

SYNTHESIS AND ANTIMICROBIAL ACTIVITY OF SOME 2-PYRAZOLINE DERIVATIVES

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Summary

Some new pyrazoline derivatives were synthesized by reacting chalcones of 2-acetyl thiophene with phenyl hydrazine hydrochloride in the presence of alcohol. The synthesized compounds were identified by spectral data and screened for anti-microbial activity. Some of these compounds showed moderate to considerable anti-microbial activity.

Key words: Synthesis, Pyrazolines, Anti microbial activity.

Introduction

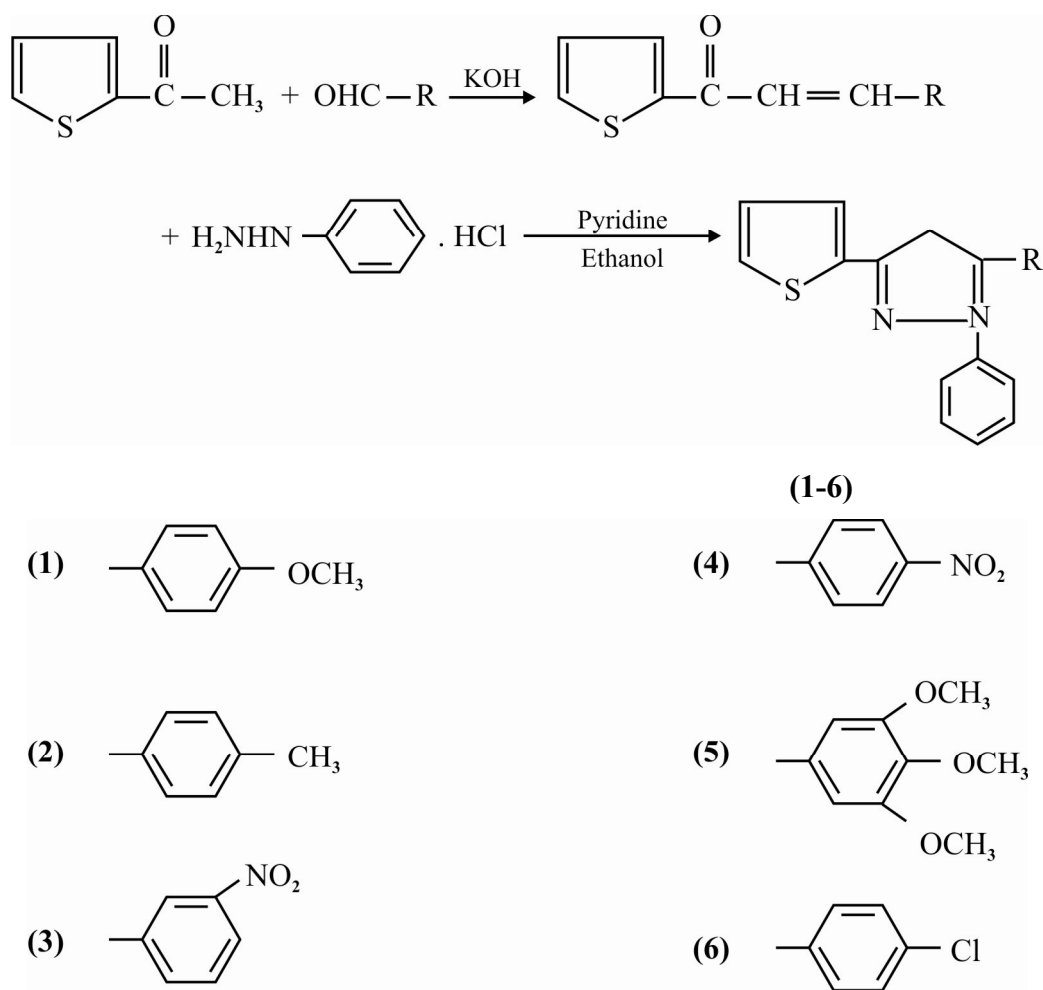
Compounds with pyrazoline structures are known to possess antimicrobial^{1,2,3}, anti-inflammatory⁴, antidepressant^{5,6}, anti-tubercular⁷ activities. In the present study some new pyrazoline derivatives (1 to 6) have been synthesized by the reaction of chalcones of 2-acetyl thiophene and phenyl hydrazine hydrochloride. The structures of the various synthesized compounds are assigned on the basis of elemental analysis, IR and ¹H NMR spectral data. These compounds were also screened for their anti-microbial activity.

Experimental

Melting points were determined on a capillary melting point apparatus and are uncorrected. ¹H NMR spectra was recorded in the indicated solvent on Bruker WM 400 MHz spectrometer with TMS as internal standard. Infrared spectra were recorded in KBr on Perkin-Elmer AC-1 spectrophotometer. Microanalyses were performed on Carlo Erba EA-1108 element analyzer and were within the $\pm 0.5\%$ of the theoretical values. Column chromatography was performed on silica gel (Merck, 60-120 mesh).

General procedure for the preparation of 2-pyrazolines (1-6)

A chalcone of 2-acetyl thiophene (0.001 mol) dissolved in 20 mL of ethanol and phenyl hydrazine hydrochloride (500 mg) was added to it. To this mixture 0.3 mL of pyridine was added drop wise at room temperature. After that the mixture was refluxed for 5-6 hours and the solvent was evaporated completely. The reaction mixture was poured into ice-cold water and the solid mass that separated out was filtered, dried and purified by column chromatography with ethyl acetate/ hexane and recrystallized from chloroform.



Scheme 1 : Synthesis of some new 2-pyrazoline derivatives

Table 1. Physical data of compounds (1-6)

Compound	M.F.	M.P (°C)	Yield (%)	Elemental analyses (%)					
				C		H		N	
				Calcd	Found	Calcd	Found	Calcd	Found
(1)	C ₂₀ H ₁₆ ON ₂ S	211	68	72.28	72.12	4.81	4.72	8.43	8.06
(2)	C ₂₀ H ₁₆ N ₂ S	172	68	75.94	75.81	5.06	5.04	8.86	8.74
(3)	C ₁₉ H ₁₃ N ₃ SO ₂	205	71	65.70	65.58	3.74	3.04	12.10	12.04
(4)	C ₁₉ H ₁₃ N ₃ SO ₂	231	72	65.70	65.42	3.74	3.73	12.10	11.74
(5)	C ₂₂ H ₂₀ N ₂ SO ₃	237	71	67.34	66.81	5.10	5.04	6.12	5.74
(6)	C ₁₉ H ₁₃ N ₂ SCI	239	74	67.65	67.64	3.8	3.74	8.30	8.28

Table 2. Spectral data of the compounds (1-6)

Compound	IR (KBr, cm^{-1})	^1H NMR (CDCl_3 , ppm)
(1)	1642 (C=N), 1355 (C-N), 670 (C-S), 1160 (OCH_3).	3.13 (1H, dd, HA) 3.77 (1H, dd, HB), 5.25 (1H, dd, HX), 3.85 (3H, S, $-\text{OCH}_3$) 6.70-7.62 (12H, Ar-H).
(2)	1648 (C=N), 1352 (C-N), 660 (C-S), 1160 (OCH_3).	2.35 (3H, S, Ar- CH_3) 3.15 (1H, dd, HA), 3.83 (1H, dd, HB) 5.28 (1H, dd, HX), 6.80-7.50 (12H, Ar-H).
(3)	1648 (C=N), 1080 (C-N), 660 (C-S), 1510 (N=O).	3.15 (1H, dd, HA), 3.86 (1H, dd, HB), 5.35 (1H, dd, HX), 6.45-7.80 (12H, Ar-H).
(4)	1594 (C=N), 1104 (C-N), 693 (C-S), 1541 (N=O).	3.15 (1H, dd, HA), 3.85 (1H, dd, HB) 5.25 (1H, dd, HX), 6.50-7.60 (12H, Ar-H).
(5)	1640 (C=N), 1068 (C-N), 655 (C-S), 1180 (OCH_3).	3.5 (1H, dd, HA), 3.78 (1H, dd, HB), 5.20 (1H, dd, HX), 6.45-7.60 (10H, Ar-H).
(6)	1595 (C=N), 1097 (C-N), 689 (C-S), 822 (C-Cl).	3.18 (1H, dd, HA), 3.85 (1H, dd, HB), 5.30 (1H, dd, HX), 6.40-7.90 (12H, Ar-H).

Antimicrobial activity

Cup plate method^{8,9} using Mueller-Hinton agar medium was employed to study the preliminary antibacterial activity of (1-5) against *B. pumilis*, *B. substilis*, *E. coli* and *P. vulgaris*. The agar medium was purchased from Hi media Laboratories Ltd., Mumbai, India. Preparation of nutrient broth, subculture, base layer medium, agar medium and peptone water was done as per the standard procedure. Each test compound (5 mg) was dissolved in 5 mL of dimethyl sulfoxide (1000 $\mu\text{g}/\text{mL}$). Volumes of 0.05 mL and 0.1 mL of each compound were used for testing.

The cups each of 9 mm diameter were made by scooping out medium with a sterilized cork borer in a petri dish, which was streaked with the organisms. The solutions of each test compound (0.05 and 0.1 mL) were added separately in the cups and petri dishes and were subsequently incubated. Sparfloxacin was used as standard reference drug (200 $\mu\text{g}/\text{mL}$) and dimethyl sulphoxide as a control, which did not reveal any inhibition. Zone of inhibition produced by each compound was measured in mm and the results are presented in Table 3.

Table 3. Antibacterial activity of the compounds (1-6)

Compound	Zone of inhibition (in mm)							
	<i>B. subtilis</i>		<i>B. pumilis</i>		<i>E. coli</i>		<i>P. vulgaris</i>	
	0.05mL	0.1mL	0.05mL	0.1mL	0.05mL	0.1mL	0.05mL	0.1mL
Standard	23	25	26	28	19	24	20	23
Control	-	-	-	-	-	-	-	-
1	09	12	09	12	07	10	09	13
2	10	13	12	13	11	09	13	11
3	13	16	10	15	13	16	12	16
4	15	20	13	17	12	15	11	17
5	16	21	14	18	14	18	12	18
6	16	20	15	17	14	16	13	18

Control: (Dimethyl sulfoxide), Standard (sparfloxacin)

Results and Conclusion

Antibacterial activity: The screening results revealed that the compounds (1-6) showed significant antibacterial activity at both 0.05 mL and 0.1 mL concentration levels when compared with standard drug sparfloxacin and other compounds 3, 4 and 5 showed moderate activity.

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