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Synthesis and Antifungal Activity of N^4-(Piperazinoyl-methyl)-N,N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl)acetamide

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A series of N^4-(piperazinoyl-methyl)-N,N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl) acetamide have been synthesized by the condensation of the N,N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl) acetamide with chloroacetic acid and then with piperazine. All the synthesized compounds have been tested for their antifungal activity against *Tricophyton rubrum*, *Epidermophyton floccosum* and *Malassazia furfur*. Some of the compounds exhibited appreciable activity. The structure of the synthesized compounds **7a-c** has been established on the basis of elemental analysis and spectral data.

Key Words: N4-(Piperazinoyl-methyl)-N,N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl) acetamide, Piperazine, Chloroacetic acid, N,N-Diaryl-2-(3-oxo-1,4-benzothiazin-2-yl) acetamide.

INTRODUCTION

Antifungal agents like polyenes and azoles are available for the treatment of serious and life-threatening fungal infection, mainly caused by *Tricophyton rubrum*, *Epidermophyton floccosum* and *Malassazia furfur*[1]. However, their clinical uses are restricted due to toxicity (polyenes), fungistatic activity (azoles) and the emergence of resistant isolates (azoles). To overcome these problems, novel antifungal agents with a different mode of action are in demand.

1,4-Benzothiazines[2] and piperazines form an important class of heterocyclic system. The utility of piperazines as antifungal[3-5], antibacterial[6], anthelmintic[7], antiallergenic[8], antipsychotic[9], antiemetic[10] and psychoactive[11] is firmly established. By combining these both potential nucleuses in a single molecule, it provides very efficient antifungal agents (Fig. 1). In continuation to our work on N-(alkyl/aryl)-2-(3-oxo-1,4-benzothiazine-2-yl) acetamide[2], which are reported as antifungal agents, has secondary nitrogen in basic nucleus which is another important site for attachment of the side chain and helpful in potentiation of its activity.

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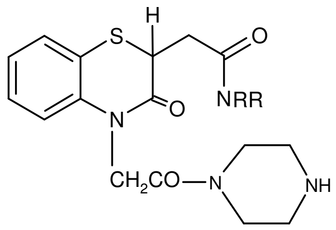


Fig. 1

EXPERIMENTAL

The melting points reported in this work are uncorrected. Thin layer chromatography was performed using silica gel coated on a glass plate and spots were visualized by exposure to iodine vapour. IR spectra in nujol were recorded on a Shimadzu IR spectrophotometer. PMR spectra in deuterium substituted DMSO on a Bruker FT-NMR spectrometer using TMS as an internal standard (chemical shifts in δ ppm).

Maleanillic acids (1a-c): To the solution of maleic anhydride (19.6 g, 0.2 mol) in diethyl ether (80 mL), the solution of diphenylamine (33.8 g, 0.2 mol) in diethyl ether (80 mL) was added. The reaction mixture was stirred at room temperature for 5 min. The precipitate was filtered, washed with ether (2 × 60 mL) and purified by recrystallization from acetone-petroleum ether (1:1) to get the compound **1a**. Yield 93 %, m.p. 147-190 ºC. The other compounds **1b-c** were also prepared in the similar way by the reaction of **2** with various secondary amines **1**.

Maleanillic esters (2a-c): To the ice-cold methanol (300 mL), phosphorus pentaoxide (42.5 g, 0.3 mol) was added in portions with stirring and the temperature was kept below 10 ºC. To the resulting solution, **1a** (40 g, 0.15 mol) was added in one portion and was mixed gently. It was then refluxed for 6 h on water bath. Excess methanol was distilled out under reduced pressure. The resulting residue was poured into crushed ice, filtered and purified by recrystallization from ethanol to get the compound **2a**. Yield 91 %, m.p. 202-204 ºC. The other compounds **2b-c** were also prepared in a similar way from **1b-c**.

Isomerized maleanillic esters (3a-c): To the solution of **2a** (28.2 g, 0.1 mol) in absolute ethanol (200 mL), a solution of redistilled aniline (4.65 g, 0.05 mol) was added. The resulting mixture was refluxed on a steam bath for 3 h. The precipitate was filtered, washed with dil. HCl (50 mL) and purified by recrystallization from ethanol to get the compound **3a**. Yield 83 %, m.p. 179-81 ºC. The other compounds **3b-c** were also prepared in the similar way from **2b-c**.

N,N-Diaryl-2-(3-oxo-1,4-benzothiazin-2-yl) acetamide (4a-c): To the solution of **3a** (21.0 g, 0.075 mol) in DMF (150 mL), a solution of *o*-aminothiophenol (*o*-ATP) (9.37 g, 0.075 mol) in DMF (30 mL) was added. The resulting mixture was refluxed for 5 h. The solution was cooled and poured into crushed ice. The solid that separated out was filtered, washed with water and purified by recrystallization from ethanol to get the compound **4a**. Yield 96 %, m.p. 178-179 ºC. The other compounds (**4b-c**) were also prepared in the similar way from **3b-c**.

N^4-(Carboxyl-methyl)-N, N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl) acetamide (5a-c): To the solution of chloroacetic acid (5.67 g, 0.06 mol) in DMF (60 mL), **4a** (22.44 g, 0.06 mol) and anhydrous sodium carbonate (13.52 g, 0.128 mol) was added. The reaction mixture was refluxed for 3 h. Cool the solution and rendered slightly acidic with conc. HCl and allowed to stand overnight. The precipitate was filtered, washed with water (2 × 60mL) and purified by recrystallization from hot water using little decolourizing carbon to get the compound **5a**. Yield 79 %, m.p. 201-203 ºC. The other compounds **5b-c** were also prepared in the similar way from **4b-c**.

N^4-(Chloro acetoxyl)-N,N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl)acetamide (6a-c): In a mortar, grinded together **5a** (17.3 g, 0.04 mol) and phosphorus trichloride (22 mL) until all solid became semisolid. Dried the content to get the compound **6a**. Yield 80 %, m.p. 213-14 ºC. The other compounds (**6b-c**) were also prepared in the similar way from **5b-c**.

N^4-(Piperazinoyl-methyl)-N,N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl)acetamide (7a-c): To the solution of piperazine (1.72 g, 0.02 mol) in DMF (30 mL), **6a** (9 g, 0.02 mol) and sodium hydroxide (0.8 g, 0.2 mol) was added. The reaction mixture was refluxed for 1 h. The precipitate was filtered and dried to get the compound **7a**. Yield 80 %, m.p. 213-214 ºC. The other compounds **7b-c** were also prepared in the similar way from **6b-c**. Anal. calcd for $C_{28}H_{28}N_4O_3S$: C, 67.18; H, 5.64; N, 11.19, found: C, 66.94; H, 5.72; N, 10.96 %; IR (Nujol, ν_{max}, cm^{-1}): 3345 (NH), (30 N), 1635 (NCO), 2560 (S-H), 1760 (C=O), 3220 (amide NH_2), 1680 (amide CO), 2950 (CH_2). The ^{1}H NMR (for **7a**): 2.9 (m, 2H, -CH_2CO), 4.6 (m, 1H, S-CH), 7.4 (m, 4H, ArH), 7.8 (m, 10H, ArH), 9.8 (bs, 1H, NH), 10.8 (bs, 1H, NH), 3.0 (s, 2H, N-CH_2CO), 3.9-4.1 (m, 8H, piperazine H); ^{1}H NMR (for **7b**): 0.9 (m, 2H, -CH_2CO), 4.6 (m, 1H, S-CH), 7.4 (m, 8H, ArH), 9.8 (bs, 1H, NH), 10.8 (bs, 1H, NH), 3.0 (s, 2H, N-CH_2CO), 3.9-4.1 (m, 8H, piperazine H); ^{1}H NMR (for **7c**): 0.9 (m, 2H, -CH_2CO), 4.6 (m, 1H, S-CH), 7.4 (m, 8H, ArH), 9.8 (bs, 1H, NH), 10.8 (bs, 1H, NH), 3.0 (s, 2H, N-CH_2CO), 3.9-4.1 (m, 8H, piperazine H).

Biological activity

Antifungal activity: The synthesized compounds **7a-c** in concentration range 0.250-0.031 µmol/mL were tested for antifungal activity against *Tricophyton rubrum*, *Epidermophyton floccosum* and *Malassazia furfur* by turbidimetric method. For comparison, ketoconazole was used as a standard. Results are presented in Table-1.

RESULTS AND DISCUSSION

N^4-(Piperazinoyl-methyl)-N,N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl) acetamide **7a-c** were prepared by the reaction of piperazine with N^4-(chloro acetoxyl)-N,N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl) acetamide **6a-c** in NaOH and DMF solvent. The reaction mixture was allowed to reflux for 1 h and the product was isolated

TABLE-1
ANTIFUNGAL ACTIVITY DATA OF COMPOUNDS **7a-c**

Concentration of compounds (1 μmol/mL) required for inhibition				
Epidermophyton floccosum				
Compounds	0.25	0.125	0.062	0.031
7a	(–)	(–)	(+)	(+)
7b	(–)	(–)	(+)	(+)
7c	(–)	(–)	(–)	(+)
Tricophyton rubrum				
Compounds	0.25	0.125	0.062	0.031
7a	(–)	(–)	(+)	(+)
7b	(–)	(+)	(+)	(+)
7c	(–)	(–)	(–)	(+)
Malassazia furfur				
Compounds	0.25	0.125	0.062	0.031
7a	(–)	(+)	(+)	(+)
7b	(–)	(–)	(+)	(+)
7c	(–)	(–)	(–)	(+)

(–) Indicates inhibition of growth, (+) Indicates presence of growth, K: ketoconazole.

TABLE-2
CHARACTERIZATION DATA OF COMPOUNDS **7b-c**

Comp.	Yield (%)	m.p. (°C)	R_f	m.f.	IR (cm^{-1})	Elemental analysis (%): Found (calcd.)		
						C	H	N
7b	79	234-236	0.44	$C_{30}H_{32}N_4O_3S$	3345 (NH), (3° N) 1635 (NCO), 2560 (S-H), 1760 cm^{-1} (C=O), 3220 (amide NH_2), 1680 (amide CO), 2950 (CH_2)	67.06 (68.16)	6.11 (6.10)	10.69 (10.60)
7c	81	207-209	0.46	$C_{28}H_{40}N_4O_3S$	3345 (NH), (3° N) 1635 (NCO), 2560 (S-H), 1760 cm^{-1} (C=O), 3220 (amide NH_2), 1680 (amide CO), 2950 (CH_2)	64.68 (65.59)	7.98 (7.86)	10.90 (10.93)

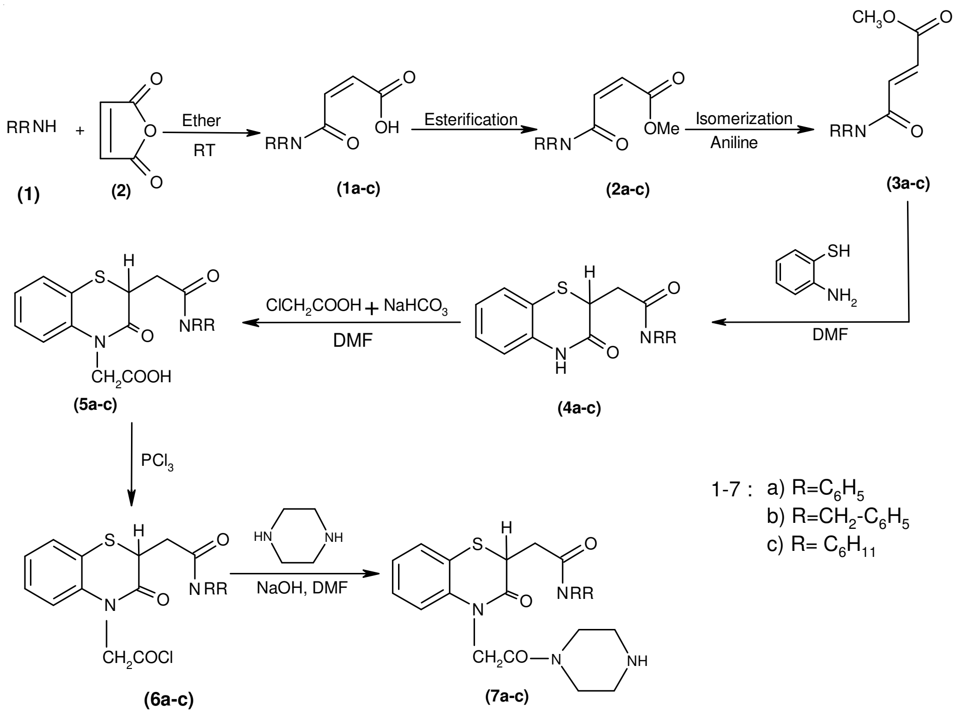
by dumping the mixture into crushed ice. The starting N^4-(chloro acetoxyl)-N,N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl) acetamide **6a-c** was prepared by chlorination of N^4-(carboxyl-methyl)-N, N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl) acetamide **5a-c**. Later compounds were prepared by reaction of chloroacetic acid with N,N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl) acetamide **4a-c** in the presence of sodium bicarbonate and DMF solvent. N,N-diaryl-2-(3-oxo-1,4-benzothiazin-2-yl) acetamide **4a-c** were prepared by the condensation of the corresponding isomerized maleanillic ester **3a-c** with *o*-ATP in DMF solvent. The reaction mixture was allowed to reflux for 5 h and the product was isolated by dumping the mixture into crushed ice. The

TABLE-3
CHARACTERIZATION DATA OF COMPOUNDS (**1-6**) **b-c**

Compound	Yield (%)					
	1	2	3	4	5	6
b	92	82	78	95	82	86
c	87	81	68	88	78	89
	m.p. (ºC)					
b	158-59	210-12	176-78	167-69	197-99	213-15
c	84-6	67-9	62-4	102-04	110-12	154-56
	R_f					
b	0.44	0.52	0.42	0.49	0.6	0.57
c	0.46	0.52	0.46	0.5	0.57	0.54

starting isomerized maleanillic ester **3a-c** was prepared by a three-step process from substituted amines **1** and maleic anhydride **2**. Maleanillic acid **1a-c** was synthesized by the reaction of substituted secondary amines **1** with maleic anhydride **2** in ether solvent at room temperature. Maleanillic ester **2a-c** was synthesized from **1a-c** by esterification with methanol in the presence of P_2O_5. Isomerized maleanillic ester **3a-c** was synthesized by isomerization of **2a-c** in the presence of aniline.

The synthesis is outlined in **Scheme-I**. The characterization data of compounds (**1-7**) **b-c** are given in Tables 2 and 3.



Scheme-I

REFERENCES

1. E. Jawetz, J. Melnick and E.A. Adelberg, *Rev. Medicinal Microbio*., **7**, 244 (1997).
2. G. Gupta and S.B. Wagh, *Indian J. Chem*., **45B**, 697 (2006).
3. R.S. Upadhayaya, N. Sinha, S. Jain, N. Kishore, R. Chandra and S.K. Arora, *Bioorg. Med. Chem*., **12**, 2225 (2004).
4. J. Heeres, R. Hendrickx and C.J. Van, *J. Med. Chem*., **26**, 611 (1983).
5. O. Kondoh, Y. Inagaki, H. Fukuda, E. Mizuguchi, Y. Ohya, M. Arisawa, N. Shimma, Y. Aoki, M. Sakaitani and T. Watanabe, *Bio. Pharm. Bull*., **28**, 2138 (2005).
6. P. Chaudhary, R. Kumar, A.K. Verma, D. Singh, V. Yadav, A.K. Chhillar, G.L. Sharma and R. Chandra, *Bioorg. Med. Chem*., **14**, 1819 (2006).
7. R.P. Tripathi, V.K. Tiwari, S. Bhattacharya, K. Tyagi, V.M. Srivastava and P.K. Murthy, *Acta Tropica*, **87**, 215 (2003).
8. S. Ozbey, E. Kendi, H. Goker and M.J. Tuncbilek, *J. Chem. Crystallogr*., **28**, 461 (1998).
9. H. Zhao, X. He, A. Thurkauf, D. Hoffman, A. Kieltyka, R. Brodbeck, R. Primus and J.W. Wasley, *Bioorg. Med. Chem. Lett.*, **12**, 3111 (2002).
10. J.B. Lucot, *Eur. J. Pharmacol.*, **253**, 53 (1994).
11. G.J. Gonzalez, L. Santana, E. Uriarate, J. Brea, M. Villazon, M.I. Loza , M. DeLuca, M.E. Rivas, G.Y. Montenegro and J.A. Fontenla, *Bioorg. Med. Chem. Lett*., **13**, 175 (2003).

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