Eur. J. Med. Chem. **24** (1989) 563–567

Original paper

Studies on antifungal agents. 27. Novel 5-{[(substituted phenyl)thio]-methyl}isoxazolidines

Thomas R. DeCORY[1], George B. MULLEN[1], Jeffrey T. MITCHELL[2], Stanley D. ALLEN[2] and Vassil St. GEORGIEV[1*]

[1]*Department of Organic Chemistry, Fisons Pharmaceuticals, Divisional Research and Development, Fisons Pharmaceuticals, Rochester, NY 14623, USA; and*
[2]*Laboratory Animal Research Center, Utah State University, Logan, UT 84322, USA*

(Received July 21, 1988, accepted February 9, 1989)

Summary — The synthesis and antifungal activity of a series of novel 5-{[(substituted phenyl)thio]methyl}isoxazolidine derivatives are discussed. The title compounds were prepared by 1,3-dipolar cycloaddition reaction of α-substituted ketonitrones with allyl phenyl sulfides to provide diastereomeric mixtures of the corresponding *cis / trans*-analogues. When tested in solid agar cultures, the title derivatives exerted moderate to potent *in vitro* antifungal activity against a number of dermatophytes and fungi causing systemic infections. The minimum inhibitory concentration (MIC) values ranged between <0.2–20.0 μg / ml.

Résumé — **Étude d'antifongiques. 27. Nouvelles 5-arylthiométhyl isoxazolidines.** *On discute la synthèse et l'activité antifongique d'une série de nouveaux dérivés 5-arylthiométhyl isoxazolidines. Ces composés ont été préparés par cycloaddition bipolaire 1,3 de cétonitrones α-substituées avec des sulfures d'allyle et de phényle pour fournir des mélanges de diastéréomères des analogues-*cis / trans *correspondants. Au cours d'épreuves en culture d'agar solide, ces dérivés ont exercé une activité* in vitro *antifongique modérée à puissante vis à vis d'un certain nombre de dermatophytes et de champignons infectieux. La valeur de concentration inhibitrice minimale (MIC) se situe entre <0,2–20,0 μg / ml.*

5-{[(substituted phenyl)thio]methyl}isoxazolidines / 1,3-dipolar cycloaddition / antifungal activity

Introduction

In recent years, the number of systemic fungal infections has reached allarming proportions. This has been especially true for large populations of immunocompromized patients as well as those suffering from various hematologic malignancies, acquired immune deficiency syndrome (AIDS) and patients undergoing organ transplantation or cancer chemotherapy [1]. In addition to widespread candidiasis, other fungal infections such as cryptococcosis, aspergillosis, zygomycosis, coccidioidomycosis, paracoccidioidomycosis and chromoblastomycosis, have shown a dramatic rise in frequency [1].

Recently, we have studied the application of 1,3-dipolar cycloaddition reactions to the synthesis of biologically active compounds [2, 3], especially in the preparation of novel isoxazolidine derivatives with potent antifungal activity against various dermatophytes and fungi causing systemic infections [4–7]. A further extension of our previous knowledge of 1,3-dipolar species was the synthesis of novel antifungal isoxazolidines by 1,3-dipolar cycloaddition of α-substituted ketonitrones with allyl phenyl sulfides. In the present communication, we discuss some of the results of these studies, namely the synthesis and antifungal activity of a series of 5-{[(substituted phenyl)thio]-methyl}isoxazolidine derivatives **5** and **6**.

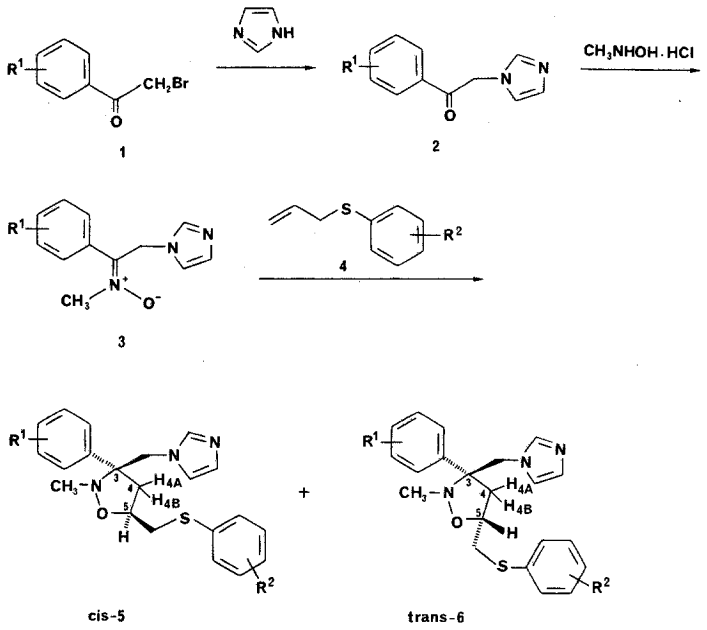
Chemistry

The synthesis of 5-{[(substituted phenyl)thio]methyl}-isoxazolidines **5** and **6** is illustrated in Scheme 1. Bromo derivatives **1** (obtained by bromination of the corresponding acetophenone precursors) were treated with imidazole

*Author to whom correspondence should be addressed.

[i]The 2-(1*H*-imidazol-1-yl)-1-(substituted phenyl)ethanones 2 were prepared by modified procedures of the method of Godefroi *et al.* [10].

[ii]The regio- and stereospecificity of the reaction is highly dependent on the structures of the nitrone and the dipolarophile and involves both electronic and steric factors. The regiospecificity of 1,3-dipolar cycloadditions of monosubstituted olefins are usually dominated by HOMO-LUMO (highest occupied molecular orbital–lowest unoccupied molecular orbital) interactions. Thus, electron-rich olefins will give exclusively the 5-substituted isoxazolidines, whereas electron-deficient olefins can give either the 5- or the 4-substituted product, or a mixture of both. The presence of a sterically demanding α-substitution pattern in the case of nitrones **3** [3-phenyl-3-(1*H*-imidazol-1-ylmethyl)], coupled with the bulky allyl phenyl sulfide **4**, will make the formation of 4-substituted isoxazolidine derivatives highly unlikely, since in the latter, the 4-phenyl ring would be positioned next to the C-3 carbon atom carrying the 3-phenyl-3-(1*H*-imidazol-1-ylmethyl) moiety.



Scheme 1.

to provide the 2-(1*H*-imidazol-1-yl)-1-phenylethanones **2**[i]. The latter, in turn, were converted into the α-substituted ketonitrones **3** by reaction with *N*-methylhydroxylamine hydrochloride under mild conditions. Next, a regiospecific 1,3-dipolar cycloaddition reaction [8][ii] of **3** and appropriate allyl phenyl sulfides **4** [9] furnished *cis* / *trans*-diastereomeric mixtures of the desired isoxazolidines **5** and **6**. In all cases the major products of the diastereomeric mixtures were the *cis*-analogues **5** which were conveniently separated from the *trans*-isomers **6** by flash chromatography on neutral silica gel (Table I).

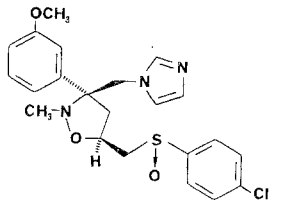
Results and Discussion

The *cis*-5-{[(substituted phenyl)thio]methyl}isoxazolidines **5** were screened for *in vitro* antifungal activity in solid agar tests performed in 24-well tissue culture plates. The minimum inhibitory concentration (MIC) values (in μg / ml) were interpreted as the lowest concentration at which no visible growth occurred. In all assays, ketoconazole was used as the positive standard drug. The obtained results are listed in Table I.

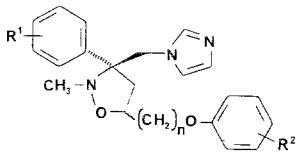
Table I. 5-{[(Substituted phenyl)thio]methyl}isoxazolidine derivatives. *In vitro* antifungal activity expressed as the minimum inhibitory concentration (MIC) (in μg / ml).

Compound	R^1	R^2	T.m.[a]	T.r.[b]	T.t.[c]	T.s.[d]	E.f.[e]	M.a.[f]	M.c.[g]	A.f.[h]	C.a.[i]	C.s.[j]
5a	H	4-Cl	nt[k]	nt	nt	nt	nt	nt	nt	nt	nt	nt
5b	H	4-CH_3		2.0						20.0	7.0	
5c	4-Cl	H	nt	nt	nt	nt	nt	nt	nt	nt	nt	nt
5d	4-Cl	4-Cl	0.7	0.7	0.7	7.0	0.7	7.0	2.0	7.0	7.0	2.0
5e	4-Cl	4-F	0.7	0.7	0.7	2.0	0.7	7.0	7.0	70.0	7.0	< 0.2
5f	4-Cl	4-CH_3	0.7	0.7	0.7	0.7	<0.2	7.0	7.0	7.0	7.0	< 0.2
				< 2.0[l]						20.0[l]	20.0[l]	
5g	4-Cl	4-NO_2	<0.2	0.7	<0.2	0.7	0.7	7.0	2.0	>70.0	7.0	< 0.2
5h	4-F	4-Cl	0.7	2.0	2.0	2.0	<0.2		0.7	20.0	2.0	2.0
5i	4-F	4-CH_3	0.7	2.0	0.7	0.7	<0.2		2.0	7.0	7.0	7.0
5j	3-OCH_3	4-Cl		< 0.2						20.0	20.0	
5k	3-OCH_3	4-OCH_3		< 2.0						20.0	7.0	
5l	3,4-Cl_2	2-OCH_3	nt	nt	nt	nt	nt	nt	nt	nt	nt	nt
6a	4-Cl	4-CH_3	nt	nt	nt	nt	nt	nt	nt	nt	nt	nt
7				>70.0						>70.0	>70.0	
Ketoconazole			2.0	0.7	0.2	0.7	0.2	7.0	2.0	7.0	20.0	20.0

[a]T.m. = *Trichophyton mentagrophytes* ATCC 9533; [b]T.r. = *Trichophyton rubrum* ATCC 18762; [c]T.t. = *Trichophyton tonsurans* ATCC 9085; [d]T.s. = *Trichophyton schoenleinii* ATCC 22775; [e]E.f. = *Epidermophyton floccosum* ATCC E-18397; [f]*Microsporum audouinii* ATCC 9079; [g]M.c. = *Microsporum canis* ATCC 44459; [h]A.f = *Aspergillus fumigatus* ATCC 28212; [i]C.a. = *Candida albicans* ATCC 10259; [j]C.s. = *Candida stellatoidea* ATCC 36232; [k]nt = not tested; [l]Data for **5f**·HCl salt.



7



8

The substitution was varied at both phenyl rings. As seen from the Table, moderate to potent activity was evident throughout the series against dermatophytes (*Trichophyton* sp. and *Epidermophyton floccosum*) and yeast (*Candida* sp.) fungi. Overall, the best activity attained was against dermatophytes with compounds **5d–g, i** being the most potent. When compared to ketoconazole, all of the analogues tested were either equipotent **(5j)** or had better activity against *Candida* sp. Seemingly, there was no great difference in potency among derivatives with a *para*-substituted halogen on the C-3 phenyl ring, but different substituents on the 5-(phenylthio)methyl group **(5d–i)**. One possible exception was the 3-(4-fluorophenyl)-5-[(4-chlorophenyl)thio]methyl analogue **5h** which was somewhat less active against *Trichophyton* sp.

When compared to the free base, the hydrochloride salt of **5f** suffered a considerable loss of potency against *T. rubrum, A. fumigatus* and *C. albicans*.

Changing the oxidation state of the sulfur in **5j** to the corresponding sulfoxide **(7)** led to a dramatic decrease in antifungal activity.

As previously reported [4], a series of 3-phenyl-5-[(phenoxy)methyl]-3-[(1*H*-imidazol-1-yl)methyl]-2-methylisoxazolidines (**8**; n=1) were prepared and tested *in vitro* for antifungal activity. When compared to compounds **5**, the phenoxymethyl analogues **8** showed, in general, higher potency against dermatophytes (*Trichophyton* sp. and *E. floccosum*) and yeast (*C. albicans*) fungi. However, against *A. fumigatus*, where **8** exerted only marginal activity, their sulfur-containing analogues **5** were significantly more effective *in vitro* agents with compounds **5d, f** and **i** demonstrating potency comparable to that of ketoconazole.

In summary, of all compounds tested, the 3-(4-chlorophenyl) analogues *cis*-**5e–g** showed a potent *in vitro* antifungal activity, especially against *C. stellatoidea*, where their potency was significantly better than that of ketoconazole. Hence, the *cis*-5-{[(substituted phenyl)thio]-methyl}isoxazolidines **5** represent a novel class of antifungal agents.

Experimental protocols

Chemistry

All melting points were determined on a Thomas–Hoover capillary melting point apparatus and are uncorrected. The IR spectra were obtained on a Nicolet MX-1 FT spectrometer as KBr discs. The ^{1}H NMR spectra were taken on a Varian EM-360 A (60 MHz) or a Bruker-IBM 200 Fourier transform (200 MHz) instruments using tetramethylsilane as the internal standard. All spectra were consistent with the assigned structures.

The (2-propenylthio)benzenes **4** were prepared by the method of Hurd and Greengard [9]:

(2-Propenylthio)benzene. (**4**; R^2=H), b.p.=65°C (0.75 Torr) [lit. [9] b.p. 104–106°C (25.0 Torr)].

1-Chloro-4-(2-propenylthio)benzene. (**4**; R^2=4-F), b.p. 90°C (0.15 Torr).

1-Fluoro-4-(2-propenylthio)benzene. (**4**; R^2=4-Cl), b.p. 52–54°C (0.15 Torr).

1-Methyl-4-(2-propenylthio)benzene. (**4**; R^2=4-CH_3), b.p. 50–53°C (0.15 Torr) [lit. [9] b.p. 123–127°C (25 Torr)].

1-Nitro-4-(2-propenylthio)benzene. (**4**; R^2=4-NO_2), b.p. 95°C (0.09 Torr).

1-Methoxy-2-(2-propenylthio)benzene. (**4**; R^2=2-OCH_3), b.p. 77–80°C (0.50 Torr).

*3-(4Chlorophenyl)-3-[1H-imidazol-1-yl)methyl]-2-methyl-5-{[(4-methylphenyl)thio]methyl}isoxazolidine (*cis-**5f** *and* trans-***6a**)*

A solution of 10.44 g (41.8 mmol) of 1-(4-chlorophenyl)-2-(1*H*-imidazol-1-yl)-*N*-methylethanimine *N*-oxide (**3**; R^1=4-Cl) and 10.54 g (64.2 mmol) of 1-methyl-4-(2-propenylthio)benzene (**4**; R^2=4-CH_3) in 250 ml of toluene was refluxed for 40 h under a nitrogen atmosphere. Upon cooling to ambient temperature, the reaction mixture was concentrated to a dark-colored oil which was dissolved in 400 ml of ethyl acetate and extracted with water (3 × 100 ml). The organic extract was dried over anhydrous magnesium sulfate and concentrated under reduced pressure. The residue was flash-chromatographed on neutral silica gel with ethyl acetate to give 11.20 g (65%) of *cis*-**5f**. mp = 98–101°C (ethyl acetate). Anal. Calcd. for $C_{22}H_{24}ClN_3OS$: C: 64.00; H: 5.84; Cl: 8.56; N: 10.15; S: 7.75. Found: C: 63.98; H: 5.98; Cl: 8.68; N: 10.11; S: 7.84. IR (KBr): 2974 (m); 2921 (m); 1505 (s); 1493 (s); 1456 (s); 1399 (m); 1280 (m); 1250 (m); 1234 (m); 1098 (m); 1079 (s); 1013 (m); 940 (m); 847 (m); 815 (s); 750 (m); and 664 (m) cm^{-1}. ^{1}H NMR (200 MHz, $CDCl_3$): 2.29 ppm (dd, 1H, J=5.5, 13.2 Hz, H_{4A}); 2.33 ppm (s, 3H, $ArCH_3$); 2.44 ppm (s, 3H, NCH_3); 2.84 ppm (dd, 1H, J=9.9, 13.2 Hz, H_{4B}); 3.07 (dd, 1H, J=8.3, 13.2 Hz, *H*CHS); 3.33 ppm (dd, 1H, J=5.5, 13.2 Hz, HC*H*S); 3.97 ppm (d, 1H, J=13.8 Hz, *H*CHN); 4.40 ppm (d, 1H, J=13.8 Hz, HC*H*N); 4.58–4.71 ppm (m, 1H, OCH); 6.47 ppm (s, 1H); 6.87–7.36 ppm (m, 10H).

Monohydrochloride salt of **5f**, mp = 200–205°C (2-propanol).

Further elution with ethyl acetate gave 2.20 g (13%) of *trans*-**6a** as a light yellow oil. Oxalate salt of **6a**, mp = 165–167°C (ethanol). Anal. Calcd. for $C_{24}H_{26}ClN_3O_5S$: C: 57.20; H: 5.20; Cl: 7.03; N: 8.34; S: 6.36. Found: C: 57.00; H: 5.30; Cl: 7.30; N: 8.31; S: 6.60. The following derivatives were prepared by similar procedures.

cis-5-{[(4-Chlorophenyl)thio]methyl}-3-[(1H-imidazol-1-yl)methyl]-2-methyl-3-phenylisoxazolidine ***5a***

Yield: 48%, mp = 132.5–133°C (ethyl acetate). Anal. Calcd. for $C_{21}H_{22}ClN_3OS$: C: 63.07; H: 5.54; Cl: 8.86; N: 10.51; S: 8.02. Found: C: 62.93; H: 5.55; Cl: 9.32; N: 10.45; S: 8.50. IR (KBr): 2972 (m); 1504 (m); 1478 (s); 1455 (m); 1251 (m); 1237 (m); 1093 (s); 1077 (m); 1009 (m); 817 (s); 735 (m); 699 (m); and 663 (m) cm^{-1}. ^{1}H NMR (200 MHz, $CDCl_3$): 2.28 ppm (dd, 1H, J=5.0, 13.8 Hz, H_{4A}); 2.47 ppm (s, 3H, NCH_3); 2.89 ppm (dd, 1H, J=9.4, 13.8 Hz, H_{4B}); 3.12 ppm (dd, 1H, J=7.2, 13.8 Hz, *H*CHS); 3.33 ppm (dd, 1H, J=5.5, 13.8 Hz, HC*H*S); 4.07 ppm (d, 1H, J=13.8 Hz, *H*CHN); 4.41 ppm (d, 1H, J=13.8 Hz, HC*H*N); 4.60–4.78 ppm (m, 1H, OCH); 6.42 ppm (s, 1H); 6.87 ppm (s, 1H); 6.97–7.04 ppm (m, 3H); 7.25–7.39 ppm (m, 7H).

cis-3-[(1H-Imidazol-1-yl)methyl-2-methyl-5-{[(4-methylphenyl)thio]methyl}-3-phenylisoxazolidine **5b**
mp = 116°C (ethyl acetate). Anal. Calcd. for $C_{22}H_{25}N_3OS$: C: 69.63; H: 6.64; N: 11.07; S: 8.45. Found: C: 69.53; H: 6.69; N: 11.01; S: 8.54. IR (KBr): 2970 (m); 1650 (m); 1630 (m); 1505 (s); 1494 (s); 1456 (s); 1399 (m); 1279 (m); 1251 (m); 1234 (m); 1077 (m); 1035 (m); 1013 (m); 938 (m); 816 (s); 744 (m); 696 (s); and 664 (m) cm^{-1}. ^1H NMR (200 MHz, $CDCl_3$): 2.29 ppm (dd, 1H, J=5.5, 13.2 Hz, H_{4A}); 2.34 ppm (s, 3H, $ArCH_3$); 2.46 ppm (s, 3H, NCH_3); 2.88 ppm (dd, 1H, J=9.9, 13.2 Hz, H_{4B}); 3.09 ppm (dd, 1H, J=7.7, 13.2 Hz, *H*CHS); 3.34 ppm (dd, 1H, J=5.5, 13,2 Hz, HC*H*S); 4.05 ppm (d, 1H, J=14.3 Hz, *H*CHN); 4.41 ppm (d, 1H, J=14.3 Hz, HC*H*N); 4.61–4.75 ppm (m, 1H, OCH); 6.41 ppm (s, 1H); 6.85–7.38 ppm (m, 11H).

cis-3-(4-Chlorophenyl)-3-[(1H-imidazol-1-yl)methyl]-2-methyl-5-{[(phenyl)thio]-methyl}isoxazolidine **5c**
mp = 81–88°C (ethyl acetate). Anal. Calcd. for $C_{21}H_{22}ClN_3OS$: C: 63.07; H: 5.54; Cl: 8.86; N: 10.51. Found: C: 62.98; H: 5.62; Cl: 9.15; N: 10.44; IR (KBr): 2970 (m); 1582 (m); 1511 (m); 1506 (m); 1494 (s); 1440 (m); 1239 (m); 1095 (s); 1080 (s); 1022 (m); 1016 (m); 835 (m); 819 (m); 750 (m); 738 (s); 690 (m); and 664 (m) cm^{-1}. ^1H NMR (200 MHz, $CDCl_3$): 2.50 ppm (s, 3H, NCH_3); 2.50 ppm (dd, 1H, J=4.4, 13.2 Hz, H_{4A}); 2.59 ppm (dd, 1H, J=7.7, 13.2 Hz, H_{4B}); 2.98 ppm (dd, 1H, J=8.8, 13.8 Hz, *H*CHS); 3.45 ppm (dd, 1H, J=5.0, 13.8 Hz, HC*H*S); 4.07 ppm (d, 1H, J=14.9 Hz, *H*CHN); 4.26 ppm (d, 1H, J=14.9 Hz, HC*H*N); 4.43 ppm (ddd, 1H, J=4.4, 5.0, 7.7, 8.8 Hz, OCH); 6.38 ppm (s, 1H); 6.90–7.45 ppm (m, 11H).

cis-3-(4-Chlorophenyl)-5-{[(4-chlorophenyl)thio]methyl}-3-[(1H-imidazol-1-yl)methyl]-2-methylisoxazolidine **5d**
mp = 118–119°C (ethyl acetate). Anal. Calcd. for $C_{21}H_{21}Cl_2N_3OS$: C: 58.07; H: 4.87; Cl: 16.32; N: 9.67; S: 7.38. Found: C: 58.12; H: 4.95; Cl: 16.29; N: 9.67; S: 7.64. IR (KBr): 2975 (m); 1505 (m); 1492 (m); 1478 (s); 1456 (m); 1407 (m); 1238 (m); 1093 (s); 1078 (m); 1008 (m); 852 (m); 817 (s); 753 (m); and 664 (m) cm^{-1}. ^1H NMR (200 MHz, $CDCl_3$): 2.29 ppm (dd, 1H, J=5.0, 13.2 Hz, H_{4A}); 2.44 ppm (s, 3H, NCH_3); 2.85 ppm (dd, 1H, J=9.9, 13.2 Hz, H_{4B}); 3.10 ppm (dd, 1H, J=7.2, 13.8 Hz, *H*CHS); 3.32 ppm (dd, 1H, J=5.5, 13.8 Hz, HC*H*S); 4.00 ppm (d, 1H, J=13.8 Hz, *H*CHN); 4.40 (d, 1H, J=13.8 Hz, HC*H*N); 4.58–4.72 ppm (m, 1H, OCH); 6.49 ppm (s, 1H); 6.89–7.39 ppm (m, 10H).

cis-3-(4-Chlorophenyl)-5-{[(4-fluorophenyl)thio]methyl}-3-[(1H-imidazol-1-yl)methyl]-2-methylisoxazolidine **5e**
mp = 106–107°C (ethyl acetate–hexane, 1:1). Anal. Calcd. for $C_{21}H_{21}ClFN_3OS$: C: 60.35; H: 5.06; Cl: 8.48; F: 4.55; N: 10.05; S: 7.67. Found: C: 60.08; H: 4.84; Cl: 8.57; F: 4.61; N: 9.99; S: 8.02. IR (KBr): 1583 (m); 1503 (m); 1491 (s); 1455 (m); 1442 (m); 1398 (m); 1230 (s); 1095 (m); 1081 (m); 1014 (m); 822 (s); 739 (m); 660 (m); and 635 (m) cm^{-1}. ^1H NMR (200 MHz, $CDCl_3$): 2.26 ppm (dd, 1H, J=5.5, 13.8 Hz, H_{4A}); 2.44 ppm (s, 3H, NCH_3); 2.84 ppm (dd, 1H, J=9.9, 13.8 Hz, H_{4B}); 3.07 ppm (dd, 1H, J=7.2, 13.2 Hz, *H*CHS); 3.29 ppm (dd, 1H, J=5.0, 13.2 Hz, HC*H*S); 3.98 ppm (d, 1H, J=13.8 Hz, *H*CHN); 4.39 ppm (d, 1H, J=13.8 Hz, HC*H*N); 4.55–4.69 ppm (m, 1H, OCH); 6.48 ppm (s, 1H); 6.89–7.47 ppm (m, 10H).

cis-3-(4-Chlorophenyl)-3-[(1H-imidazol-1-yl)methyl]-2-methyl-5-{[(4-nitrophenyl)thio]methyl}isoxazolidine **5g**
mp =179–180°C (ethanol). Anal. Calcd. for $C_{21}H_{21}ClN_4O_3S$: C: 56.69; H: 4.76; Cl: 7.97; N: 12.59; S: 7.21. Found: C: 56.47; H. 4.90; Cl: 8.21; N: 12.39; S: 7.46. IR (KBr): 1593 (m); 1580 (m); 1509 (s); 1335 (s); 1228 (m); 1097 (m); 1077 (m); 1005 (m); 854 (m); 833 (m); and 741 (m) cm^{-1}. ^1H NMR (200 MHz, $CDCl_3$): 2.32 ppm (dd, 1H, J=5.5, 13.2 Hz, H_{4A}); 2.46 ppm (s, 3H, NCH_3); 2.91 ppm (dd, 1H, J=9.4, 13.2 Hz, H_{4B}); 3.22 ppm (dd, 1H, J=6.6, 13.8 Hz, *H*CHS); 3.41 ppm (dd, 1H, J=5.0, 13.8 Hz, HC*H*S); 4.05 (d, 1H, J=14.3 Hz, *H*CHN); 4.42 ppm (d, 1H, J=14.3 Hz, HC*H*N); 4.64–4.78 ppm (m, 1H, OCH); 6.54 ppm (s, 1H); 6.92–7.45 ppm (m, 8H); 8.15 ppm (d, 2H, J=8.3 Hz).

cis-5-{[(4-Chlorophenyl)thio]methyl}-3-(4-fluorophenyl)-3-[(1H-imidazol-1-yl)methyl]-2-methylisoxazolidine **5h**
Yield: 43.6%. mp = 120.5°C (ethyl acetate). Anal. Calcd. for $C_{21}H_{21}ClFN_3OS$: C: 60.35 ; H: 5.06; Cl: 8.48; F: 4.55; N: 10.05; S: 7.67. Found: C: 60.30; H: 5.12; Cl: 8.48; F: 4.43; N: 10.04; S: 7.88. IR (KBr): 1648 (m); 1626 (m); 1600 (m); 1506 (s); 1477 (s); 1457 (m); 1235 (m); 1093 (m); 1079 (m); 1006 (m); 850 (m); 819 (s); 748 (m); and 664 (m) cm^{-1}. ^1H NMR (200 MHz, $CDCl_3$): 2.29 ppm (dd, 1H, J=5.5, 13.2 Hz, H_{4A}); 2.44 (s, 3H, NCH_3); 2.87 ppm (dd, 1H, J=9.9, 13.2 Hz, H_{4B}); 3.11 ppm (dd, 1H, J=7.2, 13.2 Hz, *H*CHS); 3.33 ppm (dd, 1H, J=5.5, 13.2 Hz, HC*H*S); 4.00 ppm (d, 1H, J=14.3 Hz, *H*CHN); 4.40 ppm (d, 1H, J=14.3 Hz, HC*H*N); 4.59–4.73 ppm (m, 1H, OCH); 6.47 ppm (s, 1H); 6.89 ppm (s, 1H); 6.96–7.39 ppm (m, 9H).

cis-3-(4-Fluorophenyl)-3-[(1H-imidazol-1-yl)methyl]-2-methyl-5-{[(4-methylphenyl)thio]methyl}isoxazolidine **5i**
mp = 109.5–110°C (ethyl acetate). Anal. Calcd. for $C_{22}H_{24}FN_3OS$: C: 66.47; H: 6.09; F: 4.78; N: 10.57; S: 8.07. Found: C: 66.39; H: 6.10; F: 5.02; N: 10.69; S: 8.24. IR (KBr): 2865 (m); 1605 (m); 1505 (s); 1437 (m); 1226 (s); 1105 (m); 1076 (m); 1050 (m); 1012 (w); 852 (m); 828 (m); 818 (m); 800 (m); 734 (m); and 661 (m) cm^{-1}. ^1H NMR (200 MHz, $CDCl_3$): 2.30 ppm (dd, 1H, J=5.5, 13.8 Hz, H_{4A}); 2.34 ppm (s, 3H, $ArCH_3$); 2.44 ppm (s, 3H, NCH_3); 2.85 ppm (dd, 1H, J=9.4, 13.8 Hz, H_{4B}); 3.08 ppm (dd, 1H, J=8.3, 13.2 Hz, *H*CHS); 3.34 ppm (dd, 1H, J=5.5, 13.2 Hz, HC*H*S); 3.98 ppm (d, 1H, J=13.8 Hz, *H*CHN); 4.40 ppm (d, 1H, J=13.8 Hz, HC*H*N); 4.59–4.73 ppm (m, 1H, OCH); 6.42 ppm (s, 1H); 6.88 ppm (s, 1H); 6.91–7.37 ppm (m, 9H).

cis-5-{[(4-Chlorophenyl)thio]methyl}-3-[(1H-imidazol-1-yl)methyl]-3-(3-methoxyphenyl)-2-methylisoxazolidine **5j**
mp = 113–114°C (ethyl acetate). Anal. Calcd. for $C_{22}H_{24}ClN_3O_2S$: C: 61.46; H: 5.63; Cl: 8.25; N: 9.77; S: 7.46. Found: C: 61.35; H: 5.65; Cl: 8.59; N: 9.71; S: 7.57. IR (KBr): 2972 (m); 1610 (m); 1584 (m); 1509 (m); 1479 (s); 1452 (m); 1435 (m); 1297 (m); 1234 (m); 1222 (m); 1093 (m); 1074 (m); 1045 (m); 1025 (m); 1008 (m); 945 (m); 821 (s); 816 (m); 785 (m); 748 (m); 705 (m); and 665 (m) cm^{-1}. ^1H NMR (200 MHz, $CDCl_3$): 2.26 ppm (dd, 1H, J=5.5, 13.2 Hz, H_{4A}); 2.48 ppm (s, 3H, NCH_3); 2.89 ppm (dd, 1H, J=9.9, 13.2 Hz, H_{4B}); 3.12 ppm (dd, 1H, J=8.3, 13.2 Hz, *H*CHS); 3.34 ppm (dd, 1H, J=5.5, 13.2 Hz, HC*H*S); 3.72 ppm (s, 3H,OCH_3); 4.01 ppm (d, 1H, J=13.8 Hz, *H*CHN); 4.42 ppm (d, 1H, J=13.8 Hz, HC*H*N); 4.61–4.74 ppm (m, 1H, OCH); 6.48–7.39 ppm (m, 11H).

cis-3-[(1H-Imidazol-1-yl)methyl]-3-(3-methoxyphenyl)-2-methyl-5-{[(4-methylphenyl)thio]methyl}isoxazolidine **5k**
mp = 107–108°C (ethyl acetate). Anal. Calcd. for $C_{23}H_{27}N_3O_2S$: C: 67.45; H: 6.65; N: 10.26; S: 7.83. Found: C: 67.37; H: 6.59; N: 10.22; S: 8.02. IR (KBr): 2977 (m); 2928 (m); 1610 (m); 1584 (s); 1508 (m); 1496 (s); 1456 (s); 1435 (s); 1298 (m); 1234 (s); 1221 (s); 1074 (m); 1045 (m); 1025 (m); 812 (s); 780 (m); 704 (m); and 664 (m) cm^{-1}. ^1H NMR (200 MHz, $CDCl_3$): 2.26 ppm (dd, 1H, J=5.5, 13.8 Hz, H_{4A}); 2.33 ppm (s, 3H, $ArCH_3$); 2.47 ppm (s, 3H, NCH_3); 2.86 ppm (dd, 1H, J=9.4, 13.8 Hz, H_{4B}); 3.08 ppm (dd, 1H, J=7.7, 13.2 Hz, *H*CHS); 3.33 ppm (dd, 1H, J=5.5, 13,2 Hz, HC*H*S); 3.70 ppm (s, 3H, OCH_3); 4.00 ppm (d, 1H, J=14.3 Hz, *H*CHN); 4.41 ppm (d, 1H, J = 14.3 Hz, HC*H*N); 4.60–4.74 ppm (m, 1H, OCH); 6.46–7.37 ppm (m, 11H).

cis-3-(3,4-Dichlorophenyl)-3-[(1H-imidazol-1-yl)methyl]-5-{[(2-methoxyphenyl)thio]methyl}-2-methylisoxazolidine **5l**
mp = 55–65°C (dec) (ethyl acetate). Anal. Calcd. for $C_{22}H_{23}Cl_2N_3O_2S$: C: 56.90; H: 4.99; Cl: 15.27; N: 9.05; S: 6.90. Found: C: 56.53; H: 5.20; Cl: 5.10; N: 8.89; S: 7.08. IR (KBr): 2966 (m); 2920 (m); 1578 (m); 1505 (m); 1478 (s); 1451 (m); 1380 (m); 1271 (m); 1243 (s); 1072 (m); 1043 (m); 1027 (s); 903 (w); 821 (w); 747 (m); and 661 (m) cm^{-1}. ^1H NMR (200 MHz, $CDCl_3$): 2.32 ppm (dd, 1H, J=5.5, 13.2 Hz, H_{4A}); 2.46 ppm (s, 3H, NCH_3); 2.81 ppm (dd, 1H, J=9.9, 13.2 Hz, H_{4B}); 3.05 ppm (dd, 1H, J=7.7, 12.7 Hz, *H*CHS); 3.32 ppm (dd, 1H, J=5.0, 12.7 Hz, HC*H*S); 3.92 ppm (s, 3H, OCH_3); 3.95 ppm (d, 1H, J= 14.3 Hz, *H*CHN); 4.39 ppm (d, 1H, J=14.3 Hz, HC*H*N); 4.56–4.70 ppm (m, 1H, OCH); 6.51 ppm (s, 1H); 6.68–7.41 ppm (m, 9H).

cis-5-{[(4-Chlorophenyl)sulfoxy]methyl}-3-[(1H-imidazol-1-yl)methyl]-3-(3-methoxyphenyl)-2-methylisoxazolidine **7**
Under a nitrogen atmosphere, a solution of 2.0 g (4.65 mmol) of **5j** in 100 ml of methylene dichloride was cooled to −78°C using a dry ice–acetone bath. To this solution was added dropwise over a period of 30 min a solution of 1.24 g (5.75 mmol) of 85% *meta*-chloroperbenzoic acid in 70 ml of methylene dichloride. The resulting solution was warmed gradually to ambient temperature, then washed repeatedly with saturated aqueous sodium bicarbonate (3 × 100 ml) and dried over anhydrous magnesium sulfate. The solvent was removed under reduced pressure and the crude product was flash-chromatographed on neutral silica

gel using 98:2 mixture of chloroform and methanol as the eluent. Yield: 0.43 g (21%). mp = 120–121°C (ethyl acetate). Anal. Calcd. for $C_{22}H_{24}ClN_3O_3S$: C: 59.25; H: 5.42; N: 9.42; S: 7.19. Found: C: 58.96; H: 5.52; N: 9.32; S: 7.41.

Pharmacology

In vitro *assay for antifungal activity*
The antifungal activity was assayed *in vitro* in solid agar tests performed in 24-well tissue culture plates. The test medium was prepared by diluting the test compound 10-fold into "Antibiotic Medium 3" + 2% agar. The testing was accomplished either by using a 4-point (70, 20, 2 and 0.2 μg/ml) or a 6-point (70, 20, 7, 2, 0.7 and 0.2 μg/ml) dilution scheme with ketoconazole being used as a positive control drug in all assays. All test organisms were grown on potato flake agar at 26°C. *Candida albicans* was grown overnight, *Aspergillus fumigatus* was grown for *ca.* 1 week, and *Trichophyton rubrum* was grown *ca.* 2 weeks. The cells were either removed from the plates with a sterile cotton swab and suspended in sterile water (*C. albicans, A. fumigatus*) or washed from the surface of the plate with sterile water and diluted in sterile water (*T. rubrum*). The actual counts were performed using a hemacytometer and the suspensions were diluted to 1×10^4 cells/ml. The test and control plates were inoculated with 0.05 ml of the fungal suspension and were incubated at 26°C until a visible growth in the compound-free control plates was evident. The minimum inhibitory concentration (MIC) values were interpreted as the lowest concentration (in μg/ml) at which no visible growth occurred.

References

1 Georgiev V.St. (ed.) (1988) *Antifungal Drugs. Ann. NY Acad. Sci.*, New York
2 Mullen G.B. & Georgiev V.St. (1986) *Heterocycles* 24, 3441
3 Mullen G.B., Swift P.A. & Georgiev V.S. (1987) *J. Pharm. Sci.* 76, 930
4 Mullen G.B., Swift P.A., Maryniak D.M., Allen S.D., Mitchell J.T., Kinsolving C.R. & Georgiev V.St. (1988) *Helv. Chim. Acta* 71, 718
5 Mullen G.B., DeCory T.R., Mitchell J.T., Allen S.D., Kinsolving C.R. & Georgiev V.St. (1988) *J. Med. Chem.* 31, 2008
6 Dormer P.A., Mullen G.B., Mitchell J.T., Allen S.D. & Georgiev V.St. (1988) *Chemotherapy* 34, 411
7 Bennett G.A., Swift P.A., Mullen G.B., Mitchell J.T., Allen S.D., Jones W.E., Kinsolving C.R. & Georgiev V.St. (1988) *Helv. Chim. Acta* 71, 1622
8 Tufariello J.J. (1984) *in: 1,3-Dipolar Cycloaddition Chemistry* (A. Padwa, ed.), Wiley & Sons, New York, pp. 83–168[ii]
9 Hurd C.D. & Greengard H. (1930) *J. Am. Chem. Soc.* 52, 3556
10 Godefroi E.F., Heeres J., Van Cutsem J. & Janssen P.A.J. (1969) *J. Med. Chem.* 12, 784[i]