

## Original Article

# Synthesis, anticonvulsant (Chemo shock) activity of Isatin Mannich bases of quinazolone derivative

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### ABSTRACT

New N-methyl isatin [N-methyl -3-aryl -3H-quinazoline-4-one] derivatives 4(a-d) were synthesized and screened for anticonvulsant activity by strychnine, isoniazid (INH) and thiosemicarbazide induced chemo shock convulsion models. The neurotoxicity of the compounds was found by rotarod test. All the compounds were reported moderate protection of mice at 30 mg/kg at 0.5-2 hr. in the entire test. No neurotoxicity was observed at the highest dose of 300 mg/kg for all the compounds.

## 1. INTRODUCTION

Epilepsy is ubiquitous disease characterized by recurrent seizures and inflicts more than 60 million people worldwide according to epidemiological studies [1,2]. Every year approximately 250000 new cases were added to their figure. It is roughly estimated that 25-30% of patients are resistant to the available medical treatment. Despite the development of several new anticonvulsants with diverse mechanism of action, the treatment of epilepsy remains still inadequate and the patients suffer from a lot of specific problems like neurotoxicity, sedation and other CNS related diseases. Moreover many antiepileptic drugs have serious side effects [3] and lifelong medication may be required. Therefore, it is essential to search for newer chemical entities for the treatment of epilepsy.

In recent years, the chemistry of quinazoline derivatives and heterocyclic annelated quinazolines are reported to be physiologically and pharmacologically active [4]. Various quinazoline derivatives exhibit anticonvulsant [5-8] activity. Methaqualone 2 (2-methyl-3-o-tolyl-4(3H)-quinazolinone) is a well known sedative-hypnotic containing quinazolin-4(3H)-one nucleus. In fact, these evidences suggest that the quinazolin-4(3H)-one nucleus possess a pharmacophoric

character for CNS activity. Thus, the quinazoline scaffold was selected for the research. Semicarbazones and related compounds have a documented and consistent role in the design of novel anticonvulsant agents, through the work of Dimmock's and Pandeya's research group. A number of semicarbazones, thiosemicarbazones, bis-carbohydrazones, aryl, arylidene, aryloxyaryl semicarbazones, acetylhydrazones and oxamoylhydrazones have been synthesized and evaluated for anticonvulsant activity [9-17].

A literature survey revealed that the presence of a substituted aromatic ring at position 3 and a methyl group at position 2 on quinazolin-4(3H)-one nucleus is a necessary requirement for CNS depression and anticonvulsant activities. Modification of the methyl group by some other chemical moiety yielded structural analogs with potent CNS activity [8]. In addition to this extensive SAR studies have led to postulating a specific binding site of semicarbazones. The proposed pharmacophoric requirements in the semicarbazone molecules are: a hydrophobic domain (HPD), hydrogen bonding donors (HBD), a two electron donor system (D) and a diastral aryl ring which affects pharmacokinetics (PKS)[18].

The design and synthesis of the titled compounds were carried out by the molecular hybridization of Methaqualone, having quinazolin-4(3H)-one nucleus and N-(substituted) semicarbazides. The objective of the above molecular hybridization was to introduce an additional hydrogen bonding domain exemplified by the presence of the –NHCO– grouping and electron donor systems in quinazolin-4(3H)-one nucleus so as to enhance the anticonvulsant potential.

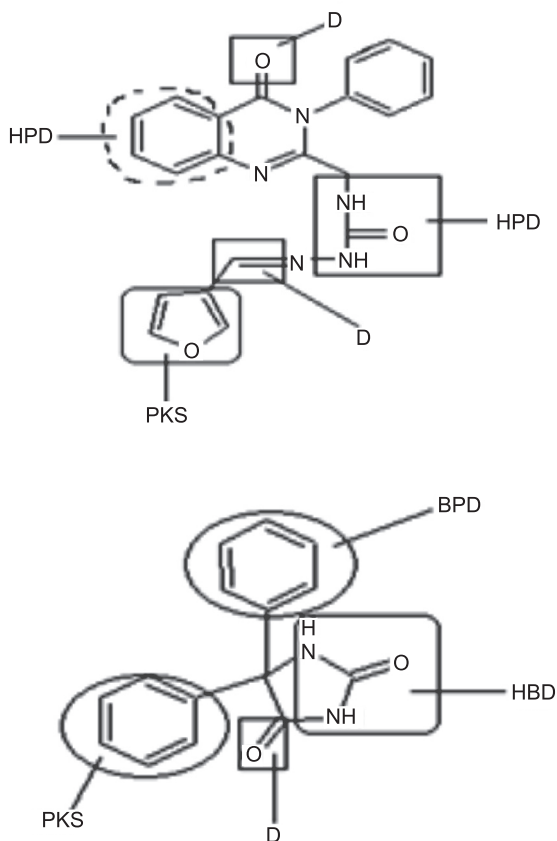


Fig. 1. Proposed pharmacophoric requirement in titled compounds.

## 2. MATERIALS AND METHODS

Melting points were determined in open capillary tubes and are uncorrected. The homogeneity of the compounds was monitored by thin layer chromatography (TLC) using silica gel G as stationary phase and visualized by iodine vapors. Solvent system was chloroform: methanol (9:1). Microanalysis of the compounds was done on Perkin-Elmer model 240 analyzer and the values found within  $\pm 0.4\%$  of the theoretical values. Their chemical structures were characterized using IR,  $^1\text{H}$  NMR and elemental analysis techniques.  $^1\text{H}$ -NMR spectra were recorded on DPX-300 NMR spectrometer and BRUKER-400 Ultra shield<sup>TM</sup> spectrometer. Chemical shifts ( $\delta$ ) are expressed in ppm relative to tetramethylsilane (TMS). The FT-IR spectra were recorded in KBr pellets on BIO-RAD FTS, FT-IR spectrophotometer. All the chemicals and solvents used were procured from Merck (India), S.D. Fine Chemicals (India) & Rankem (India). The physical constants of the synthesized compounds are presented

in Table 1. Their anticonvulsant activity was evaluated by using experimental epilepsy models, strychnine (ST) induced model, thiosemicarbazide (TCS) induced model and isoniazid (INH) induced model in mice. The rotorod assay was performed to evaluate the neurotoxicity of the compounds.

### 2.1 Procedure for synthesis of N-methyl isatin [N-methyl -3-aryl -3H-quinazoline-4-one] derivatives

0.5 gm of 2-chloromethyl-3-(H)-aryl-quinazolin-4-one 1 was refluxed with different aromatic amine derivatives solution (Equimolar amount) for 2 hrs. The product was filtered and dried. Further 0.5 gm of previously synthesized aromatic amine derivatives of 2-(chloromethyl) quinazolin-4(3H)-one 3 compound were refluxed with equimolar concentration of isatin in the presence of formaldehyde as a solvent for 3-4 hrs. The product was cooled at room temperature and then poured into ice cold water. The separated product was workup, dried and collected.

The present work reports the synthesis and anticonvulsant activity of a series of quinazolin-4(3H)-one isatin derivatives 4(a-d) according to the synthetic schemes as shown in scheme 1.

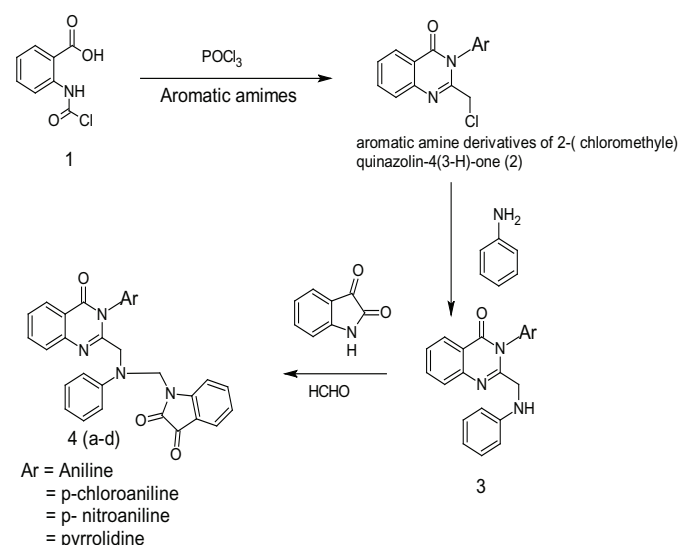


Fig. 2. Scheme for synthesis of N-methyl isatin [N-methyl -3-aryl -3H-quinazoline-4-one] derivatives

### 2.2 Spectral data of 2-(chloromethyl) quinazolin-4(3H)-one isatin derivatives 4(a-d)

#### 4a

$^1\text{H}$  (300 MHz,  $\text{CDCl}_3$ ) :  $\delta$  7.0-7.9 (s, 9H, Ar-H),  $\delta$  7.0-7.9 (s, 4H, Ar-H),  $\delta$  4.26 (s, 2H, C-CH<sub>2</sub>-N), IR (KBr)  $\nu$  = 1600, 1500 (C=C Str.), 1500 (CH; Str), 3077, 3000; (NH; Str), 1750, 1650 (C=O; Str), 1690, 1590 (C=N), calculated (%) C (74.04%) H (5.12%) N (11.28%), Found (%) C (74.24%) H (5.02%) N (11.17%).

#### 4b

$^1\text{H}$  (300 MHz,  $\text{CDCl}_3$ ) :  $\delta$  6.91-7.11 (s,9H,Ar-H ),  $\delta$  7.2-7.9 (s,4H,Ar-H ),  $\delta$  4.28 (s,2H, C- $\text{CH}_2$ -N), IR (KBr,  $\text{cm}^{-1}$ )  $\nu$  = 1603-1510(C=C Str.) ,1500 (CH; Str), 3070-3020; (NH: Str ), 1790 (C=O; Str), 1696, 1585 (C=N), 1100-1035 Ar-Cl, calculated (%) C (69.56 %) H (4.62%) N (10.36%), Found (%) C (69.46%) H (4.51%) N (10.36%).

#### 4c

$^1\text{H}$  (300 MHz,  $\text{CDCl}_3$ ) :  $\delta$  6.98-7.10 (s,9H,Ar-H),  $\delta$  7.0-7.2 (s,4H, Ar-H),  $\delta$  4.23 (s, 2H, C- $\text{CH}_2$ -N), IR (KBr,  $\text{cm}^{-1}$ )  $\nu$  = 1600-1520(C=C Str.) ,1505 (CH; Str), 3072-3010; (NH: Str), 1780 (C=O; Str), 1692, 1595 (C=N), 1372(strong),1550, Ar- $\text{NO}_2$ , calculated (%) C (68.05%) H (4.62%) N (12.70%), Found (%) C (68.12%) H (4.43%) N (12.81%).

#### 4d

$^1\text{H}$  (300 MHz,  $\text{CDCl}_3$ ) :  $\delta$  7.4-7.9 (s,9H,Ar-H ),  $\delta$  7.0-7.3 (s, 4H,Ar-H ),  $\delta$  4.25 (s, 2H, C- $\text{CH}_2$ -N),  $\delta$  3.69 (s, H, pyrrolidine)  $\delta$  1.85-3.01 (s,6H,pyrrolidine),  $\delta$  2.0 (s,NH,pyrrolidine). IR (KBr,  $\text{cm}^{-1}$ )  $\nu$  = 1601-1500(C=C Str.) ,1509 (CH; Str), 3076-3020; (NH: Str ), 1790 (C=O; Str), 1695, 1590 (C=N), calculated (%) C (70.02%) H (5.61%) N (14.38%), Found (%) C (69.84%) H (5.65%) N (14.54%).

### 2.3 Evaluation of Anticonvulsant Activity

#### 2.3.1 Animals

Ten groups of the healthy Swiss albino mice (each having 6 animals) of both sexes weighing 25-30 g were taken for the study. The animals were kept in large spacious hygienic cages during the course of experimental period. The animals had free access to standard commercial diet and water ad libitum and were kept in rooms maintained at  $22 \pm 1^\circ\text{C}$  with 12 h light dark cycle.

The synthesized compounds were dissolved in polyethylene glycol (PEG-400) and 30 mg/kg i.p. dose were administered and the activity was established using different chemical induced convulsion tests. The anticonvulsant screening of the final compounds was done according to the protocols of the anticonvulsant drug development (ADD) programme [4].

#### 2.3.2 Strychnine induced model

Mice of either sex with a weight of 25-30 g were treated with the test compounds or the standard (e.g. diazepam 30 mg/kg i.p.) by oral or intraperitoneal administration. Controls received the vehicle only.30 minute after i.p. treatment the animals

were injected with a subcutaneous dose of mg/kg strychnine. The occurrence of clonic seizures, tonic seizures and death or recovery was recorded after 0.5 hr, 1 hr, 2 hr, & 4 hr respectively.

#### 2.3.3 Thiosemicarbazide induced model

Mice of either sex with a weight of 25-30 g were treated with the test compounds or the standard (e.g. diazepam 30 mg/kg i.p.) by oral or intraperitoneal administration. Controls received the vehicle only. 30 minute after i.p. treatment the animals were injected with a subcutaneous dose of 20 mg/kg thiosemicarbazide. The occurrence of clonic seizures, tonic seizures and death or recovery was recorded after 0.5 hr, 1 hr, 2 hr, & 4 hr respectively. Not Protected means death of the rats occurs at the mentioned time.

#### 2.3.4 Isoniazid (INH) induced model

Mice of either sex with a weight of 25-30 g were treated with the test compounds or the standard (e.g. diazepam 30 mg/kg i.p.) by oral or intraperitoneal administration. Controls received the vehicle only.30 minute after i.p. treatment the animals were injected with a subcutaneous dose of 20 mg/kg isoniazid. The occurrence of clonic seizures, tonic seizures and death or recovery was recorded after 0.5 hr, 1hr, 2hr, & 4hr respectively [19].

#### 2.3.5 Neurotoxicity screening

Activity of the drugs interfering with motor coordination was checked by the rotorod test. The mice will train to stay on an accelerating rotorod that rotate at 6 revolutions per minute. The rod diameter will be of 3.2 cm neurotoxicity indicated by the inability of the animal to maintain equilibrium on the rod for at least 1 min in each of three trials. The dose, at which the animals were unable to grasp the rotorod, will determine. All the results were reported in Table 2.

**Table 1.** Physicochemical data of N-methyl isatin [N-methyl -3-aryl -3H-quinazoline-4-one] derivatives

Com-pounds	Mol. Formula	Log P	M.P. ( $^\circ\text{C}$ )	Percent-age yield	Mol. Wt.	Rf value
4a	$\text{C}_{31}\text{H}_{25}\text{N}_4\text{O}_3$	4.38	218-220	88%	501.56	0.48
4b	$\text{C}_{31}\text{H}_{24}\text{ClN}_4\text{O}_3$	3.39	272-274	70%	536.00	0.56
4c	$\text{C}_{24}\text{H}_{20}\text{N}_6\text{O}_4$	2.14	275-277	72%	456.45	0.52
4d	$\text{C}_{28}\text{H}_{27}\text{N}_5\text{O}_3$	5.11	282-284	66%	481.55	0.62

**Table 2.** Anticonvulsant activity evaluation of N-methyl isatin [N-methyl -3-aryl -3H-quinazolin-4-one] derivatives

30 mg/kg (Dose)										Neurotoxicity by Rota-Rod	
Strychnine induced convulsion			Thiosemicarbazide Induced convulsion			INH induced convulsion					
Time to peak effect	0.5 hr	1 hr	2 hr	0.5 hr	1 hr	2 hr	0.5 hr	1 hr	2 hr		
Controls	—	—	—	—	—	—	—	—	—	20 mg	300 mg
Comp code	—	—	—	30 mg	30 mg	NP	30 mg	30 mg	NP		
4a	30 mg	30 mg	NP	30 mg	NP	NP	30 mg	NP	NP	NN	NN
4b	30 mg	NP	NP	30 mg	NP	NP	30 mg	NP	NP	NN	NN
4c	30 mg	NP	NP	30 mg	30 mg	NP	30 mg	NP	NP	NN	NN
4d	30 mg	30 mg	NP	30 mg	30 mg	30 mg	30 mg	30 mg	30 mg	NN	NN
Diazepam (mg/kg)	30 mg	30 mg	30 mg	30 mg	30 mg	NP	30 mg	NP	NP	—	—

Test compounds were suspended in polyethylene glycol (PEG) and dose of 30 mg/kg was administered through intraperitoneal (i.p.) NP denoted not protected at given dose (30 mg/kg) NN (Non Neurotoxic up to the dose 300 mg/kg).

### 3. RESULTS AND DISCUSSION

All the synthesized isatin derivatives of quinazolinone 4 (a-d) were evaluated for anticonvulsant activity compound 4d was showing good activity against chemo shock models of epilepsy while 4 a-c were found to be less active. The quinazolinone scaffold can be exploited for further molecular designing of novel drugs to treat epilepsy.

### 4. CONCLUSION

New isatin derivatives of quinazolinone were synthesized and evaluated for anticonvulsant activity. The synthesized compounds 4(a-d) displayed moderate protection for anticonvulsant activity.

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### REFERENCES

- [1] Reum, V.J.; Tripathi, B.R.; Srivastava, A.K. Synthesis and antihyperglycemic activity of suitably functionalized 3H-quinazolin-4-ones. *Bioorg Med Chem.* 2003, 11, 2439.
- [2] Dander, A.; Singh, R.; Sarawagi, P. Synthesis of quinazolinones and quinazolines. *J. Fluorine Chemistry.* 2005, 126, 307.
- [3] Negwer, M.; Schornow, H. *Organic Chemistry Drugs and their synonyms*, Wiley-Vch, Weinheim 2001, 5<sup>th</sup> Ed, p. 214 .
- [4] Aziza, M.A.; Ibrahim, M.K.; El-Helmy, A.G.; Al-Azhar. Design, synthesis and antiviral activity of novel quinazolinones. *J. Pharm. Sci.* 1994, 14, 193-201.
- [5] El-Azab, A.S.; Eltahir, K.E. Design and synthesis of novel 7-aminoquinazolinone derivatives. *Bioorg Med Chem Lett.* 2012, 22, 1879-85.
- [6] Georgey, H.; Abdel-Gawad, N.; Abbas, S. Synthesis and anticonvulsant activity of some quinazolin-4. *Molecules.* 2008, 13, 2557-69.
- [7] Usifoh, C.O.; Scriba, G.K. Synthesis and anticonvulsant activity of acetylenic quinazolinone derivatives. *Arch Pharm (Weinheim).* 2000, 333, 261-6.
- [8] Kumar, P.; Shrivastava, B.; Pandeya, S.N.; Stables, J.P. Design, synthesis and potential 6 Hz psychomotor seizure. *Eur J Med Chem.* 2011, 46, 1006-18.
- [9] Dimmock, J.R.; Puthucode, R.N.; Smith, J.M.; Hetherington, M.; Quail, J.W.; Pugazhenth, U.; Lecher, T.; Stables, J.P. Solid phase synthesis of functionalized biaryl ethers. *J Med Chem.* 1996, 39, 3984-3997.
- [10] Puthucode, R.; Pugazhenth, U.; Quail, J.W.; Stables, J.P.; Dimmock, J.R.,” Design, synthesis and anticonvulsant screening of newer benzothiazole-semicarbazones”, *Eur. J. Med. Chem.* 1998, 33, 595-607.
- [11] Pandeya, S.N.; Ponnilarasan, I.; Pandey, A.; Lakhan, R.; Stables, J.P. Menthone semicarbazides and thiosemicarbazides as anticonvulsant agents. *Pharmazie* 1999, 54, 923-925.

- [12] Dimmock, J.R.; Vashishtha, S.C.; Stables, J.P. Anticonvulsant Activities of Various Series of Heterocyclic Compounds Containing Triazole, Thiadiazine, Benzo-triazole, Benzothiazole, Oxadiazole Ring Systems. *Eur J Med Chem.* 2000, 35, 241-248.
- [13] Pandeya, S.N.; Yogeewari, P.; Stables, J.P., “Synthesis and anticonvulsant activity of 4-bromophenyl substituted aryl semicarbazones”, *Eur. J. Med. Chem.* 2000, 35, 879-886.
- [14] Pandeya, S.N.; Mishra, V.; Ponnilaravasan, I.; Stables, J.P. Anticonvulsant activity of Schiff bases of isatin derivatives. *Pol J Pharmacol.* 2000, 52, 283-290.
- [15] Dimmock, J.R.; Vashishtha, S.C.; Stables, J.P. Synthesis and anticonvulsant activity of some 2-pyrazolines. *Pharmazie.* 2000, 55, 490- 494.
- [16] Pandeya, S.N.; Manjula, H.; Stables, J.P., “Design of semicarbazones and their bioisosteric analogues as potential anticonvulsants”, *Pharmazie* 2001, 56, 121-124.
- [17] Yogeewari, P.; Sriram, D.; Sunil Jit, L.R.; Kumar, S.S.; Stables J.P. Anticonvulsant and neurotoxicity evaluation of some 6-chlorobenzothiazolyl-2-thiosemicarbazones. *Eur J Med Chem.* 2002, 37, 231-236.
- [18] Pandeya, S.N.; Agarwal, A.K.; Singh, A.; Stables, J.P. Synthesis of 4-aryl substituted semicarbazones of some terpenes. *Acta Pharm.* 2003, 53, 15-24.
- [19] Vogel, H.G., *Drug Discovery and evaluation: pharmacological assay*, Berlin: Springer Verlag, New York, 2002, 696-716.