EtOAc (3 × 300 mL). The combined organic layers were washed with cold water, brine and concentrated under reduced pressure to afford 60g of crude product. The crude product was purified by column chromatography to afford of product(61% on two steps).

^{1}H NMR (300 MHz, DMSO-d_6): 7.76-7.75 (d, 1H), 7.67-7.61 (m, 1H), 7.47-7.29 (m, 3H), 7.05-7.01 (t, 1H), 3.64-3.60 (m, 2H), 3.35-3.29 (m, 2H), 3.18 (s, 2H), 1.45 (s, 9H), 0.97-0.85 (m, 4H).LCMS: 99.51% (m/z= 429.2 $[M+H]^+$).

1-(2-Fluorophenyl)-3-(4,7-diazaspiro[2.5]octan-7-yl)-1*H*-thieno[3,2-c]pyrazole HCl:4M HCl in 1,4-Dioxane was added to a solution of compound-**7**in MeOHat 0 ^{o}C and stirred at rt for 12 h. The reaction was monitored by TLC After completion of reaction, the reaction mixture was concentrated under reduced pressure afford product. The crude product was triturated with diethylether (25 mL) and filtered to affordproduct (79%).

^{1}H NMR (400 MHz, DMSO-d_6): 9.77 (brs, 2H), 7.81-7.79 (d, 1H), 7.68-7.63 (m, 1H), 7.49-7.01 (m, 3H), 7.06-7.05 (t, 1H), 3.66-3.64 (brs, 2H), 3.48 (s, 2H), 3.37-3.34 (brs, 2H), 1.14-1.11 (m, 2H), 0.97-0.94 (m, 2H).Mass: (m/z = 329.2$[(M-HCl)+H]^+$).HPLC: 98.97% (215 nm), 99.70% (254 nm).

(7-(1-(2-Fluorophenyl)-1*H*-thieno[3,2-c]pyrazol-3-yl)-4,7-diazaspiro[2.5]octan-4-yl)(pyridin-2-yl)methanone:HATU(1-[Bis(dimethylamino)methylene]-1H-1,2,3-triazolo[4,5-b]pyridinium 3-oxide hexafluorophosphate, Hexafluorophosphate Azabenzotriazole Tetramethyl Uronium)was added to a solution of Compound **8**, Pyridine-2-carboxylic acid and N,N-Diisopropylethylamine in DMF (20 mL) at 0 ^{o}C and stirred at0 ^{o}Cfor 15min then allowed to rt for 1h. The reaction was monitored byTLCafter completion of reaction, the reaction mixture was poured in ice water and extracted with EtOAc (3 × 100 mL). The combined organic layers were washed with water, brine and concentrated under reduced pressure to afford crude product. The crude product was purified by column chromatography to afford product (42%).

^{1}H NMR (300 MHz, $CDCl_3$):8.61-8.59 (d, 1H), 7.78 (brs, 1H), 7.67-7.56 (m, 2H), 7.39-7.37 (m, 2H), 7.26-7.20 (m, 3H), 6.96-6.92 (t, 1H), 3.99 (brs, 2H), 3.70-3.50 (m, 4H), 1.25 (brs, 1H), 1.11 (brs, 1H), 0.72 (brs, 1H), 0.51 (brs, 1H). Mass: (m/z = 434.2$[M+H]^+$).HPLC: 99.11% (215 nm), 99.32% (254 nm).

Table 1: Free energy of binding between the compound and tubullin

Rank	Est. Free Energy of Binding	Est. Inhibition Constant, Ki	vdW + Hbond + desolv Energy	Electrostatic Energy	Total Intermolec. Energy	Frequency	Interact. Surface	Download	
	1.	-4.46 kcal/mol	534.71 uM	-5.44 kcal/mol	+0.01 kcal/mol	-5.43 kcal/mol	100 %	556.951	

Table 2: Interaction Table

hydrogen bonds	polar	hydrophobic	pi-pi	other	
	N1 () THR70 [*3.20*] (*CB, CG2, OG1*)	O1 () GLN42 [*3.15*] (*OE1*)	C18 () PRO40 [*3.80*] (*CB*)	C20 () TYR55 [*3.78*] (*CE1*)	C19 () SER38 [*3.64*] (*OG*)
		N5 () TYR55 [*3.56*] (*OH*)	C19 () PRO40 [*3.72*] (*CB, CG*)		C22 () GLN42 [*2.95*] (*OE1*)
		N5 () ASN68 [*2.84*] (*ND2, OD1*)			C20 () TYR55 [*3.44*] (*OH*)
		N3 () ASN68 [*3.21*] (*ND2*)			C21 () TYR55 [*3.01*] (*OH*)
		N4 () ASN68 [*3.50*] (*ND2*)			S1 () GLU67 [*3.54*] (*CB, OE1*)
		O1 () ASN68 [*3.82*] (*ND2*)			C11 () GLU67 [*3.48*] (*CB*)
		N2 () THR70 [*3.65*] (*OG1*)			N5 () ASN68 [*3.64*] (*CG*)
					C12 () ASN68

					[*3.25* (*ND2*)]	
					C13 () [*3.90*] ASN6 –8 (*ND2*)	
					C14 () [*3.66*] ASN6 –8 (*ND2*)	
					C15 () [*2.89*] ASN6 –8 (*ND2*)	
					C16 () [*3.33*] ASN6 –8 (*ND2*)	
					C9 () [*3.68*–8] ASN6 (*ND2*)	
					C8 () [*3.59*–8] ASN6 (*ND2*)	
					S1 () [*3.33*–8] ASN6 (*ND2*)	
					C17 () [*3.46*] ASN6 –8 (*ND2*)	
					C21 () [*3.79*] ASN6 –8 (*ND2*)	
					N2 () [*3.46*] THR7 –0 (*CB, CG2*)	
					C5 () [*3.71*–0 (*CB,] THR7 *OG1*)	
					C4 () [*3.21*–0 (*CB,] THR7 *OG1*)	
					C9 () [*3.75*–] THR7 0 (*CG2, OG1*)	

					C8 () THR7 [*3.55*–0] (*OG1*)	
					C7 () THR7 [*3.11*–0] (*OG1*)	
					C10 () THR7 [*3.46*–0] (*OG1*)	
					C12 () THR7 [*3.45*–0] (*CG2*)	
					C3 () THR7 [*3.53*–1 (*CB*,] *CG2*)	
					C4 () THR7 [*3.84*–1] (*CG2*)	

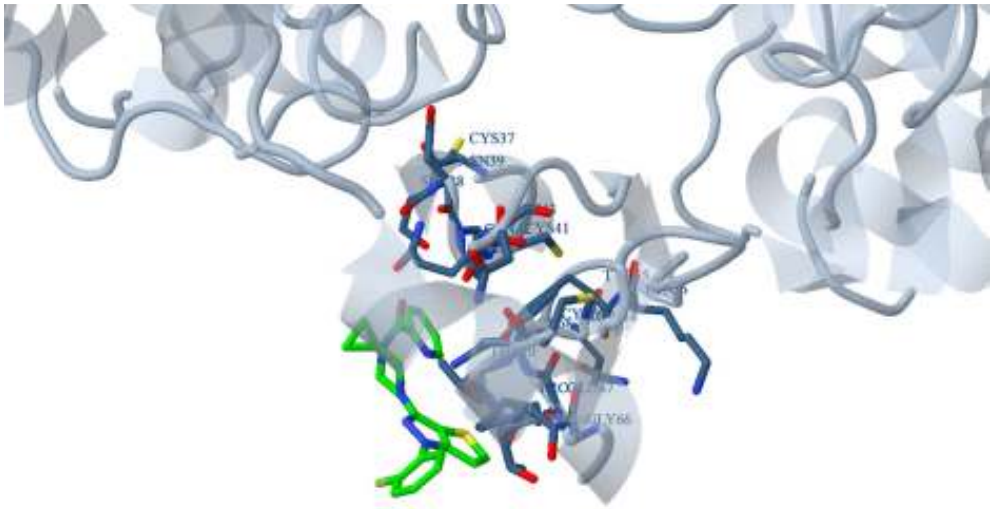


Figure 1: Bonding involved in the binding of the ligand to the engyme

References

i. W. E.Krikpatrick, T. Okabe, I.W. Hillyard, R.K. Robins, A. T. Dren, T.J. Novinson, J. Med Chem., 20, 386 (1977).
ii. A.A. Elagamy, M. A. El-Taweel, F. A. Amer, H. H. Zoorob, Arch. Pharm., 246, 320 (1987).
iii. A. N. Kost, I. I.Grandberg, In Advances in Heterocyclic Chemistry; Katritzky Ed. Academic Press: New York, 347 (1996).
iv. G. W. Raiziss, L.W. Clemence, M. Friefelder J. Am. Chem. Soc., 63, 2739 (1941).
v. Warner-Lambert Co., 236, 778 (1986); Chem. Abstr., 20, 101967h (1987).
vi. S. D. Lindell, B. A. Moloney, B. D. Hewitt, C. G.Earnhaw, P. J. Philip, J. E. Dancer, Bioorg. Med. Chem. Lett., 9, 1985 (1999).

vii. Y. S. Sanghvi, S. B. Larson, D. F. Smee, G. R.Revankar, R. K. Robins,Nucleotides., 10, 1417 (1991).
viii. M. K. Bunnage, J. P. Mathias, S. D. A. Street A. Wood ., PCT Int. Appl. WO, 14, 333, 487 (1999); Chem. Abstr., 131, 310644f (1999).
ix. A. S. Bell, N. K. Terrett, Eur. Pat. EP 526, 004(1993); Chem. Abstr., 118, 254957c (1993).
x. P.C. Doherty, V. A. Place, W. L. Smith, PCT Int. Appl. 9921, 558 (1999); Chem. Abstr., 130, 332908d (1999).
xi. G. M. Szczech, P. Furman, G. R. Painter, D. W. Barry, K. Borroto-Esoda, T. B. Grizzle,M. R. Blum, j. P. Sommadossi, R. Endoh, T. Niwa, M. Yamamoto, C. Moxham, Antimicrob. Agents Chemothe., 44, 123 (2000).
xii. A. Arnaud, L. Fontana, A. J. Angulo, A. Gil, López-Pedrosa, J. M. Clin. Nutr., 22, 391 (2003)
xiii. A. Kumar, S. P. Sinha, M. S. Chauhan, Bioorg. Med. Chem. Lett., 12, 667 (2002).
xiv. J. B. Behr, T. Gourlain, A. Helimi, G. Guillerm, Bioorg. Med. Chem. Lett., 13, 1713 (2003).

Received on May 5, 2020.