Original Research Paper

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Synthesis, characterization and microbiological activity of 1-(2,4-dinitrophenyl)-4-(substitutedphenyl)-6-{[(4-chloroyphenyl)methylene] amino}-3-methyl-1,4-dihydropyrano[2,3-c]pyrazole-5-carbonitrile

Chemistry

KEYWORDS: dihydropyrano[2,3-c]pyrazole , methoxybenzaldehyde, ethanol

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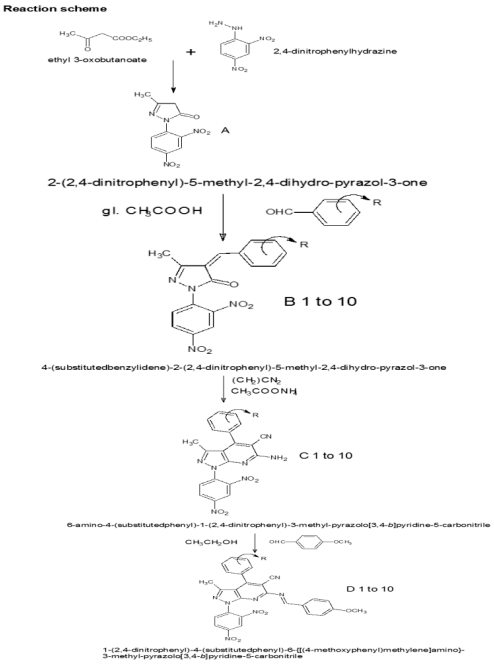
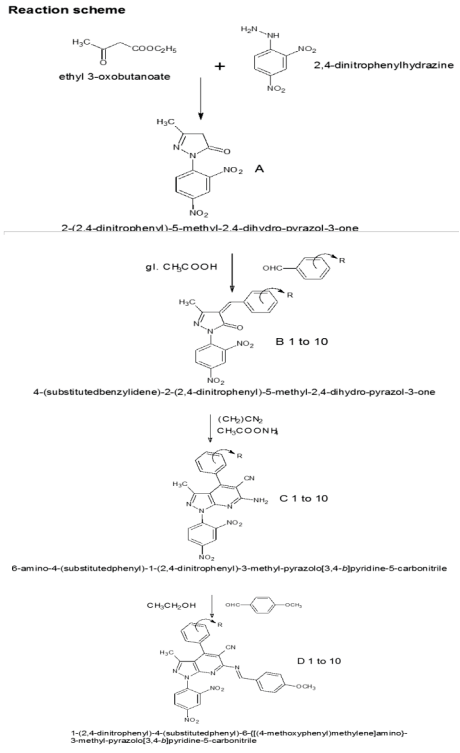
ABSTRACT

synthesis, characterization and microbiological activity of 1-(2,4-dinitrophenyl)-4-(substitutedphenyl)-6-{[(4-chloroyphenyl)methylene] amino}-3-methyl-1,4-dihydropyrano[2,3-c]pyrazole-5-carbonitrile synthesized by coupling reaction between 6-amino-4-(substitutedphenyl)-1-(2,4-dinitrophenyl)-3-methyl-pyrazolo[3,4-b]pyridine-5-carbonitrile and 4-methoxybenzaldehyde by using ethanol as a solvent. The synthesized compounds were characterized by Elemental Analysis ,Infra Red spectroscopy and NMR spectroscopy. All compound tested their microbiological activity by Cupborer method.

INTRODUCTION

The Schiff base compounds having structure, R-CH = NR^1 or R-C= NR^1 and their related derivati0ves. These compounds are known as imines, azomethines, anils or more commonly as schiff bases (where R & R^1 = alkyl or aryl) **[1]**.A large number of schiff bases are known to have useful biological activities like tuberculostatic, fungicidal **[2]** and bactericidal **[3]**.Anticonvulsant activity of the schiff base derivatives obtained from vanillin was established by Tiwari S S *et al.*[**4**]. Dash B *et al.* have synthesized and tested schiff base derivatives for fungicidal activity. **[5].** Profft Elmar *et al.* reported the synthesis and anthelmintic activity of schiff base derivatives**[6]**. Pawar R P *et al.* have synthesized schiff base derivatives and tested for their anti bacterial activity**[7]**. Modi D *et al.* have reported the synthesis and anti cancer activity of schiff bases**[8]**. Mittal has synthesized some new coumarin schiff bases, which possess anti bacterial and anti fungal activity **[9]**. Turan-Zitouni *et al.* have prepared some 5-Bromoimidazo[1,2a] pyridine-2-carboxylic acid benzylidenehydrazide and screened their antimicrobial activity **[10]**. Terzioglu and Gursoy have synthesized some novel 2,6-dimethyl-Nsubstituted phenylmethyleneimidazo[2,1-b][1,3,4]thiadiazole-5-carbohydrazides showed the most favorable cytotoxicity**[11]**. Schiff base of 4-(2-aminophenyl)morpholines (d-26) have been synthesized and studied for their analgesic, anti-inflammatory, antibacterial and antifungal activities by Panneerselvam and his co-workers**[12]**





EXPERIMENTAL WORK

For synthesis of 2-(2,4-dinitrophenyl)-5-methyl-2,4-dihydro-pyrazol-3-one mix 0.4 M of ethyl acetoacetate and 0.44 M of 2,4-dinitrophenyl hydrazine in a evoparating dish and heat it for two hours in water bath in fume cupboard with continuous stir then cool it and add 100 ml of ether and stir the mixture vigorously the product 2-(2,4-dinitrophenyl)-5-methyl-2,4-dihydro-pyrazol-3-one is obtain in 15 minute then filter it and wash with eather to remove coloured impurities.Recrystalise product of 2-(2,4-dinitrophenyl)-5-methyl-2,4-dihydro-pyrazol-3-one from hot water the yield of product was 76 % and product was melt at 95 °C

Found: C(45.42%) H(3.03%) N(21.19%), Calcd. for C10H8N4O5: C(45.46%) H(3.05%) N(21.21%) IR; (cm-1):3079(=CH), 2912(-CH, Stretch),1720(>C=O), 1600(>C=N Stretch),1499(>C=C<,aromatic ring),1557(-N=O),1463(-CH3 bend),1343(-C-N<), 1245(>N-N<).1H NMR (DMSO);: 2.55, singlate (3H) (-CH3),2.30,singlate (2H)(-CH2-), 8.16- 9.10, multiplate (3H) (Ar-H).

Preparation of 4-(substitutedbenzylidene)-2-(2,4-dinitrophenyl)-5-methyl-2,4-dihydro pyrazol -3-one (B 1 to 10)

A mixture of 2-(2,4-dinitrophenyl)-5-methyl-2,4-dihydro-pyrazol-3-one (0.01M) and substitutedbenzaldehyde (0.01M) in glacial acetic acid (25ml) was refluxed for 5 hours at a temperature of 1200C. The content was poured on to crushed ice. The isolated product was filtered, dried and crystallized from ethanol.

IR; B-1 (cm-1):3028(=CH),2913(-CH,Stretch) ,1676(>C=O), 1586(>C=N Stretch), 1487(>C=C<, aromatic ring), 1564(-N=O), 1407 (-CH3 bend), 1308(-C-N<), 1209(>N-N<), 752(-C-Cl).

1H NMR (DMSO); SP-08: 2.5871, singlate (3H)(-CH3), 2.4999, singlate (6H)[-N(CH3)2], 7.7571, singlate (1H)(Ar-CH=,Vinylic), 7.3732-9.0397, multiplate (7H) (Ar-H).

Preparation of 6-amino-4-(substitutedphenyl)-1-(2,4-dinitrophenyl)-3-methyl-pyrazolo [3,4-b]pyridine-5-carbonitrile(C 1 to 10)

A mixture of 4-(substitutedbenzylidene)-2-(2,4-dinitrophenyl)-5-methyl-2,4-dihydro-pyrazol-3-one (0.01M)react with malononitrile (0.01M) and ammonium acetate (0.08M) in absolute alcohol (30ml) and heated under refluxed for 6 hours. The content was pourer on to crushed ice. The product was isolated and crystallized from ethyl acetate.

IR ; **C-10** (cm-1): 3344(>NH-), 3063(=C-H), 2918(-C-H, str), 2192(-C_N),1574(>C=N, str), 1524(>C=C<, aromatic ring), 1495(-N=O),1414(-CH3, bend), 1397(-C-N<), 1273(>N-N<).

1H NMR (DMSO); **C-6** : 2.5989, singlate (3H)(-CH3),3.7360, singlate (3H) (-OCH3), 4.1728, singlate (2H) (-NH2), 4.9762,singlate (1H)(Ar-OH), 7.8927-8.9647,multiplate (6H)(Ar-H, >CH=CH<).

1-(2,4-dinitrophenyl)-4-(substitutedphenyl)-6-{[(4-methoxyphenyl)methylene] amino} - 3-methyl-pyrazolo[3,4-b]pyridine-5-carbonitrile

In a 250ml flask (equipped with reflux condenser) a mixture of 6-amino-4-(substitutedphenyl)-1-(2,4-dinitrophenyl)-3-methyl-pyrazolo[3,4-b]pyridine-5-carbonitrile (0.01M) , 4-methoxybenzaldehyde (0.01M) and absolute alcohol(30ml)were placed and 1 to 2 drops of hydrochloric acid was added and the mixture was then heated on water bath for 6 hours and then cooled and the precipitates were filtered off and re-crystallized from absolute alcohol.The yield of the product was 68% and the product melts at 114 °C.

Melting points were taken in open capillary tube and were uncorrected. IR spectra were recorded on I.R. Spectrophotmeter of Bruker scientific Model No. Alpha E and instrument used for NMR Spectroscopy was recorded in DMSO on Bruker Advance II 400 MHz spectrometer using TMS as an internal standard.Purity of the compounds were checked by tlc on silica- G plates.

IR ; **D-2** (cm-1): 3060**(=C-H) ,** 2910 (**-C-H (Stretch)**), 2200 (**-CN**) , 1575(**>C=N- (Stretch)**) , 1523 (**>C=C< aromatic**) , 1490 (**-N=O**) , 1405 (**-CH_3 (bend)**) , 1345 (**C-N**).

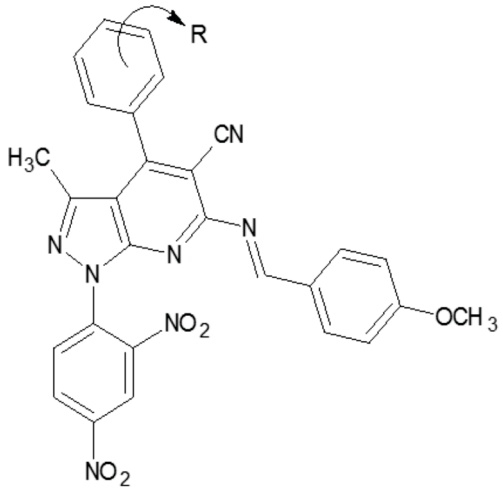
^{1}HNMR (DMSO) D-7 : δ=2.5539ppm singlet (3H)(-CH_3) , δ=3.7229 ppm singlet (3H)(-OCH_3) , δ=4.9631 ppm singlet (1H)(Ar-OH) , δ=6.8888 – 8.7067 ppm multiplate (11H)(Ar-H) , δ=8.1246 ppm singlet (1H)(Ar-CH=N-) ,

TABLE NO. 1

Physical constant of 1-(2,4-dinitrophenyl)-4-(substitutedphenyl)-6-{[(4-methoxyphenyl) methylene] amino}-3-methyl-pyrazolo[3,4-b]pyridine-5-carbonitrile

ANTI BACTERIAL CTIVITY:

synthesized compounds suggests their moderate antibacterial and antifungal activity as compared to the standard drugs Penicillin, Chloramphenlcol, Streptomycin, Tetracyclin and Amphotericin using cupboar method. Antibiotic solution is prepared in sterile distilled water.Penicillin: 12 units , Chloramphenicol: 30 µg/ml , Streptomycin: 30 µg/ml,Tetracycline: 30 µg/ml , Fluconazole: 30 µg/ml



sub	R	Formula	M.Wt g/mol	Yield %	MP °C	C %		H %		N %	
						Fou	Req	Fou	Req	Fou	Req
D-1	4-Cl	$C_{28}H_{18}ClN_7O_5$	567.93	68	114	59.16	59.21	3.15	3.19	17.20	17.26
D-2	2-Cl	$C_{28}H_{18}ClN_7O_5$	567.93	70	143	59.14	59.21	3.15	3.19	17.23	17.26
D-3	3-OCH_3 ,-4-OCH_3	$C_{30}H_{23}N_7O_7$	593.54	66	98	60.62	60.71	3.86	3.91	16.47	16.52
D-4	H	$C_{28}H_{19}N_7O_5$	533.49	70	88	63.00	63.04	3.54	3.59	18.31	18.38
D-5	2-OH	$C_{28}H_{19}N_7O_6$	549.49	67	102	61.12	61.20	3.46	3.49	17.78	17.84
D-6	3-OCH3 , 4-OH	$C_{29}H_{21}N_7O_7$	579.51	66	127	60.02	60.10	3.60	3.65	16.87	16.92
D-7	4-OH	$C_{28}H_{19}N_7O_6$	549.49	70	148	61.11	61.20	3.45	3.49	17.78	17.84
D-8	4-$N(CH_3)_2$	$C_{30}H_{24}N_8O_5$	576.56	68	108	62.40	62.49	4.15	4.20	19.35	19.43
D-9	4-OCH_3	$C_{29}H_{21}N_7O_6$	563.52	72	95	61.73	61.81	3.74	3.76	17.34	17.40
	3- NO_2	$C_{28}H_{18}N_8O_7$	578.49	69	132	58.04	58.13	3.10	3.14	19.37	19.37

Culture activation:

The culture was activated in nutrient broth and potato dextrose broth for bacteria and yeast respectively. One colony of each organism was inoculated and incubated at 37°C (bacteria) and 28°C (yeast) temperature for 24 hours. 200 µl of activated culture was inoculated in 25 ml of molted Nutrient agar and Yeast peptone agar for bacteria and yeast respectively. After proper mixing of culture it was poured in sterile 100mm Petri dish.

Over all analysis D-3,D-9 showed good anti-bacterial activity and D-6 are showed equal anti-bacterial activity for E.coli bacteria than the standard tested drugs stroptomycine against E.coli NCIM 2066. D-3 have good anti-bacterial activity then standard tested drugs Chloramphenicol and streptomycine against S.aureus MTCC 737. D-3have equal anti-bacterial activity then streptomycine and Chloramphinicol against B.spizinzenii MTCC 441. D-1 to D-10 all most compound showed very good anti-bacterial activity for P.aeruginosa than the standard tested drugs used for bio-assay against P.aeruginosa MTCC 1688. D-5 showed high anti-bacterial

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activity then the standard tested drugs used for bio-assay for S.paratyphi A. MTCC 735. D-1,D-2,D-3,D-5,D-6,D-8,D-9,D10 have High anti-bacterial activity then other standard drugs Penicillin,streptomycine and Chloramphenicol against B.pumilusMTCC 1607. D-6 have equalanti-bacterial activity for K.pneumoniae than the standard tested drug Chloramphenicol against K.pneumoniae MTCC 432. And D-1,D-9 were showed equal anti-fungal activity then the standard tested drugs used for bio-assay for C.albicans MTCC 227 Hence these compounds should be further tested under various conditions for their pharmaceutical applications.

TABLE NO. 1

No	Sample code	Microorganisms							Yeast
		E.coli NCIM 2066	S.aureus MTCC 737	B.spizinzenii MTCC 441	P.aeruginosa MTCC 1688	S.paratyphi A MTCC 735	B.pumillus MTCC 1607	K.pneumoniae MTCC 432	C.albicans MTCC 227
1	D-1	16	21	20	18	18	19	17	21
2	D-2	17	18	23	19	19	21	16	20
3	D-3	20	18	21	15	17	18	18	16
4	D-4	15	14	16	16	NI	14	15	15
5	D-5	17	18	17	18	20	17	17	17
6	D-6	19	17	18	17	18	19	19	NI
7	D-7	18	18	17	18	16	15	14	17
8	D-8	17	20	NI	18	17	17	16	18
9	D-9	20	17	20	17	15	18	15	21
10	D-10	19	22	19	16	17	19	18	17

Whwre, SD1=Penicillin, SD2=Chloramphenicol, SD3=Streptomycin, SD4=Tetracyclin SD5= Fluconazole.

Refrance :

1. Schiff H.,(1865), Ann. Chem., 3, 348.
2. Farrow W. M,Hanna C. and Scheler F. W,(1954) J. Am. Pharma.Assoc.,43,370.
3. Bahadur S, Goel A. K and Varma R. S, (1976),J. Ind. Chem. Soc.,53,590.
4. Tiwari S. S, Husain M. I and Srivastava G. C,(1981),J. Ind. Chem. Soc.,LVIII,214.
5. Dash B, Patra M. and Mahapatra P,(1983), J. Ind. Chem. Soc.,LX, 772.
6. Profft Elmar and Hogel Egon,(1974),C. A. 80,26913b.
7. Pawar R. P, Andurkar N. M,(1999), J. Ind. Chem. Soc.,76(5),271;(1999),C. A., 137,2711829y.
8. Modi D,Sabnis S. S and Deliwala C. V,(1970), J. Med. Chem.,13,935.
9. Mital A. K and Singhal U. P,(1982), J. Ind. Chem. Soc.,LIX, 373.
10. TuranZitouni G.,Blache Y and Guven K,(2001), Bull. Chim. Farm.,140,397.
11. Terzioglu N and Gursoy A,(2003), Eur. J. Med. Chem.,38,781.
12. Panneerselvam P, Priya G.M, Kumar N.R and Saravanan G.,(2009), Ind. J.Pharm.Sci.,71,428.