Pergamon

Bioorganic & Medicinal Chemistry Letters, Vol. 4, No. 17, pp. 2087-2090, 1994

0960-894X/94 $7.00+0.00

0960-894X(94)00287-8

LATENT LEISHMANICIDAL ACTIVITY OF QUINAZOLINONES AND 1,2,4-TRIAZOLOQUINAZOLINONES[1]

Vishnu Ji Ram*, Atul Goel, Monika Verma
Medicinal Chemistry Division
Central Drug Research Institute, Lucknow 226 001, India.

Indu Bala Kaul, Aruna Kapil
Pharmacology Division
Regional Research Laboratory, Jammu Tawi 180 001, India.

Abstract: Derivatives of quinazolinone **(1,2,6)** and 1,2,4-triazoloquinazolinone **(3,4,5,7,8)** have been synthesized and evaluated for in vitro leishmanicidal activity against *L. donovani* promastigotes.

Introduction: Quinazolinones and 1,2,4-triazoloquinazolinone derivatives are known for diverse pharmacological activities as neuroleptic[2], hypnotic[3], anticonvulsant[4], analgesic[5], antifertility[6] and muscle relaxant[7] but so far none had disclosed and reported the leishmanicidal activity in these class of compounds. Our past observations demonstrated that compounds possessing N-C(=N)-N or S-C(=N)-N structural unit either in flexible or in rigid forms express potent leishmanicidal activity[8]. Repetition of such unit often enhances leishmanicidal activity. The antiparasitic activity of azoles such as imidazoles, thiazoles and thiadiazoles is well documented[9,10] and their fusion with other heterocycles often ameliorate or diminish the bioresponses depending upon the nature of substituents and position of their fusion.

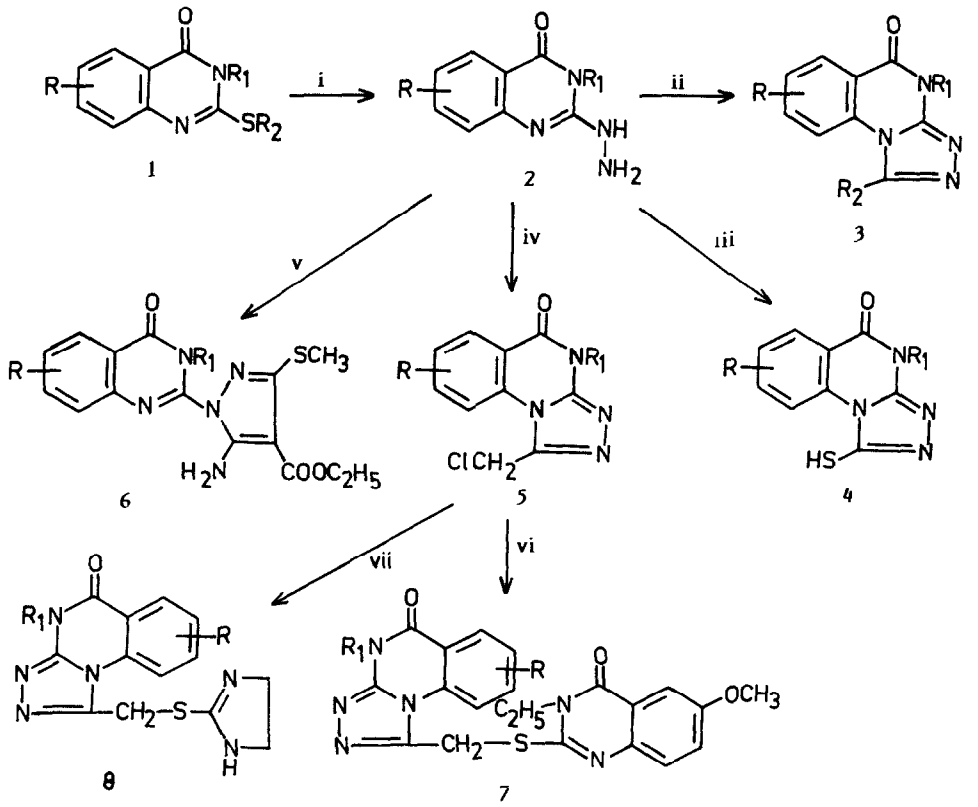
Among all the synthesized compounds of prototypes: quinazolin-4-one, 1,2,4-triazolo[4,3-a]quinazolin-5-one and 1-(4-quinazolinon-2-yl)-1,2,4-triazolo[4,3-a]quinazolin-5-one **(7)**, only compounds of prototype second displayed high order of leishmanicidal activity while others were either inactive or poorly active. The most potent compound **5b** exhibited 95% of inhibition, closer to pentamidine. The other two compounds **3c** (81%) and **4a** (80%) also demonstrated the activity nearly of the same order.

Synthesis: The precursor **(1)** used for the synthesis of title compounds was prepared from the reaction of suitable anthranilic acid and alkyl isothiocyanate[11]. The 2-hydrazinoquinazolin-4-ones **(2)** obtained by heating a mixture of **1** and hydrazine hydrate were subjected to react with ethyl orthoformate and ethyl orthoacetate separately to yield **3a-e**. Reaction of **2** with carbon disulphide yielded 1-mercapto-1,2,4-triazolo[4,3-a]quinazolin-5-one **(4a,b)** while interaction with chloroacetyl chloride, it gave 1-chloromethyl-1,2,4-triazolo[4,3-a]quinazolin-5-ones[11] **(5)** which on reaction with **1a** and imidazolidin-2-thione separately led to **7a,b** and **8a,b** respectively. The condensation-cyclization of **2** with ketene dithioacetals gave 2-(pyrazol-1-yl)quinazolin-4-ones **(6)** (Scheme 1).

Biological Activity: All the synthesized compounds listed in Table 1 were screened for their leishmanicidal activity against promastigotes of *L. donovani* in vitro as described below:

Promastigotes in the stationary phase of growth ($1x10^5$ parasite/ml) were inoculated into culture tubes containing solid part as Brain Heart Infusion-Agar (BHI-Agar) and liquid part as Hank's Balance Salt Solution (HBSS). The test chemicals and pentamidine (100 µg/ml, DMSO/PBS) were added to the above tubes respectively. The inhibitory effect of these compounds was compared with standard drug pentamidine and with untreated control cultures. The whole operation was carried out aseptically in an U.V. Chamber and tubes were inoculated for 5 days at 22°C. The antileishmanial activity of each compound was determined in triplicate by counting the number of live parasites per field microscopically and inhibition percentage was calculated and compared with standard drug pentamidine by using 'Z' statistic.

Scheme-1



Reagents/Conditions: i) N_2H_4 /C_2H_5OH/80°C, ii) $HC(OC_2H_5)_3$ or $(CH_3CO)_2O$ /PTSA/145°C, iii) CS_2 /pyridine/80°C, iv) $ClCOCH_2Cl$/DMF/100°C, v) $(CH_3S)_2$ C=C(CN)$COOC_2H_5$ /C_2H_5OH/ 80°C, vi) **1a**/K_2CO_3 /DMF/80°C, vii) Imidazolidin-2-thione/C_2H_5OH/100°C.

Table 1: In vitro antileishmanial activity of quinazolinones **(1,2,6)** and 1,2,4-triazoloquinazolinones **(3,4,5,7,8)**at 100 µg/ml concentration. Values are the average % inhibition of triplicates afforded by the test compounds.

	R	R_1	R_2	Mortality Inhibition (%)
	Pentamidine (standard drug)			100**
1a	6-OCH_3	C_2H_5	H	12
1b	7-Cl	CH_3	CH_3	0
2a	6-OCH_3	C_2H_5	--	0
3a	7-OCH_3	C_2H_5	H	39
3b	7-OCH_3	C_2H_5	CH_3	42
3c	8-Cl	CH_3	H	81*
3d	8-Cl	C_2H_5	H	19
3e	8-Cl	C_2H_5	CH_3	49
4a	7-OCH_3	C_2H_5	SH	80*
4b	8-Cl	CH_3	SH	55
4c	8-Cl	C_2H_5	SH	30
5a	7-OCH_3	C_2H_5	CH_2Cl	67
5b	8-Cl	CH_3	CH_2Cl	95**
5c	8-Cl	C_2H_5	CH_2Cl	70
6a	8-Cl	CH_3	--	0
6b	8-Cl	C_2H_5	--	0
7a	8-Cl	CH_3	--	10
7b	8-Cl	C_2H_5	--	0
8a	8-Cl	CH_3	--	35
8b	8-Cl	C_2H_5	--	24

(**$P < 0.01$; *$P < 0.05$) as compared to standard drug pentamidine.

A critical examination of structure-activity relationship in the given series revealed that the compounds only with 1,2,4-triazolo[4,3-a]quinazolinone skeleton display potent leishmanicidal activity. The order of activity varies with nature and position of the substituent. Methyl substituent at position 4 alone or together with any of the substituents CH_3, SH and CH_2Cl at position 1 ameliorate the activity as evidenced from screening data of **3c,d**, **4b,c** and **5b,c**. The degree of leishmanicidal activity due to mere change in substituent at position 1 in **5b**, **3c**, **4b** decreases in the order of $CH_2Cl > H > SH$. Presence of bulky substituent like (4-quinazolinon-2-yl)thiomethyl at position 1 in **7** led to complete loss of activity possibly due to poor solubility and steric effect. Introduction of (imidazolin-2-yl)thiomethyl moiety in place of (quinazolinon-2-yl)thiomethyl in **7** led to the product **8** with increased solubility and reduced steric effects. The enhanced leishmanicidal activity of **8a,b** supported the presumptions, responsible for activity. The nature and position of the substituent in benzene ring

is not crucial for displaying leishmanicidal activity. Compounds with quinazolinone skeleton **(1a,b,2a,6a,b)** with different substituents at different positions were almost inactive. Thus 1,2,4-triazolo[4,3-a]quinazolin-5-one skeleton with chloromethyl and methyl substituents at positions 1 and 4 respectively are optimal requirements for potent leishmanicidal activity.

Acknowledgement: A. Goel is thankful to CSIR for the award of Junior Research Fellowship and Dr. R.S. Kapil, Director, R.R.L., Jammu for leishmanicidal screening of the compounds.

References:

1. CDRI Communication No.5311.
2. Mukerji, D.D.; Nautiyal, S.R.; Prasad, C.R.; Dhawan, B.N.; *Ind. J. Med. Res.*, **1980**, *71*, 480.
3. Gujaral, M.L.; Saxena, P.N.; Tiwari, R.S.; *Ind. J. Med. Res.*, **1955**, *43*, 637.
4. Glasser, A.C.; Diamond, L.; Combs, G.; *J. Pharm. Sci.*, **1971**, *60*, 127.
5. Ram, V.J.; Srimal, R.C.; Kushwaha, D.S.; Mishra, L.; *J. Prakt. Chem.*, **1990**, *332*, 629.
6. Reif, E.; Ericson, R.J.; *Ger. Offen.* 2,118,683, 11 Nov. **1971**; *Chem. Abstr.*, **1972**, *76*, 725462.
7. Tani, T.; Yamada, Y.; Ochiai, T.; Ishida, R.; Inoue, I.; Oine, T.; *Chem. Pharm. Bull.*, **1979**, *27*, 2675.
8. Ram, V.J.; Singha, U.K.; Guru, P.Y.; *Eur. J. Med. Chem.*, **1990**, *25*, 533.
9. Gosh, B.K.; Ray, D.H.; Chatterjee, A.N.; *J. Trop. Med. Hyg.*, **1989**, *92*, 383.
10. Steck, E.A.; *Progress in Drug Research*, **1974**, *18*, 290.
11. Wagner, G.; Rothe, L.; *Z. Chem.*, **1968**, *8*, 377.

(*Received in Japan* 6 *May* 1994; *accepted* 22 *July* 1994)