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Research Paper

Synthesis and Pharmacological Aspects of Some Novel N-(4-(2-(Methyl(3-Methyl-1-Phenyl-1H-Pyrazol-5- Yl)Amino)Thiazol-4-Yl) Phenyl)(Alkyl) Amide Derivatives.

Chemistry

KEYWORDS : N-methyl-3-oxobutan-amide ,Pyrazol, Thiazol,Antibacterial activity

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ABSTRACT

(Z)-N-methyl-3-(2-phenylhydrazono)butanamide (I),has been obtained from the condensation of N-methyl-3-oxobutanamide (0.10 mol) and phenyl hydrazine (0.10 mol).The compound is than converted to its N,3-dimethyl-1-phenyl-1H-pyrazol-5-amine (II) and N-(methyl(3-methyl-1-phenyl-1H-pyrazol-5-yl)carbamothioyl) benzamide (III) by condensing with P2S5 and benzoyal chloride/ammonium thiocynate.Which on further reaction gives compound (IV).Led compound on reaction with P-Phenacyl bromid gives compound (V). In presence of iron powder in acetic acid gives (VI).Further on reaction gives(VII) and its derivates(b-j). The synthesized compounds were evaluated for their in vitro antibacterial activity against gram positive and gram negative bacterial strains and two fungal species. The structures of the compounds were elucidated with the aid of IR, 1HNMr ,13C NMR.Mass spectroscopy and elemental analysis.

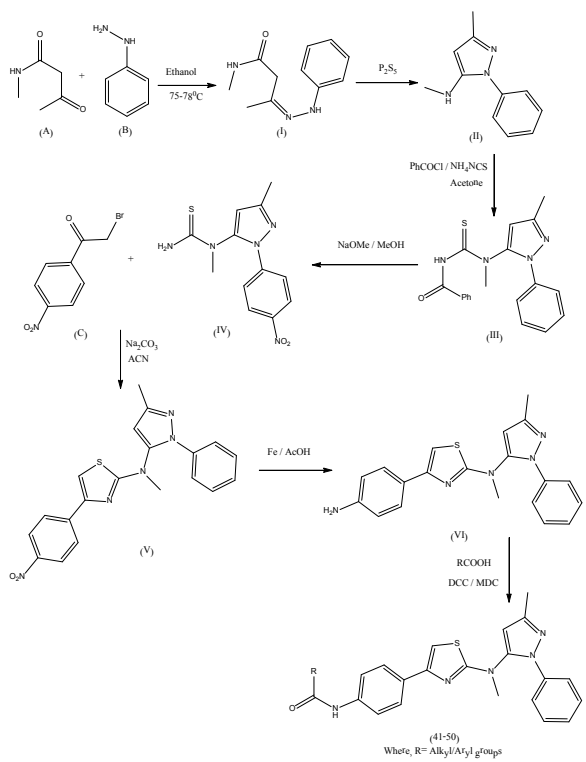
INTRODUCTION :

Heterocycles form by far the major of classical divisions of organic chemistry and are of immense use biologically and industrially. It is well known that the heterocycles are present in all kinds of organic compounds of interest in electronics, biology, optics, pharmacology, material sciences and so on. Heterocyclic nucleus imparts an important function in medicinal chemistry and serves as a key template for the development of various therapeutic agents[1].

Pyrazolone derivatives have a broad spectrum of biological activities being used as analgesic, [2]. antipyretic, [3]. anti-inflammatory, [4]. antimicrobial, [5].antimycobacterium, [6].anticancer, [7].anti-human immunodeficiency virus (HIV), [8].and antivirus. [9].The antibacterial activity caught our attention because antimicrobial resistance developed by important pathogens with available chemotherapeutic agents has increased to an alarming level. [10-11]. In view of the above mentioned factor and as a continuation of our investigations on pyrazolone derivatives recently described [12]. .

Mostly researchers have maintained their interest in sulfur and nitrogen-containing heterocyclic compounds through decades of historical development of organic synthesis[13]. There are widespread therapeutic uses of synthetic heterocycles such as antibacterial, antimycobacterial, trypanocidal, anti-HIV activity, genotoxic, herbicidal, analgesic, antiinflammatory, muscle relaxants, antileishmanial agents, anticonvulsant, anticancer, antimalarial, antifungal and lipid peroxidation inhibitor, antitubercular, hypnotics, antidepressant, antitumoral, anthelmintic and insecticidal agents[14-19].. The exploration for new biologically active heterocyclic analogues continues to be an area of intensive research in medicinal chemistry.

A series of N-(4-(2-(METHYL(3- METHYL -1-PHENYL-1H-PYRAZOL-5-L)AMINO)THIAZOL-4-YL)PHENYL)(ALKYL) AMIDE derivatives to determine the effect of various substituents at the phenyl nucleus on antimicrobial activity. Therefore, it seems that various substitutions in the pyrazolone and thiazol ring can be an effective pharmacophore in several classes of antimicrobial agents. To identify new candidates that may be of value in designing new, potent, selective and less toxic antimicrobial agents. we report herein the synthesis, in-vitro antimicrobial, screening of N-(4-(2-(METHYL(3- METHYL -1-PHENYL-1H-PYRAZOL-5-L)AMINO)THIAZOL-4-YL) PHENYL)(ALKYL)AMIDE derivatives.



Scheme : 1

EXPERIMENTAL PROCEDURE

Melting points were determined in open capillary tubes in a Hicon melting point apparatus and are uncorrected .All the fourier Transform-infa Red (FT-IR)spectra were recorded in KBr pellets on a Shimadzu-8400S spectrometer.The 1 H-NMR spectra were taken on a Bruker- Spectrospin DXC(300MHz)NMR spectrometer.Chemical shifts(9) are expressed in ppm relative to tetramethylsilane (TMS),used as an internal standard.The purity of the compounds was checked by thin layer chromatography TLC) on merck Silica Gel 60F254 precoated sheets.Iodine chamber and UV lamp were used for the visualization of TLC spots.

Evaluation of biological action

Three bacteria were used as test organisms. This bacteria were Bscilus cereus,MTTC 430, Bacillus Pumilus MTTC 1607,Escherichia coli MTTC 119. All of the synthesized compounds were screened against three bacteria using the method given in literature 22 . The bacteria strains were inoculated into nutrient

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broth for 24 and 48h, In the disc-diffusion ,sterile Mueller-Hinton agar for bacteria were inoculated with the test bacteria(105 bacteria per mL) In order to prepare the disc an part containing 1000 µg of compound from placed(10µL).Discs were applied on the solid agar medium by pressing slightly. Petri dishes were placed at 4 0C for 2 h. The bacteria samples were incubated at 35 + 0.1 0C for 24h were incubated at 25 + 0.1 0C for 72h.At the end of the period. inhibition zones were measured in millimeter. The inhibition zone of compound was calculated by subtracting that of pure DMSO from the zone of solution in DMSO of the compound.

GENERAL METHOD FOR PREPARATION OF N-(4-(2-(METHYL(3-METHYL-1-PHENYL -1H-PYRAZOL-5-YL)AMINO)THIAZOL -4- YL)PHENYL)(ALKYL) AMIDE

1. Preparation of (Z)-N-methyl-3-(2-phenylhydrazono)butanamide (I)

A solution of N-methyl-3-oxobutanamide (11.5 g, 0.10 mol) and phenyl hydrazine (10.8 g, 0.10 mol) in 20 % aqueous ethanol (60 ml) was stirred at 80-85 ^{0}C for 1 h. After 1h reaction was monitored by TLC. Solvent was evaporated completely under vacuum and water was added to residue to produce solid precipitate. Solid separated by filtration and solid was recrystalized in isopropanol giving light brown solid as compound I (16.8 g, yield 82%).

2. Preparation of N,3-dimethyl-1-phenyl-1H-pyrazol-5-amine (II)

To a suspension of (Z)-N-methyl-3-(2-phenylhydrazono)butanamide (I) (16.0 g, 0.078 mol) and sodium carbonate (16.5 g, 0.156 mol) in THF (80 ml) was stir at rt. Phosphorous pentasulfide (8.61 g, 0.039 mol) was added and stir at 80-85 ^{0}C for 2-3 h. After completion of reaction solvent evaporated completely and water was added to residue solid separated was filtered washed with water. This was further purified by crystallization in ethanol giving pure compound II (10.3 g, 68.9%).

3. Preparation of N-(methyl(3-methyl-1-phenyl-1H-pyrazol-5-yl)carbamothioyl) benzamide (III)

To a solution of NH_4SCN (4.9 g, 0.064 mol) in acetone (50 ml) was added slowly benzoylchloride (7.56 g, 0.054 mol) under dry condition within 10 min. After completion of addition reaction mixture was refluxed for 15 min. A solution of N,3-dimethyl-1-phenyl-1H-pyrazol-5-amine (10.0 g, 0.054 mol) in acetone (20 ml) was added to above stirred suspension at such a rate that reflux gently. After completion of addition reaction mixture was refluxed further for 30 min. Reaction was cooled and poured in water, and resulting yellow solid separated by filtration. Solid was recrystalized in ethanol giving light brown solid as compound III (12.7 g, yield 68.0%).

4.Preparation of 1-methyl-1-(3-methyl-1-(4-nitrophenyl)-1H-pyrazol-5- | yl)thiourea (IV)

To a suspension of N-(methyl(3-methyl-1-phenyl-1H-pyrazol-5-yl)carbamothioyl) benzamide (III) (12.5 g, 0.036 mol) in Methanol (60 ml) was added sodium methoxide (5.8 g, 0.108 mol) and stirred at rt for 1 h. Reaction mixture was poured in 5 % HCl and stirred for 10 min. Resulting solid was separated by filtration giving compound IV (7.58 g, 73.0%).

5. Preparation of N-methyl-N-(3-methyl-1-phenyl-1H-pyrazol-5-yl)-4-(4-nitrophenyl) thiazol-2-amine (V)

A mixture of 1-methyl-1-(3-methyl-1-(4-nitrophenyl)-1H-pyrazol-5-yl)thiourea (IV) (7.5 g, 0.026 mol), p-nitro phenacyl bromide (7.07 g, 0.029 mole) and sodium carbonate (5.51 g, 0.052 mol) in acetonitrile (50 ml) was heated at 60-62^0 C. under stirring for 2 h. Reaction mixture was carefully poured in ice water , stirred for 15 min. Resulting solid was separated by filtration and washed with water , hexane and dried to give compound V (7.75 g, 77.0%).

6. Preparation of 4-(4-aminophenyl)-N-methyl-N-(3-methyl-1-phenyl-1H-pyrazol-5-yl) thiazol-2-amine (VI)

To a solution of N-methyl-N-(3-methyl-1-phenyl-1H-pyrazol-5-yl)-4-(4-nitrophenyl) thiazol-2-amine (V) (7.7 g) in acetic acid (50 ml) was added iron powder (2 g) stir at room temperature for 4-5 h. Reaction was poured in water and basify by addition of sat sodium carbonate to pH 7.5-8. Solid products extracted with ethyl acetate, wash with water and distilled out to get light brown. Solid was recrystalized in methanol giving pure compound VI (4.19 g, 59.0 %).

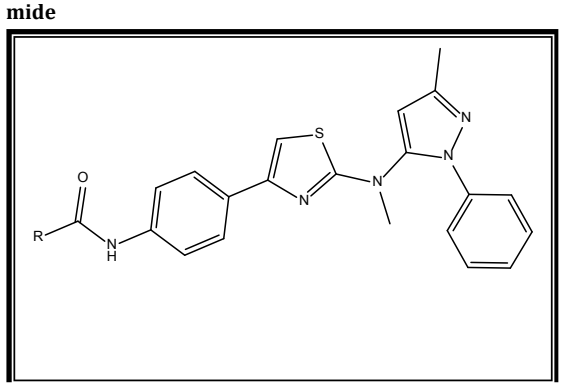
7. Preparation of N-(4-(2-(methyl(3-methyl-1-phenyl-1H-pyrazol-5-yl)amino)thiazol-4-yl)phenyl)acetamide (VII)

To a solution of 4-(4-aminophenyl)-N-methyl-N-(3-methyl-1-phenyl-1H-pyrazol-5-yl)thiazol-2-amine (VI) (200 mg, 0.554 mmol) and acetic acid (39.88 g, 0.665 mmol) in dichloromethane (10 ml) was added DCC (171.0 mg, 0.831 mmol) and stir at 5-10 ^{0}C. Reaction mixture stir at 27-30 ^{0}C for 2-3 h. After completion of reaction, solid was separated by filtration washed with DCM. Filtrate was washed with water, dried and evaporated to get yellow residue. Residue recrystallized from IPA to get off white solid as compound VIIa (98.0 mg, 43.9.0 %). Ana Obs.: C-65.40%, H-5.32%, N-17.30%; Calc. for $C_{22}H_{21}N_5OS$: C-65.49%, H-5.25%, N-17.36%.

The monitoring of reaction and purity of compounds were checked on TLC aluminium sheet silica gel 60 F245 (E.Merck) using hexane-ethyl acetae (5:5 V/V) and methanol- chloroform (2:8 V/V) as mobile phase and visualize under U.V light 254 nm.

Other compounds of the series **(7b-j)** were prepared by using similar method and their physical data are recorded in **Table-1.**

TABLE: 1
PHYSICAL DATA OF N-(4-(2-(METHYL(3-METHYL-1-PHENYL-1H-pyrazol-5-yl)amino)thiazol-4-yl)phenyl)acetamide



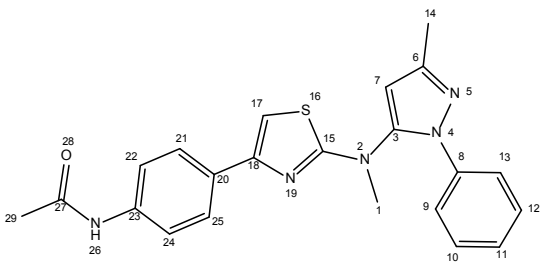
Sr. No	-R	Mol. Formula	mp °C	% yield	Elemental analysis					
					%C		%H		%N	
					Req.	Obs.	Req.	Obs.	Req.	Obs.
41.	-CH3	$C_{22}H_{21}N_5OS$	211-214	44	65.49	65.40	5.25	5.32	17.36	17.30
42.	$-C_2H_5$	$C_{23}H_{23}N_5OS$	215-217	68	66.16	66.10	5.55	5.50	16.77	16.84
43.	$-C_3H_7$	$C_{24}H_{25}N_5OS$	220-222	63	66.80	66.87	5.84	5.78	16.23	16.28
44.	$-CH(CH_3)_2$	$C_{24}H_{25}N_5OS$	211-214	61	66.80	66.86	5.84	5.78	16.23	16.29
45.	$-CH(CH_2)_2$	$C_{24}H_{23}N_5OS$	205-207	57	67.11	67.18	5.40	5.32	16.30	16.38
46.	$-C_6H_5$	$C_{27}H_{23}N_5OS$	235-237	66	69.65	69.60	4.98	4.93	15.04	15.12

47.	$-C_6H_5Cl$	$C_{27}H_{22}ClN_5OS$	244-247	61	64.86	64.80	4.43	4.49	14.01	14.10
48.	$-C_6H_5F$	$C_{27}H_{22}FN_5OS$	255-257	57	67.06	67.13	4.59	4.50	14.48	14.41
49.	$-SC_4H_3$	$C_{25}H_{21}N_5OS_2$	203-206	53	63.67	63.61	4.49	4.53	14.85	14.80
50.	$-NC_5H_5$	$C_{26}H_{22}N_6OS$	222-225	54	66.93	66.87	4.75	4.82	18.01	18.09

Result and Discussion

1.Spectral analysis

SPECTRAL DATA OF N-(4-(2-(methyl(3-methyl-1-phenyl-1H-pyrazol-5-yl)amino)thiazol-4-yl)phenyl)acetamide (41)



IR SPECTRA

Sample technique : KBr Pellet

Instrument : Perkin Elmer *Spectrum 100*

Frequency range : 4000-400 cm^{-1}

Type	Vibration mode	Frequency cm-1
Heterocyclic	-N-H Str.	3463.5
Alkyl CH3	Methyl C-H Str. Symm Methyl C-H Symm. bend	2960.6 1414.0
Alkyl CH2	Methylene C-H Str. Symm Methylene C-H Symm. bend	2930.3 1469.8
Amide	C=O Str. N-H Str.	1713.9 3312.3
Aromatic	>C=C ring skeleton vib.	1653.5 1578.0
	p-Subst bending vib. o-subst. bending vib. Mono subst. bending vib.	843.4 739.6 713.4
chloro	C-Cl str.	820.6

^{1}H NMR Spectra

Instrument : Varian *400 MHz NMR*

Standard : TMS

Solvent : DMSO d_6

Inference	δ ppm	Multiplicity	No. of protons
H-29	2.25	s	3H
H-14	2.45	s	3H
H-1	2.75	s	3H
H-26	6.50	s	1H
H-17	6.75	s	1H
H-9, H-10, H-11, H-12, H-13	7.30-7.45	m	5H
H-21, H-25	7.55-7.65	dd	2H
H-22, H-24	7.80-7.90	dd	2H

Mass Spectra:

Mol. Formula	Mol. Wt	Mass (m/z)
$C_{22}H_{21}N_5OS$	403.0	404.00

2.Antimicrobial activity

Table : 1

In-vitro antibacterial activity of synthesized compounds (1-10)

Compound Name	Inhibition Zone(in mm) against		
	E.coli	B.pumilus	B.cereus
1	11	14	12
2	11	20	15
3	16	17	17
4	18	21	11
5	15	10	12
6	14	11	21
7	10	13	14
8	16	14	11
9	15	17	15
10	11	18	10
Ciprofloxacine	30	33	17
Ampicilline	13	21	30
DMF	06	08	5
Tetracyclin	15	21	22
Gentamycin	18	20	19
Chloramphenicol	17	21	23

CONCLUSION:

It was observed that activity of all compounds can be possibly because of there selective antimicrobial effect we concluded form our investigations that major compounds may be considered promising for the development of new antibacterial agent.

REFERENCE

[1]. Kashyap, S.J., P.K. Sharma, V.K. Garg, R. Dudhe and N. Kumar, 2011. Review on synthesis and various biological potential of thiazolopyrimidine derivatives. J. Adv. Sci. Res., 2: 18-24 | [2]Gürsoy A, Demirayak S, Capan G, Erol K, Vural K. Synthesis and preliminary evaluation of new 5-pyrazolinone derivatives as analgesic agents. Eur J Med Chem 2000;35:359-64. | [3]El-Hawash SA, Badawey el-SA, El-Ashmawey IM. Nonsteroidal antiinflammatory agents-part 2 antiinflammatory, analgesic and antipyretic activity of some substituted 3-pyrazolin-5-ones and 1,2,4,5,6,7-3H-hexahydroindazol-3-ones. Eur J Med Chem 2006;41:155-65. | [4] Alam MS, Choi JH, Lee DU. Synthesis of novel Schiff base analogues of 4-amino-1,5-dimethyl-2-phenylpyrazol-3-one and their evaluation for antioxidant and anti-inflammatory activity. Bioorg Med Chem 2012;20:4103-8. | [5]Aly HM, Saleh NM, Elhady HA. Design and synthesis of some new thiophene, thienopyrimidine and thienothiadiazine derivatives of antipyrine as potential antimicrobial agents. Eur J Med Chem 2011;46:4566-72. | [6]Küçükgüzel SG, Rollas S. Synthesis, characterization of novel coupling products and 4-arylhydrazono-2-pyrazoline-5-ones as potential antimycobacterial agents. Farmaco 2002;57:583-7. | [7] Markoviæ V, Eriæ S, Stanojkoviæ T, Gligorijeviæ N, Aranðeloviæ S, Todoroviæ N, et al. Antiproliferative activity and QSAR studies of a series of new 4-aminomethylidene derivatives of some pyrazol-5-ones. Bioorg Med Chem Lett 2011;21:4416-21. | [8] Makhija MT, Kasliwal RT, Kulkarni VM, Neamati N. De novo design and synthesis of HIV-1 integrase inhibitors. Bioorg Med Chem 2004;12:2317-33. | [9] Fan X, Zhang X, Zhou L, Keith KA, Kern ER, Torrence PF. A pyrimidine-pyrazolone nucleoside chimera with potent in vitro anti-orthopoxvirus activity. Bioorg Med Chem Lett 2006;16:3224-8. | [10] Tenover FC. Mechanisms of antimicrobial resistance in bacteria. Am J Med 2006;119:S3-10. | [11] Lenski RE. Bacterial evolution and the cost of antibiotic resistance. Int Microbiol 1998;1:265-70. | [12] Sivakumar KK, Rajalakshmi G, Rajasekaran A. Synthesis, characterization and biological evaluation of Schiff bases of 5-amino-4-[2-(4-nitro-1,3-benzothiazol-2-yl) hydrazinylidene]-2,4-dihydro-3H-pyrazol-3-one derivatives. Int J Drug Des Discov 2012;3:886-97 | [13]. Valverde, M.G. and T. Torroba, 2005. Sulfur-Nitrogen heterocycles. Molecules, 10: 318-320. | [14]. Mittal, A., 2009. Synthetic nitroimidazoles: Biological activities and mutagenicity relationships. Sci. Pharm., 77: 497-520. | [15]. Nagalakshmi, G., 2008. Synthesis, antimicrobial and antiinflammatory activity of 2,5-disubstituted-1,3,4-oxadiazoles. Indian J. Pharm. Sci., 70: 49-55 | [16]. Nekrasov, D.D., 2001. Biological activity of 5- and 6-membered azaheterocycles and their synthesis from 5-aryl-2, 3-dihydrofuran-2,3-diones. Chem. Heterocycl. Compd., 37: 263-275 | [17]. Sperry, J.B. and D.L. Wright, 2005. Furans, thiophenes and related heterocycles in drug discovery. Curr. Opin. Drug Discov. Devel., 8: 723-740 | [18]. Polshettiwar, V. and R.S. Varma, 2008. Greener and expeditious synthesis of bioactive heterocycles using microwave irradiation. Pure Appl. Chem., 80: 777-790 | [19]. Katritzky, A.R., 1992. Heterocyclic chemistry: An academic subject of immense industrial importance. Chem. Heterocycl. Compd., 28: 241-259 |