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Synthesis and Evaluation of Some Novel 2-Mercapto-3-(substitutedmethyl amino)quinazolin-4(3H)-ones as Analgesic, Antiinflammatory and Antibacterial Agents

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In view of the potent analgesic, antiinflammatory and antimicrobial activities exhibited by quinazolin-4(3H)-ones, a series of novel 2-mercapto-3-(substitutedmethyl amino)quinazolin-4(3H)-ones have been synthesized. When these compounds were evaluated for their analgesic, antiinflammatory and antibacterial activities, the compounds, I, II, III, IV, V and X exhibited comparable analgesic activity with the standard diclofenac sodium and it was found to be significant as compared to control.

Bacterial infections often produce inflammation and pain. In normal practice, two groups of agents (chemotherapeutic, analgesic and antiinflammatory) are prescribed simultaneously. The compounds possessing all three activities are not common. Quinazolines and condensed quinazolines received the attention of medicinal chemists due to its wide range of biological activities which include analgesic and antiinflammatory[1-3], antibacterial[4-7], antiviral[8-9], antihistaminic[10], antihypertensive[11-12] and anticancer[13] activities. Mannich bases were reported to possess potent antibacterial activity[14]. In the present study it was envisaged that a drug molecule possessing the above mentioned pharmacophore could be of advantage since it might possess analgesic antiinflammatory and antibacterial activities. The target compounds, 2-mercapto-3-(substitutedmethyl amino)quinazolin-4(3H)-ones [Mannich bases of 3-amino-2-mercaptoquinazolin-4(3H)-one] were synthesized by condensing the active hydrogen atom of 3-amino of 3-amino-2-mercapto quinazolin-4-(3H)-one(3), with formaldehyde and the desired amines [Mannich reaction]. The starting material (3) was synthesized from anthranilic acid by a novel innovative route (Scheme I). All compounds (Table - I) gave satisfactory elemental analysis. IR and NMR spectra consistent with the assigned structure. All the synthesized compounds were tested for analgesic antiinflammatory and antibacterial activities.

Melting points were taken in open capillary tubes on a Thomas Hoover melting point apparatus and are uncorrected. IR spectra were recorded in KBr on Perkin Elmer-841 grating spectrometer (γ max in cm^{-1}, mass spectra on Varian Atlas CH-7 mass spectrometer at 70 eV and NMR spectra on Varian A-60 or EM-360 spectrometer at 60 MHZ, using TMS as internal standard.

The compound 3-amino-2-mercapto quinazolin-4-(3H)-one (3) was synthesized by adding carbondisulphide (1.6 ml, 0.026 mol) and aqueous sodium hydroxide

*For Correspondence

(1.2 ml, 20 M) dropwise to a vigorously stirred solution of anthranilic acid (2.75 g, 0.02 mol) in dimethylsulfoxide (10 ml) at room temperature. After thirty min dimethyl sulfate (2.5 g, 0.02 mol) was added dropwise under cooling with an ice bath. Stirring was continued for 3 h, the reaction mixture was then poured into ice water and extracted with chloroform. The solvent was removed by distillation under reduced pressure. Thus the obtained methyl-N-(o-carboxyphenyl)dithiocarbamate was used for further reaction without purification. Hydrazine hydrate (8.6 g, 0.2 mol 80%) was added dropwise with stirring to methyl-N-(o-carboxyphenyl)dithiocarbamate in cold condition. After completing addition stirring was continued for 1 1/2 h at 50° and then it was poured into ice water, the solid obtained was filtered, washed with water dried and recrystallized from DMF-ethanol to yield (3) as white crystaline product. Yield : 3.4 g (90%); M.P. : 236-237° (reported 236-237°)[15]; IR (KBr) : 3300, 3220 (NH_2), 2990 (CH), 2560 (SH), 1680 (C=O); NMR-($CDCl_3$) δ ppm: 3.21 (s,1H,SH), 5.12 (s,2H,NH_2D_2o exchangeable), 7.14 (m,4H,Ar-H); Anal ($C_8H_7N_3OS$) C,H,N.

The title Compound,2-mercapto-3-(N,N-dimethyl-aminomethyl amino) quinazolin-4(3H)-one (I) was synthesized by adding a mixture of formalin (1ml) and dimethyl amine (0.23 g, 0.005 mol) drop by drop with stirring to a slurry of 3-amino-2-mercapto quinazolin-4(3H)-one (3) (0.96 g, 0.005 mol) in dimethyl formamide 15 ml. The reaction mixture was then heated at 50° with stirring for thirty minutes, after cooling it was poured into ice-water the solid obtained was filtered washed with water dried and recrystallized from alcohol-chloroform mixture to Yield : 0.94 g (75%); M.P : 142°; IR (KBr); 3380 (NH), 3050(CH), 2850 (CH), 2500(SH), 1700 (C=O); NMR ($CDCl_3$) δ ppm: 1.9 (s,2H,CH_2), 2.1 (s,6H, 1-2CH_3), 7.1 (m, 4H, Ar-H), 8.6 (t, 1H, NH); Anal ($C_{11}H_{14}N_4OS$) C,H,N. Compounds (II-X) were prepared using the same methodology.

Test for analgesic activity was performed by tail flick technique[16,17] using the mouse a testing animal. Animals were divided into various group each consisting of six animals. Test compounds (I-X) and standard diclofenac sodium were administered orally at a dose of 20 mg/kg as an aqueous suspension in 1% sodium carboxymethyl cellulose (Na CMC), while the control group was fed with the same volume of 1% Na CMC suspension, the reaction time was recorded at 15 and 30 min, 1, 2 and 3 after the treatment. The per cent analgesic activity (PAA) was calculated by the following formula.

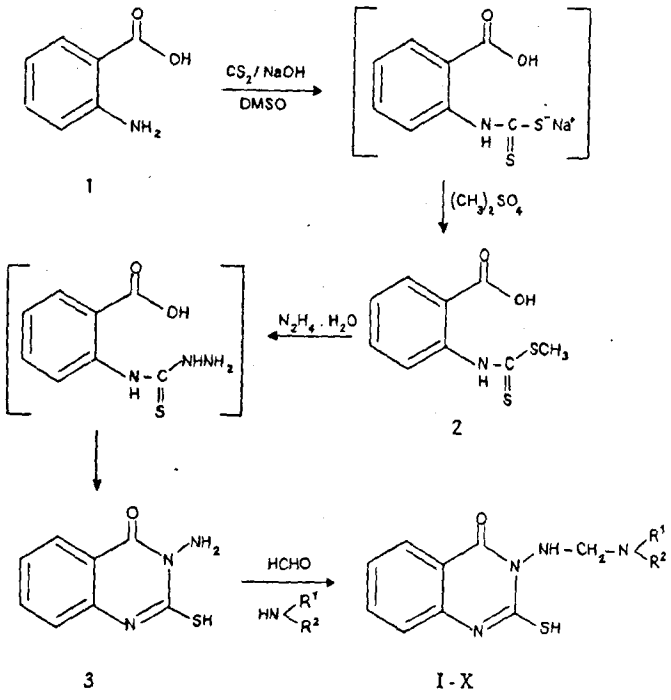
$$PAA = (T_2/T_1) \times 100$$

Where T_1 is the reaction time (s) before treatment; T_2 is the reaction time (s) after treatment. The results are presented in table-1.

Antiinflammatory activity was measured using the carrageenan-induced paw odema test in rats[18]. Animals were divided into different groups each consisting of six animals. Test compounds and standard diclofenac sodium were administered orally at a dose of 20 mg/Kg as an aqueous suspension in 1% Na CMC, while the control group was fed with the same volume of 1% Na CMC suspension. the paw volume were measured using the mercury displacement technique with the help of a plethysmometer immediately before and 15 and 30 min, 1, 2 and 3 after the carrageenan injection. The per cent inhibition of paw volume was calculated by using the following formula.

$$\text{Per cent inhibition I} = 100\left(1 - \frac{(a-x)}{(b-y)}\right)$$

Where x is the mean paw volume of rats before the administration of carrageenan and test compounds or standard compound; a stands for mean paw volume of rats after the administration of carrageenan in the control



SCHEME I

TABLE 1 : CHARACTERIZATION DATA AND PERCENT ANALGESIC ACTIVITY (±SEM) OF 2-MERCAPTO-3-(SUBSTITUTEDMETHYL AMINO) QUINAZOLIN-4(3H)-ONES (1-X)

Compd. No.	Substitution	Molecular Weight	MP°	Yield %	Percent Analgesic Activity 1st h.	2nd h.	3rd h.
I	Dimethylamino	250	142	75	296.65 ±1.04+	230.00 ±3.22+	175.75 ±2.69+
II	Diethylamino	278	149	80	299.60 ±3.32+	221.81 ±3.19+	166.15 ±2.67+
III	Pyrrolidino	276	156	77	292.10 ±4.16+	260.00 ±1.13+	175.25 ±3.76+
IV	Morpholino	292	146	73	294.10 ±1.02+	228.30 ±1.22+	165.30 ±1.48+
V	Piperazinyl	291	151	80	286.25 ±1.85+	223.75 ±1.65+	185.23 ±2.06+
VI	Anilino	298	139	76	289.11 ±2.49+	159.16 ±1.09+	123.91 ±2.79+
VII	p-Carboxyphenyl amino	342	162	79	260.00 ±3.57+	189.99 ±4.88+	168.33 ±2.25+
VIII	p-Sulphonamido phenylamino	377	169	77	225.00 ±1.81+	114.11 ±3.49+	100.00 ±2.39+
IX	2-Aminopyridinyl	299	130	81	206.61 ±1.91+	180.00 ±5.19++	172.91 ±2.23+
X	1-Benzimidazolyl	323	179	82	296.60 ±4.38	209.80 ±2.87+	165.25 ±2.71+
Control	-	-	-	-	4.44 ±1.176	3.17 ±2.97	5.98 ±1.45
Diclo-fenacsodium	-	-	-	-	312.50 ±1.93+	229.16 ±1.18+	195.23 ±3.45+

+denotes significant differences form control at $p \leq 0.05$.

group; Y is the mean paw volume of rats before the administration of carrageenan in the control group and is the mean paw volume of rats after the administration of carrageenan in the control group.

All the compounds synthesized (I-X), were screened for their antibacterial activity by agar cup-plate method[19] at a concentration of 300 µg/ml using DMF as a solvent against the following organisms. *Salmonella typhi; Escherchia coli. Vibrio cholerae, Staph. Epidermitis* and Klebsiella pneumoniae. The zone of inhibition of each strain was recorded. The activity has been compared with known standard drug ciprofloxacin at 10 µg/ml concentration. The biological results were analysed statistically by student 't' test.

While all the compounds exhibited analgesic activity (table-1) the compounds I, II, III, IV, V and X shown comparable analgesic activity with that of standard diclofenac sodium and are found to be significant when compared to control group. But none of the compounds found to be equipotent with that of standard.

Antiinflammatory activity studies indicated that compounds I, II, IV and X were found to possess good antiinflammatory activity and these compounds provided 32.17, 32.82, 34.45 and 30.46 per cent protection respectively against carrageenan-induced paw odema. While the standard, diclofenac sodium gave 45.66 per cent protection. The test of compounds exhibited moderate antiinflammatory activity.

All the test compounds exhibited mild to moderate antibacterial activity (at 300 µg/ml) while, none of the compounds exhibited comparable activity with that of standard ciprofloxacin (At 10 µg/ml). Which was effective against *S. typhi, E. Coli. V. cholerae, Staph. epidermitis, K. pneumoniae* with the zones of inhibition 23, 25, 20, 21, and 22 mm respectively. However, the compounds VII and VIII exhibited better activity against all bacteria tested.

From the results of biological activities of the test compounds, it is concluded that, these compounds possess analgesic, antiinflammatory and antibacterial activities. For some of the bacterial infections chemotherapeutic, analgesic and antiinflammatory agents are prescribed together, the compounds possessing all the three activities are not common, although the test compounds posses not only analgesic and antiinflammatory activities but also antibacterial activity, their efficacy is not enough to develop them into clinically useful agents. Hence the necessary structural modifications have to be made to improve the potency of these agents, so as to develop them into clinically useful novel class of agents that possess not only analgesic and antiinflammatory activity but also antibacterial activity.

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