Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2012, 4(5):2775-2780

Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Synthesis and screening of chromene-2-one derivatives for antipsychotic activity

Pankaj Arora[*1], M S Ranawat[2], Namita Arora[3]

[1]*Department of Pharmaceutical Chemistry, Jaipur College of Pharmacy, Jaipur, Rajasthan, India*
[2]*Department of Pharmaceutical Chemistry, B N College of Pharmacy, Udaipur, Rajasthan, India*
[3]*Department of Pharmacognosy, Jaipur College of Pharmacy, Jaipur, Rajasthan, India*

ABSTRACT

Various Coumarin derivatives (2a-2f) were synthesized and evaluated for their antipsychotic action using Apomorphine induced behavior model, 5-HTP induced behavior model, Conditioned avoidance response model and Rota rod experiment. All compounds showed significant (P<0.05) antipsychotic activity during study and it was found that target compounds have affinity for dopamine as well as for 5-HT receptors.

Keywords: Schizophrenia, Dopaminergic antagonist, Stereotype

INTRODUCTION

Schizophrenia is a psychotic disorder characterized by positive symptoms such as hallucinations and disorganized thought, and negative symptoms such as apathy and social withdrawal. This socially and economically debilitating disease is fairly common & is striking approximately 1% of the population. [1]. Classical or typical antipsychotic drugs antagonizing central dopaminergic receptors have been used for several decades in the treatment of psychiatric disorders, e.g. schizophrenia [2]. Although these drugs can reduce the positive symptoms of schizophrenia, they unfortunately often induce extrapyramidal motoric side effects and are furthermore often not able to ameliorate the negative symptoms of schizophrenia. The antipsychotic action has been suggested to be due to blockade of the mesocorticolimbic dopaminergic system, while the motoric side effects are believed to be due to antagonism of dopaminergic receptors in the nonlimbic nigro-striatal dopamine system of the brain [3]. While a diversified group of the so called atypical antipsychotic drugs express increased effectiveness in negative, affective and cognitive symptoms, including efficacy in patients resistant to standard therapy. Atypical antipsychotic drugs also have a low incidence of extrapyramidal side effects and prolactinaemia but may produce other undesirable side effects (e.g. agranulocytosis), that limit their clinical use.

Chromene-2-one & its derivatives are known to possess diverse biological activities such as antibacterial [4], anti-inflammatory [5], antineoplastic [6], antitubercular [7] & antioxidant [8]. Recently it has been reported that Chromene-2-one derivatives possess selective dopamine D_4 antagonistic activity [9] & 2-piperazinylbenzothiazoles possess high affinity for 5-HT_{1A} & 5-HT_3 receptors [10]. In this direction our efforts were devoted to combine Chromene-2-one nucleus with different benzothiazole derivatives to obtain compounds having affinities for both dopamine and 5-HT receptors.

EXPERIMENTAL SECTION

Synthetic part

In present work all synthetic reactions were monitored by TLC. All the synthesized compounds (1a-1f) were characterized by analytical and spectroscopic methods. Melting points (Table-1) were determined on Veego melting point apparatus, model no-MPI by open capillary method and are uncorrected. The FTIR spectra (Table-1) were recorded on Jasco FTIR instrument model no-5300, using KBr pellets. ^{1}H-NMR spectra (Table-2) were recorded on BRUKER AVANCE II 400 NMR spectrometer at 400 MHz, for which $CDCl_3$ was used as solvent and TMS as internal standard. Mass spectra (Table-1) were recorded on Micromass Q-Tof Micro, Mass spectrometer.

General procedure for synthesis of 7-Hydroxy-4-Methyl coumarin nucleus, (step-I)

The method of Pechmann and Duisburg was followed for the synthesis of Chromeme-2-one nucleus [11].

General procedure for synthesis of 7-(3-chloropropoxy)-4-Methyl coumarin derivatives (step-II)

3g of 7-Hydroxy-4-methyl Coumarin and 2.47 ml of 1-Bromo-3-chloro propane were added to round bottom flask containing 30ml of acetonitrile. 0.01 moles of anhydrous potassium carbonate was added to reaction mixture and refluxed for 30 hrs. The solvent was removed under vacuum and residue was dissolved in dichloromethane. Dichloromethane layer was washed with water and then with 5% w/v sodium hydroxide solution, and added with anhydrous sodium sulphate, and kept overnight. The crude 7-(3-chloropropoxy) 4-methyl Coumarin formed was collected and purified by recrystallization from ethanol [12].

General procedure for synthesis of 6-Substituted 2-aminobenzothiazole derivatives,(step-III)

0.06 moles of aniline derivative & 0.06 moles of potassium thiocyanate were added to 150 ml of glacial acetic acid (previously cooled to 5**°**C). The mixture was placed in freezing mixture of ice & salt and mechanically stirred, while bromine (0.02 moles of bromine in 10 ml glacial acetic acid) was added from a dropping funnel at such a rate that temperature does not rise beyond 0-5**°**C. After addition of bromine (105 min), the solution was stirred for an additional 2 hours at 0-10**°**C. The residue was filtered and dissolved in hot water (150 ml). The resulting solution was filtered and filtrate was neutralized with ammonia solution to pH 6.0. The precipitate was collected and crystallized with ethanol [13].

General procedure for synthesis of 6-Substituted (1'-chloroacetyl)-2-aminobenzothiazole derivatives, (step-IV)

To a stirred solution of 6-substituted 2-aminobenzothiazole (0.05 moles) and triethylamine (0.05 moles) in dry benzene (50 ml), chloroacetyl chloride (0.05 moles) was added drop wise to an ice-cold condition. The reaction mixture was stirred for about 6 hours and the separated amine hydrochloride was filtered off. The filtrate was refluxed on a water-bath for about 4 hr, concentrated at reduced pressure and the separated solid was purified over the column of silica gel using chloroform as an eluant. [14].

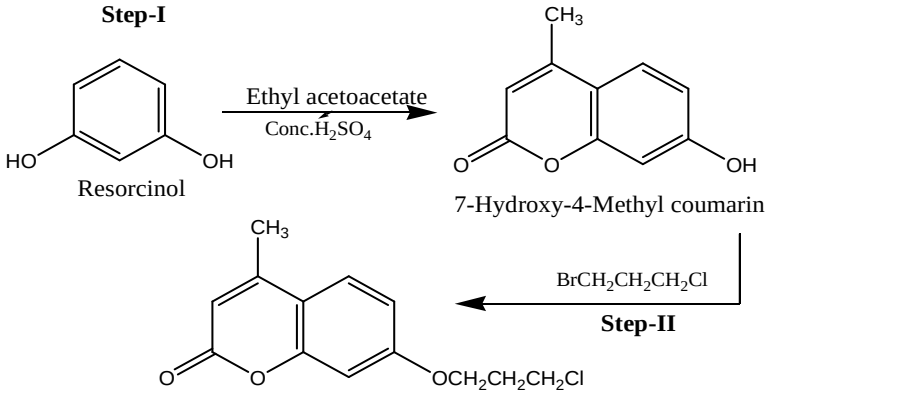
General procedure for synthesis of 6-Substituted (1'-N-Boc piperazinylacetyl)-2-aminobenzothiazole derivatives, (step-V)

The 6-Substituted (1'-chloroacetyl)-2-aminobenzothiazole derivative (20.6 mmol) was dissolved in fresh dry dichloromethane (100 ml), and triethylamine (5 ml) was added to it. N-Boc piperazine (20.6 mmol) was added slowly at 0˚C to the above reaction mixture and stirred at ambient temperature for overnight. Reaction mixture was quenched with water (100 ml) and extracted with dichloromethane (2×100 ml) and the combined organic layer was washed with water (100 ml) and brine solution (50 ml), dried over sodium sulfate, filtered and concentrated. The solid crude product was collected and recrystallized from ethanol [15].

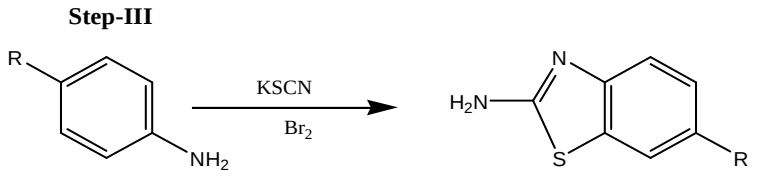
General procedure for synthesis of 6-Substituted (1'-piperazinylacetyl)-2-aminobenzothiazole derivatives, (step-VI)

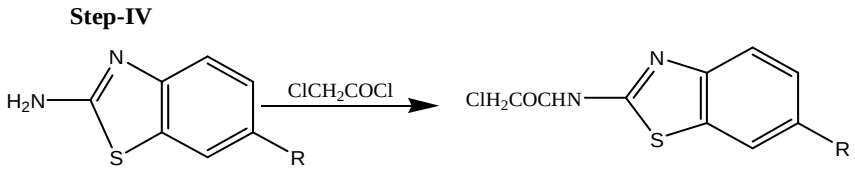
The 6-Substituted (1'-N-Boc piperazinylacetyl)-2-aminobenzothiazole derivative (12.7 mmol) was dissolved in fresh dry dichloromethane (40 ml), and trifluoroacetic acid (25.5 mmol) was added slowly at 0˚C then stirred at ambient temperature for 2 hours. Reaction mixture was quenched with water (100 ml) and basified with sodium bicarbonate solution (pH~8.0), and extracted with dichloromethane (2×150 ml). The combined organic layer was washed with water (50 ml) and brine solution (50 ml), dried over sodium sulfate and evaporated solvents under vacuum. The solid crude product was collected and recrystallized from ethanol [16].

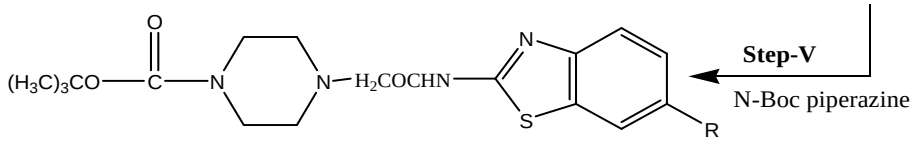
Figure 1: Scheme of synthesis

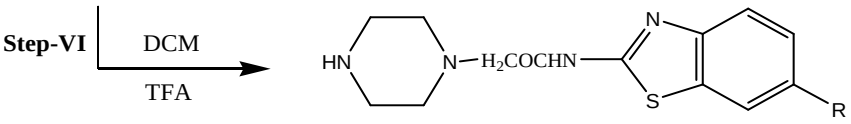


7-(3-chloropropoxy)-4-Methyl coumarin derivative **[Compound X]**

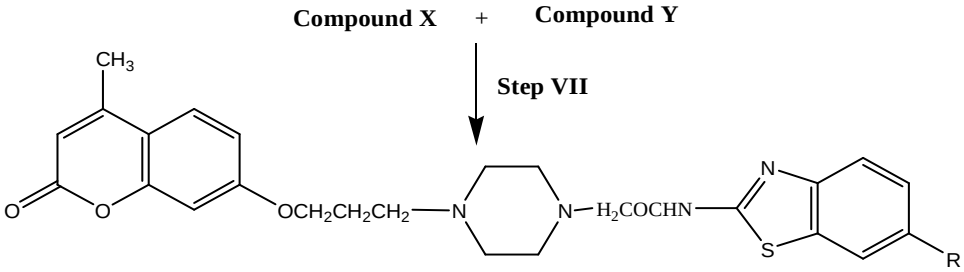








6-Substituted (1'-piperazinylacetyl)-2-aminobenzothiazole derivatives **[Compound Y]**



Compound 2a-2f

General procedure for synthesis of derivatives 2a-2f (step-VII)
The 6-Substituted (1'-piperazinylacetyl)-2-aminobenzothiazole derivative (20.0 mmol) and 7-(3-chloropropoxy) 4-methyl Coumarin (20.0 mmol) were dissolved in fresh dry dichloromethane (100 ml), and triethylamine (5 ml) was added slowly at 0˚C then stirred at ambient temperature for 12 hours. Reaction mixture was quenched with water (100 ml) and extracted with dichloromethane (2×100 ml). The combined organic layer was washed with water (100 ml) and brine solution (50 ml), dried over sodium sulfate and evaporated solvents under vacuum. The solid crude product was collected and recrystallized from ethanol.

Pharmacological Studies
All the experiments were performed as per CPCSEA guidelines, with the permission of Institutional Animal ethical committee (IAEC). Swiss albino rats (male or female, body weight: 80-120 gm) maintained under hygienic laboratory conditions were fasted for 24 hours before experimentation. All the synthesized compounds were administered through intra peritoneal (i.p.) route. Haloperidol (1mg/kg) and Risperidone (1mg/kg) were used as standard drugs.

Determination of minimum effective dose [ED_{min}]: Swiss albino rats were divided into six groups, having six animals in each group (n=6). Minimum dose [ED_{min}] required for preventing the apomorphine induced climbing response & 5-HTP induced head twitches response was recorded.

Apomorphine induced behavior: During this experiment, animals were pretreated with standard drug (1mg/kg) or test compound, 30 minutes before treatment with apomorphine (4mg/kg) [16]. Inhibition of apomorphine induced climbing behavior was recorded. Results are shown in table: 3.

5-HTP induced behavior: During this experiment, animals were pretreated with standard drug or test compound, 30 minutes before treatment with 5-HTP (50mg/kg) [16]. Inhibition of 5-HTP induced head twitches were recorded at 10 min time interval. Results are shown in table: 4.

Conditioned avoidance response using Pole climbing apparatus: Before experiment rats were trained by a shock treatment of 30 seconds duration to climb the pole. The conditioning stimulus was a buzzer. During this experiment, animals were treated with standard drug or test compound. After 30 minutes the animals were observed [17, 18]. Results are shown in table: 5.

Rota rod experiment: Before experiment rats were trained by placing them on a scraped rotating rod (25 rpm) of the Rota rod assembly to remain there on the rod at least for 3 minutes. After 30 minutes of treatment with standard drug or test compound, time of fall from the rotating rod for animals was observed [17, 18]. Results are shown in table: 5.

Statistical analysis
All the results were statistically evaluated using student's t-test and were found significant when $P<0.05$.

Table-1: Physicochemical parameters and IR spectral data of synthesized compounds (2a-2f)

Compound	-R	Yield (%)	M.P. (˚C)	R_f value	IR (KBr disc, cm^{-1})	Mass Spectra (m/z)
2a	-H	57	156-58	0.394*	3060,2955,2746,1728,1622,1524, 1470,730,1390,3355	490.8
2b	-Br	63	171-73	0.572*	3041,2932,2732,1767,1620,1538,1456,710,1295,3367,965	569.4
2c	-Cl	60	164-66	0.596*	3154,2930,2658,1714,1635,1512,1475,715,1345,3330,1050	525,2
2d	-F	65	128-30	0.524*	3088,2933,2740,1745,1628,1514,1460,714,1378,3318,1152	508.6
2e	$-CH_3$	50	139-41	0.463*	3114,2948,2664,1735,1640,1564,1472, 680,1366,3340,1438	506.1
2f	$-NO_2$	53	150-52	0.410*	3155,2976,2642,172,1655,1565,1455,756,1354,3354,1535	535.7

**(Benzene: Ethyl acetate: 3:2)*

RESULTS

All synthesized compounds were in conformity with the structure envisaged on the basis of spectral data. The minimum effective doses [ED_{min}] for title compounds (2a-2f) were recorded as 70, 50, 60, 40, 45, 65 mg/kg,

respectively. All compounds showed inhibition of apomorphine and 5-HTP induced behavior. All synthesized compounds showed less muscular rigidity than standard drug haloperidol during Rota rod experiment. During Pole climbing experiment all synthesized compounds as well as standard drugs showed tranquilizing effect.

Table-2: ^{1}H-NMR spectral data

Compound	^{1}H-NMR (δ, 400MHz, $CDCl_3$)
2a	δ 7.20-7.84 (m, 7H, Ar-H), δ 1.70 (m, 3H, -CH_3), δ 8.15 (s, 1H, -CONH), δ 2.46-3.15 (m, 12H, N-CH_2)), δ 4.20 (m, 2H, O-CH_2), δ 1.82 (m, 2H, O-(C)-CH_2), δ 5.76 (s, 1H, Pyrane-2-one ring)
2b	δ 7.10-7.90 (m, 6H, Ar-H), δ 1.71 (m, 3H, -CH_3), δ 8.12 (s, 1H, -CONH), δ 2.48-3.12 (m, 12H, N-CH_2)), δ 4.22 (m, 2H, O-CH_2), δ 1.80 (m, 2H, O-(C)-CH_2), δ 5.70 (s, 1H, Pyrane-2-one ring)
2c	δ 7.15-7.82 (m, 6H, Ar-H), δ 1.73 (m, 3H, -CH_3), δ 8.10 (s, 1H, -CONH), δ 2.40-3.10 (m, 12H, N-CH_2)), δ 4.18 (m, 2H, O-CH_2), δ 1.78 (m, 2H, O-(C)-CH_2), δ 5.66 (s, 1H, Pyrane-2-one ring)
2d	δ 7.11-7.90 (m, 6H, Ar-H), δ 1.69 (m, 3H, -CH_3), δ 8.10 (s, 1H, -CONH), δ 2.40-3.16 (m, 12H, N-CH_2)), δ 4.21 (m, 2H, O-CH_2), δ 1.71 (m, 2H, O-(C)-CH_2), δ 5.69 (s, 1H, Pyrane-2-one ring)
2e	δ 7.10-7.83 (m, 6H, Ar-H), δ 1.72-1.86 (m, 6H, -CH_3), δ 8.12 (s, 1H, -CONH), δ 2.42-3.15 (m, 12H, N-CH_2)), δ 4.12 (m, 2H, O-CH_2), δ 1.76 (m, 2H, O-(C)-CH_2), δ 5.65 (s, 1H, Pyrane-2-one ring)
2f	δ 7.08-7.88 (m, 6H, Ar-H), δ 1.70 (m, 3H, -CH_3), δ 8.16 (s, 1H, -CONH), δ 2.38-3.10 (m, 12H, N-CH_2)), δ 4.24 (m, 2H, O-CH_2), δ 1.78 (m, 2H, O-(C)-CH_2), δ 5.68 (s, 1H, Pyrane-2-one ring)

Table-3: Inhibition of Apomorphine induced climbing behavior (D_2 antagonism)

Compound	Dose	Time spent* (min)	Time spent (%)	% Inhibition
Control	-	22.42 (1.54)	74.73	-
Haloperidol	1 mg/kg	01.25 (1.40)	04.16	94.43
Risperidone	1 mg/kg	02.24 (1.34)	07.46	90.01
Compound 2a	70 mg/kg	02.55 (1.22)	08.50	88.62
Compound 2b	50 mg/kg	02.42 (1.76)	08.06	89.21
Compound 2c	60 mg/kg	02.15 (1.42)	07.16	90.41
Compound 2d	40 mg/kg	01.55 (1.12)	05.16	93.09
Compound 2e	45 mg/kg	06.10 (1.42)	20.33	72.79
Compound 2f	65 mg/kg	05.16 (1.36)	17.20	76.98

***** *The value in parenthesis indicates standard deviation (n=6, p* <0.05)*

Table-4: Inhibition of 5-HTP induced head twitches (5-HT antagonism, %)

Compound	Dose	Observation at interval of 10 minutes					
		10	20	30	40	50	60
Control	-	0.00	0.00	0.00	0.00	0.00	0.00
Haloperidol	1 mg/kg	05.00	10.52	25.00	12.5	08.33	20.00
Risperidone	1 mg/kg	25.00	21.05	45.00	43.75	41.66	70.00
Compound 2a	70 mg/kg	00.00	10.52	25.00	25.00	08.33	30.00
Compound 2b	50 mg/kg	05.00	10.52	30.00	25.00	25.00	20.00
Compound 2c	60 mg/kg	10.00	15.78	35.00	37.50	25.00	20.00
Compound 2d	40 mg/kg	20.00	26.31	50.00	37.50	33.33	60.00
Compound 2e	45 mg/kg	05.00	15.78	30.00	25.00	08.33	30.00
Compound 2f	65 mg/kg	10.00	21.05	30.00	31.25	16.66	40.00

(n=6, p <0.05)*

DISCUSSION

Apomorphine is an agonist of dopamine receptors and it induces circle movement, rearing and climbing in the animals. The inhibition of apomorphine induced behavior is a measurement of dopaminergic antagonistic activity. All synthesized compounds showed inhibition of apomorphine induced behavior which shows that all compounds have dopaminergic antagonistic activity. 5-HTP is an agonist of 5-HT receptors and it induces head weaving and head twitching in the animals. The inhibition of 5-HTP induced behavior is a measurement of 5-HT antagonistic activity. During present work all synthesized compounds showed inhibition of 5-HTP induced behavior which shows that all compounds have 5-HT antagonistic activity. Pole climbing experiment is performed to check conditioned avoidance response as a measurement of the tranquilization. All compounds showed the tranquilizing

effect. Antipsychotic drugs produce extra pyramidal side effects like rigidity of muscles; therefore to determine these side effects Rota rod experiment was performed. During experiment it was observed that synthesized compounds showed less muscular rigidity than standard drug haloperidol.

Table-5: Pole climbing and Rota rod Experiment

Compound	Pole climbing experiment	Rota rod experiment
	Time taken for pole climbing (seconds)	Time of fall from rod* (seconds)
Normal control	18	112 (1.88)
Haloperidol	Unable to climb	02(1.21)
Risperidone	Unable to climb	03(1.32)
Compound 2a	Unable to climb	06(1.22)
Compound 2b	Unable to climb	05(1.30)
Compound 2c	Unable to climb	05(1.20)
Compound 2d	Unable to climb	04(1.36)
Compound 2e	Unable to climb	05(1.30)
Compound 2f	Unable to climb	03(1.42)

****** The value in parenthesis indicates standard deviation (n=6, p* <0.05)*

CONCLUSION

In present investigation it was observed that all synthesized compounds showed promising antipsychotic effect and have dopaminergic as well as 5-HT receptor antagonistic activity with lesser extra pyramidal side effects. Therefore these compounds can be placed between classical and atypical antipsychotic drugs.

REFERENCES

[1] Reynolds G.P. *Trends Pharmacol. Sci*. 13; **1992**: 116-21.
[2] Seeman P. *Pharmacology Review*. 32; **1980**: 230-313.
[3] Gudelsky, G.A. et al. *Psychopharmacology* (Berlin). 99; **1989**: S13-S17.
[4] Hanmantgad S.S. et al. *Indian J Chem.* 24B; **1985**: 454.
[5] Ghate Manjunath et al. *Eur. J. Med. Chem*. 40; **2005**: 882-87.
[6] Kostava Irena, Momekov Georgi. *Euro. J. Med. Chem*. 20; 2007: 1-11.
[7] Sanghi Y. S. et al., *J. Med Chem*. 32 (5); 1989: 945-51.
[8] Torres Rene, Faini Francesca. *Phytochemistry*. 67; **2006**: 984-87.
[9] Gonzalez-Gornez J. C. et al. *Bioorg Med Chem Lett*. 13(2); **2003**: 175-78.
[10] Diouf O. et al. *Eur. J. Med. Chem*. 30; **2005**: 715-19.
[11] Furniss B S, Hannaford A J, Smith P W J, Tatchell A R, Vogel's Text book of Practical Organic Chemistry. Fifth edition. Singapore: Pearson Education (P) Ltd. **2004**.
[12] Li J.J., Johnson D.S., Sliskovie D.R. and Roth B.D. Contemporary Drug Synthesis. USA: A John Willey and Sons, Inc., Publication. **2004**.
[13] Yadav V. P. et al, *Indian Drugs.* 45(8); **2008**: 655-58.
[14] Srivastava S.D., Sen J.P. *Indian J Chem.* 47B; **2008**: 1583-86.
[15] Dhayanithi V et al. *Org. Commun*. 3(3); **2010**: 45-56.
[16] Turner A.R., Screening methods in pharmacology, London: Academic press. **1965**.
[17] Kulkarni S.K., Handbook of Experimental Pharmacology, Third edition reprint. India: Vallabh Prakashan, Delhi. **2007**.
[18] Ghosh M.N., Fundamentals of Experimental Pharmacology, Third edition. India: Hilton and company Publishers, Kolkata. **2005**.